



Otsuka
Medical Device Division of
Otsuka America Pharmaceutical, Inc.



1 Intended Use

The BreathTek® UBT for *H. pylori* Kit (BreathTek UBT Kit) is intended for use in the qualitative detection of urease associated with *H. pylori* in the human stomach and is indicated as an aid in the initial diagnosis and post-treatment monitoring of *H. pylori* infection in adults, and pediatric patients 3 to 17 years old. The test may be used for monitoring treatment if used at least 4 weeks following completion of therapy. For these purposes, the system utilizes an Infrared Spectrophotometer for the measurement of the ratio of $^{13}\text{CO}_2$ to $^{12}\text{CO}_2$ in breath samples, in clinical laboratories and point-of-care settings. The Pediatric Urea Hydrolysis Rate Calculation Application (pUHR-CA), provided as a web-based calculation program, is required to obtain pediatric test results.

The BreathTek UBT Kit is for administration by a health care professional, as prescribed by a physician.

2 Summary and Explanation

Since the isolation of the spiral urease-producing *Helicobacter pylori* (*H. pylori*) bacteria in 1983 by Drs. Marshall and Warren¹, a significant body of evidence has accumulated indicating that the bacteria is an important pathogen in the upper GI tract of humans.^{2,3} *H. pylori* is associated with a number of GI conditions including chronic gastritis, peptic ulcer disease, and gastric malignancy.^{4,5} Methods available for detecting current infection of the human stomach by *H. pylori* are generally divided into two (2) general types: Invasive and Non-invasive.

Invasive methods are so named because they include, as a first step, an esophagogastroduodenoscopy ("EGD") with collection of gastric biopsies. These biopsies are then examined by one or more detection methods: histological examination of stained tissue, microbiological culture of the organism, or direct detection of urease activity in the tissue. Biopsy based methods are expensive, entail some patient risk and discomfort and may give false negative results due to sampling errors when colonization of the gastric mucosa is patchy.⁶

Non-invasive methods include serological testing, fecal antigen test, and urea breath test. Several serological tests that detect serum antibodies to *H. pylori* are commercially available. A positive result with a serologic test cannot distinguish between current infection and past exposure to infection and, therefore, is not a conclusive indicator of current gastrointestinal colonization by *H. pylori*. Urea breath tests are a non-invasive method for detecting current *H. pylori* infection.

3 Principle of the BreathTek UBT for *H. pylori*

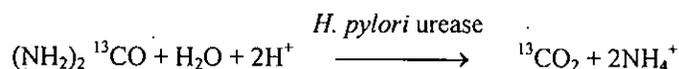
3.1 Description of the Pranactin®-Citric Diagnostic Drug Component

The diagnostic drug component of the kit is ^{13}C -urea, a synthetic urea contained in a granulated powder (Pranactin-Citric) for reconstitution with potable water to provide a clear solution for oral administration. The carbon in the drug component is predominantly Carbon-13, a stable, naturally occurring, non-radioactive isotope of carbon; the relative abundance of Carbon-13 is greater than or equal to 99%.

Each 3 gram dose of Pranactin-Citric is supplied in a polyethylene-lined foil pouch and contains 75 mg of ^{13}C -urea, citric acid⁷, aspartame and mannitol. ^{13}C -urea is the diamide of ^{13}C -carbonic acid and is highly soluble in water (1 gram per mL at 25°C). It has the following chemical formula: $^{13}\text{CH}_4\text{N}_2\text{O}$. An average adult body normally contains about 9 grams of urea, which is a product of protein metabolism. Urea in the body is referred to as natural isotopic abundance urea since it is composed of 98.9% ^{12}C -urea and 1.1% ^{13}C -urea.

3.2 Principle of the Test

Pranactin-Citric drug product is a component of the BreathTek UBT Kit. Three (3) g of reconstituted Pranactin-Citric containing 75 mg of ^{13}C -urea is ingested by the patient. In the presence of urease associated with gastric *H. pylori*, ^{13}C -urea $[(\text{NH}_2)_2^{13}\text{CO}]$ is decomposed to $^{13}\text{CO}_2$ and NH_4^+ according to the following equation:



The $^{13}\text{CO}_2$ is absorbed in the blood, and then exhaled in the breath. It results in an increase in the ratio of $^{13}\text{CO}_2$ to $^{12}\text{CO}_2$ in a POST-DOSE breath sample taken after the Pranactin-Citric solution was consumed, compared to a BASELINE sample taken before the Pranactin-Citric solution was consumed. Analysis of the breath samples is performed by UBiT[®]-IR300 Infrared Spectrophotometer or POCon[®] Infrared Spectrophotometer [located at your clinical laboratory and point-of-care settings]. For pediatric patients, the UBiT-IR300 Infrared Spectrophotometer should be used for the analysis of breath samples.

In the absence of gastric *H. pylori*, the ^{13}C -urea does not produce $^{13}\text{CO}_2$ in the stomach. The ratio of $^{13}\text{CO}_2$ in the POST-DOSE breath sample remains essentially the same as the BASELINE.

