

Part Number: FQS-047

Please read this package insert carefully **BEFORE** any use of the FQS HY-LiTE[®] fuel test kit. Should you have any questions or need assistance please contact FQS by phone at (770) 967-9790 or by email at <u>technical@fqsinc.com</u>/sales@fqsinc.com.

Disclaimer

Important Notice

The test kit should only be used as part of an investigatory process into microbiological contamination in petroleum products (i.e., gasoline, Jet A/A1, JP 8, diesel, ultra low sulfur diesel, home heating oil, F76 and biodiesel) and associated water.

The test kit must only be used and tests performed strictly in accordance with the instructions in this documentation. When testing aviation fuels follow the current relevant International Air Transport Association (IATA) "Guidance Material on Microbiological Contamination in Aircraft Fuel Tanks". When testing hydrocarbon fuels as part of an ASTM program follow the current relevant version of ASTM D7463, Standard Test Method for Adenosine Triphosphate (ATP) Content of Microorganisms in Fuel, Fuel Water Mixtures and Associated Water. Unless this is the case, the results of any tests performed cannot be considered to be valid.

It is important to note that results from detection kits are an indication of microbial contamination. Physical inspection is the only definitive method to determine the condition of the fuel tank. The result of any test performed relates only to the sample tested and not necessarily to other petroleum product contained within the system under investigation. Although guidance may be sought by the user on sampling and the interpretation of results, the responsibility for carrying out the sampling and test procedures correctly is that of the end user and not FQS or its affiliates or appointed distributors.

The test kit is designed to indicate presence of metabolically active biological material but it is the nature of biology that there may be organisms present in the sample that are not detected by the test procedure. In particular, the results of any test procedure cannot be interpreted to indicate sterility of the sample tested.

FQS, its affiliates, and appointed distributors do not accept any liability for any decision or assessment taken or made as a consequence of using this test kit.

1. Method

Micro-organisms and water soluble metabolites (including ATP) are captured from the fuel phase and concentrated in the blue Capture Solution phase. Adenosine-tri-phosphate (ATP) is detected specifically by reaction with a Luciferin/Luciferase mixture in a buffered solution. The light emitted in the process is detected and displayed by the HY-LiTE® luminometer (Part No. FQS-046 & FQS-046A). This method is in accordance with ASTM D7463.

2. Detection limit and number of determinations

Detection limit*	Number of determinations
1.0 x 10 ⁻¹⁴ mol ATP	20

^{*}ATP per milliliter capture solution or per ml bottom water

3. Applications

Detection of total microbiological contamination in gasoline, Jet A/A1, JP 8, diesel, ultra low sulfur diesel, home heating oil, F76, biodiesel fuel samples, fuel and water mixtures and associated water.

4. Limits

IATA "Guidance Material on Microbiological Contamination in Aircraft Fuel Tanks" suggests the following:

	HY-LiTE®	
Negligible:	<1000	RLU/litre fuel
Moderate:	1000-5000	RLU/litre fuel
Heavy:	>5000	RLU/litre fuel

The above limits suggested apply only to Aircraft Wing and Center Tank samples, and are based on comparison with other routinely IATA approved microbial test methods. These limits are a suggestion only and may need to be adapted for other types of fuel samples and according to individual circumstances, frequency of testing, history of contamination, etc.

5. Interference with foreign substances

Chemical additives and contaminants such as FSII and anticorrosive agents may interfere with the efficiency of the Capture Solution and the HY-LiTE® reaction and cause lower than expected readings. Biocides used for treatment of contaminated fuel may interfere with the reaction chemistry, depending on the concentration and type of biocide in the fuel.

The effect of FQS 1.5 Microbicide® at 100 ppm (v/v) and Biobor JF at 270 ppm (wt/wt) has been tested in fuel and causes no significant interference on the HY-LiTE® test. The effects of other isothiazolinone biocides substantially similar in composition, concentration and use should behave the same such as FQS 1.5 Microbicide®.

6. Reagents and test format

The reagents in the test are stable up to the date stated on the label posted on the box and on the label for each pack when stored closed at $+2^{\circ}$ C to $+8^{\circ}$ C (35.6°F to 46.4 °F). The shelf life includes a period of transport or storage of up to three weeks at room temperature. Ambient Temperature is defined as 23°C (74.3°F)

The HY-LiTE® Fuel Test Pen is for use between the temperatures of +5°C to +35°C (41°F to 95°F). Remove the Fuel Test Pen(s) from the refrigerator and box and maintain at ambient temperature for approximately 30-60 minutes in order for the pen to achieve a use temperature between +5°C to +35°C.

Package contents:

- 20 Individually sealed test packets containing:
 - ✓ 1 HY-LiTE® pen for fuel testing
 - ✓ 1 small pipette for transfer of Capture Solution from sample tube to fuel sample
 - ✓ 1 large pipette for transfer of Capture Solution from fuel sample to HY-LiTE® sample tube reservoir.

Test format:

The HY-LiTE® pen is a ready prepared test cuvette designed for use in a HY-LiTE® luminometer and consists of the following parts:

- A Sample Tube cap and reservoir with **Blue Capture Solution** (Patent pending).
- A white sampling stick (covered by the protection cap) for accurate sampling of the liquid to be tested. The stick is coated with an extractant, which releases ATP from cellular material. The stick also transfers the sample into the cuvette and in a later step opens the reagent chamber.
- A cuvette filled with test buffer for dilution, buffering and neutralization of the sample.
- A reagent chamber sealed with aluminium foil, containing freeze-dried and stabilized Luciferin/Luciferase mixture.

