Vitamin D and Autoimmunity

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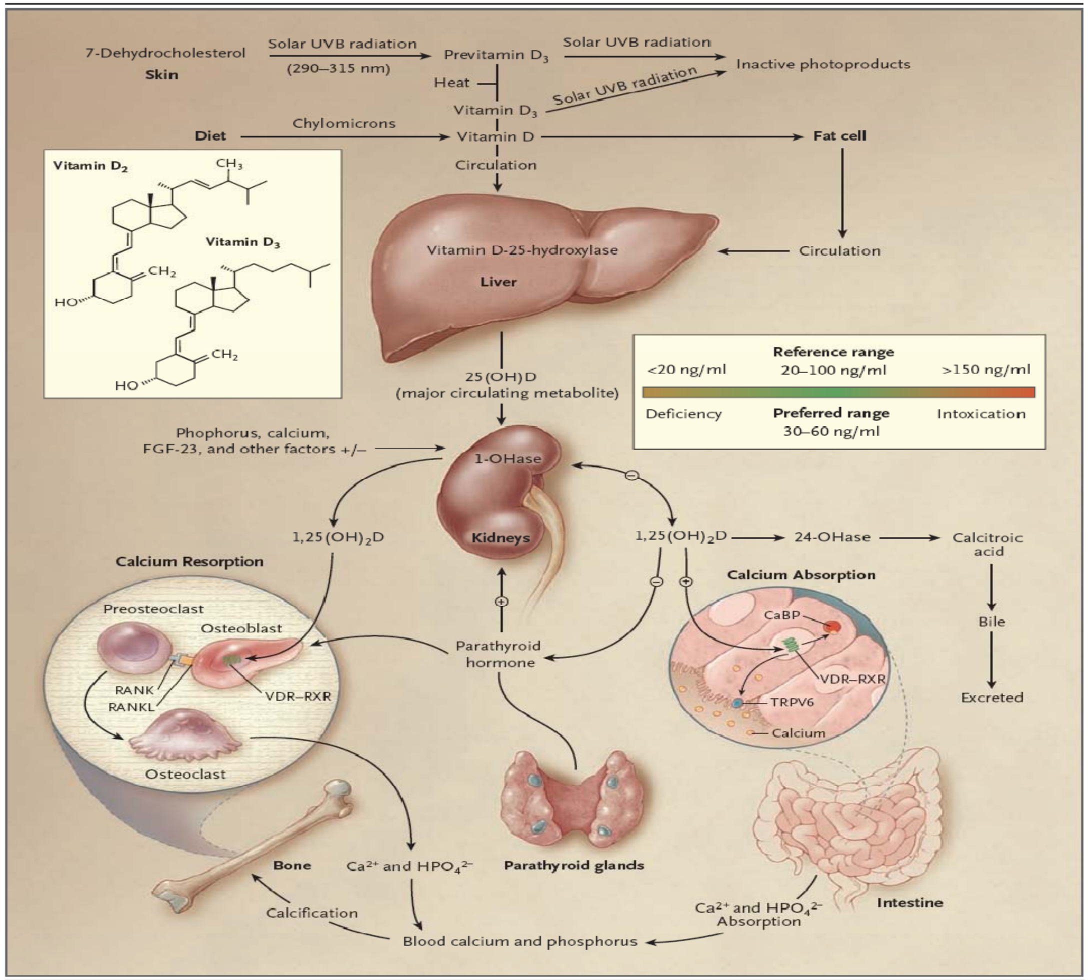
Overview

- Non-classical vitamin D actions recognised >30 years ago
- Immune system has potential to synthesis 1,25D and elicit autocrine and paracrine effects
- Mechanisms involved need elucidation
- Effects of vitamin D on immune function and secondary infection will be explored
- Association between vitamin D and immune disease plus vitamin D treatment effects will be explored
- Is vitamin D a modifiable environmental risk factor for autoimmune disease?

