The Effect of Monolaurin on Staphylococcus aureus and Escherichia coli

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Abstract: Microbial resistance to antibiotics, especially among staphylococcal strains, is a major threat to public health. The aim of this study was to assess the anti-bacterial effect of monolaurin on Staphylococcus aureus and Escherichia coli. In this study, colonies were cultured in nutrient broth media and incubated at 37°C for 24 h. Then, tubes were chilled in the laboratory environment. Three different concentrations of monolaurin prepared and added to culture media containing BHI agar media. Then, these plates were incubated at 37°C for 24 h and prepared to count. After 24 h, plates were out and colonies were counted and multiplied in 100. Data were analyzed by SPSS software Version 16. Based on data revealed that at 60°C and more. There was no observed any microorganisms colonization either Staphylococcus aureus or Escherichia coli but at the 55°C, there was observed more decrease in colonization than 50°C. This finding suggests that chilling is one of the most important ways to maintenance of foods. The results also showed that temperature also increases inhibitory properties of monolaurin. The results of the study suggest that monolaurin have anti-bacterial effect against Staphylococcus aureus and Escherichia coli.

Key words: Monolaurin, Staphylococcus aureus, Escherichia coli, temperature, microorganisms colonization, BHI agar media

INTRODUCTION

Microbial resistance to antibiotics, especially among staphylococcal strains, is a major threat to public health. Since, resistance by certain strains of Staphylococcus to multiple antibiotics like methicillin emerged in the late 1970’s (Ayliffe, 1997), many strategies to control antibiotic resistance have been proposed (Cunha, 2002). Considering current therapeutic regimens, vancomycin usage has proven to be the most reliable to treat resistant staphylococcal infections (Edmond et al., 1996; Waldvogel, 1999; Burnie et al., 2000). However, some staphylococcal strains have become resistant, at least to some extent even to vancomycin indicating a dire need for new alternative therapeutic approaches (Burnie et al., 2000; Hiramatsu et al., 1997; Denis et al., 2002).

One specific strategy has been to use multiple antibiotics of the same spectrum and low resistance potential when single antibiotic substitutions are not effective. However, researchers have examined yet another strategy; use of natural products with low potential for the development of resistance. The anti-viral, anti-bacterial and anti-protozoal properties of lauric acid and monolaurin have been recognized for nearly 3 decades by only a small number of researchers. Their research, however, has resulted in >50 research papers and numerous US and foreign patents. Kabara (1967) performed the original seminal research in this area of fat research. Kabara first patented certain Fatty Acids (FAs) and their derivatives, e.g., Monoglycerides (MGs) can have adverse effects on various microorganisms. While nontoxic and approved as a direct food additive by the FDA, monolaurin adversely affects bacteria, yeast, fungi and enveloped viruses.

Kabara found that the properties that determine the anti-infective action of lipids are related to their structure, e.g., free fatty acids and monoglycerides. The monoglycerides are active; diglycerides and triglycerides are inactive. Of the saturated fatty acids, lauric acid has greater anti-viral activity than either Caprylic acid (C-8), Capric acid (C-10) or myristic acid (C-14).

Fatty acids and monoglycerides produce their killing/inactivating effects by several mechanisms. An early postulated mechanism was the perturbing of the plasma membrane lipid bilayer. The anti-viral action attributed to monolaurin is that of fluidizing the lipids and phospholipids in the envelope of the virus causing the disintegration of the microbial membrane. More recent studies indicate that one anti-microbial effect in bacteria is related to monolaurin's interference with signal transduction/toxin formation. Another anti-microbial effect in viruses is due to lauric acid's interference with

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virus assembly and viral maturation. The third mode of action may be on the immune system itself (Witcher et al., 1996). Hierholzer and Kabara (1982) first reported the anti-viral activity of the monoglyceride of laurie acid (monolaurin) on viruses that affect humans.

They showed virucidal effects of monolaurin on enveloped RNA and DNA viruses. This research was done at the Center for Disease Control of the US Public Health Service. This study was carried out using selected virus prototypes or recognized representative strains of enveloped human viruses. All these viruses have a lipid membrane.

The presence of a lipid membrane on viruses makes them, especially vulnerable to laurie acid and its derivative monolaurin. These initial findings have been confirmed by many other studies. Research has shown that enveloped viruses are inactivated by added fatty acids and monoglycerides in both human and bovine milk (Isaacs and Thornar, 1991). Others (Isaacs and Thornar, 1986) have confirmed Kabara’s original statements concerning the effectiveness of monolaurin.

Some of the viruses inactivated by these lipids are the measles virus, herpes simplex virus (HSV-1 and -2), herpes family members (HIV, hepatitis C, Viscerelt, Stomatitis Virus (HSV), visna virus and Cyte Megalo Virus (CMV). Many of the pathogenic organisms reported to be inactivated by these anti-microbial lipids are those known to be responsible for opportunistic infections in HIV positive individuals. For example, concurrent infection with cytomegalovirus is recognized as a serious complication for HIV positive individuals.

These anti-microbial fatty acids and their derivatives are essentially nontoxic to man. According to the published research, laurie acid is one of the best inactivating fatty acids and its monoglyceride is even more effective than the fatty acid alone (Kabara, 1978; Fletcher et al., 1985; Kabara, 1985). The lipid-coated (envelope) viruses, bacteria and other microorganisms are dependent on host lipids for their lipid constituents. The variability of fatty acids in the foods of individuals as well as the variability from de novo synthesis accounts for the variability of fatty acids in their membranes.