3.3 Adjustment of Endogenous CO_2 Production with UHR Calculation in Pediatric Patients

The measured difference between the ratios of $^{13}\text{CO}_2/^{12}\text{CO}_2$ values before and after administration of Pranactin-Citric solution is referred to as Delta over Baseline (DOB). DOB is the primary outcome measure reported in adults. It is known that the measured Delta over Baseline (DOB) is a function of anthropometric variables, which determine the rate of CO_2 production.⁸

While the effect of the CO_2 production rate is small between adults, it can be significant in pediatric patients. Therefore, in performing the BreathTek UBT on pediatric patients, the primary outcome measure reported for the BreathTek UBT is the UHR. The UHR is calculated as shown below:

$$\text{UHR } (\mu\text{g}/\text{min}) = \text{DOB} \times \text{CO}_2 \text{ Production Rate} \times 0.3427$$

4 Warnings and Precautions

- 4.1 For in vitro diagnostic use only. The Pranactin-Citric solution is taken orally as part of the diagnostic procedure.
- 4.2 Phenylketonurics: Contains Phenylalanine (one of the protein components of Aspartame), 84 mg per dosage unit. (For reference, 12 ounces of typical diet cola soft drinks contain approximately 80 mg of Phenylalanine.)
- 4.3 Blood glucose: Use with caution in diabetic patients. Pranactin Citric contains Aspartame.
- 4.4 A negative result does not rule out the possibility of *H. pylori* infection. False negative results do occur with this procedure. If clinical signs are suggestive of *H. pylori* infection, retest with a new sample or an alternate method.
- 4.5 False negative test results may be caused by:
 - Ingestion of antimicrobials, proton pump inhibitors, or bismuth preparations within 2 weeks prior to performing the BreathTek UBT
 - Premature POST-DOSE breath collection time for a patient with a marginally positive BreathTek UBT result
 - Post-treatment assessment with the BreathTek UBT less than 4 weeks after completion of treatment for the eradication of *H. pylori*.
- 4.6 False positive test results may be caused by:
 - Urease associated with other gastric spiral organisms observed in humans such as *Helicobacter heilmannii*.
 - Achlorhydria.⁹

- 4.7 If particulate matter is visible in the reconstituted Pranactin-Citric solution after thorough mixing, the solution should not be used.
- 4.8 Hypersensitivity: Patients who are hypersensitive to mannitol, citric acid or Aspartame should avoid taking the drug solution as this drug solution contains these ingredients. Swollen lip and rash were reported in the pediatric clinical studies.
- 4.9 Risk of Aspiration: Use with caution in patients with difficulty swallowing or who may be at high risk for aspiration due to medical or physical conditions.
- 4.10 Pregnancy: No information is available on use of the Pranactin-Citric solution during pregnancy.
- 4.11 For pediatric test results, the UHR results must be calculated. The DOB results are only used to calculate the UHR metrics to determine *H. pylori* infection in pediatric patients. DOB results cannot be used to determine the infection status of pediatric patients.
- 4.12 Safety and effectiveness has not been assessed in children below the age of 3 years.

5 Adverse Events

5.1 Adults-Postmarketing Experience

During post-approval use of the BreathTek UBT, the following adverse events have been identified: anaphylactic reaction, hypersensitivity, rash, burning sensation in the stomach, tingling in the skin, vomiting and diarrhea. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to establish a causal relationship to drug exposure.

5.2 Pediatrics-Clinical Experience

In two clinical studies conducted on 176 (analyzed) pediatric patients ages 3 to 17 years to determine the initial diagnosis and post treatment monitoring of *H. pylori* infection, the following adverse events experienced by $\geq 1\%$ of these patients were: vomiting (5.1%), oropharyngeal pain (4.5% to include throat irritation, sore throat, throat burning), nausea (2.3%), restlessness (2.3%), stomach ache/belly pain (1.1%), and diarrhea (1.1%). Most of the adverse events were experienced by the patients within minutes to hours of ingestion of the Pranactin-Citric solution.

6 Shelf Life and Storage

The BreathTek UBT Kit should be stored at 15°-30°C (59°-86°F). Pranactin-Citric has an expiration date of 48 months at 15°-30°C (59°-86°F). Do not use beyond the expiration date stated on the label.

7 Patient Preparation

- 7.1 Remind the patient that Pranactin-Citric contains phenylalanine (one of the protein components of Aspartame). Phenylketonurics restrict dietary phenylalanine.
- 7.2 The patient should have fasted at least 1 hour before administering the BreathTek UBT.
- 7.3 The patient should not have taken antimicrobials, proton pump inhibitors, or bismuth preparations within 2 weeks prior to administering the BreathTek UBT.

