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Photo of town center and Roman aqueduct in Segovia Spain © 2016 by Dr Vasquez

Expert Perspectives • Clinical Nutrition • Research Methodology • Publication Analysis

How to Understand, Refute, and Plan Studies Using Vitamin D

Alex Vasquez DO ND DC FACN

Defining the problems

1. **The (primary) problem:** Most doctors and researchers have zero expert-level training in Nutrition (let alone Clinical Nutrition, Therapeutic/Interventional Nutrition, Functional Nutrition) and therefore the studies they design using vitamin D are methodologically flawed, as described below.
2. **The (secondary) problem:** Too many studies using vitamin D (cholecalciferol) have used vitamin D in 1) doses that are inadequate, 2) for durations that are inadequate, and thus these studies are therapeutically underpowered, tending to lead to lackluster or negative (inefficacious) results, thereby leading to the false conclusion that vitamin D is ineffective when in fact it either *is* or *might be* effective.
3. **The (tertiary) problem:** As a result of therapeutically underpowered studies, too many research articles paint a false picture of inefficacy when in fact vitamin D is or may be highly efficacious; as a result, patients are denied a safe and effective therapeutic route that offers low-cost efficacy, high safety, and numerous collateral benefits.
4. **The (quaternary) problem:** Another major problem is that too many doctors and researchers are unaware of the major paradigm-shifting studies that should have resulted in major acceptance of vitamin D utilization in preventive public health and clinical medicine; as a result of this ignorance, too many research projects are essentially starting from zero or a very shallow foundation rather than progressively building on a foundation of good science and appropriate pattern recognition. Researchers who have not studied the history of nutrition and the decades of literature are essentially ignorant of the history and direction of the

field into which they enter; one can be amused by the prospect of a researcher placed in a position of authority to shape and define the direction of a field which he/she has never studied, ie, many researchers quite obviously wear no clothes.

Guidelines for vitamin D clinical trials were broadly published in 2004 and 2005

In 2004 and 2005, I was the principal author on several publications published in peer-reviewed journals, and in each of these I listed criteria for the design and therefore evaluation of studies using vitamin D; I will list these publications here with hyperlinks to their full text and then describe these criteria with any updates.

1. Vasquez, Manso, Cannell. The clinical importance of vitamin D (cholecalciferol): a paradigm shift with implications for all healthcare providers. *Altern Ther Health Med* 2004 Sep¹: [PDF](#), [PMID 15478784](#)
2. Vasquez, Cannell. Calcium and vitamin D in preventing fractures: data are not sufficient to show inefficacy. *British Medical Journal* 2005 Jul²: [PDF](#), [PMID 16002891](#)
3. Vasquez. Subphysiologic doses of vitamin D are subtherapeutic: comment on the study by the Record Trial Group. *TheLancet.com* 2005 May PDF

According to the pioneering clinical trial by Heaney et al (*Am J Clin Nutr* 2003 Jan³), “Healthy men seem to use 3000–5000 IU cholecalciferol/d”; a daily dose of 3,000–5,000 IU cholecalciferol/d corresponds to a serum 25-OH-vitamin D of 60 ng/ml (150 nmol/L). However, according to this study, serum 25-OH-vitamin D should be equal to or greater than 80 ng/ml (200 nmol/L) in order to alleviate secondary relative hyperparathyroidism; the daily dose of vitamin D₃ required to lower/normalize

serum parathyroid hormone (PTH) is 10,000 IU (250 mcg) per day. Therefore, we can roughly conclude that a reasonable daily dose of vitamin D ranges from 4,000–10,000 IU per day, and that the lowest acceptable serum 25-OH-vitamin D levels corresponding with adequate supplementation is 60 ng/ml (150 nmol/L) whereas a level of 80 ng/ml (200 nmol/L) is required to alleviate secondary (relative) hyperparathyroidism. Several of my publications (listed as #4 and #5 below) have also included a description of the minimal values and optimal therapeutic ranges for serum 25-OH-vitamin D; the perhaps obvious importance of these ranges is to define effective treatment (ie, sufficient vitamin D supplementation/nutriture) and to therefore differentiate adequate from inadequate supplementation dosages.