7. Procedure

Preparation of measurement sample (pictured step-by-step guide):

		Remove the HY-LiTE® test pen from the refrigerator and allow temperature to rise to +5°C to +35°C (41°F to 95°F); approximately 30 minutes to 1 hour.
	1	Collect one litre of a fuel sample in a suitable plastic container (such as Nalgene Cat. No. 2104-0032 or 332189-0032). The fuel sample should be taken from the bottom of the tank when possible. (D7463 Precision and Bias is based on 500mL samples but does apply to 1000mL samples)
	2	Remove the small pipette from the pack bulb-end first. Take care not to touch the tip of the pipette. With the bulb squeezed, insert the pipette tip into the Blue Capture Solution in the HY-LiTE® sample tube reservoir. Release the bulb to collect all of the Blue Capture Solution in the pipette. Add the Blue Capture Solution to the fuel sample. Insert the small pipette back into the plastic sleeve for future use.
4	3	Close the lid to the sample container tightly and shake horizontally for 10-20 seconds in a manner that produces maximum shear of the Blue Capture Solution
	4	Leave the Blue Capture Solution treated fuel sample to stand for at least 5 minutes to allow the capture solution to settle at the bottom of the sample bottle.
	5	After 5 minutes, tap the bottom edge of the bottle to coalesce the Blue Capture Solution droplets into a single large drop.
	6	Remove a large pipette from the pack bulb-end first. Take care not to touch the tip of the pipette. With the bulb squeezed, insert the pipette tip into the Blue Capture Solution at the bottom of the fuel sample. Slowly release the bulb to collect all of the Blue Capture Solution in the pipette.

7	Using care, transfer the Blue Capture Solution to the HY-LiTE® sample tube reservoir. The level of liquid must at least reach the bottom of the bowl. Avoid transferring fuel phase to the Sample Tube. Discard the large pipette. If a fuel layer is seen or suspected to be present in the Blue Capture Solution, use the small pipette to remove the Capture Solution from the tube reservoir and slowly bubble air through the pipette to separate the two fluids. Then slowly return the Blue Caption Solution to the reservoir holding back the fuel layer in the pipette.
8	Remove the Pen carefully from the sample tube cap.
9	Dip the white sampling stick completely into the Blue Capture Solution for about 1 second.
10	Carefully remove the white stick from the Blue Capture Solution. The seven rings should contain the Blue Capture Solution. DO NOT SHAKE THE STICK or try to remove any liquid that may be dangling from the tip as this may result in loss of sample.
11	Press the stick vertically (in order to avoid breakage of the stick) onto a hard surface and under continuous pressure until the sample stick is completely retracted into the cuvette chamber .
12	Under pressure, twist the upper translucent collar clockwise 1-2 revolutions until it stops. If the collar stops twisting in less than 1 revolution, then back out the collar counter clockwise and rotate clockwise again to ensure cutting of the foil barrier. This opens the aluminium foil in between the reagent chamber and end cap by slitting it with the sharp end of the stick.
13	Grasping the top of the cuvette chamber between the thumb and forefinger, shake the pen <i>vertically</i> (along the length of the pen) vigorously for 10-20 seconds. This will mix the sample with the reagent (foam and colour change to more yellow-green color will appear). Tap the black cap 2-3 times on a solid surface to dislodge any chemical buffer from the top of the cuvette.



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Insert the pen without delay into the reading chamber of the **HY-LiTE**® **luminometer** and close the lid to start measurement.

Do not force the lid closed. If the lid does not close easily check to ensure the white stick is completely enclosed in the cuvette chamber and the cap is fully twisted closed.

8. Notes

- Since ATP is a commonly occurring substance, care must be taken not to touch the following parts with the fingers or to contaminate them in any other way: the tip of the pipettes, the lid or the edge of the protection cap containing the Blue Capture Solution, and the white sampling stick.
- To minimize risk of contamination of the pipettes, remove the pipette by pushing the bulb end of the pipette out of the protective plastic before gripping it.
- Do not put lettering or a label on the clear and transparent cuvette, as this is the reading window for the light reaction.
- In order to avoid damage to the luminometer: press the white sampling stick completely into the pen and twist both parts of the pen together completely until contact.
- Do not hold or turn the black cap of the reagent chamber, otherwise the pen could leak.
- Gloves can be worn while handling the fuel samples and performing the capture treatment. *However, do not wear gloves while handling the HY-LiTE® sampling pen (step 8-14).*
- The test is developed to work with specific sample bottles (Nalgene Cat. No. 2104-0032 or 332189-0032). Other sample bottles may work, but FQS has not verified the performance where different bottles are used. The use of a glass bottles is forbidden.
- HY-LiTE® pens can be disposed of as non-microbiological waste, in accordance with local regulation.
- Technical modifications of the test or kit in compliance with technical progress reserved.

References:

- 1. ASTM D7463Standard Test Method for Adenosine Triphosphate (ATP) Content of Microorganisms in Fuel, Fuel/Water Mixtures and Fuel Associated Water.
- 2. International Air Transport Association guidance (IATA) "Guidance Material on Microbiological Contamination in Aircraft Fuel Tanks" ISBN 978-92-9252-781-5.

For additional assistance or to reorder please contact:

Fuel Quality Services, Inc. P.O. Box 1380 Flowery Branch, GA 30542 Phone: (770) 967-9790 / FAX: (770) 967-9982

Email: sales@fqsinc.com
Technical Support: technical@fqsinc.com
Website: www.fqsinc.com

Reviewed by: Edward W. English II & Debra Chesneau

Date: April 2020