CMIT/MIT – Isothiazolone Biocide Assessment

September 17, 2020
Patrick M. Eakins, Ph.D.
Technical Director
peakins@biobor.com

Hammonds Fuel Additives, Inc.
6951 W. Little York
Houston, Texas 77040

Summary

CMIT/MIT is a biocide formula used in a shocking number of products as a preservative, considering its known toxicity and potential for serious health and environmental issues. Since its introduction in the late 1970's as a preservative and popularization in the 1980's, CMIT/MIT has been used in varied applications including water and fuel treatment. This broad use has provided researchers an opportunity to study its effect on humans and the environment leading some to refer to it as the new DDT. The American Contact Dermatitis Society named MIT the "allergen of the year" in 2013. More recently, CMIT/MIT has been associated with severe lung injury attributed to the death of a previously healthy young girl. (Lee, Park, Do, Jang, & Hong, 2019) As a fuel biocide, the product has been touted by some as the most widely used and most effective. While its wide range of use is acknowledged, this assessment will look at its effectiveness and application as a fuel biocide. CMIT/MIT efficacy, kinetics and toxicity will be assessed.

Introduction

Dow Chemical Company and its strategic partners produce CMIT/MIT based products marketed under a number of different trade names. The products contain a mixture of the active ingredients (CMIT) 5-chloro-2-methyl-2H-isothiazol-3-one and (MIT) 2-methyl-2H-isothiazol-3-one in a 3 to 1 ratio, referred to as CMIT/MIT. They are a member of a group of chemicals known as isothiazolinone. The fuel biocide mixture includes the active substances as well as magnesium dinitrate (salt), magnesium chloride (salt), water and dipropylene glycol (DPG). According to the manufacturer's information, the active ingredients have a pH of 1.9. They are classified as ultra-acidic and are more acidic than sulfuric acid. The actives are fully water soluble with some solubility in fuel. DPG is also completely water soluble with little to no fuel solubility. This will be discussed later in more detail. One of the active ingredients, MIT is considered a cytotoxin and is the subject of serious study and concern. This will also be discussed in further detail.

Efficacy

To understand the actual efficacy of CMIT/MIT, recommended dose rates and soak times will be examined. Manufacturer literature claims the product eradicates microbial growth in contaminated fuel systems in 24 to 72 hours. (Rohm and Haas, 2020) The efficacy of the product will be challenged based on this claim. According to the manufacturers product information, there are three effective dose rates. (Dow Chemical Company, 2013)

- 100-150ppmv – preventative/maintenance dose – for fuels that lack microbial contamination. The literature notes to exercise "extreme care" "to avoid the addition of

a preventative/maintenance level dosage" to heavily contaminated fuel systems. Other manufacturers literature notes that this dosage should never be used if microbial contamination is present at any level.

- 200-300ppmv – curative dose – with the evidence of microbial contamination.
- 400-1000ppmv – shock dose – for fuels with heavy microbial contamination.

A strict maximum of 100ppmv is allowed for aviation application, which according to all of the manufacturers data is insufficient to eradicate microbial contamination. *Table 1*, taken from an original study indicates the minimal biostatic dose rate for individual microbial organisms tested. (Rohm and Haas, 2020) Only one out of forty-two is within the 100ppmv dose rate limit for aviation use, a clear indication that CMIT/MIT will never be able to eradicate microbial contamination as claimed. The concern for any sub-lethal dosing of a biocide is microbial adaptation, resistance and microbial recontamination. There are well documented cases of CMIT/MIT resistance. Research shows the regular application of sub-lethal doses of a biocide leads to microorganism resistance, rendering the product ineffective. Documented research appears to validate the concerns. (Rushton, et al., 2013)

