Enhanced Lymphatic Absorption of Vitamin D3 Delivered in Mealworm (Tenebrio molitor) Oil Compared to Lichen- and Lanolin-Based Commercial Oils in a Rat Model

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Abstract

Background: The intestinal absorption of vitamin D3 (cholecalciferol) is strongly influenced by the lipid matrix in which it is delivered. Insect-derived oils have emerged as novel nutritional lipid sources and may enhance the bioavailability of fat-soluble vitamins.

Objective: To compare the lymphatic absorption of vitamin D3 delivered via Tenebrio molitor (mealworm) oil with two commercial oils enriched with vitamin D3 from lichen or lanolin.

Methods: Adult male Wistar rats underwent thoracic lymph duct cannulation and received gastric intubation with: (1) vitamin D3–rich mealworm oil (TM-A), (2) lower-dose mealworm oil (TM-E), (3) vitamin D3 from lichen in rapeseed oil, or (4) vitamin D3 from lanolin in rapeseed oil. Lymph was collected for 6 h and analyzed by HPLC for cholecalciferol and by GC for fatty acid content.

Results: Absorption rates ranged from 13–22%. TM-A (21.8%) and lichen oil (19.4%) showed significantly higher vitamin D3 absorption than lanolin oil (13.1%). TM-A produced 2–3-fold higher lymphatic enrichment of vitamin D3 compared with reference oils. TM-E (half-dose) produced lymphatic concentrations comparable to full-dose lichen and lanolin oils. Mealworm oil showed 100% fatty acid absorption and significantly higher lymph lipid content versus reference oils.

Conclusion: Mealworm oil markedly enhances vitamin D3 absorption compared to lichen and lanolin carriers. Its MUFA-rich matrix appears to optimize intestinal uptake, suggesting mealworm oil as a promising nutraceutical delivery system for fat-soluble vitamins.

Keywords: vitamin D3, bioavailability, lymphatic absorption, Tenebrio molitor, insect oil, fatty acids, nutraceuticals

1. Introduction

Vitamin D_3 plays a multifaceted physiological role that extends far beyond calcium and bone homeostasis, encompassing immune modulation, endocrine regulation, cardiometabolic signaling, and cellular differentiation. Recent epidemiological evidence links suboptimal serum 25(OH) D levels with increased risk of autoimmune disorders, metabolic syndrome, infectious diseases, and impaired musculoskeletal performance. Consequently, improving the bioavailability of dietary vitamin D_3 has become an important public health objective. However, the extent to which vitamin D_3 is absorbed and utilized is contingent upon the lipid matrix through which it is delivered, as micellarization, enterocyte uptake, and chylomicron secretion are highly dependent on fatty acid composition and matrix polarity.

Lipid structural features, including chain length, degree of unsaturation, and stereochemical orientation, have been shown to influence the kinetics of intestinal solubilization and subsequent lymphatic transport of fat-soluble nutrients. Research indicates that MUFA-rich matrices tend to facilitate micelle formation and absorption, whereas certain PUFA-dominant oils may exert competitive effects on lipid transport pathways. Against this biochemical backdrop, edible insect oils have emerged as novel, sustainable lipid sources containing unique fatty acid profiles capable of enhancing nutrient delivery.

Mealworm (Tenebrio molitor) oil, in particular, is characterized by a favorable MUFA-to-SFA ratio and bioactive lipid fractions that may synergistically enhance micellar incorporation of cholecalciferol. Yet, despite rising interest in entomological oils as functional carriers, limited empirical evidence exists regarding their capacity to modulate intestinal absorption of vitamin D_3. Accordingly, the present study evaluates the lymphatic bioavailability of vitamin D_3 delivered in mealworm oil relative to two commercial matrices derived from lichen and lanolin, using a lymph-cannulated rat model - a gold-standard method for quantifying lipid transport.