Vitamin D metabolism

- 2 critical sites of activation
- Importance of cellular metabolism not emphasised



The immune system

- Humans have inherited an innate immune system from invertebrates
- Innate IS consists of macrophages, dendritic cells and Natural killer lymphocytes
- Cells recognise specific pathogen-associated molecular patterns highly conserved among microbes
- Adaptive immune system only found in vertebrates
- Requires Ag receptor generation on T and B lymphocytes that provide specificity and maintain tolerance (non-reactivity) to self
- Adaptive IS also provides memory
- T cells provide cellular immunity and B cells humoral immunity

Vitamin D effects on immune system

Vitamin D initiates effects on both Innate and Adaptive immunity

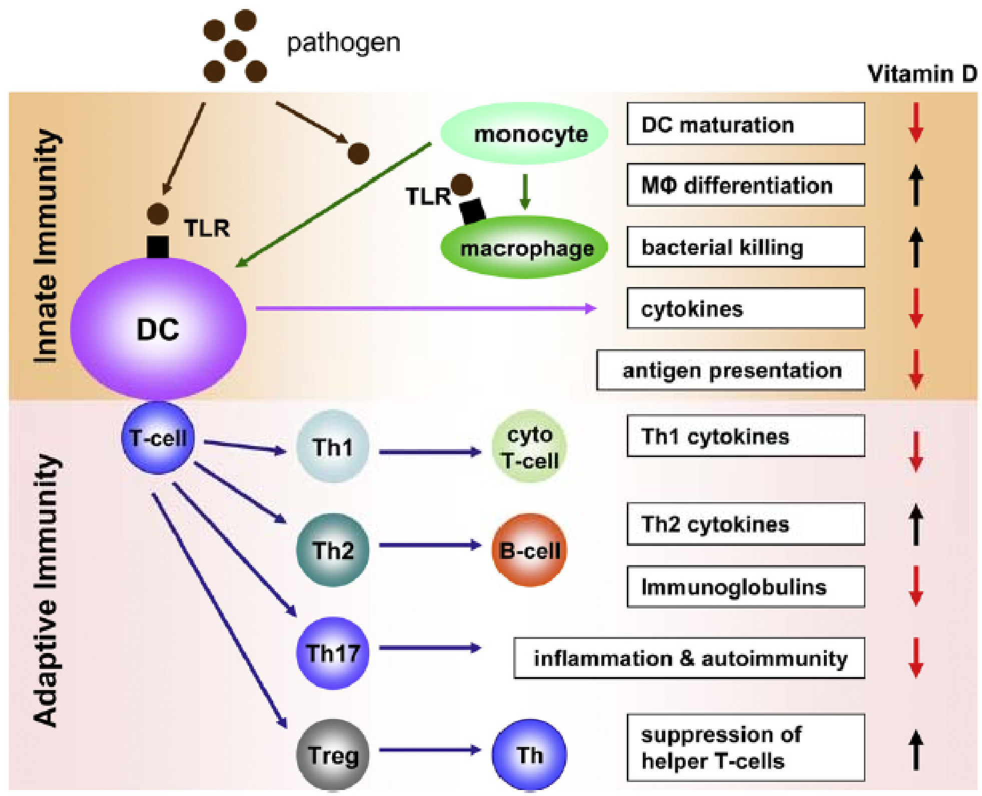


Fig. 1. Effects of vitamin D on innate and adaptive immunity. The principal innate and adaptive immune responses to a pathogenic challenge, and the positive or negative regulation of these responses by vitamin D. B-cell, B lymphocyte; cyto T-cell, cytotoxic T cell; DC, dendritic cell; MΦ, macrophage; T-cell, T lymphocyte; TLR, toll-like receptor; Treg, regulatory T cell.

