



Scientific Summary

Table of Contents

Horopito – the ancient herb.....	2
Traditional Use.....	2
Anti- <i>Candida</i> Discovery.....	2
Kolorex® Horopito.....	3
Antifungal and Antibacterial Activity.....	3
Mode of Action.....	4
Toxicology and Mutagenicity.....	4
Safety.....	5
<i>in vitro</i> Efficacy.....	5
<i>in vivo</i> Studies.....	9
Stability Studies.....	10
Clinical Studies.....	12
Kolorex® Products.....	14
References.....	15



Horopito – the ancient herb

Horopito (*Pseudowintera colorata*) only grows in New Zealand. This ancient shrub is a member of the primitive Winteraceae family, common to the Southern Pacific. It has features of the earliest evolved flowering plants, and appears in the fossil record over 65 million years ago. It is a very slow growing plant that lacks the specialist water conducting tubes found in nearly all other flowering plants. It grows well only in damp areas, especially under temperate rainforest¹. The red colouration on the leaves may give protection against harsh ultraviolet radiation² and the pungent taste is a deterrent to insects and animals³

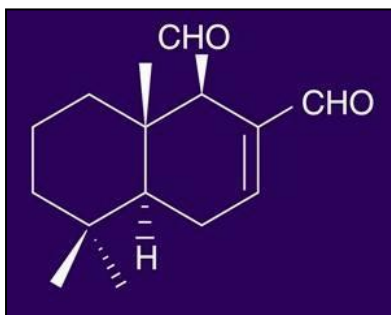
Traditional Use

Horopito has a long history of medicinal use by New Zealand's indigenous Maori population. The leaves were bruised, steeped in water and used for paipai (a skin disease), and venereal diseases. The leaves were chewed for toothache⁴. It was also used for skin diseases like ringworm. A decoction of the leaves was often used to allay inward pain and is honoured with the name 'Maori painkiller'⁵.

Anti-*Candida* Discovery

In 1982 Professor J.R.L. Walker and his team at New Zealand's University of Canterbury isolated a sesquiterpene dialdehyde called polygodial in the leaves of Horopito (see Figure 1).

Figure 1. Chemical structure of Polygodial



In vivo laboratory testing demonstrated that an extract from Horopito had strong anti-fungal activity against the yeast *Candida albicans*⁶. The activity of polygodial from the extract was compared with that of Amphotericin B (a proprietary pharmaceutical product used to treat systemic mycoses) and found to be stronger and faster acting. The polygodial extract from Horopito gave larger zones of inhibition against *C. albicans* and was effective from day one whereas the inhibitory effect of Amphotericin B against *C. albicans* required three to four days incubation to become effective⁷. See Table 1 for test results.

Scientific Summary

Table 1. Comparison of antibiotic activity of polygodial and Amphotericin B against *C. albicans*. Inhibition expressed as mean diameter (mm) of zone of inhibition⁷

Concentration (ug/disc)	Time (days)				
	1	2	4	8	11
Polygodial					
10	17.1	16.5	15.2	14.7	14.4
5	15.0	13.4	11.2	11.5	10.5
1	9.0	8.0	7.3	7.0	7.0
Amphotericin B					
100	F	F	8.2	8.3	8.3
10	F	F	6.3	6.5	6.4
1	F	X	X	X	X

F = Faint zone of inhibition; x = no inhibition

Kolorex® Horopito

Terpenes are a large class of chemical compounds found in natural products. Many of them have important biological properties.⁸ Kolorex® Horopito leaves contain at least 21 terpenes (of which the sesquiterpene polygodial is dominant) and at least 4 flavonoids⁹.



Horopito grows wild through much of New Zealand's elevated and high rainfall regions but varies in appearance and growth habit. In 1998 Forest Herbs Research Ltd commissioned research to compare all the major population groups. Leaf samples were extracted and assayed for polygodial content and their effectiveness against *C. albicans*. Antifungal activity testing (using the zone of inhibition test) against *C. albicans* showed a five-fold difference between the most active and least active subspecies¹⁰. Kolorex®

Horopito is derived from only the most active plants.

Recent research by New Zealand *P. colorata* experts has provided further evidence that polygodial is present in higher concentrations in red leaves¹¹. The interiors of red margined leaves were also found to be richer in polygodial than those of green leaves. The colour of dried Kolorex® Horopito leaves is distinctively red/purple when compared with many populations in the wild.

Antifungal and Antibacterial Activity

In 1998 Professor Kubo's team at Berkeley University used *in vitro* macrobroth dilution testing to demonstrate that polygodial had strong antifungal activity against the yeast like fungi *C. albicans*, *C. utilis*, *C. krusei*, *Cryptococcus neoformans*, *S. cerevisiae* and also filamentous fungi including *T. mentagrophytes*, *T. rubrum* and *Penicillium marneffeii*. They found polygodial's antifungal activity was strongly increased under acidic conditions while variation in incubation temperature and

Scientific Summary

inoculum size had little effect. Unlike Amphotericin B, polygodial did not show any hemolytic activity (rupture of red blood cells) and its mode of action was concluded to be different from that of existing antifungal drugs (see below)¹².

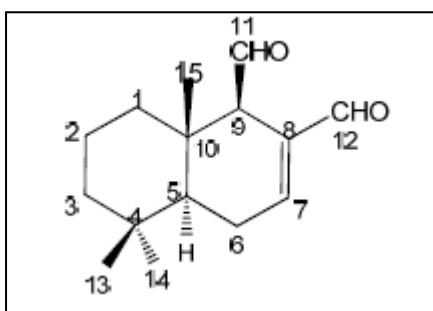
In the area of food preservation polygodial exhibits synergistic fungicidal properties. Sorbic acid is considered one of the least harmful preservatives in use but high concentrations are necessary for fungicidal activity. The fungicidal activity of sorbic acid against *S. cerevisiae* was enhanced 64 fold and that of benzoic acid 400 fold when these common preservatives were combined with half the minimum fungicidal concentration of polygodial¹³. EDTA is another food preservative upon which polygodial exerts a synergistic effect, presumably by facilitating its transport into yeast cells¹⁴. *Zygosaccharomyces baillii* is a spoilage yeast that can survive in acid media with ethanol such as wine, however polygodial controls it at very low concentrations¹⁵. Research in this area highlights the potential of Kolorex® Horopito extracts to be used as natural preservatives.

In addition to its antifungal activities, polygodial has moderate antibacterial activity against both gram positive bacteria (including *Bacillus subtilis* and *Staphylococcus aureus*) and gram negative bacteria (including *Escherichia coli* and *Salmonella choleraesuis*) with minimum bactericidal concentrations ranging from 100–400ug/ml¹⁶.

Mode of Action

Using *S. cerevisiae* as a model, polygodial was found to act as a potent antifungal. It uses various processes but polygodial's primary antifungal action is as a nonionic surfactant. It damages the permeability barrier of yeast cells.¹⁷ Disruption of the cell membrane surface induces cell leakage in the human neuroblastoma cells¹⁸. It is also likely that polygodial permeates by passive diffusion across the plasma membrane, and once inside the cells may react with a variety of intracellular compounds¹⁹.

