Sample Stability
Sample stability studies were conducted in house. Human hemoglobin-free stool was spiked with a known level of human hemoglobin to result in the following concentrations: 0, 5, 8, 10, 12, 15, 400 µg/g stool that are equivalent to 0, 25, 40, 50, 60, 75 and 2000 ng/mL sampling buffer. The samples in sampling bottles are stored at 15, 25, and 30℃ for 15 days, and at 2, 4, and 8℃ for 30 days. Overall percent agreements against expected results were favorable at all the temperatures tested.

(1) Stored at room temperature for 15 days

Day 15	Actual Results: OneStep Pro+ FIT	Expected Results: Positive Results	Expected Results: Negative Results	Expected Results: Total Results	Overall Percent Agreement
15℃	Positive Results	284	3	287	99.3%
	Negative Results	0	154	154	
	Total Results	284	157	441	
25℃	Positive Results	284	2	286	99.5%
	Negative Results	0	155	155	
	Total Results	284	157	441	
30℃	Positive Results	283	0	283	99.8%
	Negative Results	1	157	158	
	Total Results	284	157	441	

(2) Stored at refrigeration for 30 days

Day 30	Actual Results: OneStep Pro+ FIT	Expected Results: Positive Results	Expected Results: Negative Results	Expected Results: Total Results	Overall Percent Agreement
2℃	Positive Results	283	6	289	98.4%
	Negative Results	1	151	152	
	Total Results	284	157	441	
4℃	Positive Results	284	4	288	99.1%
	Negative Results	0	153	153	
	Total Results	284	157	441	
8℃	Positive Results	280	0	280	99.1%
	Negative Results	4	157	161	
	Total Results	284	157	441	

Interference Testing
Cross-reactivity studies were performed by adding non-human hemoglobin (Hb) and tissue extracts to OneStep Pro+ FIT. Hb of bovine, equine, goat, porcine, rabbit, sheep, turkey, and fish were added to the test to determine if non-human Hb has cross-reactivity or interference with OneStep Pro+ FIT. The same tests were performed with addition of tissue extracts from beef, horse, goat, pork, rabbit, sheep, chicken, and fish. All the tests resulted in no cross-reactivity or interference to the test.

Dietary Testing
A potential interference of dietary substances on OneStep Pro+ FIT was assessed. Aqueous extracts of raw broccoli, cauliflower, cantaloupe, horseradish, red radish, parsnip, and turnip were added to the test to determine if vegetable extracts cross-react with the test. The extracts were prepared by homogenizing raw vegetable in a food processor and then subsequently centrifuging the extract to separate the solid and liquid phases. Dietary Iron and Vitamin C supplements were also tested for cross-reactivity. No cross-reactivity was evident.

Comparison Study
OneStep Pro+ FIT was compared with a commercially available predicate device, OC-Light® FOBT with 953 specimens. The study was performed at three POL sites and three PML (Professional Medical Laboratory) sites. The overall percent agreement between OneStep Pro+ FIT and the predicate OC-Light® FOBT was 99.9%, with positive percent agreement of 100.0% and negative percent agreement of 99.9%, demonstrating that the analytical performance of the device is substantially equivalent to the predicate.

		Positive Results	Negative Results	Total Results	Overall Percent Agreement (95% CI)	Positive Percent Agreement (95% CI)	Negative Percent Agreement (95% CI)
OneStep Pro+ FIT	Positive Results	121	1	122	99.9% (99.4%–100.0%)	100.0% (97.0%–100.0%)	99.9% (99.3%–100.0%)
	Negative Results	0	831	831			
	Total Results	121	832	953			

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Index of Symbols

IVD — For *In Vitro* diagnostic use
REF — Catalog number
LOT — Batch code
Caution
Instructions for Use
Use by
2ºC 30ºC — Temperature limitation
Manufacturer
Single Use

Distributed by (in U.S. only):
HENRY SCHEIN INC.
135 DURYEA ROAD
MELVILLE, NY 11747 USA

Made in Japan
Rev. 2017-03

HENRY SCHEIN **Instructions for Use**

OneStep Pro+ FIT (Fecal Immunochemical Test)

Caution: For *in vitro* diagnostic use only. Rx Only.
United States Federal law restricts this device to sale and distribution to or on the order of a physician or to a clinical laboratory; and use is restricted to, by, or on the order of a physician.