Hewison, Endo Clin NA, 2010

Innate Immunity: Macrophages and vitamin D

- Vitamin D stimulates monocytes to produce more mature phagocytic macrophages
- Macrophages produce 1,25D when stimulated with IFN gamma
- Macrophages express VDR
- Antibacterial protein cathelicidin is regulated by vitamin D and promotes monocytic killing of TB
- Thus vitamin D activates the cathelicidin gene to produce antimicrobial peptides which kill TB baccilli

TB and autoimmune disease

- Autoimmune phenomenon is seen in patients with active TB. Eg. A non-infective monopolyarthritis
- Poncet's disease is a non-infective polyarthritis with periarticular osteoporosis, erosions and loss of joint space (similar to RhA)
- TB and SLE share similar features: fever, myalgia, arthritis

Innate Immunity: Dendritic cells

- Innate immunity also involves APC's
- Dendritic cells (DC) express VDR
- Vitamin D inhibits maturation of monocyte derived dendritic cells (thus reducing APC activity)
- Reciprocal VDR and 1AH expression as DC mature
- Mature DC relatively insensitive to 1,25D so allowing initial T cell response but acts on immature DC to prevent continued maturation

Adaptive Immunity: Vitamin D and T cells

- Resting T cells express low amounts of VDR
- Ag stimulated T cells express higher amounts of VDR
- 1,25D inhibits T helper 1 cells but promotes TH2 cells
- 1,25D inhibits TH17 expression (and thus IL17 levels)
- CD8 suppressor T cells express VDR

Adaptive Immunity: vitamin D and B cells

Vitamin D directly inhibits:

- B lymphocyte proliferation
- Generation of memory B cells
- Plasma cell differentiation
- Immunoglobulin production

Vitamin D deficiency and autoimmune disease

Autoimmune condition	Prevalence of vitamin D deficiency	Reference
Inflammatory Bowel disease	34%	
Multiple Sclerosis	23%	
Rheumatoid arthritis	64%	
Type 1 Diabetes	36%	
Systemic Lupus Erythematosis	67%	

Autoimmune diseases are the 3rd commonest cause of morbidity and Mortality in industrialised countries after cancer and heart disease 1.

Harel et al, Ann NY Acad Sci 2006

Vitamin D deficiency and observed rates of Type 1 DM in Finnish infants

- 12,055 Northern Finish women giving birth enrolled in 1966
- 2000IU daily cholecalciferol recommended supplement
- 91% followed up at age 1 years with 81 cases of T1M

	Type 1 diabetes	Time at risk (years)	Incidence per 100 000 years at risk	RR (95% CI)	Adjusted RR (95% CI)*
Use of vitamin D supplements					
None	2	981	204	1 (reference)	1 (reference)
Irregularly	12	36 143	33	0·16 (0·04–0·72)	0·16 (0·04–0·74)
Regularly	67	276 235	24	0·12 (0·03–0·47)	0·12 (0·03–0·51)
Dose of vitamin D†					
Low	2	2 093	96	1 (reference)	1 (reference)
Recommended	63	259 779	24	0·20 (0·05–0·84)	0·22 (0·05–0·89)
High	2	13 245	15	0·14 (0·02–0·97)	0·14 (0·02–1·01)
Suspected rickets‡					
No	77	306 945	25	1 (reference)	1 (reference)
Yes	4	6 414	62	2·6 (1·0–7·2)	3·0 (1·0–9·0)

*Adjusted for sex, neonatal (parity, gestational and maternal age), length of maternal education, social status, and standardised birth weight, and growth rate in infancy (suspected rickets adjusted in addition to the increased dose of vitamin D); †In children recieving vitamin D supplementation regularly.

Table 2: **Incidence rate and rate ratio (RR) of type 1 diabetes by the use of vitamin D supplements and suspected rickets in infancy**

Hypponen et al, The Lancet 2001

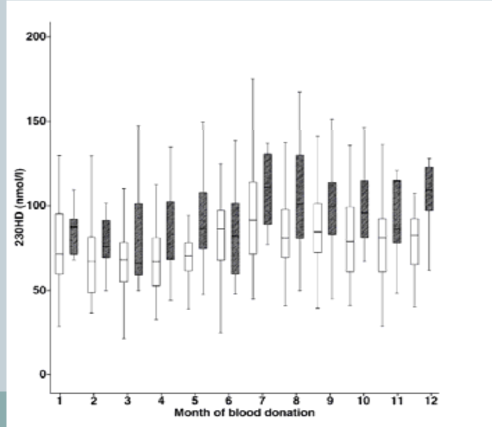
Vitamin D deficiency and observed rates of Type 1 DM in Swedish children/young adults