2. Materials and Methods

2.1 Study design

An established lymph-cannulated Wistar rat model was used to enable continuous, temporally resolved collection of postprandial lymph following administration of vitamin D_3-enriched oils. This model is widely recognized as the most physiologically accurate system for characterizing lipid-phase nutrient absorption because it captures chylomicron-mediated transport prior to hepatic first-pass metabolism.

Oils were fully characterized by HPLC-DAD for vitamin D_3 concentration and by GC-FID for fatty acid composition to ensure mechanistic interpretation of absorption differences. Animals underwent a standardized fasting protocol to normalize intestinal transit and received a single intragastric dose calibrated to deliver either 50 µg or 25 µg vitamin D_3 depending on group assignment. Lymph was collected continuously for 6 h into chilled, EDTA-treated tubes to prevent lipid oxidation.

Vitamin D_3 quantification followed validated extraction workflows employing saponification, non-polar phase separation, and spectrally verified chromatographic detection. Fatty acid analysis used reference standards and calibration curves to ensure accuracy. The absorption coefficient (T) was computed using tracer-normalized reference values to yield quantitative estimates of intestinal bioavailability. Statistical comparisons employed ANOVA followed by Tukey's test, with significance defined as $p < 0.05$.

2.2 Test oils

Four formulations were evaluated: (1) TM-A, a mealworm oil enriched to 24.6 µg/mL vitamin D3; (2) TM-E, a mealworm oil with 11.2 µg/mL vitamin D3; (3) a lichen-derived vitamin D3 formulation diluted in virgin rapeseed oil to 28.6 µg/mL; and (4) a lanolin-derived vitamin D3 formulation diluted in virgin rapeseed oil to 23.5 µg/mL. A control group received physiological saline. The fatty acid compositions of the oils included MUFA, PUFA (n-6 and n-3), and saturated fatty acids (SFA).

Table 1. Vitamin D3 content and fatty acid composition of the test oils.

Oil	Vit D3 (µg/mL)	MUFA (%)	PUFA n-6 (%)	PUFA n-3 (%)	SFA (%)
TM-A	24.6	42.7	28.0	1.4	27.6
TM-E	11.2	58.6	20.6	9.7	11.0
Lichen-D3	28.6	58.5	20.7	9.7	11.1
Lanolin-D3	23.5	59.2	20.8	10.1	10.7

2.3 Dosing and administration

All treatment groups received an oil volume calibrated to deliver 50 µg vitamin D3, except the TM-E group, which received 25 µg vitamin D3 to explore a potential dose–response effect. Oils were administered once by gastric intubation following a fasting period.

2.4 Lymph collection and analysis

Thoracic lymph was collected continuously for 6 h after intubation. Vitamin D3 (cholecalciferol) concentrations in lymph were quantified using high-performance liquid chromatography equipped with diode-array detection. Fatty acid profiles were determined by gas chromatography with flame ionization detection.

2.5 Calculation of absorption rate

The intestinal absorption rate (T) of vitamin D3 was expressed relative to a lipid tracer standard assumed to have 100% absorption, according to the equation:

$$T = (RL / RH) \times 100$$

where RL is the amount of vitamin D3 recovered in lymph relative to the reference and RH is the

amount of vitamin D3 ingested relative to the reference dosage.

3. Results

3.1 Lymphatic vitamin D3 concentrations and absorption

Lymphatic vitamin D3 concentrations and calculated absorption rates are summarized in Table 2. Mealworm oil formulations (TM-A and TM-E) showed higher absorption rates than the lanolin-based formulation. TM-A produced 2–3-fold higher lymphatic enrichment compared with the commercial reference oils, despite equivalent dosing.

Table 2. Lymphatic vitamin D3 and absorption rate for each group.