CMIT/MIT Minimal Biostatic Dose

Organism	Minimum CMIT/MIT ppmv
Flavobacterium suaveolens	600
Aspergillus niger	600
Chaetomium globosum	600
Gliocladium fimbriatum	600
Aspergillus foetidus	530
Cellulomonas sp.	400
Aspergillus repens	400
Gleophylum trabeum	400
Sarcina lutea	330
Azotobacter vinelandii	330
Enterobacter aerogenes	330
Escherichia coli	330
Proteus vulgaris	330
Pseudomonas aeruginosa	330
Pseudomonas oleoverans	330
Salmonella typhosa	330
Aspergillus oryzae	330
Candida albicans	330
Hormonoconis resinae	330
Mucor rouxii	330
Penicillium funiculosurn	330
Aureobasidium pullulans	300
Rhizopus stolonifer	300
Lentinus lepidus	270
Alternaria dianthicola	200
Phoma glomerata	200
Desulfovibrio desulfuricans	170
Bacillus cereus var. mycoides	130
Bacillus subtilis	130
Brevibacterium ammoniagenes	130
Staphylococcus aureus	130
Staphylococcus epiderm idis	130
Achromobacter parvulus	130
Alcaligenes faecalis	130
Pseudomonas fluorescens	130
Shigella sonnei	130
Fusarium Sp.	130
Penicillium variabile	130
Phoma herbarum	130
Rhodotorula rubra	130
Saccharomyces cerevisiae	130
Trichosporon	130
Streptomyces albus	70

Table 1

The efficacy of a biocide is dependent on proper application which includes both adequate mixing and soak time. CMIT/MIT is highly water soluble. However, its fuel solubility is a matter of question likely due to the carrier, dipropylene glycol (DPG). (Rohm and Haas, 2020) DPG is not miscible in kerosene. (Huntsman Corporation, 2013) Miscibility is a specific term that relates to a liquids ability to blend or solubilize in another liquid. In the case of DPG which represents approximately 90% of the CMIT/MIT fuel biocide mixture, it is not miscible – therefore not soluble.

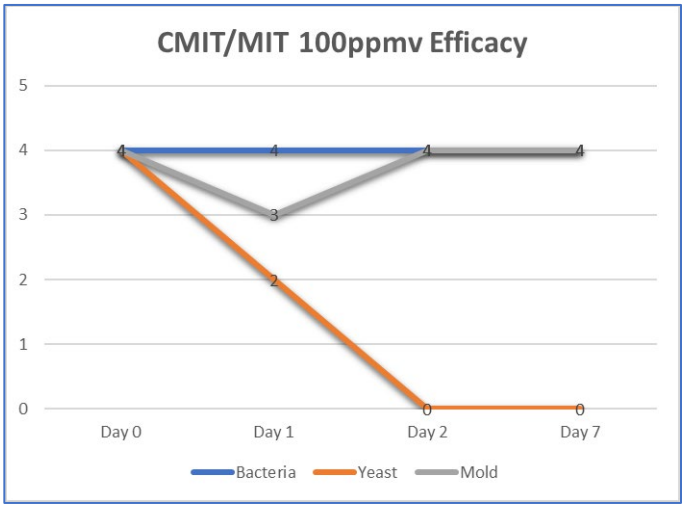
With any additive that exhibits solubility issues, product mixing is of paramount importance, likely the reason product instructions specifically relate effectiveness with mixing. Product information notes "that effective treatment will only be achieved with efficient mixing." (Dow Chemical Company, 2013) A biocide that has a solubility gap will have an efficacy issue regardless the dose rate. Solubility gaps and inadequate mixing will result in unprotected fuel. If a product is prone to settle out of solution, the only area being protected is the small area where the product has settled. The rest of the fuel and system are left without adequate treatment where microorganisms will multiply and flourish. It is also possible to have inaccurate test results if the biocide has settled to a low point of access where samples are regularly taken. That location would have a higher concentration of biocide than the rest of the tank (potentially a majority of the biocide), showing

a more efficacious result, but only for that very small area in the fuel system. The rest of the fuel and system are left unprotected.

Inadequate mixing can also affect soak times. The published soak time for CMIT/MIT is 24 to 72 hours depending on the level of microbial contamination. The product information specifically states that mixing methods can "extend the treatment time required." (Dow Chemical Company, 2013) The following efficacy data reveals the necessity for extended soak times and higher dose rates under all conditions tested. Efficacy *Graph 1* shows the lack of efficacy when dosed at 100ppmv. This is not surprising given the aforementioned biostatic information in *Table 1*. In its study, Dow does note that 100ppmv is a "maintenance level" dosage, not capable of eradicating microbial contamination. It is the dosage maximum for aviation use. *Table 2* shows Dow's microbial scoring methodology and *Graph 1* illustrates the efficacy based on Dow's testing score. Note that little has changed with a 100ppmv dose of CMIT/MIT. In the study, Dow administers an additional 100ppmv dose at day 14 with similar results. It isn't until an additional 400ppmv dose is added on day 21 that any efficacious results are seen. Even with a total of 600ppmv over 21 days, the biocide still could not totally eradicate all of the microbial growth. The only success the biocide had was on yeast at 48 hours soak time.