- Collected samples at diagnosis of 459 patients in 1987/8
- Compared with 208 age, sex and residence matched subjects
- Effect unaffected by location or month of year

Table 2 Clinical characteristics and plasma 25OHD at diagnosis in young adults with type 1 diabetes vs control subjects (mean±SEM)

Characteristic	Patients (n=459)	Control subjects (n=208)	Test of difference between patients and controls (p value)
Males/females	1.6	1.1	0.016
Age (years)	24.4±0.3	25.6±0.5	0.027
25OHD (nmol/l)			
All	82.5±1.3	96.7±2.0	<0.0001
Males	77.9±1.4[a]	93.9±2.7[b]	<0.0001
Females	90.1±2.4	99.7±2.9	0.014

Test of difference by sex: [a] $p<0.0001$; [b] $p=0.15$



VDR polymorphisms and type 1 DM

- 203 Japanese T1M and 220 controls examined
- Increased frequency of B Allele in T1M independent of HLA type
- VDR genotype may be relevant to vitamin D supplementation

TABLE 1. Distribution of VDR gene polymorphism in patients with type 1 diabetes overall and controls

	Patients (n = 203)	Control (n = 222)	*P*
Genotype frequencies			
BB	12 (5.9%)	1 (0.5%)	
Bb	64 (31.5%)	49 (22.0%)	0.0010[a]
bb	127 (62.6%)	172 (77.5%)	
Allele frequencies			
B allele	0.217	0.115	<0.0001
b allele	0.783	0.885	

[a] Statistical analysis was performed between BB + Bb and bb.

TABLE 4. VDR gene polymorphism in subgroups of patients with type 1 diabetes

	Group			Control
	A	B	C	
Number	51	91	61	222
Onset	Acute	Acute	Slow	
Autoantibody	Negative	Positive	Positive	
Mean onset age (yr)	27.8	31.3	44.8	
Allele frequencies				
B allele	0.255	0.236	0.156	0.115
b allele	0.745	0.764	0.844	0.885
P value (*vs.* control)	0.0007	0.0002	0.2182	

Motohashi et al, JCEM 2003

Vitamin D deficiency and Rheumatoid Arthritis

- Prospective cohort study of older women
- 152 cases of RhA in cohort of 29,000 women over 11 years
- Greater intake of vit D associated with lower risk of RhA

Table 2. Relative risks (RRs) and 95% confidence intervals (95% CIs) for risk of rheumatoid arthritis according to intake of vitamin D from food and supplements, Iowa Women's Health Study, 1986–1997

