



## Correct Biocide Dosing is Critical for Effective Protection of Fuels

Laboratory efficacy tests demonstrate that maintenance dosing of biocides protects clean fuel from spoilage, but shock doses are required to effectively treat heavily fouled fuel systems.

### Overview:

The protection of fuels from microbial spoilage is determined by two critical factors: 1) the biocide product to be used for treatment, and 2) the dosage level of the biocide product. The proper dosage of a biocide can mean the difference between survival and death of microorganisms that spoil the fuel system. The data summarized herein illustrate the importance of distinguishing between a “maintenance dose” and a “shock dose” when treating a fuel system. A “maintenance dose” is necessary to protect a clean, uncontaminated system from microbial contamination. A “shock dose,” is required to decontaminate an already contaminated system.

KATHON™ FP 1.5 and FUELSAVER™ biocides are two effective, well-recognized, and trusted products that are used to protect fuel products including diesel, low sulfur diesel (LSD), ultra low sulfur diesel (ULSD), unleaded gasoline, marine, and biodiesel fuels from microbial spoilage. In order for KATHON FP 1.5 and FUELSAVER to eliminate microbes in fuels effectively, proper dose levels must be used.

### Laboratory study:

A recent Dow laboratory study demonstrates both the effectiveness of properly dosing the KATHON FP 1.5 and FUELSAVER biocide products, as well as the pitfalls of under-dosing a biocide.

#### Treating contaminated fuel:

During this study, ULSD fuel was contaminated with high levels of industrially relevant microbes ( $5 \times 10^6$  colony forming units per milliliter (CFU/mL) of bacteria and  $5 \times 10^5$  CFU/mL of fungi). Organisms used included the bacterium *Pseudomonas aeruginosa* (ATCC# 33988) and the fungal organisms *Hormoconis resinae* (ATCC# 20495) and *Yarrowia tropicalis* (formerly *Candida tropicalis*) (ATCC# 18138) which are used in the ASTM E1259 method entitled, *Standard Practice for Evaluation of Antimicrobials in Liquid Fuels boiling below 390°C*.

ULSD fuel samples were inoculated with microorganisms on day 0 and then a shock dosage level or a maintenance dosage level of KATHON FP 1.5 (as specified by the EPA label) was applied to the 50 mL fuel samples (45 mL fuel and 5 mL sterile water) beginning on day 7 and continuing on days 14 and 21. The level of contamination was determined at various time points during the study and scored on a numerical scale of 1 to 4. A score of 2 (which indicated contamination of at least  $1 \times 10^2$  CFU/mL) and above was considered to be a failing score. Failing scores are shown in red, and passing scores are shown in green. No additional biocides were dosed into the fuel samples after day 7; therefore, biocidal efficacy observed throughout the study is the result of the original biocide dosage.

#### Treating clean, uncontaminated fuel:

During this study, clean ULSD fuel was first dosed with a maintenance level of either KATHON FP 1.5 (100 ppm) or FUELSAVER (125 ppm), prior to inoculation of the fuel samples on day 0 with microorganisms ( $5 \times 10^4$  CFU/mL of bacteria and  $5 \times 10^3$  CFU/mL of fungi). The fuel samples were challenged again with microorganisms on day 7. No additional biocides were dosed into the fuel samples after day 0; therefore, biocidal efficacy observed throughout the study is the result of the original biocide dosage on day 0.



Results:

Table 1 shows the results of dosing the ULSD with either maintenance level (100 ppm) or shock level (400 ppm) dosages of KATHON™ FP 1.5. Maintenance level dosages are recommended as a preventative measure to protect clean, uncontaminated fuels from future microbial exposure. **Maintenance level dosages of biocide products should not be used to treat heavily fouled systems.** As shown in Table 1, two maintenance dosages (days 7 and 14) were not sufficient to decontaminate the fuel samples; however, a shock dosage of KATHON FP 1.5 (400 ppm) on day 7 was rapidly effective (within 24 to 48 hours) for decontamination of the ULSD. In Table 2, maintenance dosages of FUELSAVER™ and KATHON FP 1.5 biocides were effective against two microbial challenges of the fuel. The fuel samples that were not pretreated with biocides were susceptible to heavy contamination.

