Technical Report

Significance of Grapefruit Seed Extract in CanXida Remove (Formula RMV)

45% standardized grapefruit seed extract in CanXida Remove (Formula RMV) provides a full-spectrum antimicrobial effect against a wide range of bacteria and fungi. It is also rich in antioxidants and bioflavonoids which relieve inflammation and promote wound healing.

Contents

Executive Summary

Grapefruit seed extract is a full-spectrum natural antibacterial for oral use as a dietary supplement or as a natural food additive. It is non-toxic to human cells and is classified as a GRAS substance. It is effective against a wide range of bacterial and fungal species. Among bacteria, it is effective against foodborne pathogens such as Shigella, *Klebsiella, Salmonella, and Staphylococcus* species while *Candida albicans* and its related pathogenic strains are its target among fungal pathogens of the gastrointestinal tract. It is also rich in its signature bioflavonoids and sterols which act as antioxidants and anti-inflammatory*.

CanXida Remove (Formula RMV) contains 45% standardized grapefruit seed extract optimized for bacterial and fungal pathogens of the gut including Candida, E.coli, and other foodborne pathogens. It retains its antimicrobial activity even at lower concentrations. The grapefruit seed extract in the CanXida Remove formulation also provides relief from inflammatory stresses and promotes wound healing in the gut that is constantly insulted by pathogenic microbes of bacterial and fungal origin.

** These statements have not been evaluated by Food and Drug Administration. This product is not intended to diagnose, treat, cure or prevent any disease.*

1. Introduction

Grapefruit seed extract is extracted from grapefruit seeds, white membranes, and non-juicy pulp. It is extremely effective as an antimicrobial with proven antibacterial, antiviral, and antifungal effects. It is also rich in flavonoids, contributing to its antioxidant and anti-inflammatory properties. Other biologically significant components in grapefruit seed extract include ascorbic acid (vitamin C), tocopherols, sterols, citric acid, limonoids, and minerals.

Grapefruit seed extract has been shown to positively regulate microbial flora in the gut by limiting the activity of pathogenic bacteria and fungi (Heggers et al 2002; Reagor et al 2002). It is an essential part of naturopathic remedies and food supplements in most of the commercially available products. It is also included in the food product to discourage the growth of foodborne pathogens. It effectively inhibits the growth of bacterial pathogens including *Listeria, Clostridium, E. coli, and Salmonella* at concentrations as low as 0.2%. Among fungal pathogens, *Candida albicans* is one of the strongly affected yeast.

As consumers are becoming increasingly inclined to natural antimicrobials, grapefruit seed extract is widely accepted as an effective antimicrobial. It is classified as GRAS (Generally Recognized as Safe) by the FDA and the WHO (CFR 180.20). It is commonly added to food products to limit the activity of bacteria and fungi. Pure grapefruit seed extract is non-toxic, and it is safe to use as a supplementary food product or as an antimicrobial agent (Roy et al 2023). Its extraction methods are environmentally friendly and simple, producing a chemical-free food-grade pure product. Owing to its origin from citrus fruits, it is rich in

Grapefruit Seed Extract

Extract Type:	Purified Natural Substances
Composition:	Bioflavonoids, Polyphenols, Polysterols, Tocopherols, minerals.
Bioactivity:	Antimicrobial, Antiviral, Antioxidant
Source:	Plant-based, Citrus Fruits
FDA Status:	Approved
CFR category:	GRAS
Toxicity:	Non-toxic
Classification:	Food Additive, Food Supplement, Natural Antimicrobial
CFR Code:	180.20

antioxidants such as ascorbic acid, flavonoids, and carotenoids. The antioxidants relieve inflammatory stresses and accelerate the healing process.

2. Bioactive Compounds in Grapefruit Seed Extract

Grapefruit seed extract is obtained from the seeds and non-juicy pulp (white membranes, peels) of grapefruits. Most of the extract from seed contains essential oils, fats, and fat-soluble bioactive compounds, ranging up to 40%. Among fats are fatty acids such as oleic acid (20 – 25%), palmitic acid (23 – 25%), stearic acid (5–10%) and linoleic acid (nearly 40%). Among the fat-soluble portions are carotenoids, tocopherols, and polysterols. Tocopherol concentration in the grapefruit seed extract is around 380 mg/kg. Tocopherols act as antimicrobials as well as antioxidants.