The structural features that are responsible for the biological activity of polygodial and similar drimane compounds have been the subject of much research. Originally the activity was thought to be due to the aldehyde groups especially the one at C8¹⁷. The latest contribution to this debate gives evidence that the double bond in the drimane skeleton (between C7 and C8) is a necessary structural feature. The molecular electrostatic potential was also found to be a feature. This work also confirmed the minimum fungicidal concentrations of many naturally occurring and semi-synthetic drimanes similar to polygodial. Polygodial showed the best minimum fungicidal concentration of all the compounds tested²⁰.



Toxicology and Mutagenicity

Toxicological assessment of Kolorex® Horopito & Aniseed capsules (175mg of each) indicates that this formula is not toxic following acute exposure up to the level of 2 grams per kg bodyweight. At this dosage satisfactory body weight gains were maintained and macroscopic examination of the abdominal and thoracic cavities revealed no abnormalities²¹.

Scientific Summary

In contrast to compounds of a similar structure with strong biological activity, polygodial has been shown to be nonmutagenic (Ames and V79/HGPRT assay), and exhibit the least cytotoxicity²¹.

Plants containing polygodial have been traditionally used as foods or medicines in Japan²², South America and Africa. There is no documented historical evidence of toxicity of Horopito by either oral ingestion or topical application.

Safety

Approximately one million Kolorex[®] capsules containing milled Horopito have been sold annually per year since 1997. Over this time Forest Herbs Research has documented fourteen adverse reactions. Half of these relate to nausea or vomiting and half to rashes. Well over a million Kolorex[®] softgel capsules containing Horopito extract are sold annually. Only three adverse reactions of transient nausea have been reported.

Kolorex[®] Intimate Care cream has been on the market for 20 years, during which seven incidents of adverse reactions have been reported. Two were severe allergic reactions. No adverse reactions have been reported for Kolorex[®] Foot & Toe Care cream.

Although there is no evidence of teratogenicity, as a precaution it is suggested that pregnant women and small children do not take the oral formulations.

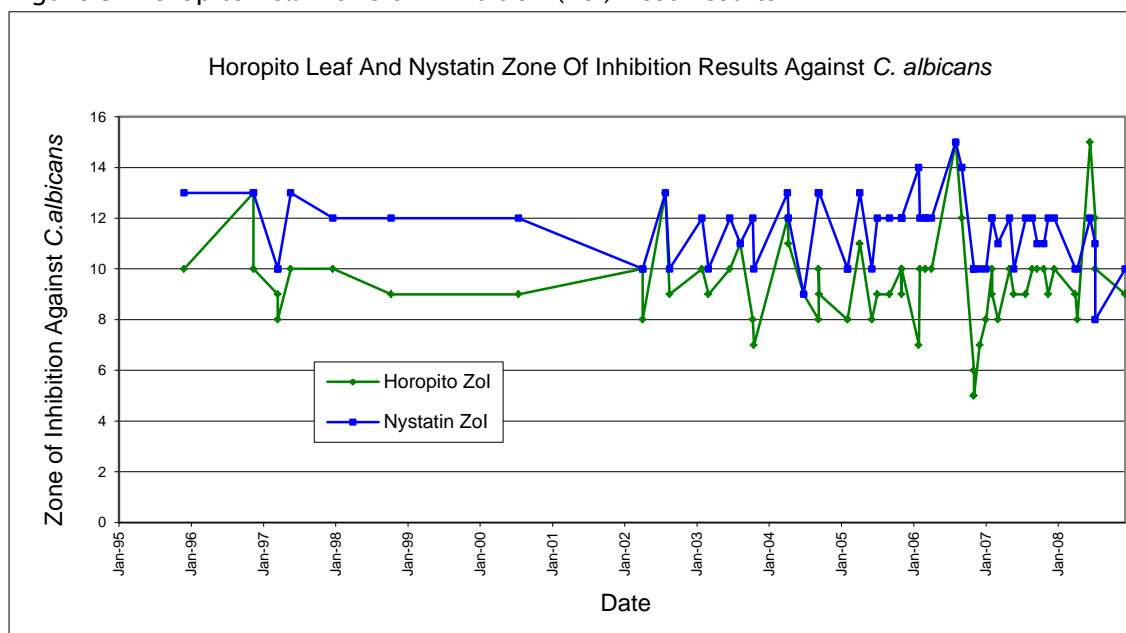
in vitro Efficacy

Kolorex[®] Horopito leaf

Each batch of Horopito leaves is independently assayed for antifungal activity against *Candida albicans*.

Historical leaf testing results for the inhibitory activity of Kolorex[®] Horopito leaf samples against *C. albicans* compared with the antifungal treatment drug Nystatin are shown in Figure 3.

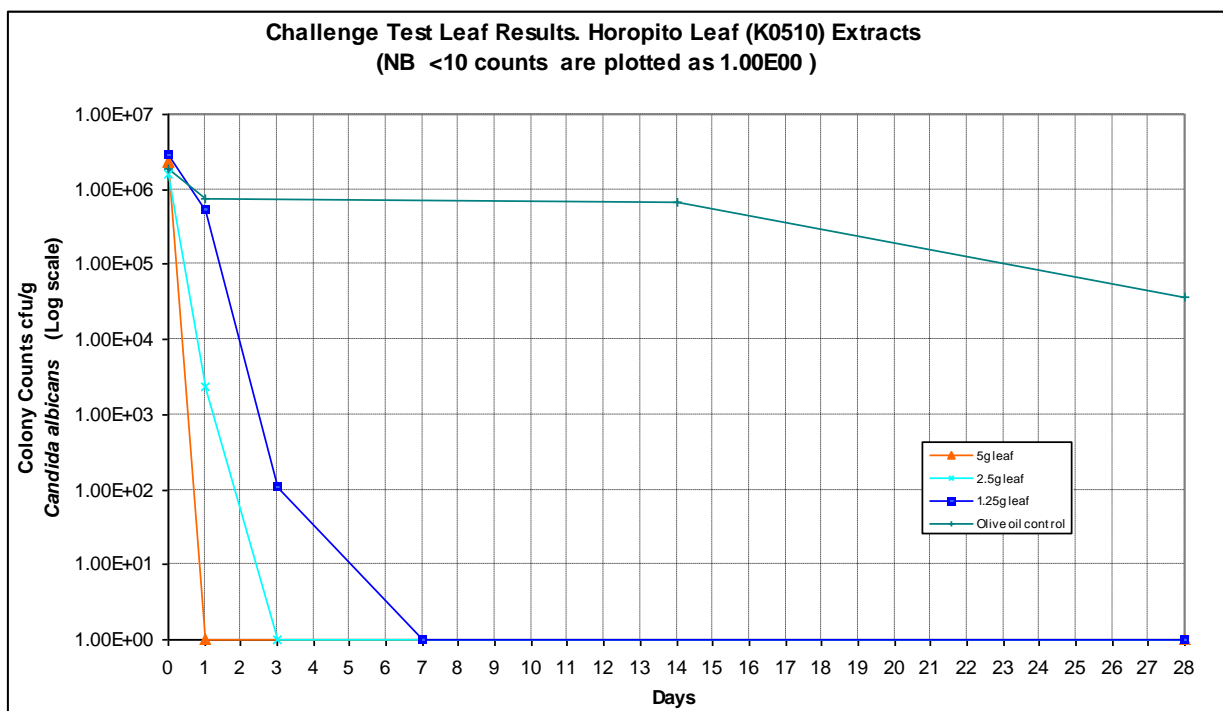
Figure 3. Horopito Leaf Zone of Inhibition (Zol) Test Results²³



Scientific Summary

Zone of Inhibition testing was stopped after the University of Canterbury discontinued its testing service. A new antifungal test based on the challenge test (European Pharmacopoeia General Text 5.1.3) was developed to monitor the antifungal capacity of Horopito leaf used in Kolorex® products. The antifungal efficacy of the infusion can be seen in Figure 4, where the *C. albicans* colonies are quickly reduced to zero²⁴.