8 Procedure for Collecting Breath Samples Using BreathTek UBT Kit, for Analysis by Infrared Spectrophotometer

8.1 Materials

8.1.1 Materials provided

Each sealed single-patient BreathTek UBT Kit contains:

- One (1) "How To" guide
- Test instructions

- One (1) pouch of Pranactin-Citric powder (3 g)
- A set of four (4) self-adhesive bar-code stickers. All bar-codes should bear the same number.
- Two (2) breath collection bags, one (1) blue bag for the BASELINE sample and one (1) pink bag for the POST-DOSE sample.
- One (1) sample transport bag
- One (1) plastic straw
- One (1) plastic drinking cup

8.1.2 *Materials needed but not provided*

- A timer capable of timing an interval up to 15 minutes

8.1.3 *Instruments and Software*

- In adult patients, an Infrared Spectrophotometer (UBiT-IR300 or POCono, Otsuka Pharmaceutical Co., Ltd.) is required for analysis of breath samples.
- In pediatric patients,
 - Use the UBiT-IR300 Infrared Spectrophotometer to analyze the breath samples.
 - Use of the Pediatric Urea Hydrolysis Rate Calculation Application (pUHR-CA), as a web-based calculation program, is required to obtain the test result.
 - Go to: <https://pUHRCA.Otsuka-Us.com/pUHR-CA> to use the web-based pUHR-CA to calculate the UHR and obtain pediatric test results.

8.2 Step-By-Step Procedure

Time intervals listed in the following step-by-step procedure are critical. They are highlighted by the timer icon:

- 8.2.1 Verify that the patient has been prepared for the test as specified in Section 7.
- 8.2.2 Open the BreathTek UBT Kit, which should contain all the materials listed in Step 8.1.1. Label each breath collection bag to maintain patient identification using the bar-code labels provided, or according to your laboratory or office procedure.
- 8.2.3 Collect the BASELINE breath sample according to the following procedure:
- a. Pick up the blue breath collection bag.
 - b. Remove the pull-off cap from the mouthpiece of the breath collection bag.
 - c. Instruct the patient to: (1) breathe normally; (2) take a deep breath then pause momentarily; (3) exhale into the mouthpiece of the bag.
 - d. Replace the cap firmly until it clicks on the mouthpiece of the bag.
- 8.2.4 Prepare the Pranactin-Citric solution *no more than 60 minutes before administering it to the patient. Urea slowly decomposes in water.*
- a. Pick up the Pranactin-Citric pouch. Tap the upright packet of Pranactin-Citric to settle the contents in the bottom half.
 - b. Tear off the top of the packet and carefully empty the contents into the drinking cup provided, making sure to transfer all of the contents by tapping on the bottom of the pouch.
 - c. Add drinking water to the fill line indicated on the outside of the cup by a raised plastic ridge.

- d. Replace the lid securely and swirl the mixture for up to 2 minutes to dissolve the packet contents; typically, only 1 minute is required for complete dissolution. *The resulting drug solution should be clear with no particulate matter. If particulate matter is present after thorough mixing, the drug solution should not be used.*
- 8.2.5 Instruct the patient, including pediatric patients aged 3-17 regardless of age and body weight, to drink all of the drug solution with the straw provided, without stopping. Advise the patient NOT to 'rinse' the inside of his/her mouth with the drug solution before swallowing. Discard the straw after the patient has finished drinking the drug solution.
- 8.2.6 Set the timer for 15 minutes. The patient should sit quietly and should not eat, drink or smoke during the 15 minute interval.
- 8.2.7 After 15 minutes have elapsed, pick up the pink breath collection bag. Collect the POST-DOSE breath sample according to the procedure described in Steps 8.2.3 b through 8.2.3 d.
- 8.2.8 Store the specimens at 15°-30°C (59°-86°F) until analysis is performed.
- 8.2.9 Perform breath sample analysis within 7 days of breath sample collection. If desired, use the plastic sample transport bag for transport of the breath samples.

9 Quality Control

Complete operating information, including self-diagnostic instrument routines and user maintenance procedures provided in the Instruction Manuals for the UBiT-IR300 Spectrophotometer, the UBiT-AS10 Autosampler, the POCone Infrared Spectrophotometer and the POC-AS10 Autosampler, respectively. Additionally, each office laboratory or test facility should follow its own internal procedures for quality control.

10 Test Results

10.1 Adults

10.1.1 The Test Method

The ratio of $^{13}\text{CO}_2$ to $^{12}\text{CO}_2$ in breath samples is determined by Infrared Spectrophotometer, either UBiT-IR300 or POCone.

10.1.2 Calculation of Results

The result is provided as the Delta over Baseline (DOB) which is the difference between the ratio of $^{13}\text{CO}_2 / ^{12}\text{CO}_2$ in the POST-DOSE sample and the corresponding ratio in the BASELINE sample. No calculations are required by the user.

10.1.3 Determination of the Cutoff Point

The DOB cutoff value is 2.4 as determined in a controlled study of 66 infected and 53 uninfected asymptomatic, apparently healthy volunteers. Histological examination of biopsy tissue was used as the reference standard.

Meretek UBT™

Meretek UBT is an earlier version of the BreathTek UBT. The drug component of the test contained 125 mg of ^{13}C -urea. Analysis of the breath samples was performed by gas isotope ratio mass spectrometry (GIRMS). DOB values for the Meretek UBT were determined in a controlled clinical study of 66 infected and 53 uninfected asymptomatic, apparently healthy volunteers. Histological examination of biopsy tissue was used as the reference method in the determination of infection in this study. The Meretek UBT DOB values for the uninfected group ranged from 0.0 to 2.2. The DOB cutoff value for Meretek UBT was determined to be 2.4 in this study. Distribution of Meretek UBT DOB values in infected and uninfected groups in this study is shown in Figure 1a.