INTENDED USE
OneStep Pro+ FIT (Fecal Immunochemical Test, also known as iFOBT, immunochemical fecal occult blood test) is a qualitative test intended for the immunochemical detection of fecal occult blood (FOB) by professional laboratories and physician office laboratories. Measurement of FOB is useful as an aid to detect blood in stool when gastrointestinal (GI) bleeding may be suspected.
OneStep Pro+ FIT is recommended for use in routine physical examinations.

SUMMARY
Presence of fecal occult blood in stool is associated with gastrointestinal disorders such as diverticulitis, polyps, and Crohn’s disease, that may lead to colorectal cancer if not treated. Early diagnosis by fecal occult blood screening and treatment of these problems has been shown to significantly reduce mortality from colorectal cancer. Conventional test methods used for the detection of fecal occult blood do not provide a high degree of accuracy. Immunological tests developed to detect human hemoglobin are more accurate and do not require special dietary restrictions on patients.

PRINCIPLE
OneStep Pro+ FITis an *in vitro* diagnostic device, a qualitative test designed for the immunochemical detection of human hemoglobin (hHb) in stool specimens. When the sample end of the test strip is dipped in the fecal extract, the liquid fecal extract wicks through a series of absorbent materials and contacts colloidal gold conjugated with monoclonal antibodies specific to hHb. If hHb is present in the sample, it reacts with the antibodies on the colloidal gold. When the gold conjugate with hHb reaches the test region of the membrane, it binds with the immobilized antibodies also specific to hHb to form a visible reddish/pink line. The procedural control region of the membrane contains immobilized anti-mouse antibodies that capture the conjugate independent of the presence of the hHb, thereby always producing a distinct reddish/pink line. The reddish/pink line in the procedural control region demonstrates the validity of the test, and assures the operator that the device is working properly.

REAGENTS AND MATERIALS PROVIDED
• Test Strip
• Sampling Bottle containing 2mL of collection buffer (HEPES buffer with stabilizers)

ORDERING INFORMATION
OneStep Pro+ FIT Manual Kit (570-0609)
50 Test Strips
50 Sampling Bottles
50 Collection Papers
OneStep Pro+ FIT Test Strips (570-0610)
50 Test Strips
OneStep Pro+ FIT Personal Use Kit (570-0607)
20 Personal Use Kits

PRECAUTIONS
• For *in vitro* diagnostic use only.
• For professional and laboratory use.
• The directions for use must be followed carefully for accurate results.
• Do not reuse Test Strips, and Sampling Bottles.
• Do not use Test Strips if canister is damaged; does not seal.
• Do not touch the reagent area of Test Strip, or disassemble.
• In case of contact with collection buffer to eye, mouth, or skin, wash thoroughly with plenty of water and see a doctor for proper treatment if necessary.
• Treat the sample dissolved solution (fecal extract) and used Test Strips as if they are potentially infectious.
• Wear disposable gloves when performing the test to avoid infection.
• In case of sample spill, bleach is recommended for cleaning and disinfecting.
• Do not use beyond the labeled expiration date. The expiration date can be found on the carton and vial labels.
• Do not use sampling probe directly on a human body.
• Dispose of used Sampling Bottles and Test Strips in accordance with Federal, State, and Local requirements.

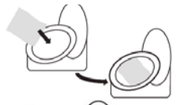
STORAGE AND STABILITY
Store OneStep Pro+ FIT at 2 - 30°C (36 - 86°F). DO NOT FREEZE. OneStep Pro+ FIT is stable when stored at these temperatures until the expiration date printed on the label.

MATERIALS REQUIRED BUT NOT PROVIDED
• Timing device • Gloves • Rack (item number 123-6830) • External Controls (item number 123-6822)

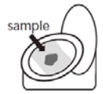
SAMPLE COLLECTION AND HANDLING
Collect stool sample from Collection Paper following the instruction below. Contamination from toilet water should be avoided.