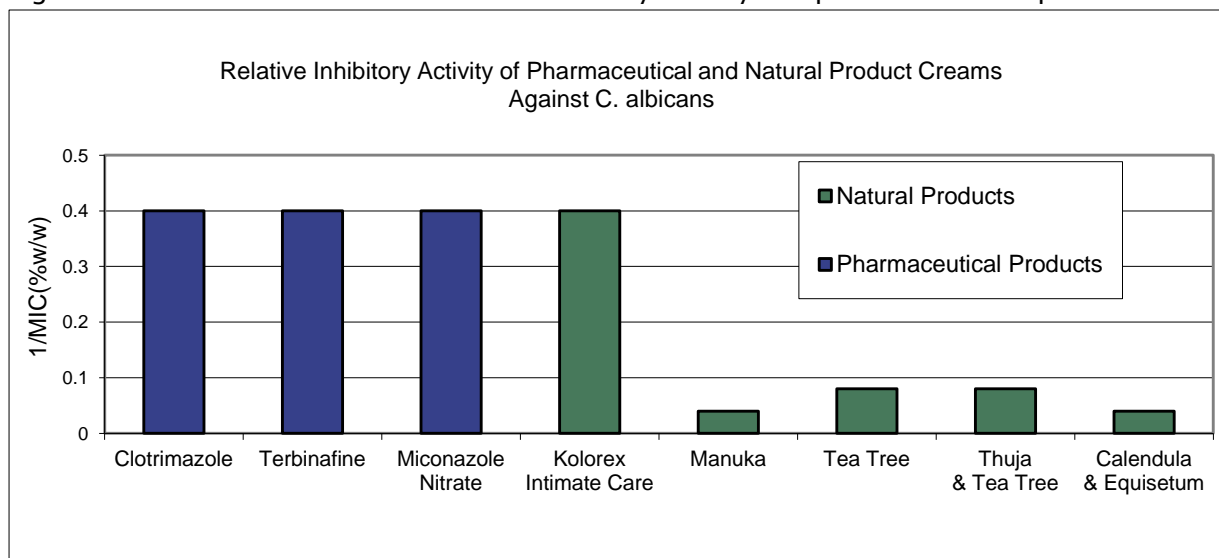
Figure 4. Challenge test results for various concentrations of Kolorex® Horopito Leaf Extracts



Kolorex® Intimate Care Cream

Kolorex® Intimate Care cream is more effective than other natural products and as effective as the pharmaceutical products tested (see Figure 5).

Figure 5. Kolorex® Intimate Care cream inhibitory activity compared with other products²⁵.

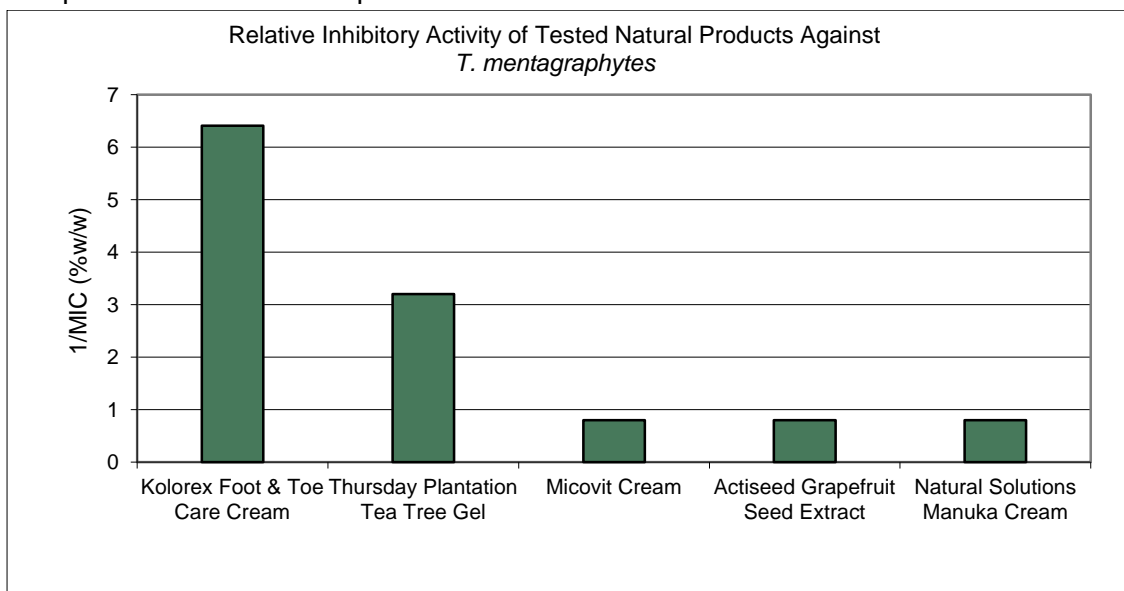


Scientific Summary

Kolorex® Foot & Toe Care Cream

The testing of Kolorex® Foot & Toe Care cream (See Figure 6 below) demonstrates that it has higher antifungal activity against one of the main athlete foot fungi (*T. mentagraphytes*), than any other natural product that was tested²⁶.

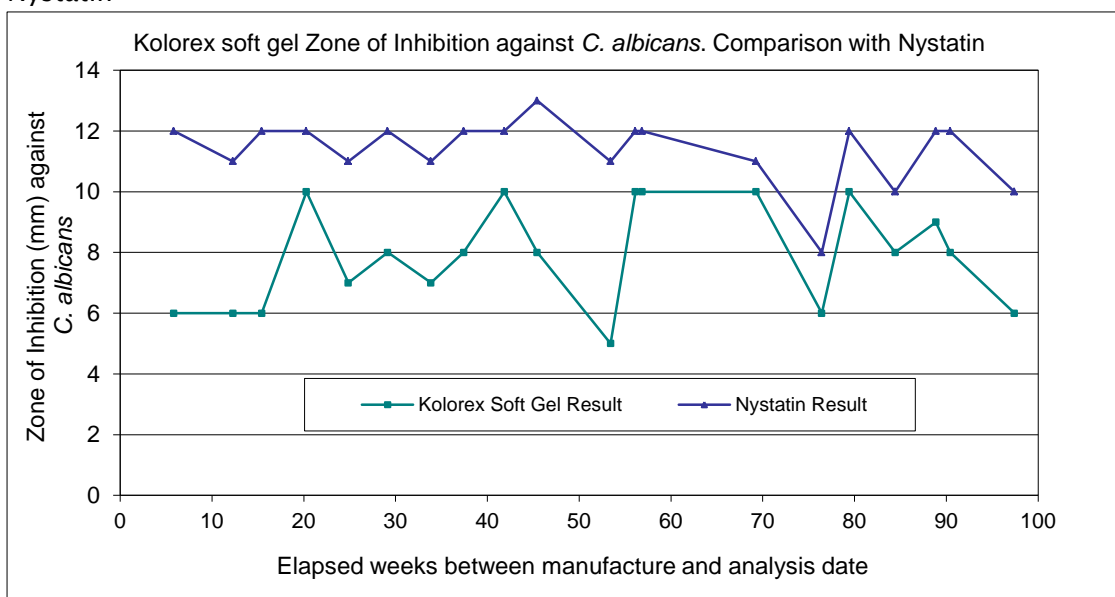
Figure 6. Relative inhibitory activity against *T. mentagraphytes* of Kolorex® Foot & Toe Care cream compared to other natural products tested.



