



# In vitro Demodicidal Activity Using High Concentration Terpinen-4-ol (T4O) Encapsulated in Nano-lipidic Particle Emulsion



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## Background

*Demodex folliculorum* is a translucent mite measuring 0.3 to 0.4 mm in length with four pairs of clawed legs.<sup>1</sup> It tends to cluster near the lash root and follicle, where it feeds on sebum and follicular epithelial cells<sup>2</sup> and can lead to the development of *Demodex* blepharitis.<sup>1</sup> Tea tree oil (TTO) products are available in many forms and have been proven effective in improving both the signs and symptoms of blepharitis.<sup>1</sup> Terpinen 4-ol (T4O) is a derivative of tea tree oil (TTO) that has been shown to exhibit broad-spectrum antimicrobial activity against both bacteria and fungi.<sup>1</sup> It has also demonstrated acaricidal properties.<sup>3</sup> However, the precise mechanism of action against *Demodex* has yet to be identified.<sup>3</sup>

## Purpose

High-concentration TTO products (>50%) have proven effective for mite eradication,<sup>4</sup> but they are not safe for home use.<sup>1</sup> It is uncertain whether lower concentrations can achieve mite eradication.<sup>4</sup> This study seeks to describe the demodicidal activity of 5% terpinen-4-ol (T4O) encapsulated in nano-lipidic particle emulsion lotion on *Demodex folliculorum*.

## Ingredients

Water, 4-Terpineol, Alcohol, Butyrospermum Parkii (Shea) Butter, Polysorbate 80, Olea Europaea (Olive) Fruit Oil, Cetearyl Alcohol, Squalane, Cetyl Alcohol, Cyclopentasiloxane, Glyceryl Stearate, PEG-100 Stearate, Sea Whip Extract, Camellia Sinensis Leaf Extract, Phenoxyethanol, Ethylhexylglycerin, Lecithin, Di-PPG-3 Myristyl Ether Adipate, Polysorbate 60, Hydrogenated Polyisobutene, Propylene Glycol, Carbomer, Sodium Hydroxide, PEG-150 Stearate, Steareth-20, Dimethicone Crosspolymer-3, Bisabolol

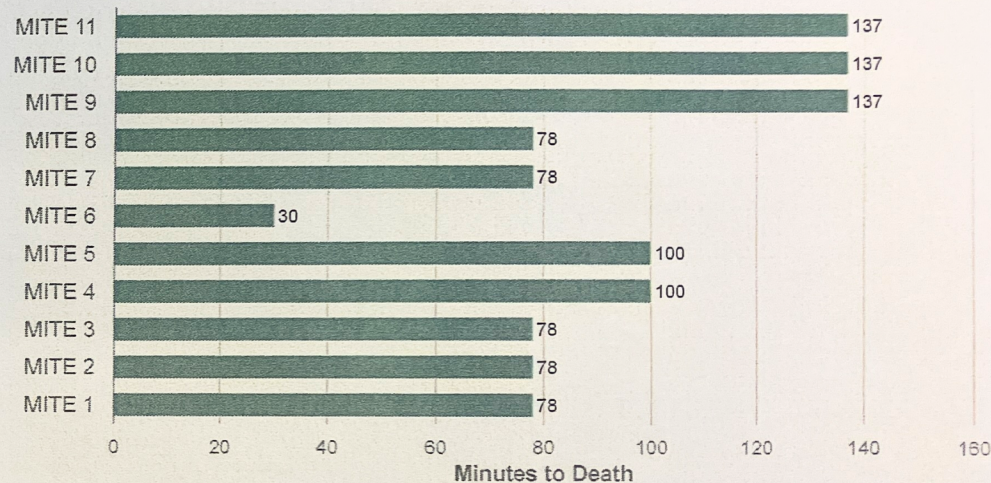
## Methods

- Live *Demodex* mites were obtained from human volunteers by epilating two eyelashes from the upper and lower eyelid of both eyes.
- The mites were immersed in a test solution: 0.07 grams of 5% terpinen-4-ol (T4O) encapsulated in nano-lipidic particle technology. The test solution was diluted with 1 ml of phosphate buffered saline (1x) pH 7.4 for visibility purposes.
- Samples were observed under the microscope for up to 137 mins, with time-to-death recorded every 30 minutes.
- Kill time was defined as the elapsed time between the addition of test solution and all cessation of movement of the body, legs, mouth and pedipalps for a minimum of 5-10 minutes.

## Results

The 5% terpinen-4-ol (T4O) encapsulated in nano-lipidic particle emulsion utilized in this proof-of-concept study demonstrated demodicidal activity as early as 30 minutes with 100% of *Demodex* mites reaching zero movement or death in an average of 96.6 minutes, even when placed in a saline-diluted solution as was required for visibility purposes.

## Mite Eradication



## Discussion

*Demodex* is the most common ectoparasite found on humans and is frequently implicated in blepharitis.<sup>1</sup> Consequently, 25 million Americans are affected by *Demodex* blepharitis lid disease.<sup>1</sup> Particularly in mild to moderate cases, many patients desire a natural remedy that can be acquired without a prescription. However, balancing efficacy with safety and tolerability has been challenging. To that end, next-generation formulations have been developed utilizing encapsulated in nano-lipidic particle emulsion. The current *in vitro* study demonstrates promise with regard to the efficacy of the study agent. Future research is needed to explore the side-effect profile and overall tolerability of this lotion-based emulsion for use on human lids.

## Conclusion

The 5% terpinen-4-ol (T4O) encapsulated in nano-lipidic particle lotion effectively killed all mite samples within 137 minutes of exposure. These proof-of-concept results demonstrating kill times provide the basis for future testing to determine whether the encapsulated nano-particle formulation can mitigate discomfort associated with high-concentration terpinen-4-ol (T4O) application.

## Sources

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