Monolaurin does not appear to have an adverse effect on desirable gut bacteria but rather on only potentially pathogenic microorganisms. For example, Isaacs and Schneidman (1991) reported no inactivation of the common Escherichia coli or Salmonella enteritidis by monolaurin but major inactivation of Hemophilus influenza, Staphylococcus epidermis and group B gram positive streptococcus. The potentially pathogenic bacteria inactivated by monolaurin include Listeria monocytogenes, Staphylococcus aureus, Streptococcusagalactiae, groups A, streptococci-gram-positive organisms and some gram-negative organisms (Vibrio parahaemolyticus and Helicobacter pylori). Decreased growth of Staphylococcus aureus and decreased production of toxic shock syndrome toxin-1 was shown with monolaurin (Holland et al., 1994).

Monolaurin was 5000 times more inhibitory effect against Listeria monocytogenes than ethanol. In vitro monolaurin rapidly inactivate Helicobacter pylori. Of greater significance, there appears to be very little development of resistance of the organism to the bactericidal effects of these natural anti-microbials.

A number of fungi, yeast and protozoa are also inactivated or killed by monolaurin. The fungi include several species of ringworm (Isaacs and Thornar, 1991). The yeast reported to be affected is Candida albicans (Isaacs and Schneidman, 1991), the protozoan parasite Giardia lamblia is killed by monoglycerides from hydrolyzed human milk (Isaacs and Thornar, 1991). Chlamydia trachomatis is inactivated by monolaurin. Hydrogels containing monocaprin/monolaurin are potent in vitro inactivators of sexually transmitted viruses such as HSV-2 and HIV-1 and bacteria such as Neisseria gonorrhoea. The aim of this study was to assessment of anti-bacterial effect of monolaurin on Staphylococcus aureus and Escherichia coli.

MATERIALS AND METHODS

In this study, colonies are cultured in nutrient broth media and incubated at 37°C for 24 h. Then, this culture is divided into the 3 separate tubes including:

- First tube incubated at 50°C for 10 min
- Second tube incubated at 55°C for 10 min
- Third tube incubated at 60°C for 10 min

Then tubes chilled in the laboratory environment. Three different concentrations of monolaurin prepared and added to culture media containing BHI agar media. Then these plates are incubated 37°C for 24 h and prepared to count. After 24 h, plates were out and colonies are counted and multiply in 100. Data were analyzed by SPSS software Version 16.

RESULTS AND DISCUSSION

Results are shown in Table 1 and 2. Based on Table 1 and 2 revealed that at 60°C and more, there was no observed any microorganisms colonization either Staphylococcus aureus or Escherichia coli but at the 55°C, there was observed more decrease in colonization.
than 50°C. This finding suggests that chilling is one of the most important ways to maintain the quality of foods. The results also showed that temperature also increases inhibitory properties of monolinor.

The carriage and subsequent dissemination of antibiotic-resistant *Staphylococcus aureus* by hospital staff and patients is a recognized risk for nosocomial infections (Carson et al., 1995). Although, use of antibiotics has generally been tempered to avoid antibiotic resistance, the development of resistance was inevitable. Therefore what alternatives now exist to treat antibiotic resistant organisms? Essential oils, especially Origanum oil and the fat monolinor are natural substances reported to have the ability to kill *Staphylococcus aureus* and other microbes in culture (Carson et al., 1995; Hitokoto et al., 1980; Ismaiel and Pierson, 1990; Mansour et al., 1999; Kim et al., 1995; Kivanc et al., 1991). Nevertheless, in vivo studies designed to test the anti-microbial effects of various essential oils and monolinor are lacking.

Many essential oils have been shown to be effective against a number of organisms. Origanum oil, cinnamon and clove were judged very active by examining their inhibitory effects on *Clostridium botulinum* 33A (Ismaiel and Pierson, 1990). In addition to effects against *Klebsiella pneumoniae* and *Staphylococcus aureus*, Origanum oil is fungicidal (Manuchar et al., 2001; Hitokoto et al., 1980). Also, anti-viral actions of Origanum and clove oils against RNA and DNA viruses have been reported (Siddiqui et al., 1996). As a potential mechanism of action, the outer protective membrane of the viruses disintegrated after exposure to the Origanum oil when viewed by electron microscopy (Siddiqui et al., 1996). Importantly, most essential oils of spices are classified as GRAS (Generally Recognized as Safe) indicating that consumers can eat them reasonably without fear (Ismaiel and Pierson, 1990). Accordingly, the benefits/risks ratio of essential oils in treating microbes would seem to be very high. Considering other natural products with anti-microbial properties, Kabara championed the use of certain lipids. He measured the anti-microbial activity of fatty acids and their corresponding monoglycerides and reported that the optimum chain length was C12 (Kabara et al., 1977). Of the saturated fatty acids, lauric acid (C12) has greater anti-viral activity than caprylic acid (C8), capric acid (C10) or myristic acid (C14). In contrast to monolinor, the dilaurin derivative was inactive. It is now generally accepted that monoglycerides are active: diglycerides and triglycerides are inactive. When the anti-chlamydial effects of several fatty acids and monoglycerides were studied by incubating Chlamydia trachomatis bacteria, the results indicate that the lipid kills the bacteria, possibly by disrupting the membranes of the elementary body (Bergson et al., 1998). Corroborating evidence is available from viral studies suggesting that the bactericidal effects are via disintegration of membranes by fatty acids (Thomar et al., 1987; Isaacs and Thomar, 1991) similar to a report of the action of Origanum oil on viruses (Siddiqui et al., 1996).
CONCLUSION

The results of the study suggest that monolaurin have anti-bacterial effect against *Staphylococcus aureus* and *Escherichia coli*.

REFERENCES