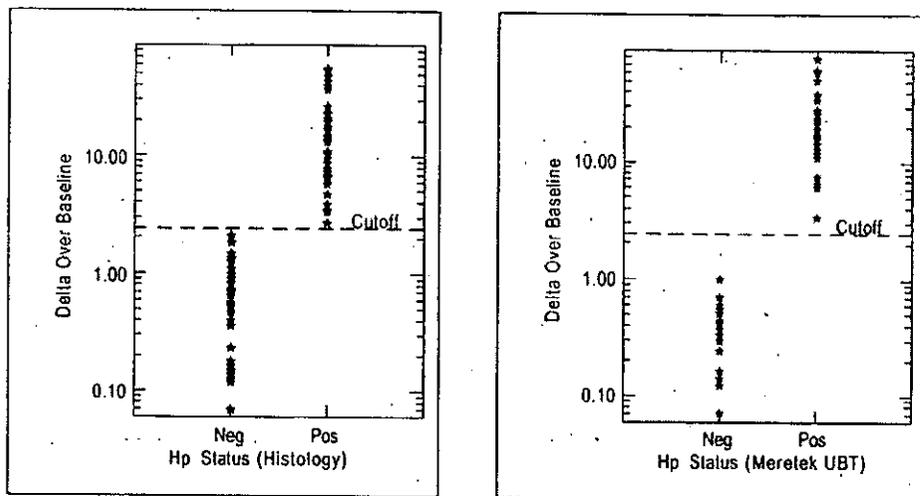
The Meretek UBT was subsequently validated in clinical trials of patients with documented duodenal ulcer disease (see Section 13.5).

BreathTek UBT

For the BreathTek UBT, the DOB cutoff values was determined to be 2.4 in a controlled study of 26 infected and 23 uninfected adult volunteers. Test subjects were judged to be in acceptable health based on the results of a medical history and physical examination and demonstrated no uncontrolled clinically significant abnormality other than, for some, symptoms of peptic ulcer. The Meretek UBT was used as the reference standard. The range of BreathTek UBT DOB values for the uninfected group was determined to be 0.0 to 1.0. The cutoff value was calculated by determining the BreathTek UBT result level at which negative and positive subjects were best distinguished by co-optimization of relative sensitivity and specificity. Distribution of BreathTek UBT DOB values in infected and uninfected groups in this study is shown in Figure 4b.

The 2.4 cutoff point for the BreathTek UBT was validated in an independent study by retrospective analysis of Clinical Field Trial data collected on 145 *H. pylori* negative and 105 *H. pylori* positive test subjects using the original Meretek UBT as a reference (see Section 13.4.2).

Figure 1a. Data Distribution before the Cutoff for Meretek UBT Figure 1b. Data Distribution before the Cutoff for BreathTek® UBT



10.1.4 Interpretation of Results for Adults

A DOB value of ≥ 2.4 is interpreted as diagnostically positive indicating the presence of urease associated with *H. pylori*. A DOB value of < 2.4 is interpreted as diagnostically negative indicating the absence of urease associated with *H. pylori*. The same DOB cutoff value applies to both initial diagnosis and post-treatment monitoring of *H. pylori* infection. The infrared spectrophotometer provides the interpretation of the DOB result on the test strip.

10.2 Pediatrics

10.2.1 The Test Method

The ratio of $^{13}\text{CO}_2$ to $^{12}\text{CO}_2$ in breath samples from children aged 3 - 17 years is determined by the UBiT-IR300 Infrared Spectrophotometer. Although the DOB result of the BreathTek UBT is provided by the UBiT-IR300 Infrared Spectrophotometer, urea hydrolysis rate (UHR) using the $\rho\text{UHR-CA}$, a web-based calculation program, is required to obtain the test results in pediatric patients.

10.2.2 Calculation of Results

The web-based *p*UHR-CA converts DOB to the UHR result in pediatric patients. The calculation incorporates the patient's anthropometric data (i.e., age, gender, height, and body weight) to calculate the CO₂ production rate in that patient. The UHR is calculated as shown below:

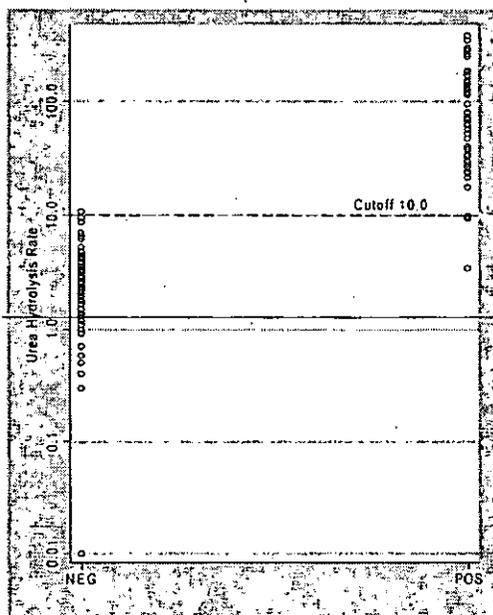
$$\text{UHR } (\mu\text{g}/\text{min}) = \text{DOB} \times \text{CO}_2 \text{ Production Rate} \times 0.3427$$

10.2.3 Determination of the Cutoff Point

UHR values from pediatric patients were first determined in a group of 312 asymptomatic preschool and school-age children aged 1 - 10 years in the Houston, Texas, area.⁸ An UHR cut-off value was determined to be 10.0 μg/min.