Kolorex® Advanced Intestinal Care softgels

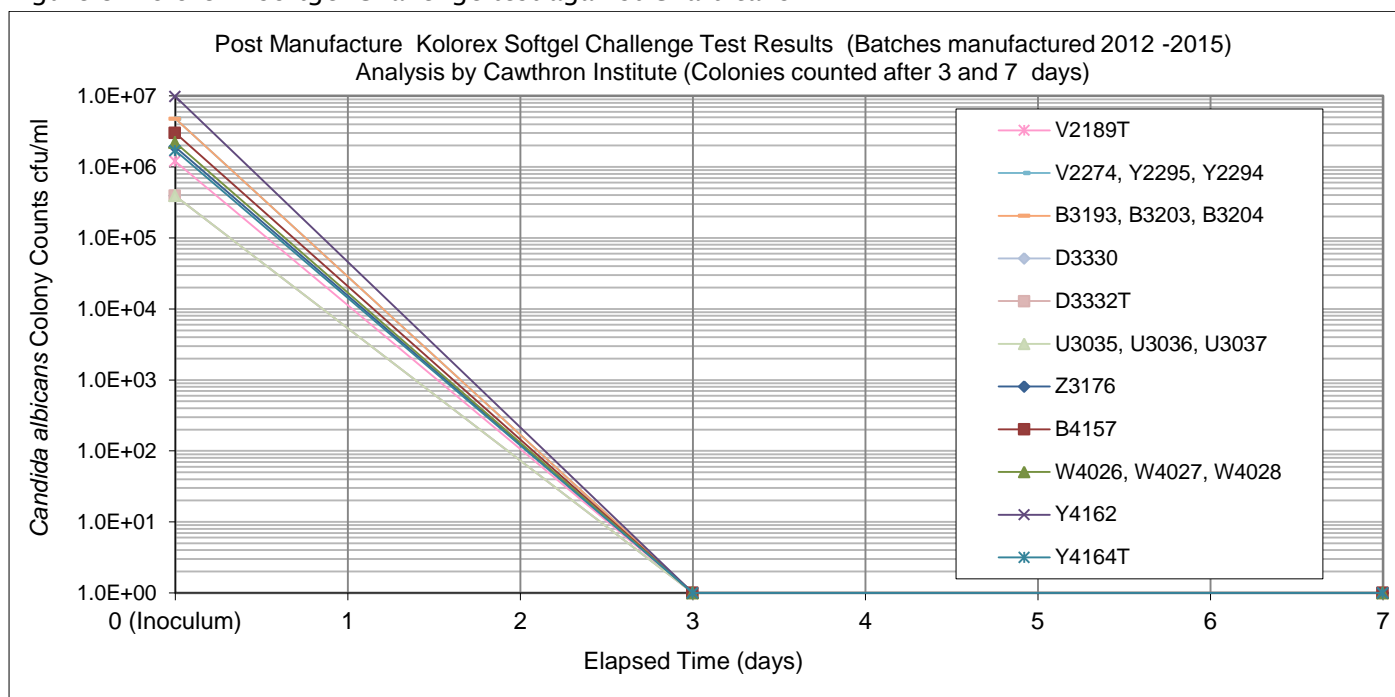
The first production batch of Kolorex® softgels was assayed for its effectiveness against *Candida albicans* on a monthly basis for almost two years, using the pharmaceutical anti-yeast product Nystatin as a control. See Figure 7. After 94 weeks the softgels are inhibiting the growth of *C. albicans* as effectively as when they were manufactured²⁷.

Figure 7. Kolorex® softgel Zone of Inhibition against *C. albicans* test results compared with Nystatin



Scientific Summary

Figure 8. Kolorex® softgel Challenge test against *C. albicans*



The Challenge test is used to test Kolorex® softgels against *Candida albicans*. An aliquot of oil removed from the softgel oil is inoculated with approximately one million colonies of *Candida albicans*. Figure 8 shows that the softgel oil rapidly and effectively destroys *Candida albicans* colonies.

Kolorex® Horopito & Aniseed Capsules

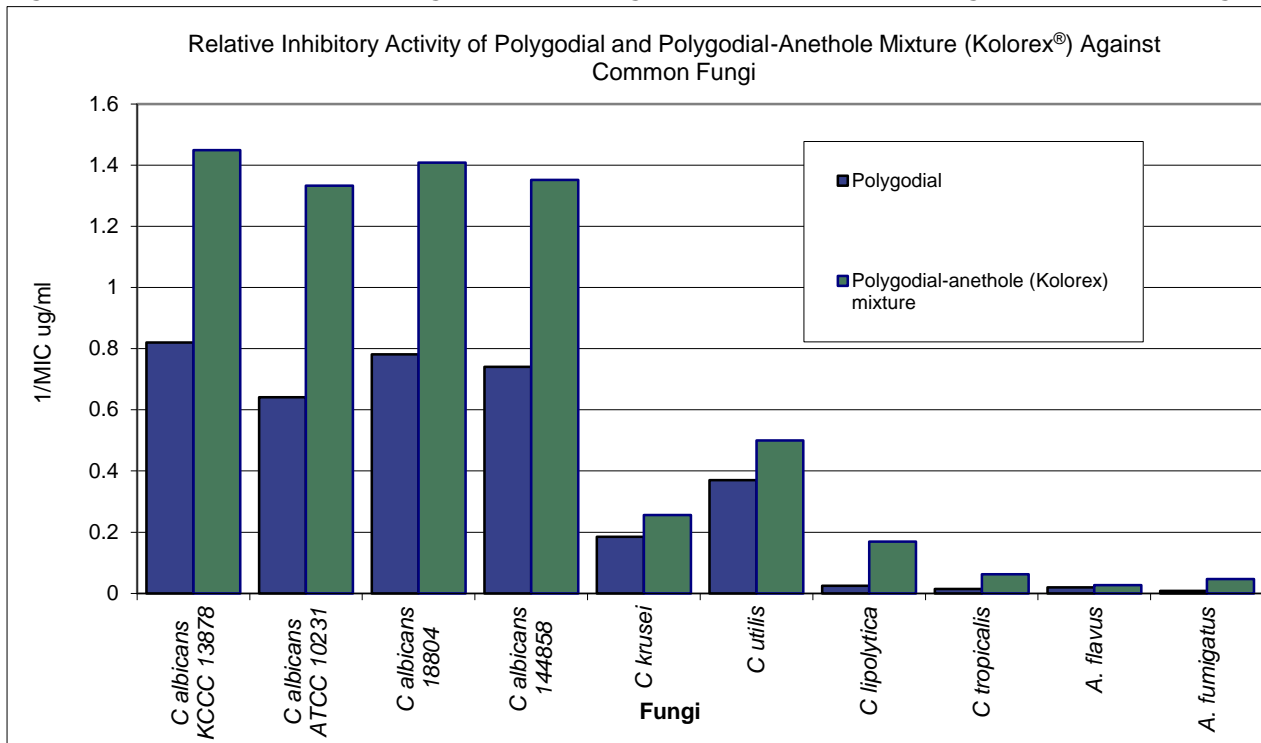
Kolorex® Horopito & Aniseed capsules exhibit a wide profile of antifungal activity. Published results of the testing of Kolorex® capsules (175mg horopito and 175mg aniseed) show that the remarkable *in vitro* antifungal activity of the Kolorex® mixture was better than polygodial alone (as illustrated in Figure 9). Furthermore results showed that there was no significant toxicity associated with Kolorex®. The most commonly used antibiotics however, do have the disadvantage of toxic side-effects. The study concluded that “Kolorex® is ... expected to be a promising compound for the development of therapeutic regimens acting through a synergistic effect”.²⁸