4. Vasquez. Musculoskeletal Pain: Expanded Clinical Strategies, continuing medical education (CME) monograph commissioned and published by the Institute for Functional Medicine 2008 PDF*

5. Vasquez. Revisiting the five-part nutritional wellness protocol: the supplemented Paleo Mediterranean diet. *Nutritional Perspectives* 2011 Jan PDF*

This article from 2011 is excerpted from my 2016 textbook [Inflammation Mastery, 4th Edition](#) to provide necessary updates; this article also describes the clinical use of vitamin D within the context of a foundational clinical nutrition protocol.

“This insight also illuminates a double-standard in research: whereas no legitimate drug study would use a subtherapeutic dose of a pharmaceutical agent and then (falsely) assert inefficacy, poorly designed and therapeutically underpowered (eg, using 10% of the known effective dose) nutrition studies are published and make headlines and shape policy (mostly by maintaining the status quo of nutritional inaction and ignorance) on weekly basis. For example, a study using an antibiotic or antiseizure drug that failed to administer a therapeutic dosage or achieve a therapeutic serum level would never be accepted for publication in a headlining medical journal; yet, underdosed nutrition studies are commonly published in headlining journals and then reported to mainstream media as proof of the inefficacy of nutritional intervention.”

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vitamin D3 per day. Therefore, intervention trials with supplemental vitamin D should use between 4,000 IU/day, which is presumably sufficient to meet physiologic demands, and 10,000 IU/day, which is the physiologic dose attained naturally via full-body sun exposure within a short period of time outdoors. Also, the higher dose of 10,000 IU/day is necessary in some patients who have absorption defects and therefore need a higher oral dose to "force absorption" and/or who are obese and therefore need a higher dose to achieve tissue saturation for a larger body mass. Based on these physiologic criteria, we see that the majority of intervention studies in adults have used inadequate, subphysiologic doses of vitamin D. Therefore, many studies that failed to identify therapeutic benefits from vitamin D supplementation were flawed due to insufficient therapeutic intervention—the dose of vitamin D was too low. This insight also illuminates a double-standard in research: whereas no legitimate drug study would use a subtherapeutic dose of a pharmaceutical agent and then (falsely) assert inefficacy, poorly designed and therapeutically underpowered (eg, using 10% of the known effective dose) nutrition studies

are published and make headlines and shape policy (mostly by maintaining the status quo of nutritional inaction and ignorance) on weekly basis. For example, a study using an antibiotic or antiseizure drug that failed to administer a therapeutic dosage or achieve a therapeutic serum level would never be accepted for publication in a headlining medical journal; yet, underdosed nutrition studies are commonly published in headlining journals and then reported to mainstream media as proof of the inefficacy of nutritional intervention.

2. Vitamin D supplementation must be continued for at least 5-9 months for maximum benefit: Since serum 25(OH)D levels do not plateau until after 120 days or 4 months of supplementation, and we would expect clinical and biochemical changes to become optimally apparent some time after the attainment of peak serum levels, any intervention study of less than 6-9 months is of insufficient duration to determine either maximum benefit or inefficacy of vitamin D supplementation. Conversely, since vitamin D supplementation can alter intracellular metabolism within minutes of administration, benefits seen in short-term studies should not be inaccurately

Past and Future Vitamin D Studies: Critique and Design

A large percentage of published clinical trials have suffered from flawed design, including inadequate dosing, inadequate duration, wrong type of vitamin D (ie, ergocalciferol, D2), failure to test serum vitamin D levels, and/or failure to ensure that serum vitamin D levels entered into the optimal range. The following guidelines have been provided for clinicians and researchers using vitamin D in clinical practice and research to improve the quality of research and patient care.