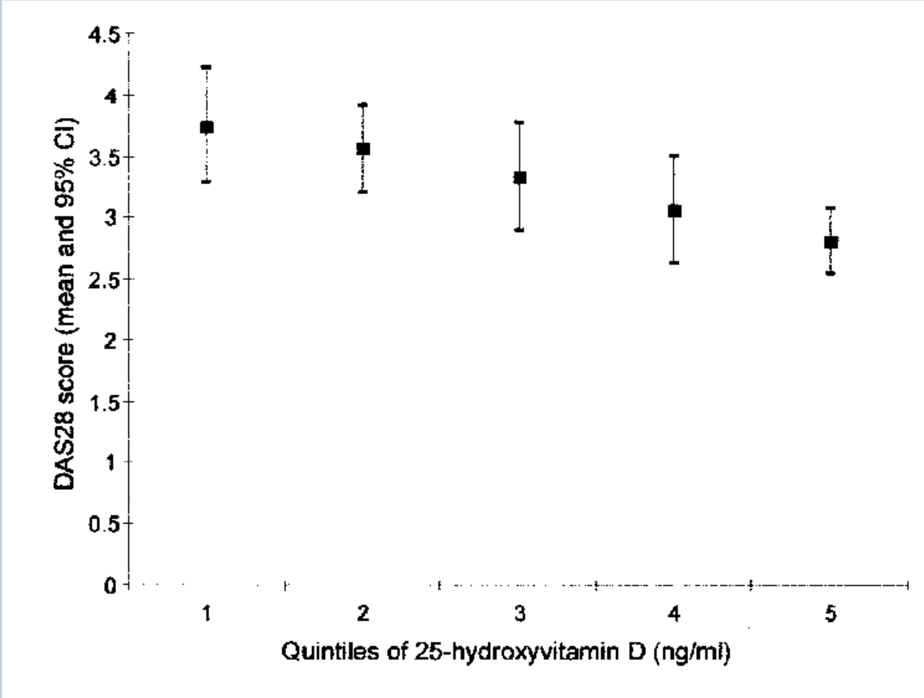
Vitamin D measure, IU/day*	Cases	Person-years	Age-adjusted RR (95% CI)	Multivariable-adjusted RR (95% CI)†
Total				
<221.4	64	103,613	1.00 (referent)	1.00 (referent)
221.4–467.6	42	103,741	0.64 (0.43–0.97)	0.67 (0.45–1.01)
≥467.7	46	106,827	0.67 (0.45–1.00)	0.67 (0.44–1.00)
P for trend			0.05	0.05
Dietary				
<169	59	103,586	1.00 (referent)	1.00 (referent)
169–289.9	50	103,750	0.81 (0.55–1.21)	0.87 (0.58–1.29)
≥290	43	106,845	0.68 (0.44–1.06)	0.72 (0.46–1.14)
P for trend			0.09	0.16
Supplemental				
Nonusers	109	200,008	1.00 (referent)	1.00 (referent)
<400	13	37,423	0.64 (0.36–1.14)	0.65 (0.36–1.15)
≥400	30	76,750	0.69 (0.46–1.04)	0.66 (0.43–1.00)
P for trend			0.05	0.03

* Tertiles of standard supplemental dose.
† Adjusted for age, caloric intake, smoking status, hormone replacement therapy, decaffeinated coffee consumption, and β-cryptoxanthin intake.

Merlino et al, Arthritis & Rheum 2004

Vitamin D deficiency and inflammatory arthritis

- 206 consecutive patients with RhA studied within 6/12 of disease onset
- Not on steroids
- Disease activity score associated with 25OHD
- Every 25nmol/L increment in 25OHD associated with reduction of DAS by 0.3 and CRP by 25%



Patel et al, Arthritis & Rheum 2007

Vitamin D deficiency and SLE

- 123 recently diagnosed SLE in Carolina, USA
- c/w 240 age and sex matched
- Caucasians sig lower 25OHD with SLE (p= 0.04)
- Expected seasonal and racial differences in 25OHD also seen

Table 1
Severe vitamin D deficiency, defined as 25-hydroxyvitamin D below 10 ng/ml, associated with SLE classification criteria present at time of study enrollment

	Adjusted OR	95% CI	*p*-value
Discoid rash	1.4	0.2–8.3	0.72
Photosensitivity	12.9	2.2–75.5	<0.01*
Arthritis	0.3	0.1–1.4	0.12
Renal disease	13.3	2.3–76.7	<0.01*

All values are adjusted for race, season, age, and smoking. (OR, odds ratio; CI, confidence interval).

Kamen et al, Autoimmunity rev 2006

Vitamin D deficiency and SLE

- Cross-sectional study of 92 SLE patients (90% women, 98% caucasian)
- 69 (75%) patients 25OHD < 75 nmol/L (photosensitivity predicted)
- 14 (15%) patients 25OHD < 25 nmol/L (photoprotection predicted)
- Fatigue was more common if 25OHD <25nmol/L
- Vitamin D had no relation with SLE disease severity