Significant amounts of phenolic compounds are also present in the grapefruit seed extract. Prominent phenolic acids in the grapefruit seed extract include trans-ferulic acid, trans-2-hydroxycinnamic acid, and rosmarinic acid. These compounds have potent antibacterial properties. Some extraction methods and extract maturation processes also produce secondary phenolic compounds that also contribute to the antimicrobial profile of grapefruit seed extract (Rashid et al 2013). Prominent phenols and flavonoids in the grapefruit seed extract include gallic acid, rhoifolin, ponicirin, and taxifolin.

Limonoids, the characteristics of oxygenated terpenoids of citrus fruits, are also present in the grapefruit seed extract. These compounds are often indicators of the purity of the commercially available extracts. Furthermore, grapefruit seed extract also contains a special class of coumarins called furanocoumarins, such as bergamotin, epoxybergamotin, and dihydroxybergamotin. Coumarins are medically important compounds with a wide range of physiological benefits (Hung et al 2017). Trace amounts of quinones are also present. Quinones have anti-protozoal properties.

The amount of different bioactive compounds in the grapefruit seed extract is given below in the Table.

Table 1: *Major classes of bioactive compounds found in the grapefruit seed extract.*

Bioactive Compound	Amount in Grapefruit Seed Extract
Flavonoids	29.79%
Phenolic Acids	14.1%
Alkaloids	8.91%
Lipids	8.64%
Organic Acids	8.24%
Amino Acids & Derivatives	7.85%
Lignans & Coumarins	6.65%
Saccharides	4.39%
Nucleotides and Derivatives	3.56%
Terpenoids	1.6%
Vitamins	1.06%
Quinones	0.93%
Others	3.9%

3. Health Benefits of Grapefruit Seed Extract

Grapefruit seed extract has a well-established antibacterial, antifungal, and antimicrobial profile. It also provides anti-inflammatory support to deal with chronic health conditions of the digestive tract such as ulcers, inflammation, bacterial and yeast overgrowth, and foodborne ailments. Early reports on the antimicrobial efficacy of the grapefruit seed extract suggest that it is effective against 794 bacterial strain types and 93 fungal strain types (Ionescu et al 1990). Tables 2 and 3 show the effect of grapefruit seed extract on major pathogenic fungi and bacteria.

3.1. Antibacterial and Antifungal Effects

Due to increasing strains of multi-drug resistance bacteria, natural remedies like grapefruit extract are getting a renewed interest among researchers and clinicians. Grapefruit seed extract has shown a growth inhibitory effect on the bacterial colonies in in vitro studies.

An open-label clinical trial conducted by Ionescu and colleagues showed that grapefruit seed extract was effective against intestinal pathogens such as *Candida, hemolytic E. coli,* and *Geotrichum* without affecting the good probiotic bacterial species of *Bifidobacterium* and *Klebsiella.* All patients showed improved gut health without any side effects.

Bioflavonoid naringin and hesperidin are major contributors to the antibacterial activity. In 2004, Cvetnić and Vladimir-Knezevic published a research report in Acta Pharmaceutica showing that grapefruit seed extract was effective against 20 strains of bacteria and 10 strains of pathogenic yeasts (Cvetnic & Vladimir-Knezevic 2004).

Table 2: *Grapefruit seed extract and its effect on the growth of different candida species and pathogenic fungal strains. (Cvetnic & Vladimir-Knezevic 2004)*

Fungi	**Minimum Inhibitory Concentration of Grapefruit Seed Extract (% m/v)**				
	16.50	**8.25**	**4.13**	**2.06**	**1.03**
Candida albicans	-	+	+	+	+
Candida. krusei	-	-	+	+	+
Candida tropicalis	-	+	+	+	+
Candida parapsilosis	-	+	+	+	+
Saccharomyces cerevisiae	-	-	+	+	+
Kluyveromyces maxianus	-	+	+	+	+

Table 3: *Grapefruit seed extract and its effect on the growth of different bacterial pathogens of the gastrointestinal tract. (Cvetnic & Vladimir-Knezevic 2004)*