This UHR cut-off value was subsequently validated in two multi-center, controlled clinical studies of dyspeptic children aged 3 - 17 years using the BreathTek UBT kit and the UBiT-IR300 Infrared Spectrophotometer (see Section 13.5 for more information). *H. pylori* infection was established with an endoscopic composite reference method criteria consistent with the FDA guidance.^{10,11} Of the 176 analyzed study subjects, the range of UHR values were 0.0 - 10.9 μg/min for the 128 uninfected children and 3.4 - 403.8 μg/min for the 48 infected children. Distribution of the UHR values is shown in Figure 2. Note that the UHR scale is logarithmic; therefore, in displaying negative UHR values on a logarithmic scale, value between -5 and 0 were assigned a value of 0.01.

Figure 2: Data Distribution and Cutoff for UHR



10.2.4 Interpretation of Results for Pediatrics

A UHR value of $\geq 10 \mu\text{g}/\text{min}$ is interpreted as diagnostically positive indicating the presence of urease associated with *H. pylori*. A UHR value of $< 10 \mu\text{g}/\text{min}$ is interpreted as diagnostically negative indicating the absence of urease associated with *H. pylori*. The same UHR cutoff value applies to both initial diagnosis and post-treatment monitoring of *H. pylori* infection in children. The web-based *p*UHR-CA program provides the interpretation of the UHR result on the output of the calculation.

Go to: <https://pUHRCA.Otsuka-Us.com/pUHR-CA> to use the web-based *p*UHR-CA program.

11 Limitations of the Test

- 11.1 The BreathTek UBT should not be used until 4 weeks or more after the end of treatment for the eradication of *H. pylori* as earlier post-treatment assessment may give false negative results.
- 11.2 The performance characteristics for initial diagnosis and post-treatment monitoring for pediatric patients < 3 years of age have not been established for this test.
- 11.3 The specimen integrity of breath samples and reference gases stored in breath bags under ambient conditions has not been determined beyond 7 days.
- 11.4 A correlation between the number of *H. pylori* organisms in the stomach and the BreathTek UBT result has not been established.
- 11.5 Do not use DOB to determine the *H. pylori* positive or negative results in pediatric patients. Use the web-based pUHR-CA to calculate the UHR to obtain pediatric test results. Go to: <https://pUHRCA.Otsuka-Us.com/pUHR-CA>
- 11.6 The web-based pUHR-CA to calculate the UHR to obtain pediatric test has only been tested with Firefox and Internet Explorer.
- 11.7 No information is available in using POCone Infrared Spectrophotometer in analyzing pediatric samples.

12 Expected Values

12.1 Adults

DOB values for the BreathTek UBT were determined in a controlled clinical study of 26 infected and 23 uninfected adult volunteers. The Meretek UBT, an earlier version of the BreathTek UBT, was used as the reference method in the diagnosis of infection. The range of BreathTek UBT DOB values for the uninfected group was determined to be 0.0 to 1.0 (see Figure 1b).

12.2 Pediatrics

Of the 176 analyzed study subjects described in Section 10.2, the range of UHR values were 0.0 - 10.9 µg/min for the uninfected children and 3.4 - 403.8 µg/min for the infected children (see Figure 2).

13 Performance Characteristics

- 13.1 The primary outcome measure for clinical validation of both the Meretek UBT and the BreathTek UBT is a composite reference method consisting of histology and *H. pylori* culture of endoscopically-obtained gastric biopsies as well as a urease detection assay.^{10, 11}
- 13.2 *Analytical Performance Characteristics for the UBiT-IR300 Infrared Spectrophotometer. Refer to the Instruction Manual for the instrument.*
- 13.3 *Analytical Performance Characteristics for the POCone Infrared Spectrophotometer. Refer to the Instruction Manual for the instrument.*
- 13.4 Clinical Performance in Clinical Trials for Adults

13.4.1 Comparison of Meretek UBT with the Composite Reference Method in the Adult Population

a. Experimental Design

The clinical performance data presented here were collected from two (2) independent double-blind clinical field trials which involved treatment of *H. pylori* infection. The studies included 499 adult patients with duodenal ulcer disease at 75 clinical sites in the United States. Patients were tested for *H. pylori* infection initially by the composite reference method; histopathology, microbiological culture, urease detection test compared to the Meretek UBT, and at various post-treatment intervals throughout the study (using histopathology, microbiological culture, and the Meretek UBT). In these clinical trials, patients were treated with various combinations of clarithromycin, omeprazole and placebo. Note, however, that there is no evidence that differing treatment regimens affect the performance of the Meretek UBT.

1. **Histopathology**

Biopsy specimens, fixed with 10% buffered formalin were cut into 4-mm sections, stained with Genta stain and examined by an experienced pathologist.