Sample Deposit

1. Place supplied collection paper inside toilet bowl on top of water.



2. Deposit stool sample on top of collection paper.

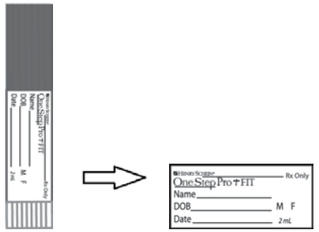


3. Collect sample from stool before paper sinks and stool sample touches water.

4. Flush. Collection paper is biodegradable and will not harm septic systems.

Sample Collection

1. Fill in all required information on the Sampling Bottle.

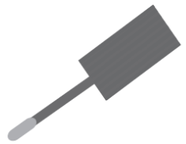


2. Open green cap by turning to the left and pulling upwards.

3. Scrape the surface of the fecal sample with the sample probe.

Scrape the surface of the stool widely or stab it at 5-6 different points.

4. Cover the grooved portion of the sample probe completely with stool sample.



Loose stool sample should be collected in the same way by completely covering the grooved portion of the probe.

5. Close Sampling Bottle by inserting the sample probe and screwing cap on tightly to the right. Do not reopen.

6. The sample may be stored at room temperature for up to 15 days or can be refrigerated at 2 - 8°C (36 - 46°F) for up to 30 days.

TEST PROCEDURE
Refer to Figure 1.

1. Bring Test Strips and the Sampling Bottle containing patient's fecal sample to room temperature at 20 - 30°C (68 - 86 °F). Shake the Sampling Bottle vigorously.
2. Remove a OneStep Pro+ FIT Test Strip from the canister. Minimize the amount of time that the canister is left open and assure that the canister is securely closed after opening.
3. Remove the white cap on the Sampling Bottle. Drop the sample end of the Test Strip into the extraction vial.
4. Start a timer.
5. When the timer reaches 5 minutes, read results. Read results as shown under "Interpretation of Results".

NOTE: Specimens with high concentrations of Hb may produce positive results in as little as 1 minute. Confirm negatives at 5 minutes.

Figure 1

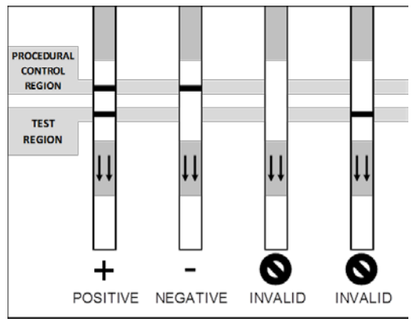
INTERPRETATION OF RESULTS
Refer to Figure 2.

POSITIVE
Carefully look for the appearance of a test line in the Test Region. ANY reddish/pink colored line in the Test Region along with a reddish/pink colored line in the Procedural Control Region is a positive result. Neither the intensity nor the color of the line in the Test Region should be compared to that of the Procedural Control line.

NEGATIVE
If no reddish/pink line appears in the Test Region and one reddish/pink line appears in the Procedural Control Region the result is negative.

INVALID
If no reddish/pink line appears in the Procedural Control Region, the test is invalid and must be repeated with a new strip.

Figure 2



QUALITY CONTROL
Good laboratory practices recommend the use of appropriate controls. There are two types of controls for OneStep Pro+ FIT, the internal procedural control and external controls.

Procedural Control
The Procedural Control is found in the Procedural Control Region of the Test Strip. This control assures the operator that (A) sample addition and migration through the Test Strip has occurred and that (B) the control anti-mouse antibody and the reporter MAb are intact and functional. This control does not ensure that the capture antibody is accurately detecting the presence or absence of Hb in the sample.

External Control
External controls are used to assure the operator that the capture and conjugated antibodies are present and reactive. External controls will not detect an error in performing the patient test procedure. Controls should be assayed once per kit lot, following the local and state guidelines. To use, unscrew the white cap on the sample bottle. Add four drops of the control. Replace the white cap and shake vigorously. Follow step two through five of the test procedure. If controls do not perform as expected, do not use the test results. Repeat the test or call Polymedco Technical Services at 800-431-2123.