Photo: Kolorex® Horopito plants at Forest Herbs Research Organic Farm used for the manufacture of Kolorex® products

Scientific Summary

Figure 9. Relative activity of polygodial and polygodial-anethole mixture against common fungi²⁸



Recent research using a Horopito and aniseed mixture containing 50mg ground Horopito (*P. colorata*), 41.5mg Aniseed (*P. anisum*), 2.9mg *L. acidophilus* and 5.6mg vitamin C was tested on patients with operated gastrointestinal cancer. The patients had a history of ongoing candiduria (urinary *Candida* infection). It was found that a significant percentage developed urinary *Candida* infections following chemotherapeutic treatment. The Horopito formulation was used without any side effects and was successful in treating the infections in the majority of cases. The researchers recommend this natural antifungal phytochemical and think consideration should be given for its use as a prophylactic²⁹.

Note:

Forest Herbs Research considers Horopito the active ingredient in Horopito & Aniseed capsules. Aniseed is an excipient based primarily on its flavour and digestion aiding properties. If anethole from the aniseed exerts a synergistic effect this is an additional benefit but not relied upon in the formulation.

Kolorex® Digestive Care Tea

Results from research by Nakajima and colleagues³⁰ indicate that a Kolorex® product (containing a polygodial and anethole combination) applied to the oral cavity inhibits the growth of different *Candida* yeasts. The combination of Kolorex® Horopito leaf and aniseed used in Kolorex® Digestive Care Tea is designed for the cleansing of mucosal surfaces.

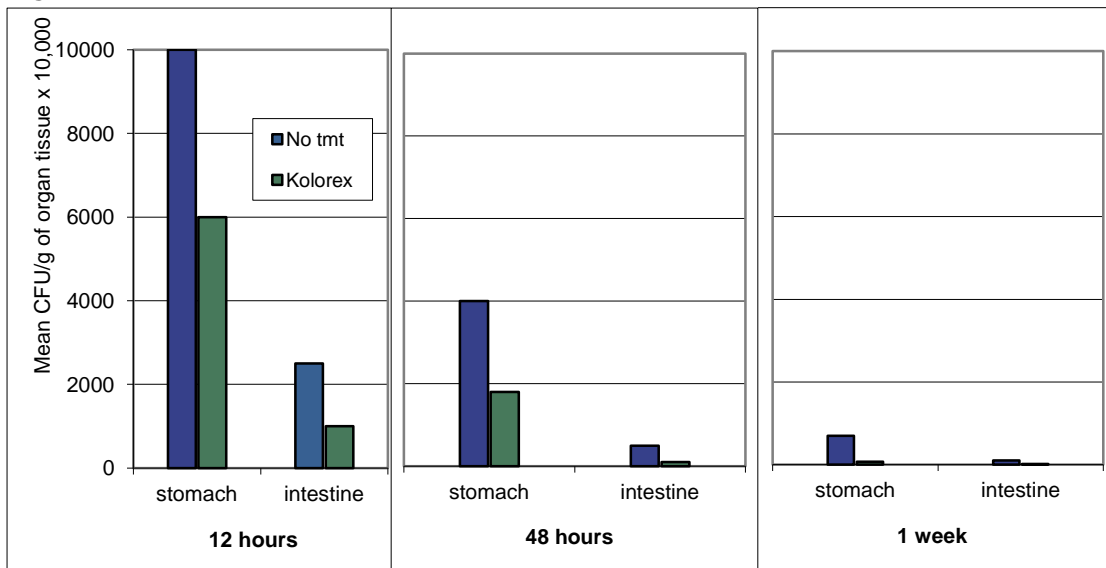
in vivo Studies

In 2001 Naito and his team³¹ investigated the ability of a Kolorex® product containing polygodial and anethole to protect the gut of mice from colonization and dissemination of *Candida albicans*.

Scientific Summary

After mice were inoculated with *C. albicans* and treated with Kolorex[®], testing of intestinal samples showed that Kolorex[®] treated mice had a much reduced concentration of *C. albicans* per gram of tissue (see Figure 10). The data suggested that the Kolorex[®] product might exert an early competitive effect against colonisation. Chemically synthesised antifungal treatments have been devised but they have the possible drawbacks of toxic effects and bacterial overgrowth. With the natural Kolorex[®] product no toxicity or bacterial dissemination occurred during the observation period.

Figure 10. *C. albicans* concentration in tissue tested: Effect of 48hr Kolorex[®] pre-treatment³¹



Further research by Marotta and associates³² in 2006 replicated the effect of Kolorex[®] on reducing the concentration of *C. albicans* in organs inoculated with the fungus. Their research was carried out using conditions of protein-calorie malnutrition often experienced by the elderly. It showed that Kolorex[®] treatment significantly decreased the absolute number of organs infected and enabled complete clearance of *C. albicans* in the lungs. The study concluded that Kolorex[®] exhibited potential clinical interest for specific conditions of calorie-protein malnutrition.

An *ex vivo* study by Nakajima³⁰ who used a Kolorex[®] Horopito and aniseed mixture to inhibit the growth of *C. albicans* in the oral cavity has already been mentioned in connection with Kolorex[®] Digestive Care Tea. This research concluded that in contrast to the commonly used oral antiseptics containing chlorhexidine, the antifungal action of Kolorex[®] was more constant against all species tested (including *C. albicans*, *C. tropicalis*, *C. glabrata*, *C. guilermanii*, *C. parapsilosis* and *C. krusei*) with a minimum inhibitory concentration of 1:20 of Kolorex[®] diluted with sterilised distilled water.