Bacteria	**Minimum Inhibitory Concentration of Grapefruit Seed Extract (% m/v)**				
	16.50	**8.25**	**4.13**	**2.06**	**1.03**
Bacillus cereus	-	±	+	+	+
Bacillus subtilis	-	±	+	+	+
Sarcina flava	-	±	+	+	+
Sarcina lutea	-	-	+	+	+
Staphylococcus aureus	-	-	+	+	+
Staphylococcus epidermidis	-	-	+	+	+
Streptococcus faecalis	-	-	-	+	+
Streptococcus sp.	-	-	-	+	+
Listeria monocytogenes	-	-	-	+	+
Escherichia coli	-	±	±	+	+
Shigella sonnei	-	-	+	+	+
Salmonella enteritidis	-	-	-	-	+
Yersinia enterocolitica	-	-	+	+	+
Citrobacter freundi	-	+	+	+	+
Klebsiella oxytoca	-	-	+	+	+
Proteus mirabilis	-	+	+	+	+
Proteus vulgaris	-	+	+	+	+
Pseudomonas aeruginosa	-	-	+	+	+

The grapefruit antimicrobial activity was effective from 4.13% to 16.5% for all pathogenic species of bacteria and fungi. Most of the bacteria studied were Gram-positive bacteria. Similar results were reproduced in the latest study by Faleye and colleagues (Faleye et al 2012). The grapefruit seed extract was effective against *Proteus vulgaris*, *Staphylococcus aureus*, *Candida albicans,* and *Salmonella enteritidis.* The purified extract was effective in quantities as low as 0.1% to inhibit the growth of pathogenic microorganisms*.

Another study conducted on 200 different strains of pathogenic yeast and fungi, including *Candida albicans*, showed that a 33% concentration of grapefruit seed extract was effective against different types of pathogenic fungi (Krajewska-Kułak et al 2001). Similar results were reported elsewhere showing that grapefruit seed extract was more potent than similar types of antifungal agents and extracts (Ignaci & Thai, 2005). The extract was effective in arresting the growth of candida yeast in the range of 100 – 120 µg/mL concentration*.

In a recent study published in the journal Antibiotics, Han and colleagues demonstrated that grapefruit seed extract demonstrated a strong inhibitory effect for multidrug-resistant bacteria (Han et al. 2021). The antibacterial activity was present even at a very low dose. The antimicrobial activity of Grapefruit seed extract is due to its effects on bacterial and fungal cell membranes due to its diverse group of bioactive compounds.

It can be observed from Tables 2 and 3 that grapefruit seed extract has a growth inhibitory effect at concentrations of 16.5% and above showing bacteriostatic and fungistatic effects. However, concentrations in the range of 33% or above are required for the effective elimination of pathogens. Canxida Remove (Formula RMV) contains 45% standardized grapefruit seed extract for optimal results.

3.2. Anti-Inflammatory Effect

Plants are rich in antioxidant and anti-inflammatory flavonoids in general, but some flavonoids are only found in grapefruits and citrus plants. Grapefruit seed extract is rich in bioflavonoids that are well known for their anti-inflammatory properties. Some of the prominent flavonoids are hesperidin, neo-hesperidin, naringin, and narirutin. Chronic infections and imbalance in the intestinal microbiome often leave gut walls with inflammation and ulcers, which act as niches for the growth and proliferation of pathogenic microbes. As the site of inflammation is constantly under attack from the immune system as well as from the pathogens, it does not find enough time for healing and regeneration.

Grapefruit seed extract is rich in antioxidants and anti-inflammatory bioactive compounds such as ascorbic acid, fumaric acid, omega-6 fatty acids, and flavonoids which help in fighting inflammation and promoting the healing process for regeneration. These substances in combination with antimicrobial bioactive compounds provide a synergistic effect for wound healing for ulcers present in the small intestine by eliminating pathogen colonies and reducing inflammation*.

Inflammation is often triggered by the innate immune system which attacks the site of infection indiscriminately, damaging its own cells along with pathogenic microbes. It results in weakened cells and swollen blood

vessels with insufficient adaptive immune response. Reduction in inflammation promotes adaptive immune cells (B and T cells) which damage only pathogenic cells with higher accuracy and efficiency. Grapefruit seed extract reduces inflammation and weakens the pathogenicity of microbes allowing full-spectrum immune clearance for a complete and long-term healing process.

4. Biosafety Profile

Table 4: *List of antioxidant bioactive compounds (sterols and flavonoids)which are generally found only in citrus plants.*

Bioactive Antioxidant	Quantity (mg/100g)
Flavonoid	
Hesperidin	6
Hesperitin	0.4
Narirutin	90
Natsudaidain	–
Neohesperidin	14
Neoponcirin	–
Nobiletin	1.6
Poncirin	10.6
Rhoifolin	4.7
Rutin	–
Taxifolin	7.8
Gallic acid	6.5
Phytosterol	
Campesterol	3.6
Didymin	7.5
Diosmin	0.3
Eeodiosmin	0.5
Neoeriocitrin	0.2
Eriocitrin	0.4

Grapefruit seed extract is classified as a GRAS (generally recognized as safe) substance by the FDA and WHO. The extraction process for the extract is simple and chemical-free producing a non-toxic and all-natural product. It is being used in several dietary supplements and nutraceutical labels that are being sold either as a pure grapefruit seed extract or in combination with other products. FDA also approves grapefruit seed extract to control foodborne pathogens and as an edible antimicrobial.