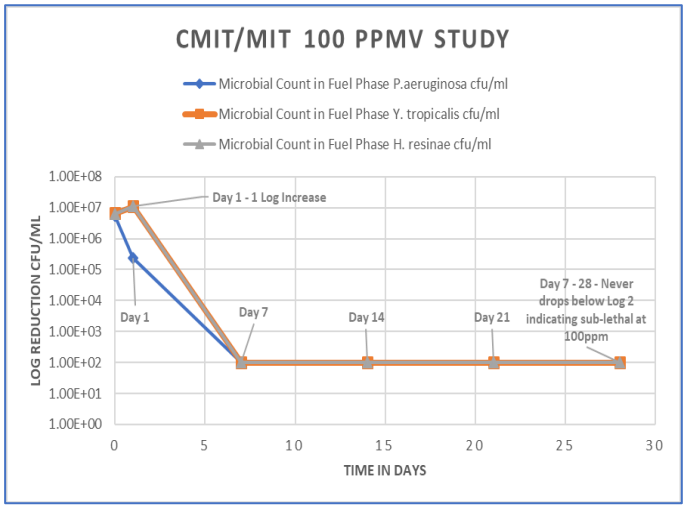
Score	# of colonies per plate	Approximate CFU/mL
0	0	$<1 \times 10^1$
1	1 to 9	$1 \times 10^1 - 9 \times 10^1$
2	10 to 99	$1 \times 10^2 - 9.9 \times 10^2$
3	100 to 300	$1 \times 10^3 - 3 \times 10^3$
4	> 300	$> 3 \times 10^3$

Table 2



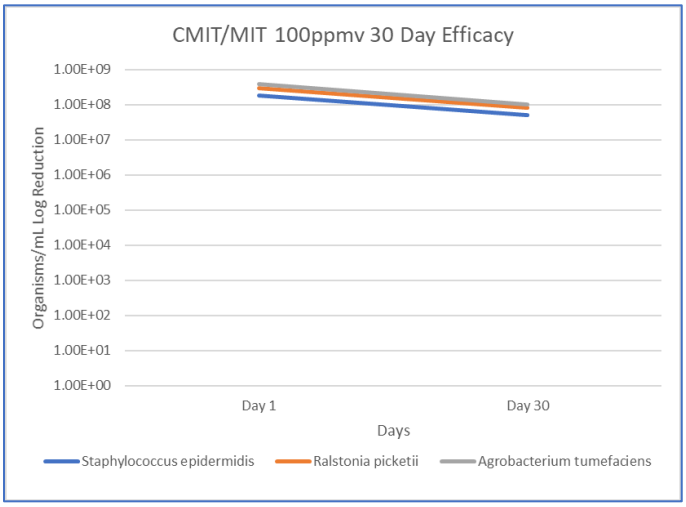
Graph 1

Additional studies reveal the same sub-lethal results. *Graph 2* illustrates another study completed on three specific microorganisms. The information provided in *Table 1* serves to confirm the poor efficacy results shown in *Graph 2*. A 100ppmv dose does not eradicate the microbes. The ongoing concern when using CMIT/MIT in continued sub-lethal doses has to be adaptation or resistance. Even with increased soak times, microbial infestation still remains at high enough levels to create concern and cause problems. In aviation use, a 100ppmv treatment still allows for biomass generation, airframe and engine damage, among other serious consequences associated with a sub-lethal dose.



Graph 2

Other studies confirm the ineffectiveness of CMIT/MIT at the 100ppmv dose level. One such study challenged CMIT/MIT in JP-8 fuel with a high level of contamination. (Raikos, et al., 2012) The results are seen in *Graph 3*. The biocide could only affect a <1 Log reduction over the 30-day period representing a 77.6% reduction. While this may sound good, at the level of contamination, it leaves the fuel with over 10,000,000 organisms/mL. This does not bode well for fuel protection in the long-term storage of aircraft. That level of microbial contamination left in the fuel tanks will undoubtedly increase both maintenance cost and risk.