2. **Microbiologic culture**

Culture was performed using fresh blood-based media, both selective and non-selective, at 37°C in 12% CO₂ in air with 98% humidity. *H. pylori* were identified by Gram stain, typical colony morphology, and biochemical properties (production of oxidase, catalase and urease).

3. **Urease detection test**

A biopsy specimen was tested for urease activity with the urease detection test according to the instructions in its package insert.

4. **The Meretek UBT for *H. pylori***

The diagnostic Meretek UBT was performed in accordance with procedures described in its package insert.

b. **Results**

Clinical performance results are presented in two-way contingency tables. In Table 1, the Meretek UBT results are compared with the composite reference method results (urease detection test, histology, and culture) for the initial patient visit¹⁰. In the same study, the Meretek UBT results were also compared with urease detection test and histology. The relative sensitivity and specificity of Meretek UBT for initial visit are 92.8% (95% CI: 90, 95) and 94.1% (95% CI: 71, 100), respectively, compared to urease detection test, and are 95.2% (95% CI: 93, 97) and 90.0% (95% CI: 74, 98), respectively, compared to histology. In Table 2, the Meretek UBT results are compared with the composite reference method results (histology and culture) for the post-treatment visits which occurred 4 weeks or more after end of treatment.

The exact binomial distribution was used to calculate the lower and upper limits of the 95% confidence intervals of the performance statistics. The confidence intervals are entered in parentheses following the point estimate of the statistic.

Table 1. Comparison with Composite Reference Method* in Adult Patients for Initial Visit (pre-treatment)

Endoscopy	Meretek UBT Results		
	Positive	Negative	Total
Positive	395	20	414
Negative	3	26	29
Total	398	46	444

* Composite reference method^{10,11} includes the urease detection test, histology, and culture for pre-treatment diagnosis

Sensitivity: 95.2% [95 % CI: (93, 97)]
Specificity: 89.7% [95% CI: (73, 98)]

Table 2. Comparison with Composite Reference Method* in Adult Patients for Post-Treatment Visits (4 weeks or more after End of Treatment (EOT))

	Meretek UBT Breath Test Results							
	1 Month EOT		3 Months EOT		6 Months EOT		1-6 Months Combined	
Endoscopy	Pos	Neg	Pos	Neg	Pos	Neg	Pos	Neg
Positive	187	6	123	8	91	5	401	19
Negative	5	97	4	87	2	80	11	264
Sensitivity (95% CI)	96.9 (93, 99)		93.9 (88, 97)		94.8 (88, 98)		95.5 (93, 97)	
Specificity (95% CI)	95.1 (89, 98)		95.6 (89, 99)		97.6 (92, 100)		96.0 (93, 98)	

* Composite reference method^{10,11} includes histology, urease detection test and culture for post-treatment monitoring.

Please note that the post-treatment performance characteristics at 1, 3 and 6 months after therapy are not statistically different. Therefore, the single best estimates of sensitivity and specificity are presented in the 1-6 Months Combined column.

Negative Predictive Value (NPV) for Post-Treatment Monitoring

Given the post-treatment sensitivity (95.5%) and specificity (96.0%) observed in these studies, and assuming a treatment efficacy of 90% (10% prevalence of residual *H. pylori* infection), the NPV of the Meretek UBT is greater than 99%. When efficacy of treatment drops to 50%, the NPV is still greater than 95%.

13.4.2 Comparison of the BreathTek UBT with the Meretek UBT in the Adult Population

a. Experimental Design

The clinical performance data presented here were collected from a prospective, cross-over clinical field trial designed to validate the BreathTek UBT test procedure and to examine the effect of pre-test fasting time on test performance. The study included 252 adult test subjects from Houston and Galveston, Texas. Subjects were judged to be in acceptable health based on the results of a medical history and physical examination and demonstrated no uncontrolled clinically significant abnormality other than, for some, symptoms of dyspepsia. Test subjects were tested for *H. pylori* infection using the Meretek UBT according to established procedure and with the BreathTek UBT under differing conditions of pre-test fasting times. Otherwise, no special instructions were given to subjects beyond those listed in the step-by-step procedures for administration of the Meretek UBT and BreathTek UBT. To minimize potential bias due to test order, the sequence of urea breath tests administered to each subject was randomized. All breath tests were administered to a given individual within 14 days of one another, most often and at a minimum, on successive days.

b. Results

It was demonstrated in the field trial that the BreathTek UBT may be administered at any time beyond 1 hour after consuming solid and/or liquid food.

Point estimates of Percent Agreement of the BreathTek UBT with Meretek UBT positive and negative results are listed in the contingency table (Table 3). The comparative method for determining the true diagnosis was the predicate device (Meretek UBT) rather than endoscopic methods. The exact binomial distribution was used to calculate the lower and upper limits of the

95% confidence intervals of the performance statistics. The confidence intervals are entered in parentheses following the point estimate of the statistic.