Table 1. Shock dosing of KATHON FP 1.5 biocide is required to rapidly decontaminate ULSD fuel.

Contamination on Day 0 (10 <sup>6</sup> Bacteria/10 <sup>5</sup> Fungi)	Organism Type	Growth Score at Time Point (Day) After Microbial Challenge on Day 0																	
		0	1	2	7	Day 7 Treatment			Day 14 Treatment			Day 21 Treatment			22	23	28	29	30
						8	9	14	15	16	21	22	23	28	29	30			
	Bacteria	4	4	4	4	Maintenance Dose: 100 ppm KATHON™ FP 1.5			Maintenance Dose: 100 ppm KATHON FP 1.5			Shock Dose: 400 ppm KATHON FP 1.5			0	0	0	0	0
	Yeast	4	4	4	4	Maintenance Dose: 100 ppm KATHON™ FP 1.5			Maintenance Dose: 100 ppm KATHON FP 1.5			Shock Dose: 400 ppm KATHON FP 1.5			0	0	0	0	0
	Mold	4	4	4	4	Maintenance Dose: 100 ppm KATHON™ FP 1.5			Maintenance Dose: 100 ppm KATHON FP 1.5			Shock Dose: 400 ppm KATHON FP 1.5			0	1	0	0	0
	Organism Type	0	1	2	7	Day 7 Treatment			Day 14 Treatment			Day 21 Treatment			22	23	28	29	30
	Bacteria	4	4	4	4	Shock Dose: 400 ppm KATHON FP 1.5			None			None			0	0	0	0	0
	Yeast	4	4	4	4	Shock Dose: 400 ppm KATHON FP 1.5			None			None			0	0	0	0	0
	Mold	4	4	4	4	Shock Dose: 400 ppm KATHON FP 1.5			None			None			0	1	0	0	0

Note: Fuel samples that were not treated with a biocide remained contaminated (scores of 4) throughout the 31-day test.

Table 2. Maintenance treatment with FUELSAVER or KATHON FP 1.5 prevents contamination of ULSD fuel.

Biocide (Dosage ppm product)	Organism Type	Day 0	Growth Score at Time Point								
			Challenge 1 (10 <sup>4</sup> Bacteria/10 <sup>3</sup> Fungi)	Cycle I			Challenge 2 (10 <sup>4</sup> Bacteria/10 <sup>3</sup> Fungi)	Cycle II			
				Day 1	Day 2	Day 7		Day 8	Day 9	Day 14	
Maintenance Dose: 125 ppm FUELSAVER™	Bacteria	0	Challenge 1 (10 <sup>4</sup> Bacteria/10 <sup>3</sup> Fungi)	2	0	0	Challenge 2 (10 <sup>4</sup> Bacteria/10 <sup>3</sup> Fungi)	0	0	0	
	Yeast	0		0	0	0		0	0	0	
	Mold	0		1	0	1		2	0	0	
Maintenance Dose: 100 ppm KATHON™ FP 1.5	Bacteria	0		0	0	0		0	0	0	
	Yeast	0		0	0	0		0	0	0	
	Mold	0		0	0	0		0	0	0	
Unpreserved Control	Bacteria	0		4	4	4		4	4	4	
	Yeast	0		2	2	4		4	4	4	
	Mold	0		3	2	4		4	4	4	



Table 3. Growth scoring scale for efficacy testing results.

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	$> 3 \times 10^3$

**Summary:**

When dosed appropriately, KATHON™ FP 1.5 and FUELSAVER™ biocides are extremely effective for both the decontamination of heavily fouled fuel systems (Table 1) and the prevention of future microbial spoilage (Table 2). Biocide treatment helps maintain the integrity of fuels at all points during storage, distribution, and usage. KATHON FP 1.5 and FUELSAVER biocides effectively preserve fuel systems and help prevent machine malfunction, engine failures, and impaired water removal from storage tanks, while protecting the fuel performance properties such as color, heat of combustion, pour point, and cloud point.

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