Stability Studies

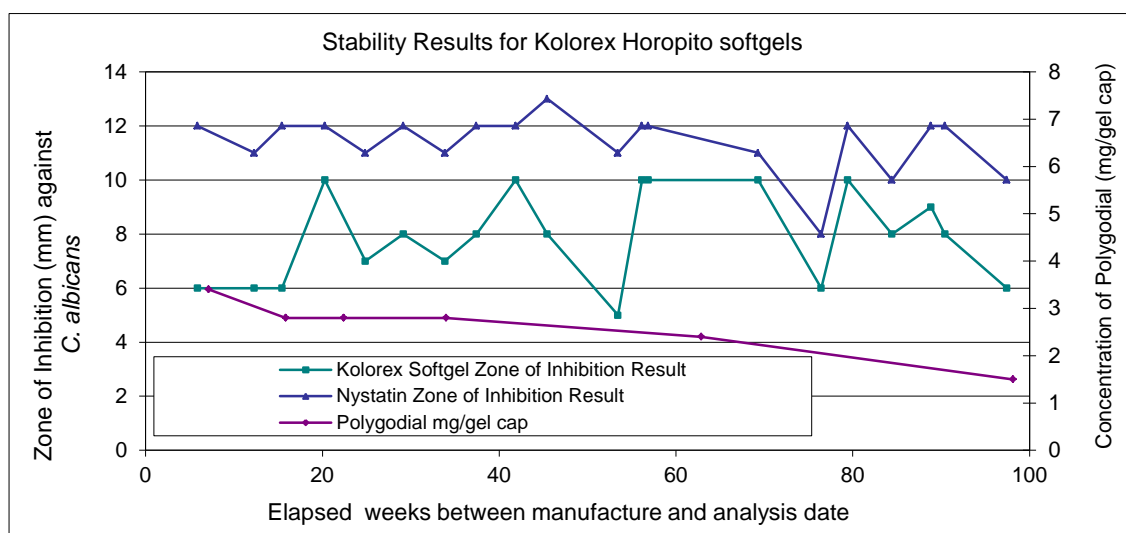
Kolorex[®] Horopito softgels

The softgels are as effective *in vitro* against *Candida albicans* at 94 weeks, as they were immediately after manufacture³³ (see zone of inhibition results Figure 11).

The stability of the active ingredient, polygodial, has been monitored under controlled storage conditions by Southern Cross University, Australia. The results are also graphed in Figure 11.

Scientific Summary

Figure 11. Kolorex® Advanced Intestinal Care 500mg softgel Capsule Stability Over 100 Weeks. Zone of Inhibition Results against *C. albicans* compared with Nystatin and Concentration of Polygodial (mg/gel cap)



Stability of the antifungal activity of Kolorex® softgels is confirmed by challenge testing of historical softgels stored at Forest Herbs Research. The results (see Figure 12) shows that even after two and a half years Kolorex® softgels continue to rapidly kill *Candida albicans* colonies (see Figure 12)

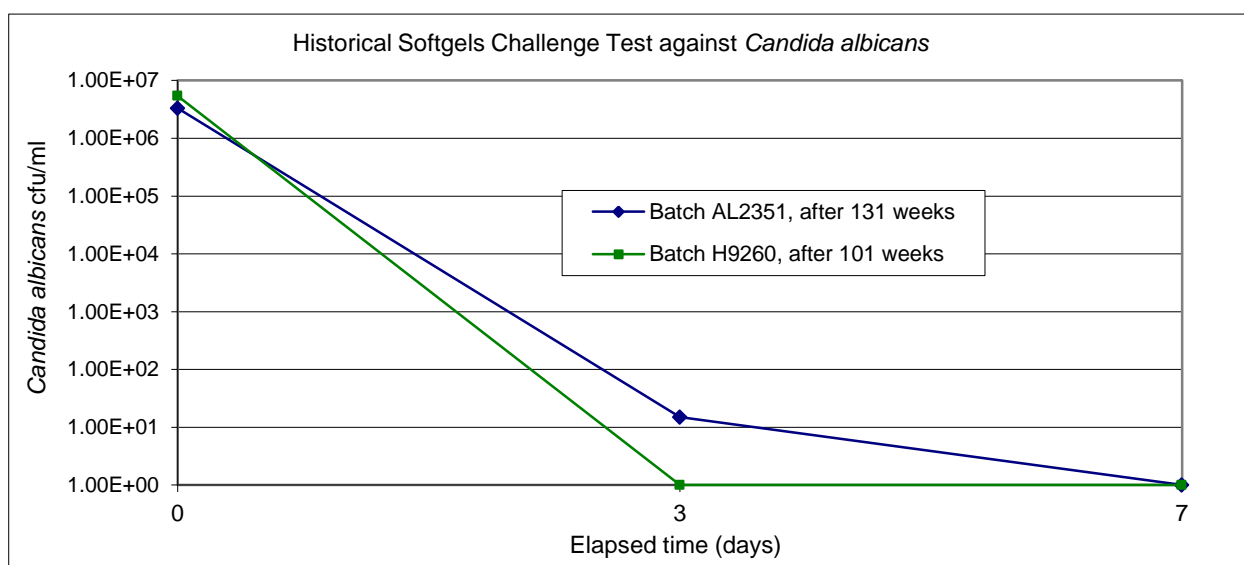


Figure 12. Antifungal activity of Kolorex® softgels analysed after 101 and 131 weeks

Clinical Studies

Kolorex® Herbal Capsules

An open study conducted by eleven NZ naturopaths in 1992 for Forest Herbs Research examined the therapeutic effect of capsules containing milled Kolorex® Horopito (300mg) in patients diagnosed with chronic intestinal candidiasis. This study demonstrated a symptom improvement rate in 76% of cases³⁴.

In 1997 the Pavlodar City Centre for Clinical Immunology and Reproduction carried out a study on patients diagnosed with chronic recurring intestinal candidiasis. It compared 22 patients taking capsules containing milled Kolorex® Horopito (300mg) with 10 patients administered fluconazole (Diflucan, Pfizer). All patients taking fluconazole and 90% of patients taking Kolorex® capsules showed a significant improvement after 7 and 14 days respectively³⁵.

Kolorex® Intimate Care Cream

A clinical study on the efficacy of Kolorex® Intimate Care cream against relapsing bacterial vaginosis (gardnerellosis) was conducted by the Pavlodar City Centre for Clinical Immunology and Reproduction in 2000. This vaginal infection is prone to relapses in more than 30% of treated cases. Twenty two women completed treatment for two months without any relapses³⁶.