Table 3. Comparison of BreathTek UBT (≥ 1 -hour fast) with Meretek UBT

Meretek UBT	BreathTek UBT Results		
	Positive	Negative	Total
Positive	105	1	106
Negative	1	145	146
Total	106	146	252

Percent Agreement with Meretek UBT positive subjects: 99.1 % [95% CI: (94.9, 100.0)]
 Percent Agreement with Meretek UBT negative subjects: 99.3% [95% CI: (96.2, 100.0)]

13.4.3 Comparison of Gas Isotope Ratio Mass Spectrometry (GIRMS) and UBiT-IR300 Infrared Spectrophotometry Method in the Adult Population

A multi-center prospective-clinical-trial was conducted to compare the UBiT-IR300 method with the traditional GIRMS method. The study included a total of 320 adult test subjects enrolled at 4 physicians' office laboratory (POL) settings and at a clinical laboratory. The results of the clinical trial are provided in the Instructional Manual for the UBiT-IR300 Infrared Spectrophotometer (refer to the Application Note, ^{13}C -Urea Breath Test using the UBiT-IR300 Infrared Spectrophotometry System).

Table 4 shows the percent agreement of the UBiT-IR300 results as compared to the GIRMS method. Overall agreement was excellent at 99.06 percent.

Table 4. Agreement of UBiT-IR300 and GIRMS for ^{13}C urea breath test

UBiT-IR 300 Results	GIRMS Results		
	Positive	Negative	Total
Positive	115	1	116
Negative	2	202	204
Total	117	203	320

Percent Overall Agreement: 99.06% [95% CI: (97.35, 99.74)]
 Percent Positive Agreement: 98.29% [95% CI: (94.26, 99.70)]
 Percent Negative Agreement: 99.51% [95% CI: (97.49, 99.97)]

13.4.4 Comparison of UBiT-IR300 and POCone Infrared Spectrophotometry Methods in the Adult Population

A multi-center, prospective study was conducted to compare the POCone Infrared Spectrophotometer to the UBiT-IR300 Infrared Spectrophotometer for measuring $^{13}\text{CO}_2$ enrichment in breath. The study included a total of 220 adult test subjects enrolled at 5 physicians' office laboratory (POL) and point of care (POC) settings. The results of the clinical trial are provided in the Instruction Manual for the POCone Infrared Spectrophotometer (refer to the Application Note, ^{13}C -Urea Breath Test using the POCone Infrared Spectrophotometry System).

Table 5 shows the percent agreement of the POC-one results with the UBiT-IR300 results. Overall agreement was 99.55 percent.

Table 5. Agreement of POConc and UBiT-IR300 for the ¹³C urea breath test

POConc Results	UBiT-IR300 Results		
	Positive	Negative	Total
Positive	86	1	87
Negative	0	133	133
Total	86	134	220

Percent Overall Agreement: 99.55% [95% CI: (97.67, 99.98)]

Percent Positive Agreement: 100.00% [95% CI: (95.90, 100.00)]

Percent Negative Agreement: 99.25% [95% CI: (96.27, 99.96)]

13.5 Clinical Performance in Clinical Trials for Initial Diagnosis in Pediatric Patients

13.5.1 Experimental Design

The clinical performance data were collected from a multi-center, open-label study designed to compare the BreathTek UBT with endoscopic methods for the initial diagnosis of *H. pylori* in pediatric population. Subjects were symptomatic pediatric patients 3 to 17 years old undergoing diagnostic upper endoscopy at the determination of their treating pediatric gastroenterologist. Study enrollment was based on esophagogastroduodenoscopy (EGD) performed on each subject in proximity to the administration of the BreathTek UBT test.

The study enrolled 206 pediatric patients at five (5) U.S. investigational sites (New Orleans, Louisiana, Miami, Florida, Houston, Texas, Huntington, West Virginia and Detroit, Michigan) of which 176 subjects were evaluable for analysis. The reasons for exclusion are shown in Table 6 below.

Table 6. Reasons for subject exclusion¹

Reason	Number excluded
EGD not done	10
Culture and Histology not done	1
BreathTek UBT not done	3
Invalid UBT Results	10
Prohibited medication Use ²	4
Consent withdrawn	1
Lost Records	1
Total	30

Notes: ¹There were more than 1 reason for some subjects; however, they are counted once in this table.

²Proton pump inhibitor use N=3 subjects; Antibiotic use N=1.

13.5.2 Results - Comparison of BreathTek UBT UHR to the Composite Reference Method Criteria

The primary endpoint analysis was conducted to determine the sensitivity and specificity of the BreathTek UBT UHR to the composite reference method criteria for the 176 evaluable cases. Table 7 demonstrates the diagnostic performance of the BreathTek UBT (expressed as UHR) compared to the composite reference method criteria in pediatric patients aged 3-17 years old.