1. Dosages of vitamin D must reflect physiologic requirements and natural endogenous production and should therefore be in the range of 3,000–10,000 IU per day: The physiologic requirement for vitamin D is 3,000–5,000 IU per day in adult males. Full-body exposure to ultraviolet light (eg, sunshine) can produce the equivalent of 10,000–25,000 IU of

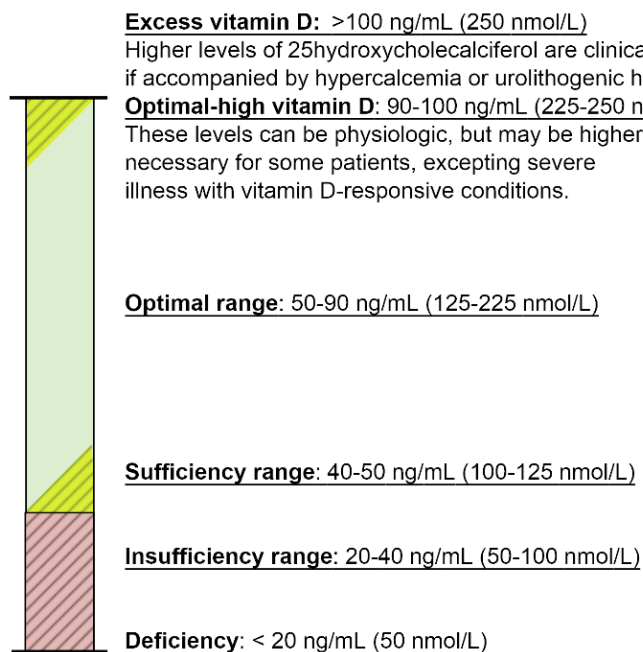
attributed to statistical error or placebo effect. The vitamin D trial does not begin with the initiation of supplementation but rather the study begins after the achievement of vitamin D sufficiency (defined below).

3. Supplementation should be performed with D3 rather than D2: Although cholecalciferol (vitamin D3) and ergocalciferol (vitamin D2) are both used as sources of vitamin D, D3 is the human nutrient and is much more efficient in raising and sustaining serum 25[OH]D levels. Vitamin D2 is a fungal metabolite and has been associated with adverse effects due to contamination and altered pharmacokinetics. The type of vitamin D must always be clearly stated in published research reports.
4. Supplements should be tested for potency: Some products do not contain their claimed amount. This problem was illustrated in the study by Heaney et al³ who found that the vitamin D supplement they used in their study, although produced by a well-known company, contained only 83% of its stated value. To ensure accuracy and consistency of clinical trials, actual dosages must be known.
5. Effectiveness of supplementation must include evaluation of serum vitamin D levels: Supplementation does not maximize therapeutic efficacy unless it raises serum 25(OH)D levels into the optimal range. To assess absorption, compliance, and safety, serum 25(OH)D levels must be monitored in clinical trials involving vitamin D supplementation. Assessment of serum levels is important also to determine the relative dose-

effectiveness of different preparations of vitamin D, as some evidence suggests that emulsification facilitates absorption of fat-soluble nutrients. Measurement of 1,25-dihydroxyvitamin (calcitriol) is potentially misleading and is not recommended for the evaluation of vitamin D status; however, measurement of calcitriol levels is increasingly used clinically to evaluate for the severity or presence of inflammatory and malignant diseases, as discussed in [Inflammation Mastery \(2016\)](#).

6. Serum vitamin D levels must enter the optimal range: The majority of clinical intervention studies using vitamin D have failed to use supplementation of sufficient dosage and duration to attain optimal serum levels of vitamin D. Our proposed optimal range for 25(OH)D is 50-100 ng/mL (see updated figure and [PDF excerpt](#)).
7. Patients must be taken from a state of absolute or relative deficiency to absolute sufficiency: If patients are deficient at the start and the end of the study, then no adequate treatment has taken place. If patients were not deficient at the start of the study, then little improvement would be expected in moving them from "vitamin D adequate" to "vitamin D supra-adequate" in most cases.

The above-mentioned criteria will aid future researchers in designing interventional studies that can accurately evaluate the relationship between vitamin D status and human illness. Furthermore and by extension, these criteria help us form a checklist with which to evaluate planned and published research.



Interpretation of serum 25-hydroxy-cholecalciferol levels: Interpretation of any laboratory variable requires clinical contextualization; assessing renal function and measuring 1,25-dihydroxy-cholecalciferol prior to the initiation of vitamin D3 supplementation is reasonable, especially in patients with higher probability of renal insufficiency or granulomatous/malignant/inflammatory disease, respectively.

