Vitamin D3V® Human Intervention Bioavailability Trial

In 2020 we completed a human study to qualify Vitamin D3V® as a bioavailable source of Vitamin D3. The study was completed in Ireland through Research & Development business AnaBio Technologies Ltd together with University College Dublin.

Study Highlights

- ✓ The study was performed on 10 healthy volunteers using a daily dose of 600iu Vitamin D3V®.
- ✓ All study participants saw a significant increase in plasma Vitamin D levels over the baseline.
- ✓ The average plasma Vitamin D increases were from 43.43 to 77 nmol/L (33nmol/L). This marks an average increase of approximately 77.3% above baseline for participants.
- ✓ The results were statistically significant, with a p-value using a paired t-test of 0.002.

The study confirmed that Vitamin D3V® is a bioavailable source of Vitamin D3, supporting the existing analytical data.

Study Details

Sample size:
10 subjects.

Study Objective:
The main objective of the study involved assessment of the impact of Vitamin D3V® on adults that are "insufficient" in Vitamin D. While there is no universal consensus on Vitamin D levels and status, the following table (Figure 1) provides the commonly accepted ranges as discussed in several publications[1,2,3].

Figure 1: Vitamin D Status

Plasma Vitamin D level (nmol/L)	Vitamin D Status
<25	Deficient
25-50	Insufficient
50-70	Normal / Sufficient
70-80	Optimal

Vitamin D3V® was given to healthy volunteers in order to observe the effects after ingestion (endpoint: Vitamin D3 in peripheral plasma).

Blood samples were taken on Day 0 and Day 7 of supplementation. Each participant represented their own control.

Study Power & Recruitment:
Based on the study design, 10 participants were recruited using the Study Criteria (Figure 2).

Participants:
All participants were healthy and physically active as defined by habitually performing at least 30 minutes of exercise on five or more days each week. Each participant provided written informed consent prior to participation, with age and personal details. During the initial phase of the trial, participants attended a screening visit to confirm their eligibility according to the inclusion and exclusion criteria. Each participant was given a personal study code number (001, 002, etc.) which was used throughout the study and in the analysis of data. Coded data were treated with confidentiality when undergoing analysis.

All researchers completed the Data Protection Guidance confirmation form provided by the HSE / TGL to ensure compliance with EU data protection legislation.

Figure 2. Study Criteria

Inclusion Criteria	**Exclusion Criteria**
18 to 65 years old (male or female)	Pregnant or breastfeeding
Healthy and without prescribed Pharmaceuticals (anti-conceptive drugs allowed)	Any allergies towards standard meals or treatments
No symptoms associated with gastrointestinal dysfunction	Previous substance abuse
	Professional athletes
	Current or previous history of Vitamin D supplementation (within 12 months of the study)

Characteristics of trial participants:
Healthy adults (18-65) were recruited, considering the following exclusion criteria: metabolic disease diagnosed (cardiovascular diseases, obesity, resistance to insulin or diabetes, high blood pressure, atherogenic dyslipidaemia), endocrine dysfunction, pregnancy, or post-menopause.

Project Design:
Participants were asked to abstain from caffeine and alcohol and refrain from strenuous exercise for 24-hours prior to the lab visit and were asked to keep a 2-day portion size estimate food diary prior to their visit (analysed using the Nutritics Professional Diet Analysis Software package[4]).

Sampling:
Day 0 Baseline (before initiate supplementation) followed by a 7-day measurement. A defined study dinner and an evening snack was provided the evening prior the study Day 0 as per internal procedure by the clinical team and food diary was provided.

Dosing:
Sachets were prepared, each containing 300iu of Vitamin D3V®. Each study participant was given 2 x sachets to take per day, thus subjects consumed 600iu of Vitamin D3 per day during the study.

Analytics:
The following testing were completed during on each lab visit on Day 0 and Day 7:

- Measurement of Vitamin D3 in peripheral bloods using HPLC derivatisation method.
- Total glucose using a blood-stick[5].
- Urine was analysed for Osmolality (T0; T12) (Osmolality / Tonicity Meter).
- Completion of a Sensory, Satiety and Cognitive Questionnaire.

Biomarker and Quantification: 25(OH)D2 and 25(OH)D3:
25-hydroxivitamin D is the key Vitamin D metabolite that reflects the level of Vitamin D in the body (Aissou et al., 2019). This serum biomarker is used to diagnose vitamin insufficiency and deficiency in the Vitamin.