Graph 3

Are higher doses of CMIT/MIT more effective? Does the 400-1,000ppmv shock dose eradicate microbes in the fuel? The manufacturer states that "a shock dose is required to decontaminate an already contaminated system." (Dow Chemical Company, 2010) This should be enough to dissuade the use of anything less when microbes are present. However, the question still remains, "Will a shock dose of CMIT/MIT eradicate microbial contamination?" In a 60-day efficacy study, it was found the biocide had only "a transitory inhibitory effect." (Zimmer, et al., 2013) At 400ppmv it took over 7 days for the biocide to begin working on various biodiesels with low (Log 3) to high (Log 8) contamination levels containing natural water bottoms. Results showed only an inhibitory effect in all fuel types. The study used the reduction of biomass production to determine the biocides effectiveness. *Table 3* shows the results by product type for both the low and high challenge. While there was a reduction in biomass, neither the 400ppmv or 1,000ppmv dose rates were able to eradicate the microbial contamination as illustrated by the continued biomass production. The aggregate reduction percentage was only 51% indicating a biostatic effect. Neither the 400ppmv or 1,000ppmv dose rates were capable of eradication as indicated by the continued biomass production. The researchers noted the "poor performance" of the biocide.

Percentage of Reduction of Biomass by CMIT/MIT

	Low Contamination Level		High Contamination Level	
Fuel Type	**400ppmv**	**1,000ppmv**	**400ppmv**	**1,000ppmv**
B0	71%	53%	36%	59%
B7	7%	40%	43%	25%
B10	46%	40%	58%	51%
B100	72%	67%	78%	68%
Average Reduction	49%	50%	54%	51%

Table 3

The manufacturer's claim that CMIT/MIT eradicates microbial growth is overstated at best and unfounded in both its own data and third-party studies. According to the data, serious consideration should be given prior to any sub-lethal use of the product on existing microbial contamination. The

results provided illustrate the limited efficacy at 100ppmv and demonstrates the potential for adaptation or resistance. The biostatic information in *Table 1* reinforces the limitation of the product. The 100ppmv dosage does not appear to be effectively biostatic. Additionally, at the recommended shock dose levels, the product did not perform as stated.

Kinetics

The stability of any product is important. Kinetics involves the measurement and study of reactions that affect the stability and degradation of the biocide. The two most prevalent concerns affecting CMIT/MIT are pH and temperature. Both are shown to influence accelerated degradation, deactivation and violent thermal decomposition of the active ingredients.

Influence of pH

According to ASTM D6469, "fuel tank bottom-water pH is usually between 6 and 9." CMIT/MIT's high-water solubility tends to drive the product to any bottom-water. In the case of fuel tanks, this is where the biocide settles. Research shows that CMIT/MIT does not do well in alkaline environments. A pH of 7 is neutral, lower pH is acidic and higher pH is alkaline. The effectiveness of a biocide is determined by its ability to control microbial contamination. Any degradation over time reduces its effectiveness. There have been several kinetic studies on the matter. Some have recorded deactivation at a pH as low as 7. A detailed study showing the effect of pH on the degradation of isothiazolone biocides was completed in 1992. (Barman & Preston, 1992) It notes "the active component undergoes degradation in alkaline solutions, and the rate of degradation is faster with an increase in pH." It further suggests that the degradation of the biocide "results from hydrolysis of the chlorinated isothiazolone" (CMIT) as a result of the reaction to an alkaline pH. The half-life value of CMIT/MIT at a pH of 8.5 was found to be 47 days. As pH rose, the time to half-life was reduced. At a pH of 10, the half-life was just 2 days. The active ingredients were degraded by 50% in a very short period of time. The effect of pH continued to reduce the effectiveness of the biocide until complete deactivation.