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Vitamin D-responsive conditions*

- Depression
- Autism
- Seizures/epilepsy
- Musculoskeletal pain, especially low-back pain and "fibromyalgia"
- Opioid dependence for pain
- Hypertension
- Autoimmunity such as systemic lupus erythematosus and multiple sclerosis
- Migraine
- Diabetes and insulin resistance
- Polycystic ovarian syndrome
- Cancer, especially prostate cancer
- Infectious diseases, especially including viral and bacterial infections

*following correction of deficiency

How to Critique Vitamin D Studies—A Checklist

1. Did the study subjects receive at least 4,000-10,000 IU per day? If not, then the study likely used inadequate dosage to produce optimal physiologic effects.
2. Is the duration of the study at least 6-9 months? If not, then body stores of vitamin D were likely not replaced in time for clinical effect to occur. Daily supplementation with vitamin D requires 120 days (4 months) to reach plateau of serum 25-OH-vitamin D levels; therefore, the replenishment or “induction” phase of any clinical trial must have a duration of at least 4 months or—alternatively—use supranormal doses of vitamin D3 in order to more rapidly achieve optimal serum levels and tissue saturation.
3. Did the study use vitamin D3 (cholecalciferol) rather than fungus-derived ergocalciferol? Ergocalciferol is not a human nutrient, and it is more toxic and less effective than is cholecalciferol.
4. Was the product validated for potency? If not, then the intervention may have failed due to an erroneously produced or falsely labeled product.
5. Were serum 25-OH-vitamin D levels measured? If not, the product potency and nutrient absorption were not ensured.
6. Did serum 25-OH-vitamin D levels enter the optimal range at least 2-6 months before the end of the study? If not, then the patients may have been vitamin D deficient for the entire duration of the study.
7. Were the patients deficient at the start of the study and then robustly replaced with vitamin D? If not, then “deficiency→deficiency” is not a competent study design and intervention, nor is “replete→replete.” The appropriate intervention is to change deficiency to repletion.
8. Vitamin D supplementation should be stopped for roughly 20-30 days before serum testing because 25-hydroxyvitamin D3 (calcidiol) has a half-life of 15 days.⁴ The goal with serum testing of 25-OH-vitamin D levels is to assess tissue saturation, not acute absorption. Testing vitamin D serum levels within a few days of vitamin D supplementation is more likely to reflect absorption and hepatic conversion rather than providing the more important and more accurate assessment of vitamin D tissue stores.

Obviously, clinical trials need to control for factors that increase vitamin D status (eg, sun exposure, fish oil especially cod liver oil) and those which promote vitamin D deficiency, especially antiepileptic drugs, cholestyramine. Research and editorial integrity cannot be assumed in mainstream headlining journals.⁵

Clinical take-home

Clinicians, who are not conducting research but rather are interested in attaining clinical improvement in their patients, should follow the above guidelines when using vitamin D supplementation in patients, while remembering to monitor for toxicity with the triad of clinical assessments, serum 25(OH)D, and serum calcium. Clinicians and researchers need to remember, however, that optimal clinical effectiveness often depends on synergism of diet, lifestyle, exercise, emotional health, and other factors. Single intervention studies are a reasonable research tool only for evaluating cause-and-effect relationships based on the presumption of a simplistic, linear model that is generally inconsistent with the complexity and multiplicity of synergistic and interconnected factors that determine health and disease. Thus, single intervention studies with vitamin

D supplementation will be useful from an intellectual standpoint insofar as they will help us to further define the role of vitamin D in human physiology and pathophysiology. However, optimal clinical results with individual patients are more easily attained with the use of multicomponent treatment plans that address many facets of the patient’s health.

A reasonable goal with vitamin D supplementation is the downward normalization of parathyroid hormone (PTH) levels; relative elevations of PTH (excluding pathologic and primary elevations of PTH) signify compensation for insufficient intake and/or absorption of calcium. According to the clinical trial by Heaney et al³, the dose required to achieve this is 10,000 IU (250 mcg) per day corresponding to serum 25-OH-vitamin D of 80 ng/ml (200 nmol/L). Therefore, and also given that such levels are physiologically attained with sun exposure, a target of 80 ng/ml (200 nmol/L) is quite reasonable.