LIMITATIONS
- OneStep Pro+ FIT is intended only for the detection of human hemoglobin in feces. It is not advised for use in patients suspected of upper GI bleeding.
- Patients with the following conditions should not be considered for testing as these conditions may interfere with the test results:
  - Bleeding hemorrhoids
  - Constipation bleeding
  - Urinary bleeding
  - Menstrual bleeding
- Certain medications such as aspirin and non-steroidal anti-inflammatory drugs may cause gastrointestinal irritation and subsequent bleeding in some patients and cause positive results.
- As with any occult blood test, results obtained with OneStep Pro+ FIT should not be considered conclusive evidence of the presence or absence of GI bleeding or pathology. It is not intended to replace other diagnostic procedures such as colonoscopy, sigmoidoscopy, and double contrast barium x-ray.
- Because gastrointestinal lesions may bleed intermittently and blood in feces is not distributed uniformly, a negative test result does not assure absence of lesion.
- Urine and excessive dilution of samples with water from the toilet bowl may cause erroneous test results. For best result, use Collection Paper. OneStep Pro+ FIT is not for use in testing urine, gastric specimens, or other body fluids.
- Use of stool samples that are not collected in a provided sampling bottle following bowel movement may affect the result due to instability of Hemoglobin in stool.
- Fecal occult blood testing is recommended annually by the American Cancer Society (2008) for average-risk women and men, 50 years of age and older. However, patients with significant risk factors such as family history of colorectal cancer should be screened earlier and more often.

EXPECTED VALUES
OneStep Pro+ FIT detects Hb in feces at levels as low as 10 µg hemoglobin/g stool (50 ng/mL).

PERFORMANCE CHARACTERISTICS
Sensitivity
The sensitivity of OneStep Pro+ FIT is 10 µg hemoglobin/g of stool, or 50 ng/mL of buffer.

The ability of OneStep Pro+ FIT to detect human hemoglobin variants was determined by testing HbS and HbC, in comparison to a reference, HbA0. OneStep Pro+ FIT detected the presence of all variants, HbA0, HbS and HbC

Reproducibility
Reproducibility studies were conducted at three Physician Office Laboratories (POL). Human hemoglobin-free stool was spiked with a known level of human hemoglobin to result in the following concentrations: 0, 5, 8, 10, 12, 15, 400 µg/g stool that are equivalent to 0, 25, 40, 50, 60, 75 and 2000 ng/mL sampling buffer. Total of nine operators participated in the study for over twenty days of testing, utilizing three test kit lots. Overall percent agreement, positive percent agreement, and negative percent agreement were 98.9%, 99.8%, and 97.1%, respectively

Reproducibility Studies	Actual Results: OneStep Pro+ FIT	Expected Results: Positive Results	Expected Results: Negative Results	Expected Results: Total Results	Overall Percent Agreement (95% CI)	Positive Percent Agreement (95% CI)	Negative Percent Agreement (95% CI)
Repeatability	Positive Results	95	1	96	99.3% (96.2% - 100.0%)	100.0% (96.2% - 100.0%)	98.1% (89.5% - 100.0%)
	Negative Results	0	51	51			
	Total Results	95	52	147			
Lot-to-Lot Reproducibility	Positive Results	284	6	290	98.6% (97.0% - 99.5%)	100.0% (98.7% - 100.0%)	96.2%
	Negative Results	0	151	151			
	Total Results	284	157	441			
Between-run Reproducibility	Positive Results	283	0	283	99.8% (98.7% - 100.0%)	99.6% (98.0% - 100.0%)	100.0% (97.7% - 100.0%)
	Negative Results	1	157	158			
	Total Results	284	157	441			
Between Device Reproducibility	Positive Results	284	6	290	98.6% (97.0% - 99.5%)	100.0% (98.7% - 100.0%)	96.2% (91.8% - 98.6%)
	Negative Results	0	151	151			
	Total Results	284	157	441			
Between-site Reproducibility	Positive Results	283	6	289	98.4% (96.7% - 99.4%)	99.6% (98.0% - 100.0%)	96.2% (91.8% - 98.6%)
	Negative Results	1	151	152			
	Total Results	284	157	441			
Combined Reproducibility	Positive Results	1134	18	1152	98.9% (98.3% - 99.3%)	99.8% (99.4% - 100.0%)	97.1% (95.5% - 98.3%)
	Negative Results	2	610	612			
	Total Results	1136	628	1764			

CI: Confidence Interval