Grapefruit seed extract can arrest bacterial and fungal growth in very low quantities with no side effects at all. Heggers and colleagues from the School of Medicine, University of Texas Medical Branch, Galveston, USA, demonstrated that grapefruit seed extract is an effective antibacterial at very diluted concentrations (1: 512) and ability to kill bacteria within 15 minutes of exposure to grapefruit seed extract (Heggers, et al 2002). This shows the high specificity of grapefruit seed extract for pathogens. They also observed that the concentration of grapefruit seed extract which can kill bacteria was completely harmless to human cells upon incubation.

Grapefruit seed extract kills bacteria and fungi by disrupting the cell membranes of these pathogens which makes it a rapid-action antimicrobial. It also explains its non-toxic behavior to human cells because the cell membranes of bacteria and fungi are different from the human cells. Furthermore, its rapid-action antimicrobial effect does not require its longer circulatory time in the organs and blood, giving it a short exposure time for body organs which further reduces any negative effect to a minimum.

5. Effective Targets

Grapefruit seed extract is a broad-acting antimicrobial which is effective against bacterial and fungal pathogens alike. Multiple research reports testify that grapefruit seed extract is effective in over 794 bacterial strains and over 93 fungal strains. Some of the major pathogenic strains of the gastrointestinal tract are given below.

Candida Yeast: *Candida albicans, Candida. Krusei, Candida tropicalis*, and *Candida parapsilosis* are subgroups of candida yeast which are common pathogens in humans. Candida albicans are part of normal microbial flora in more than 55% of the healthy human population. However, it is an opportunistic pathogen and develops virulence when conditions become favorable such as overgrowth of bacteria in the gut or inflammation and ulcers which encourage its pathogenicity. Once virulent, it is hard to eliminate. Grapefruit seed extract is efficient in dealing with the pathogenic candida species.

Escherichia coli: E. coli is a commonly found bacteria in the human gut. It is also a commensal bacteria like candida yeast, but it can develop virulence when it comes in contact with other pathogenic bacteria, especially drug-resistant strains via transfer of genetic material. One of its pathogenic forms, *hemolytic E. coli* produces disruptive proteins causing damage to the intestinal wall, leading to blood loss and abnormal bowel conditions.

Foodborne Bacteria: Grapefruit seed extract is effective in eliminating foodborne pathogenic bacteria such as *Listeria, Salmonella, Klebsiella, Shigella, and Staphylococcus* species. These bacteria produce strong endotoxins which damage normal intestinal homeostasis. It leads to weakened intestinal walls which then harbor other pathogenic microbes and disturb microbial flora of probiotic strains.

6. Significance of Grapefruit Seed Extract in CanXida Remove

CanXida remove (Formula RMV) contains 45% standardized grapefruit seed extract for a full-spectrum antimicrobial effect. This concentration is potent enough to not only inhibit the growth of bacteria and fungi but also provides bactericidal and fungicidal effect to the pathogenic strains. As the minimum concentration reported for a bactericidal/fungicidal effect in various research reports is above 35 – 40%, a standardized 45% concentration was chosen as an optimal and effective dose. It is worth mentioning that for bacteriostatic and fungistatic effects (growth inhibitory effect but does not kill the pathogen), the concentration of grapefruit seed extract of 16.5% or above has been reported effective, as shown in Tables 2 and 3. 45% standardized grapefruit seed extract also provides enough antioxidant bioflavonoids and phytosterols for antioxidant activity and wound healing in the gastrointestinal tract.

7. Conclusion

The grapefruit seed extract is a highly efficient antimicrobial that is effective against a wide range of bacteria and fungi. It is also a source of potent antioxidants which are found only in grapefruit and related plants. These antioxidants are important mediators in the healing process. CanXida Remove (Formula RMV) contains standardized grapefruit seed extract in concentrations which is sufficient to kill the pathogen and are non-toxic to the human body.

8. References

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