2017 vitamin D supplementation guidelines

In early 2017, “vitamin D supplementation guidelines” were published⁶ endorsing the following supplementation regimens:

- Neonates (i.e. younger than one month): 1,000 IU/day (25 mcg/day),
- Infants older than 1 month and toddlers: 2000-3000 IU/day (50-75 mcg/day),
- Children and adolescents aged 1 to 18 years: 3000-5000 IU/day (75-125 mcg/day),
- Adults and the elderly: 7000-10,000 IU/day (175-250 mcg/day) or 50,000 IU/week (1250 mcg/week).

The authors also note that obese patients need up to 300% more vitamin D than do persons of normal weight, and that—as noted previously and consistently throughout the literature—“the dose of 10,000 IU/d was also found as the no-observed-adverse-effect level (NOAEL).” Consistent

“The vitamin D trial does not begin with the initiation of supplementation but rather the study begins after the achievement of minimal vitamin D sufficiency, as documented by a serum 25-OH-vitamin D level of at least 50 ng/ml or 125 nmol/L.”
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with the clinical guidelines that I have published since 2008, these 2017 guidelines state “It is generally accepted that a serum 25(OH)D concentration of up to 100 ng/mL (250 nmol/L) is safe for children and adults, with the exception of those who have a hypersensitivity to vitamin D.” They further note that “The Endocrine Society guidelines concluded that vitamin D toxicity is not only extremely rare, but 25(OH)D concentration of at least 150 ng/mL (375 nmol/L) is required before there would be evidence of vitamin D toxicity.”

Vitamin D's safety and efficacy have already been established, justifying routine use; to continue inertia and inaction is actually dangerous and unethical

We established the safety, efficacy, and clinical imperative of vitamin D supplementation in our landmark review in 2004 by Vasquez, Manso, and Cannell, *Altern Ther Health Med* 2004 Sep¹:

"As a medically valid diagnosis (ICD-9 code: 268.9 Unspecified vitamin D deficiency) with a high prevalence and clinically significant morbidity, vitamin D deficiency deserves equal attention and status with other diagnoses encountered in clinical practice. Given the depth and breadth of the peer-reviewed research documenting the frequency and consequences of hypovitaminosis D, failure to diagnose and treat this disorder is ethically questionable (particularly in pregnant women) and is inconsistent with the delivery of quality, science-based healthcare. Failure to act prudently based on the research now available in favor of vitamin D supplementation appears likely to invite repetition analogous to the previous failure to act on the research supporting the use of folic acid to prevent cardiovascular disease and neural tube defects—a blunder that appears to have resulted in hundreds of

thousands of unnecessary cardiovascular deaths and which has contributed to incalculable human suffering related to otherwise unnecessary neural tube defects, cervical dysplasia, cancer, osteoporosis, and mental depression. ... Of course, additional lives may be saved and suffering reduced by alleviating the morbidity and mortality associated with hypertension, autoimmune disease, depression, epilepsy, migraine, diabetes, polycystic ovary syndrome, musculoskeletal pain, osteoporosis, and cardiovascular disease."

Given cholecalciferol's low cost, high safety, and numerous direct and collateral benefits, no legitimate reason exists for routinely denying vitamin D3 supplementation to patients; vitamin D supplementation (and/or sun exposure) should be recommended and supported routinely in virtually all patients

"Until proven otherwise, the balance of the research clearly indicates that oral supplementation in the range of 1,000 IU/day for infants, 2,000 IU/day for children, and 4,000 IU/day for adults is safe and reasonable to meet physiologic requirements, to promote optimal health, and to reduce the risk of several serious diseases. Safety and effectiveness of supplementation are assured by periodic monitoring of serum 25(OH)D and serum calcium."¹¹

According to the 2011 clinical trial by Hollis et al⁷, “Vitamin D supplementation of 4,000 IU/day for pregnant women was safe and most effective in achieving sufficiency in all women and their neonates regardless of race.” A 2016 review supported the same dose of 4,000 IU/d for pregnant women.⁸

For active hyperlinks, associated PDF articles and videos, and any updates, please see: <http://www.ichnfm.org/d> ☒

History of this publication: Posted online 12 Feb 2017, updated 19 Feb, updated 23 Feb to include discussion of the recently released 2017 vitamin D supplementation guidelines.