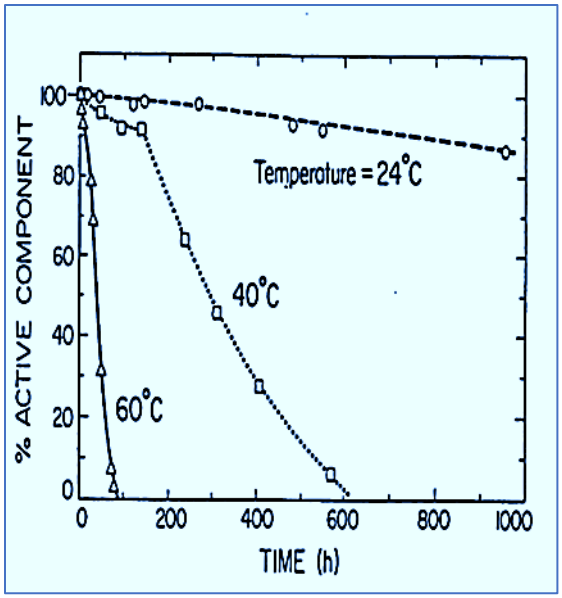
Influence of Temperature

Fluid and atmospheric temperature play a significant role in the degradation and deactivation of CMIT/MIT. The manufacturer recommends avoiding temperatures above 40˚C (104˚F). (Dow Chemical Corporation, 2015) It goes on to warn "the active ingredients can decompose violently at temperatures above 50˚C (122˚F). Generation of gases (hydrogen chloride, nitrogen oxides, sulfur oxides) can cause rapid pressure buildup in closed systems," such as fuel systems. The dangers are obvious in the manufacturer's warning. The deactivation requires further explanation. In a study on the influence of temperature on degradation of isothiazolone biocides, it was discovered that temperature played a significant role in the deactivation of the active ingredients of the biocide. (Barman B. N., 1993)

It was found that the active ingredients became unstable and began to deactivate at temperatures as low as 24˚C (75˚F). As the temperature went up, the degradation and deactivation of the actives accelerated. It appears that thermal decomposition breaks the chemical bonds, destabilizing the active

substances and inactivating them. *Graph 4* illustrates the deactivation of CMIT/MIT based on temperature and thermal decomposition. (Barman B. N., 1993) According to the graph, the active components would be completely deactivated at 60˚C in four days. At 40˚C deactivation would take just over 25 days and at 20˚C (75˚F) the deactivation would be 100% in one-year, possibly the reason the manufacturer has a published one-year shelf life on the product.

Graph 4



This data is significant especially for the protection of fuel in above ground storage tanks or the long-term parking of aircraft. The correlation between high temperature exposure in these situations and the deactivation of the product is important to understanding. The reduced effectiveness of the biocide and the storage limitations as it pertains to higher temperatures play a role in whether or not it works effectively or potentially not at all. The higher the temperature, the shorter the shelf life of the product due to thermal decomposition. It is not uncommon to have fuel temperatures in parked aircraft well above the flash point of the fuel 52˚C (126˚F) reaching temperatures above 60˚C. The same is true for fuels stored in above ground storage tanks. Thermal decomposition will happen with CMIT/MIT. Higher temperatures will deactivate the active components reducing its efficacy or rendering it completely ineffective.

Toxicity

How toxic is too toxic – a question often asked about biocides. CMIT/MIT is under global scrutiny for its toxicity and serious implications to human health and the environment. Product documentation warns of its danger including chemical burns, allergic reactions and respiratory and lung problems due to exposure. (Dow Chemical Corporation, 2015) There are hundreds of studies and peer reviewed journal articles documenting the serious health concerns attributed to exposure and use of the chemical. Over the last few years, there has been a growing concern among medical professionals and researchers due to the rapid increase of serious health issues associated with CMIT/MIT. The following are a few of the most significant.

Contact Dermatitis

The biocide has been clearly identified as a contact allergen. Between 2010 and 2012 nationwide testing in France was performed on over 7874 patients. (Hosteing & et al., 2014) The study showed a significant increase in positive test results for contact dermatitis indicating a rapidly emerging allergen. It reported “similar observations in other European countries.” As a result of this study and others, regulatory intervention has taken place with restrictions for the use of the chemicals. More restrictions are expected.

Peripheral Airway Dysfunction and Lung Injury

A 2017 study confirmed the significant damage to the airways due to exposure to CMIT/MIT inhalation. (Cho & et al., 2017) Twenty-four patients were identified as having been exposed to the chemical. None had underlying diseases. Inhalation exposure of CMIT/MIT is just now coming to the forefront. There have been over 500 additional studies exposing the inhalation danger. It was determined that inhalation exposure to the chemical was a direct cause of fatal lung injury. The results were reported in Korea from 2006 to 2011. Since then numerous other reports have been made on the deleterious effects of CMIT/MIT on humans.

Neurodegenerative Disorders

The occupational use of CMIT/MIT has been tied to neurotoxicity. (Du, McLaughlin, Pal, & Aixenman, 2002) The chemical is highly toxic to human neurons (the basic working unit of the brain, the spinal cord, the central neural tracts and the peripheral nerves). Both acute and chronic consequences have been reported for humans exposed to the toxins. In the study, the chemical induced cell death as well as excessive DNA damage. It was postulated that the adverse neurological consequences would surface as a result of occupational or environmental exposure to the biocide.