TABLE 2. Clinical and laboratory variables by vitamin D status

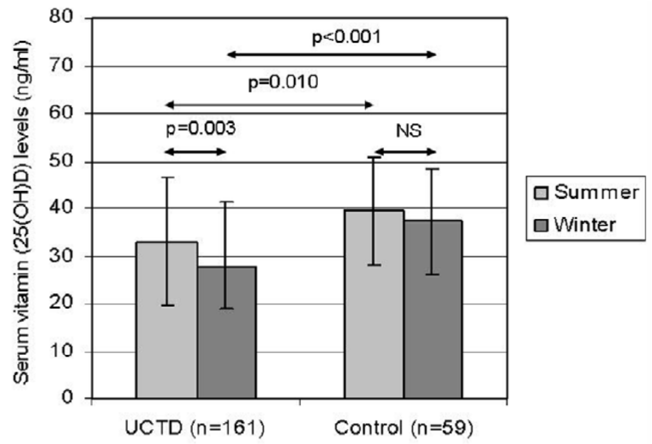
	25(OH)D > 30 (n = 23)	25(OH)D < 30 (n = 69)	25(OH)D < 10 (n = 14)
Age, mean (S.D.) (yrs)	40 (14)	41.4 (14)	36.6 (9.5)
Female sex, n/N(%)	23/23 (100)	60/69 (87)	13/14 (93)
Disease duration, median (range) (yrs)	6 (1–26)	8 (0–30)	6 (1–21)
Photosensitivity, n/N(%)	13/23 (56)	50/69 (72)	12/14 (85)
Photoprotection, n/N(%)	15/23 (65)	52/69 (75)	13/14 (93)
Anti-Ro antibodies, n/N(%)	7/23 (30)	29/69 (42)	7/14 (50)
Current smoker, n/N(%)	5/23 (22)	17/69 (25)	3/14 (21)
Current prednisolone, n/N(%)	14/23 (61)	34/69 (49)	8/14 (57)
Prednisolone dose, median (range) (mg/day)	5 (2.5–30)	5 (2.5–10)	5 (2.5–10)
Current HCQ, n/N(%)	19/23 (83)	54/69 (78)	10/14 (71)
Current calcium + vitamin D, n/N(%)	14/23 (61)	31/69 (45)	5/14 (35)
SLEDAI = 0, n/N(%)	12/23 (52)	37/69 (54)	7/14 (50)
SLEDAI 1–3, n/N(%)	4/23 (17)	22/69 (32)	3/14 (21)
SLEDAI >3, n/N(%)	7/23 (31)	10/69 (14)	4/14 (29)
SDI = 0, n/N(%)	14/23 (61)	43/69 (62)	8/14 (57)
SDI 1 or 2, n/N(%)	8/23 (35)	21/69 (31)	5/14 (36)
SDI >2, n/N(%)	1/23 (4)	5/69 (7)	1/14 (7)
Fatigue scale, mean (S.D.)	4.5 (3.4)	4.13 (2.9)	5.3 (2.3)

Ruiz-Irastoraza, Rheum 2008

Vitamin D and undifferentiated connective tissue disease

- 161 UCTD patients studied compared with controls
- 25OHD significantly lower in summer as well as winter
- Photosensitivity, discoid rash and pleuritis associated with lower 25OHD
- Patients who progressed to differentiated CTD had lower 25OHD

Figure 1



Comparison of vitamin D of undifferentiated connective tissue disease (UCTD) patients with healthy controls during the summer and winter months. NS, not significant.

Table 3

Comparison of 35 patients who developed established connective tissue disease with 126 patients who remained in the stable stage of undifferentiated connective tissue disease

	Patients with evolution into defined CTD n = 35	Patients with 'stable' UCTD n = 126	*P* value
Age, years (mean ± standard deviation)	43.85 ± 11.1 range: 21–67	44.9 ± 12.7 range: 17–78	0.651
Duration of follow-up, years	2.31 ± 1.2 range: 0.5–4	4.09 ± 2.36 range: 0.5–9	0.0006113
Vitamin D serum levels, ng/mL	14.7 ± 6.45 range: 4.7–25.2	33.0 ± 13.4 range: 6–88.9	0.0001

CTD, connective tissue disease; UCTD, undifferentiated connective tissue disease.