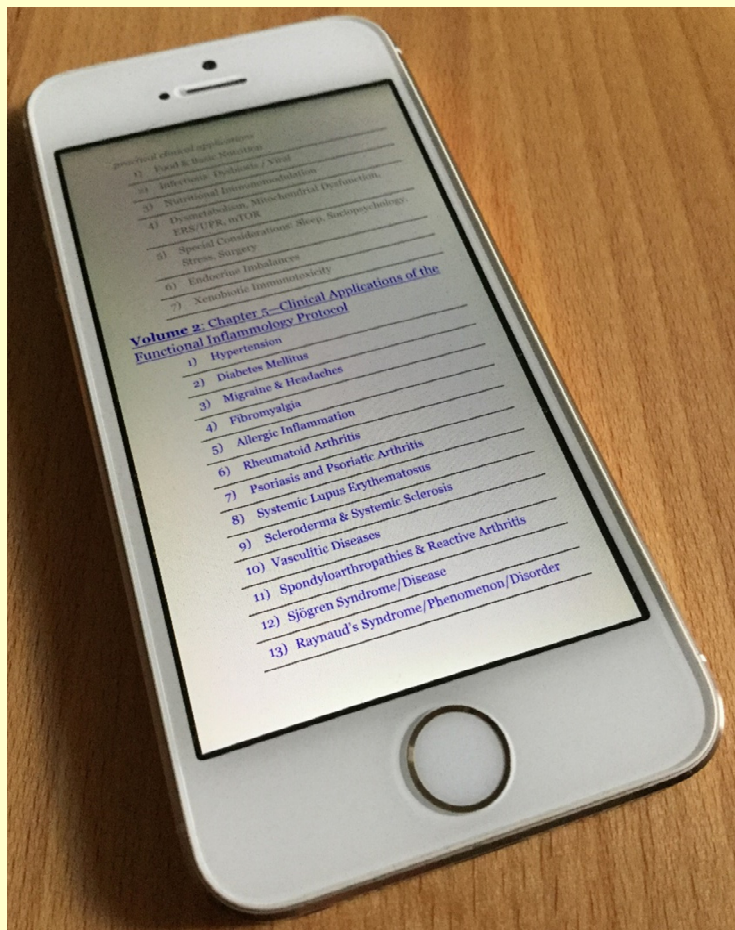
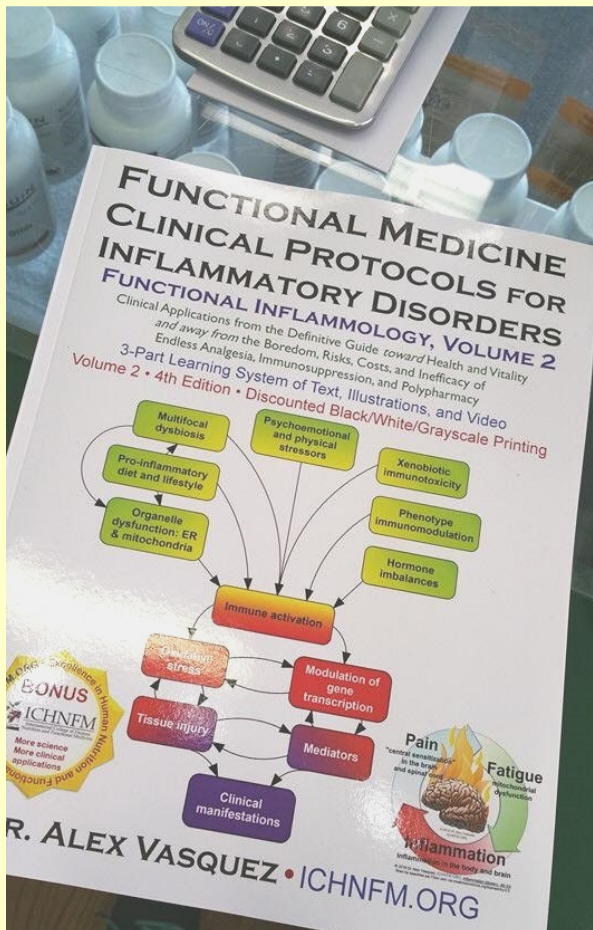
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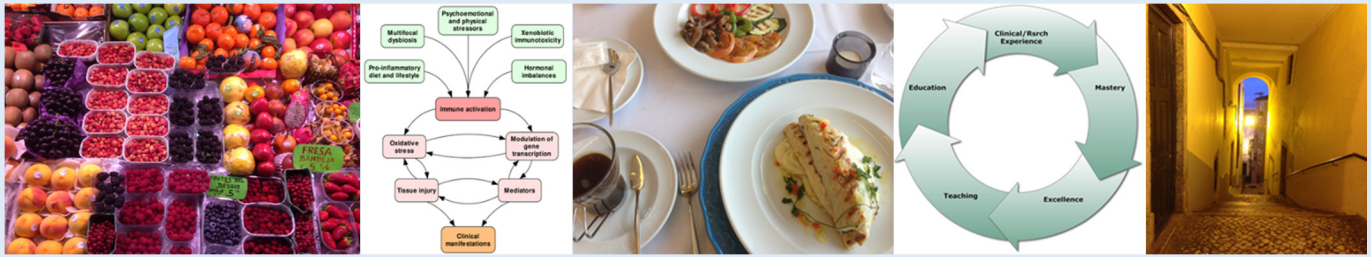
About the author and presenter: Dr Alex Vasquez holds three doctoral degrees as a graduate of University of Western States (Doctor of Chiropractic, 1996), Bastyr University (Doctor of Naturopathic Medicine, 1999), and University of North Texas Health Science Center, Texas College of Osteopathic Medicine (Doctor of Osteopathic Medicine, 2010). Dr Vasquez is the author of many textbooks, including *Integrative Orthopedics* (2004, 2012), *Musculoskeletal Pain: Expanded Clinical Strategies* (published by the