There is no doubt that the evidence is mounting on the toxicity of CMIT/MIT. Other effects of exposure to the chemical include hair loss - a sign of endocrine hormone disruption, inflammatory disease, and neuromuscular paralysis. Physical contact with CMIT/MIT, including inhalation puts the user at risk.

Conclusion

The assessment of CMIT/MIT's efficacy, kinetics and toxicity reveal a product that is long on promises and short on delivery. The manufacturers claim that the product eradicates microbial contamination is troubling. The results show limited biostatic affect in recommended dose rates. Even at a shock dose, the efficacy was not as expected from the claims made. Product stability is another concern. Both pH and temperature have a significant degradational effect. Dramatic increases in degradation are compounded when both pH and temperature work together to deactivate the biocide. (Barman B. N., 1993) Finally, toxicity has to play a role in determining the risk associated with its handling and use. Exposure to a chemical so toxic increases the likelihood for heath related incidents. When a less toxic, proven biocide is available, why use this one?

References

Barman, B. N. (1993). Influence of temperature on the degradation of isothiazolone biocides in aqueous media and in metalworking fluid concentrate. *Journal on the Science of Tribologists and Lubrication Engineers*, 351-355.

Barman, B., & Preston, H. (1992). The effects of pH on the degradation of isothiazolone biocides. *Thribology International*, 281-287.

Cho, H.-J., & et al. (2017). Effects of a mixture of chloromethylisothiazolinone and methylisothiazolinone on periperal airway disfunction in children. *Plos One*, 1-16.

Dow Chemical Company. (2010, October 8). Correct biocide dosing is critical for effective protection of fuels. *Form No. 253-02788-10/08/10 PS*. Dow Chemical Company.

Dow Chemical Company. (2013, March 25). Kathon FP 1.5 Fuel Biocide Product Information. *Form No. 253-03153-03/25/13 PS*. Dow.

Dow Chemical Corporation. (2015, June 30). Product Safety Assessment DOW™ 5-Chloro-2-Methyl-2H-Isothiazol-3-one/2-Methyl-2H-Isothiazol-3-one (CMIT/MIT)-Based Antimicrobial Products. Dow Chemcial Corporation.

Du, S., McLaughlin, B., Pal, S., & Aixenman, E. (2002). In vitro neurotoxicity of methylisothiazolinone, a commonly used industrial and household biocide, proceeds via a zinc and extracellular signal-regulated kinase mitogen-activated protein kinase-dependent pathway. *The Journal of Neuroscience*, 7408-7416.

Hosteing, S., & et al. (2014). Outbreak of contact sensitization to methylisothiazolinone: an analysis of French data from the REVIDAL-GERDA network. *Contact Dermatitis*, 262-269.

Huntsman Corporation. (2013). Miscibility predictor for organic liquids. Huntsman Corporation.

Lee, S.-Y., Park, D.-U., Do, K.-H., Jang, S.-J., & Hong, S.-J. (2019). The pathological findings of chloromethylisothiazolinone and methylisothiazolinone-associated lung injury. *Journal of Korean Medical Science*.

Raikos, V., Vamvakas, S., Sevastos, D., Kapolos, J., Karaiskakis, G., & Koliadima, A. (2012). Water content, temperature and biocide effects on growth kinetics of bacteria isolated from JP-8 aviation fuel storage tanks. *Fuel*, 559-566.

Rohm and Haas. (2020, September 17). *Market applications oil and gas*. Retrieved from Bencide - Dow Biocides: http://www.bencide.co.kr/data/KATHON%20FP15.pdf

Rushton, L., Sass, A., Baldwin, A., Dowson, C., Donoghue, D., & Mahenthiralingam, E. (2013). Key role for efflux in the preservative susceptibility and adaptive resistance of burkholderia cepacia complex bacteria. *Antimicrobial agents and chemotherapy*, 2972-2980.

Zimmer, A., Cazarolli, J., Teixeira, R. M., Viscardi, S., Cavalcanti, E., Gerbase, A., . . . Bento, F. (2013). Monitoring of efficacy of antimicrobial products during 60 days storage simulation of diesel (B0), biodiesel (B100) and blends (B7 and B10). *Fuel*, 153-162.