Zold, Arthritis Research & Therapy, 2008

Randomised trial data:
Vitamin D and respiratory tract infections

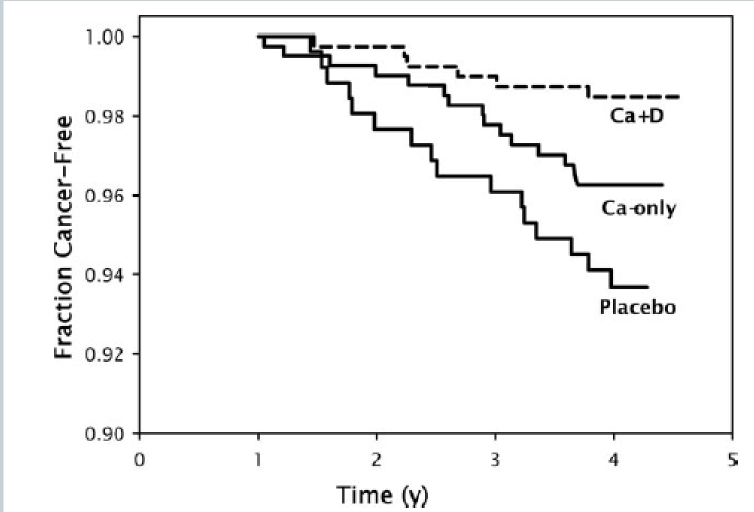
- 164 voluntary Finnish military recruits
- Randomised to 400IU D3 or placebo from Oct to March
- Primary outcome= mean number of days absent from duty

Variable	All subjects (*n* = 164)	Vitamin D supplementation group (*n* = 80)	Placebo group (*n* = 84)	*P*
Days absent from duty, mean days (± SD)				
Overall	2.6 ± 3.6	2.2 ± 3.2	3.0 ± 4.0	.096
1–6 Weeks	1.1 ± 2.4	0.7 ± 2.1	1.4 ± 2.6	.060
7–14 Weeks	0.7 ± 1.8	0.7 ± 1.4	0.8 ± 2.1	.903
15–20 Weeks	0.5 ± 1.0	0.4 ± 1.0	0.5 ± 1.1	.120
21–24 Weeks	0.4 ± 1.5	0.4 ± 1.8	0.3 ± 1.1	.311
No days absent from duty[a]				
Overall	71 (43.3)	41 (51.3)	30 (35.7)	.045
1–6 Weeks	121 (73.8)	64 (80.0)	57 (67.9)	.077
7–14 Weeks	121 (76.1)	61 (77.2)	60 (75.0)	.845
15–20 Weeks	106 (75.7)	58 (82.9)	50 (69.4)	.077
21–24 Weeks	84 (80.7)	47 (79.7)	37 (82.2)	.284

Laaksi et al, JID 2010

Randomised trial data:
Vitamin D and cancer risk

- 4 year study 1179 > 55 yo women
- Randomised placebo, Ca carb 1500mg or citr 1400mg and Ca plus 1000 IU cholecalciferol
- 86% completed study
- 25D measured by IDS RIA
- 2nd outcome measure = Ca
- Ca +D Ca RR c/w placebo 0.402 (P =0.013)
- independent risk of Ca:
  Baseline 25 OHD (P<0.002)
  12/12 25 OHD (P< 0.03)



- Multiple logistic regression:
  Baseline 25OHD
  12/12 25 OHD
  35% risk reduction for each 25 nmol/L inc in 25D

Lappe JM et al, Am J Clin Nutr 2007: 85: 1586-1591

Conclusions

- Vitamin D effects on immune function are biologically plausible
- Vitamin D may exert anti-infectious or anti-inflammatory properties via effects on innate and adaptive immune function
- Majority of studies so far are association and observational
- Only 2 randomised clinical studies show a beneficial effect of vitamin D but only as 2nd outcome events
- Until larger studies demonstrate effectiveness of intervention it is premature to recommend vitamin D supplementation for the prevention of autoimmune disease or infection
- However, in symptomatic patients or those with a high risk of falls or fractures, it is appropriate to measure 25OHD and supplement those individuals at the highest risk of adverse events (25D < 50 nmol/L)