Group	Vit D3 lymph (arbitrary units)	Absorption (%)
TM-E	21.7 ± 8.1	21.7
TM-A	21.8 ± 1.3	21.8
Lichen-D3	19.4 ± 2.8	19.4
Lanolin-D3	13.1 ± 3.6	13.1
Control	ND	—

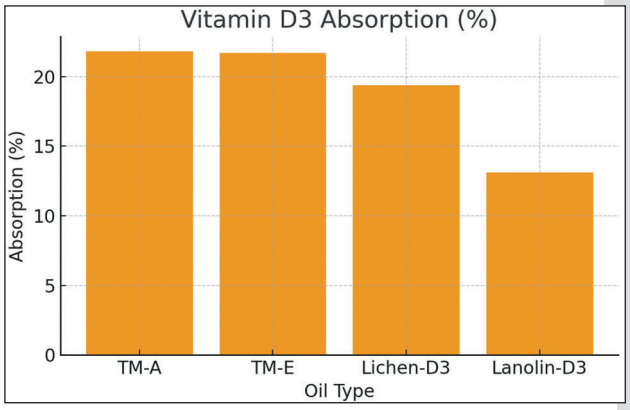

Figure 1. Vitamin D3 Absorption by Oil Type

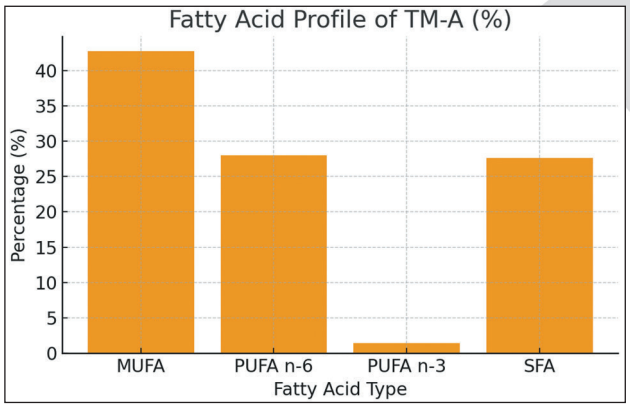

Figure 2. Fatty Acid Composition of TM-A Oil

3.2 Lymphatic fatty acid absorption

Total fatty acid absorption from mealworm oils reached 100%, as reflected by the high concentration of lymphatic fatty acids in TM-A and TM-E groups compared to lichen- and lanolin-based formulations. The lymphatic fatty acid profile closely mirrored the composition of the ingested oils.

4. Discussion

The findings of this study reveal a pronounced enhancement of vitamin D_3 absorption when delivered through mealworm oil matrices, with TM-A demonstrating the highest bioefficacy. From a mechanistic perspective, the MUFA-enriched lipid environment likely improves micelle fluidity, reduces steric interference during solubilization, and enhances substrate affinity for enterocyte transporters, including SR-B1 and NPC1L1. Moreover, the comparatively lower n-3 PUFA content of mealworm oil may reduce competitive interference during microsomal triacylglycerol resynthesis—one of the rate-limiting steps in chylomicron assembly. The observation that TM-E, despite its substantially reduced vitamin D_3 content, achieved lymphatic concentrations similar to the commercial lichen and lanolin oils indicates that matrix-driven enhancements can compensate for dose reductions. This aligns with studies demonstrating that nutrient–lipid interactions, rather than nutrient mass alone, can govern absorption kinetics. Furthermore, the complete fatty acid recovery from the mealworm treatments supports the notion that entomological lipids undergo efficient enzymatic hydrolysis and assimilation, consistent with emerging research on their digestive compatibility. These results collectively strengthen the rationale for incorporating mealworm oil into fortified food systems aimed at improving global vitamin D nutritional status. Future work should include structured lipidomics profiling, human pharmacokinetic trials, and exploration of synergistic formulations combining mealworm oil with emulsifiers or bioactive phospholipids to further potentiate vitamin D absorption.

5. Conclusion

Mealworm (Tenebrio molitor) oil significantly enhances the lymphatic absorption of vitamin D3 relative to lichen- and lanolin-based oils in a lymph-cannulated rat model. Its lipid profile appears to optimize intestinal uptake, making it a promising delivery platform for fat-soluble vitamins. Further clinical studies in humans are warranted to confirm these preclinical observations and to explore applications in fortified foods and dietary supplements.

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