Institute for Functional Medicine, 2008) now updated and expanded to 1,200 pages as Inflammation Mastery 4th Edition (published as a two-volume set as Textbook of Clinical Nutrition and Functional Medicine), Chiropractic and Naturopathic Mastery of Common Clinical Disorders (2009), Integrative Medicine and Functional Medicine for Chronic Hypertension (2011), Brain Inflammation in Migraine and Fibromyalgia (2016), and Mitochondrial Nutrition and Endoplasmic Reticulum Stress in Primary Care (2014). "DrV" has also written more than 100 letters and articles for professional magazines and medical journals such as *Nature Reviews Rheumatology*, *British Medical Journal (BMJ)*, *TheLancet.com*, *Annals of Pharmacotherapy*, *Journal of Clinical Endocrinology and Metabolism*, *Journal of the American Medical Association (JAMA)*, *Journal of the American Osteopathic Association (JAOA)*, *Alternative Therapies in Health and Medicine*, *Nutritional Perspectives*, *Journal of Manipulative and Physiological Therapeutics (JMPT)*, *Current Allergy and Asthma Reports*, *Integrative Medicine*, *Complementary Therapies in Clinical Practice*, and *Arthritis & Rheumatism*—Official Journal of the American College of Rheumatology. Dr Vasquez lectures worldwide to healthcare professionals and provides expert consultations to physicians and patients internationally. As the former Editor of *Naturopathy Digest* and a reviewer for *Journal of Naturopathic Medicine*, *Alternative Therapies in Health and Medicine*, *PLoS-ONE*, *Neuropeptides*, *International Journal of Clinical Medicine*, *Autoimmune Diseases*, Dr Vasquez is currently the Chief Editor of *International Journal of Human Nutrition and Functional Medicine*.[®] Additional information with updates and blogs is available at ICHNFM.ORG. All of DrV's books are available at Amazon.com, videos at Vimeo.com/DrVasquez, and selected lecture recordings at iTunes. Dr Vasquez has served as a consultant researcher and lecturer for Biotics Research Corporation in the United States.

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Notable publications in 2014—① Lab Fraud in Functional Medicine, ② ISIFMC Position on HPS2-THRIVE; ③ Unified Antiviral Strategy, ④ Metabolic Correction: www.ichnfm.org/publications/IJHNF2014_review.pdf

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Chapter XVIII; testimony of Howard Roark in *The Fountainhead* by Ayn Rand



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