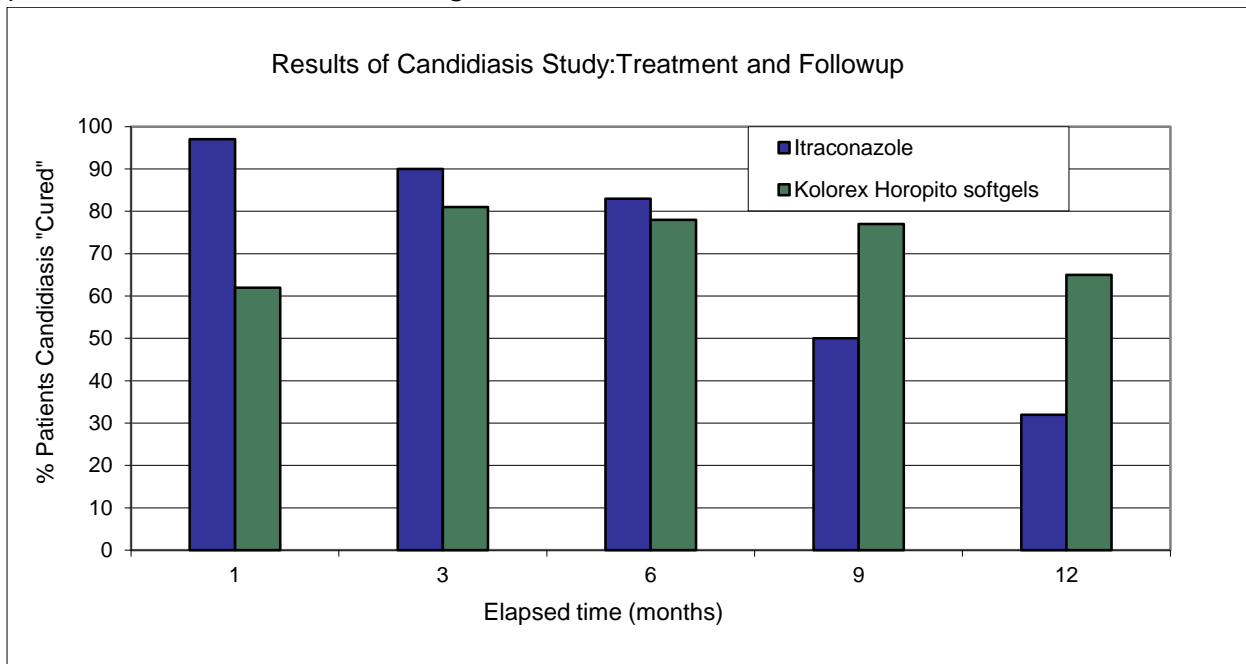
In 1995 a clinical study by NZ naturopaths was conducted on the efficacy of Kolorex® Intimate Care cream on twenty-six women suffering from vaginal candidiasis. Twenty-three of the women (89%) reported relief when the cream was applied 2 or 3 times daily for a week. Of the remaining three women, two reported a gradual increase in symptoms and 1 found the cream too hot to use a second time³⁷.

Kolorex® Horopito Softgels

In 2011 a randomised clinical study was carried out by Kumari³⁸ and colleagues. Eighty two women with recurrent vulvo-vaginal candidiasis underwent an oral treatment regime of either Kolorex® softgels or the antifungal pharmaceutical itraconazole for a period of 6 months. This was followed by observation for 6 months when no treatment was given. Despite itraconazole patients benefiting from earlier symptom relief, after 6 months the results were similar. After a total of 12 months (6 month treatment followed by 6 months observation) there were about twice as many Kolorex® treated patients who were free of *Candida* infection compared with itraconazole patients. (See Figure 13) This lower recurrence rate is a very positive outcome. Furthermore the study showed that the use of Kolorex® also reduces the growth of azole-resistant *Candida* species which are present in the itraconazole recurrent patients.

Scientific Summary

Figure 13. Number of mycologically “cured” patients during 12-month treatment and follow-up period (from Kumari et al 2011, Figure 2)



A further randomised 2 year clinical study of 122 women suffering from recurrent vulvo-vaginal candidiasis was carried out by the same research group and published in 2013³⁹. This time the study included 122 women and assessed the clinical efficacy of using Kolorex[®] as a maintenance treatment against recurrence. This was compared with treatment using itraconazole. Recurrent vulvo-vaginal candidiasis describes the condition when a woman experiences at least three episodes of candidiasis (that are unrelated to antibiotic use) in a year. Because of the recurrent nature this condition is very difficult to manage as although candidiasis usually responds to treatment, if there is no maintenance therapy a majority of women experience recurrence⁴⁰. For this study one group of patients were given maintenance therapy of Kolorex[®] softgels (one softgel twice a day for one week every month), while the other were given itraconazole (200mg orally once a week). The Kolorex[®] regime was well tolerated and had fewer side effects than itraconazole. Kolorex[®] softgels had significantly less relapses compared to itraconazole (22 compared with 39). The conclusion of the 2 year study was that Kolorex[®] softgels were equally effective as itraconazole for the overall treatment of recurrent vulvo-vaginal candidiasis. Furthermore Kolorex[®] softgels had a better preventative effect and lower relapse rate as well as less susceptibility and growth of azole-resistant species.

Scientific Summary

Kolorex® Products

The leading products that Forest Herbs Research Ltd has developed from Kolorex® Horopito are:



Kolorex® Advanced Candia/Candida Care softgels for intestinal dysfunction and Candidiasis



Kolorex® Foot and Toe Care cream for athlete's foot and nail fungi



Kolorex® Intimate Care cream for discomfort in intimate areas and vaginal thrush



Kolorex® Digestive Care tea for oral thrush, mouth hygiene and as a digestion aid

References

- ¹ Webb, C., Johnson, P., & Sykes, B. (1990). Flowering Plants of NZ, *DSIR*, p104.
- ² Brockie, B. 'Native plants and animals – overview – Unusual plant features', *Te Ara – the Encyclopedia of New Zealand*, updated 1-Mar-09
URL: <http://www.TeAra.govt.nz/en/native-plants-and-animals-overview/7> (Accessed 09/08/2011)
- ³ Orwin, J. '*Shrubs and small trees of the forest – Pepper trees: horopito and kawakawa*', *Te Ara – the Encyclopedia of New Zealand*, updated 1-Mar-09
URL: <http://www.TeAra.govt.nz/en/shrubs-and-small-trees-of-the-forest/12> (Accessed 09/08/2011)
- ⁴ Brooker, S. G., Cambie, R. C., & Cooper, R. C. (1987). New Zealand Medicinal Plants, *Heinemann*, p240.
- ⁵ Riley, M. (1994). Maori Healing and Herbal, *Viking Sevenses*, pp146–148.
- ⁶ Perry, N. 'Plant extracts – Medicines', *Te Ara – the Encyclopedia of New Zealand*, updated 2-Mar-09
URL: <http://www.TeAra.govt.nz/en/plant-extracts/3> (Accessed 09/08/2011)
- ⁷ McCallion, R. F., Cole, A. L., Walker, J. R., Blunt, J. W., & Munro, M. H. (1982). Antibiotic Substances from New Zealand Plants, *Planta Medica*, *44*(3), 134–138.
- ⁸ Gershenzon, J., & Dudareva, N. (2007). The Function of Terpene Natural Products in the Natural World. *Nature Chemical Biology*, *3*(7), 408–414.
- ⁹ Larsen, L. A. (2001). Literature Survey of the Constituents of *Pseudowintera colorata*. *NZ Institute for Crop and Food Research* (FHR on File).
- ¹⁰ Mylek, M. (1999). Development and Analysis of Extracts of Polygodial from Horopito, *Industrial Research Ltd*, Report No.904.
- ¹¹ Cooney, L. J., van Klink, J. W., Hughes, N. M., Perry, N. B., Schaefer, H. M., Menzies, I. J., & Gould, K. S. (2012). Red Leaf Margins Indicate Increased Polygodial Content and Function as Visual Signals to Reduce Herbivory in *Pseudowintera colorata*. *New Phytologist*, *194*: 488–497. doi: 10.1111/j.1469-8137.2012.04063.x
- ¹² Lee, S. H., Lee, J. R., Lunde, C. S., & Kubo, I. (1999). In Vitro Antifungal Susceptibilities of *Candida albicans* and other Fungal Pathogens to Polygodial, a Sesquiterpene Dialdehyde, *Planta Medica*, *65*, 205–208.
- ¹³ Kubo, I., & Lee, S. H. (1998). Potentiation of Antifungal Activity of Sorbic Acid. *J. Agric. Food Chem.*, *46*, 4052–4055.
- ¹⁴ Kubo, I., Lee, S. H., & Ha, T. J. (2005). Effect of EDTA Alone and in Combination with Polygodial on the Growth of *Saccharomyces cerevisiae*. *J. Agric. Food Chem.*, *53*(5), 1818–1822.
- ¹⁵ Fujita, K., & Kubo, I. (2005). Naturally Occurring Antifungal Agents Against *Zygosaccharomyces bailii* and Their Synergism, *J. Agric. Food Chem.*, *53*, 5187–5191.
- ¹⁶ Kubo, I., Fujita, K., Lee, S. H., & Ha, T. J. (2005). Antibacterial Activity of Polygodial, *Phytotherapy Research*, *19*, 1013–1017.
- ¹⁷ Taniguchi, M., Yano, Y., Tada, E., Ikenishi, K., Oi, S., Haraguchi, H., Hashimoto, K. & Kubo, I. (1988). Mode of Action of Polygodial, an Antifungal Sesquiterpene Dialdehyde. *Agricultural and Biological Chemistry*, *52*(61), 1409–1414.