Blood Sampling:
Blood samples were centrifuged at 2000 g for 10 minutes at 4°C. The recovered serum was divided into aliquots then frozen at -20°C and protected from light for a period of 3 to 8 months.

Results:

- Assessment was completed upon 10 healthy volunteers.
- Each daily dose comprised 600iu of Vitamin D3V®.
- In all treatment cases the plasma levels of vitamin D increased significantly over baseline.
- The average increases were from 43.43 to 77 nmol/L (33nmol/L). See Figure 3 below. This marks an average increase of approximately 77.3% above baseline for participants.
- The results were statistically significant, with a p-value using a paired t-test of 0.002.

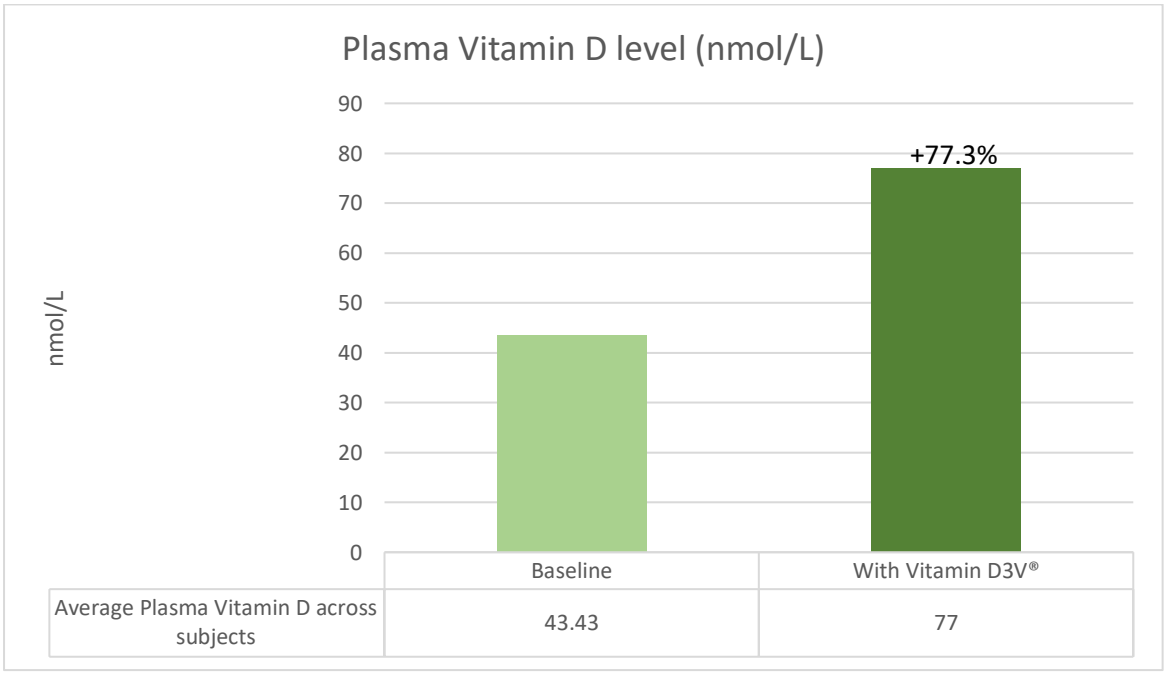

Figure 3: Chart showing the average Plasma levels of Vitamin D across the subjects, comparing between baseline (initial) and following 7 days supplementation with 600iu per day of Vitamin D3V®. The average increase was 77.3%.

References:

1. Pearce SH, Cheetham TD. Diagnosis and management of vitamin D deficiency. BMJ.340:b5664
2. SACN. Update on Vitamin D: Position Statement by the Scientific Advisory Committee on Nutrition. London: TSO; 2007.
3. Bischoff-Ferrari HA, Giovannucci E, Willett WC, Dietrich T, Dawson-Hughes B. Estimation of optimal serum concentrations of 25-hydroxyvitamin D for multiple health outcomes. Am J Clin Nutr. 2006 Jul;84(1):18-28.
4. https://www.nutritics.com/p/clinical
5. https://myfreestyle.ca/en/products/precision_neo

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