Table 7. Comparison of Composite Reference Method Criteria and BreathTek UBT (UHR) in Pediatric Patients for Initial Diagnosis

Endoscopic Composite Reference Method	¹³ C-UBT UHR							
	Age 3-5 Years		Age 6-12 Years		Age 13-18 Years		All Age Groups Combined	
	Pos	Neg	Pos	Neg	Pos	Neg	Pos	Neg
Infected	3	0	21	0	22	2	46	2
Not Infected	0	17	0	62	1	48	1	127
Sensitivity (95% CI)	100.0 (29.2, 100.0)		100.0 (83.9, 100.0)		91.7 (73.0, 99.0)		95.8 (85.7, 99.3)	
Specificity (95% CI)	100.0 (80.4, 100.0)		100.0 (94.2, 100.0)		98.0 (89.0, 100.0)		99.2 (95.7, 100.0)	

13.6 Clinical Performance in Clinical Trials for Post Treatment Monitoring in Pediatric Patients

13.6.1 Experimental Design

The study was a multi-center, open-labeled study designed to compare the BreathTek UBT with endoscopic methods for the post treatment monitoring of *H. pylori* in the pediatric population. Pediatric patients 3 to 17 years old who enrolled in this study had participated in the initial diagnosis study described above, and were diagnosed by upper endoscopy to be infected with *H. pylori* using the composite reference method criteria (e.g., histology, culture and urease test).

The study enrolled 22 pediatric patients at three (3) U.S. investigational sites (Houston, Texas, Detroit, Michigan and Huntington, West Virginia) of which 20 subjects were evaluable for analysis. The reasons for data exclusion were due to invalid UBT results and EDG was not performed.

The primary outcome variable of the BreathTek UBT was the UHR in comparison to the endoscopic findings of the composite reference method criteria. To determine the infection status following eradication therapy, these criteria were interpreted to include test results for all three *H. pylori* testing methods (histology, culture, rapid urease test). Results for all three *H. pylori* testing methods were available for all of the 20 evaluable cases. The primary endpoint analysis was conducted to determine the sensitivity and specificity of the BreathTek UBT (UHR) to the endoscopic composite reference method criteria for the 20 evaluable cases.

13.6.2 Results – Comparison of BreathTek UBT (UHR) to the Composite Reference Method Criteria

The observed sensitivity for UHR when compared to the composite reference method criteria is 83.3%, and the observed specificity is 100% (Table 8). Because of the small sample size, the results, including the 95% confidence intervals around the sensitivity and specificity, should be interpreted with caution.

Table 8. Comparison of Composite Reference Method Criteria and BreathTek UBT UHR in Pediatric Patients for Post Treatment Monitoring

		¹³ C-UBT UHR	
		N=20	
		Pos	Neg
Composite Reference Method Criteria	Infected	5	1
	Eradicated	0	14
Sensitivity		83.3% -[95% CI: (40.2, 99.2)]	
Specificity		100% [95% CI: (77.0, 100.0)]	
PPV		100%	
NPV		93.3%	

14 Bibliography

1. Marshall, B.J., Warren, J.R. Unidentified curved bacilli on gastric epithelium in active chronic gastritis, *Lancet*, June 4: 1273-1275; 1983.
2. Northfield T.C., Mendall M., Goggin P.M., (Eds), *Helicobacter pylori* infection. Pathophysiology, Epidemiology and Management. Kluwer Academic Publisher (1993).
3. Rathbone B.J., Heatley R.V., (Eds) *Helicobacter pylori* and Gastroduodenal Disease, Blackwell Scientific Publications, 2nd edition (1992).
4. Centers for Disease Control and Prevention. "The Key to Cure." Sept. 28, 2006. <http://www.cdc.gov/ulcer/keytocure.htm>. May 3, 2007.
5. W. D. Chey, B. C.Y. Wong, American College of Gastroenterology Guideline on the Management of *Helicobacter pylori* Infection, *Am J Gastroenterol* 2007;102:1808-1825.
6. Northfield T.C., Mendall M., Goggin P.M., (Eds), *Helicobacter pylori* infection. Pathophysiology, Epidemiology and Management. Kluwer Academic Publisher (1993, page 113).
7. Graham, D.Y., Runke, D., Anderson, S., Malaty, H.M., and Klein, P.D. Citric Acid as the Test Meal for the ¹³C-Urea Breath Test. *American Journal of Gastroenterology*, 5, 1214-1217; 1999.
8. Klein PD, Malaty HM, Czinn SJ, Emmons SC, Martin RF, and Graham DY. Normalizing Results of ¹³C-Urea Breath Testing for CO Production Rates in Children, *J Pediatr Gastroenterol Nutr* 1999;29:297-301.
9. Borriello, S.P., Reed, P.J., Dolby, J.M., Barclay, F.E. and Webster, A.D.B. Microbial and metabolic profile of achlorhydric stomach: comparison of pernicious anemia and hypogammaglobulinaemia. *J. Clin. Pathol.* 38, 946-953; 1985.
10. Guidance for Industry: *Helicobacter pylori*-Associated Duodenal Ulcer Disease in Adults: Developing Drugs for Treatment. October 2009. <http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM184500.pdf>, assessed October 2011.

11. Draft Guidance for Industry and FDA Staff: **Establishing the Performance Characteristics of *In Vitro* Diagnostic Devices for the Detection of *Helicobacter pylori***. September 23, 2010.
<http://www.fda.gov/downloads/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/UCM227110.pdf>, assessed October 2011.

15 Name and Place of Business

The BreathTek UBT for *H. pylori* Kit is manufactured for Medical Device Division of Otsuka America Pharmaceutical, Inc., 2440 Research Boulevard, Rockville, MD 20850.

For additional information, please call 1.888.637.3835 or visit www.BreathTek.com.

16 Trademarks

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17 Labeling Revision Information

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