Scientific Summary

- ¹⁸ Forsby, A., Walum, E., & Sterner, O. (1992). The Effect of Six Sesquiterpenoid Unsaturated Dialdehydes on Cell Membrane Permeability in Human Neuroblastoma SH-SY5Y cells. *Chemico-Biological Interactions*, *84* (1), 85–95.
- ¹⁹ Kubo, I., Fujita, K., & Lee, S. H. (2001). Antifungal Mechanism of Polygodial. *Journal of Agricultural and Food Chemistry*, *49*(3), 1607–1611.
- ²⁰ Derita, M., Montenegro, I., Garibotto, F., Enriz, R. D., Fritis, M. C., & Zacchino, S. A. (2013). Structural Requirements for the Antifungal Activities of Natural Drimane Sesquiterpenes and Analogues, Supported by Conformational and Electronic Studies. *Molecules*, *18*(2), 2029–2051. doi:10.3390/molecules18022029.
- ²¹ Winkelman, E. (2008). Toxicological Assessment of Horopito (*Pseudowintera colorata*). GlaxoSmithKline Memorandum.
- ²² Jansen, B.J. & de Groot, A. (2004). Occurrence, Biological Activity and Synthesis of Drimane Sesquiterpenoids. *Natural Product Reports*, *21*(4), 449–477.
- ²³ Forest Herbs Research. Horopito Leaf Zone of Inhibition against *C. Albicans* Data on File.
- ²⁴ Forest Herbs Research. Horopito Challenge Test Results. Cawthron Institute Report Reference S50250–1.
- ²⁵ Forest Herbs Research. Kolorex Intimate Care Cream MIC Data on File.
- ²⁶ Forest Herbs Research. Kolorex Foot and Toe Cream MIC Data on File.
- ²⁷ Forest Herbs Research. Kolorex Horopito Softgel Zone of Inhibition Data on File.
- ²⁸ Metugriachuk, Y., Kuroi, O., Pavasuthipaisit, K., Tsuchiya, J., Minelli, E., Okura, R., Fesce, E., & Marotta, F. (2005). In View of an Optimal Gut Antifungal Therapeutic Strategy: and in vitro Susceptibility and Toxicity Study Testing a Novel Phyto-compound. *Chinese Journal of Digestive Diseases*, *(6)*, 98–103.
- ²⁹ Marotta, F., Naito, Y., Bishier, M. P., Jain, S., Yadav, H., Minelli, E., Kumari, A., Solimene, U., & Sollano, J. (2010). Subclinical candiduria in patients with gastrointestinal malignancies: a preliminary study on the protective effect of a natural phytocompound. *Journal of Biological Regulators and Homeostatic Agents*, *24*(3), 317–324.
- ³⁰ Nakajima, J., Papaah, P., Yoshizawa, M., Marotta, F., Nakajima, T., Mihara, S., & Minelli, E. (2007). Effect of a novel phyto-compound on mucosal candidiasis: Further evidence from an ex vivo study. *Journal of Digestive Diseases*, *8*, 48–51.
- ³¹ Naito, Y., Wu, C. C., Seal, M. G., Gelosa, F., Yoshioka, M., Safran, P., & Marotta, F. (2001). Protective Effect of a Polygodial/Anethole-Containing Natural Product against *Candida albicans* Gastrointestinal Colonization and Dissemination. *International Medical Journal*, *8*(1), 3–9.
- ³² Marotta, F., Barreto, R., Kawakita, S., Minelli, E., Pavasuthipaisit, K., Lorenzetti, A., Nishiwaki, M., Gelosa, F., Fesce, E., & Okura, R. (2006). Preventative Strategy for *Candida* Gut Translocation During Ischemia-Reperfusion Injury Supervening on Protein-Calorie Malnutrition. *Chinese Journal of Digestive Diseases*, *7*, 33–38.
- ³³ Forest Herbs Research. Soft Gel Stability Trial Data on File.
- ³⁴ 1992 NZ Naturopath Study. Data on File, Forest Herbs Research Limited, Nelson, New Zealand.
- ³⁵ 1997 Pavlodar study. Pavlodar City Centre for Clinical Immunology and Reproduction. Head Physician: O. Ogorodnikova. Pavlodar. Data on File, Postgraduate Physicians' Training Faculty Assistant: M. Valivach.
- ³⁶ Forest Herbs Research. Kolorex Cream 2000 Pavlodar Clinical Study Data on File.

Scientific Summary

- ³⁷ Forest Herbs Research. Kolorex Cream 1995 Clinical Study Data on File. Training Faculty Assistant: M. Valivach.
- ³⁸ Kumari, A., Bishier, M. P., Naito, Y., Sharma, A., Solimene, U., Jain, S., Yadava, H., Minelli, E., Momella, C., & Marotta, F. (2011). Protective effect of an oral natural phytonutrient in recurrent vulvovaginal candidiasis: a 12-month study. *Journal of Biological Regulators and Homeostatic Agents*, 25 (4), 543-551.
- ³⁹ Chopra V., Marotta F., Kumari A., Bishier M.P., He F., Zerbinati N., Agarwal C., Naito Y., Tomella C., Sharma A., Solimene U. (2013) Prophylactic strategies in recurrent vulvovaginal candidiasis: a 2-year study testing a phytonutrient vs itraconazole. *Journal of Biological Regulators and Homeostatic Agents*, 27(3):875-82.
- ⁴⁰ Sobel, J. D., Wiesenfeld, H. C., Martens, M., Danna, P., Hooton, T. M., Rompalo, A., Sperling, M., Livengood, C., Horowitz, B., von Thron, J., Edwards, L. Panzer, H., Chu, T.-C. (2004). Maintenance fluconazole therapy for recurrent vulvovaginal candidiasis. *The New England Journal of Medicine*, 351(9), 876-883.