

Submission ID #: 68219

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Title: Rapid Detection of Fecal Antigen of *Helicobacter pylori* Infection Based on Double Antibody Sandwich Detection Technology

Authors and Affiliations:

Xiuxian Chen[#], Caiping Gong[#], Xiaoxiao Wang, Ziyang Li, Xiaozhen Jiang, Jinxin Lai, Yanfei Luo

Laboratory Medicine, Guangdong Provincial People's Hospital (Guangdong Academy of Medical Sciences), Southern Medical University

[#]These authors contributed equally

Corresponding Authors:

Yanfei Luo 13544596975@163.com

Email Addresses for All Authors:

Xiuxian Chen	157292285@qq.com
Caiping Gong	caipinggong@163.com
Xiaoxiao Wang	916092097@qq.com
Ziyang Li	liziyang19970610@163.com
Xiaozhen Jiang	jiangxiaozhen@gdph.org.cn
Jinxin Lai	laijinxin@gdph.org.cn
Yanfei Luo	13544596975@163.com

Author Questionnaire

- 1. Microscopy:** Does your protocol require the use of a dissecting or stereomicroscope for performing a complex dissection, microinjection technique, or something similar? **No**

- 2. Software:** Does the part of your protocol being filmed include step-by-step descriptions of software usage? **No**

- 3. Filming location:** Will the filming need to take place in multiple locations? **No**

Current Protocol Length

Number of Steps: 8
Number of Shots: 14

Introduction

Videographer: Obtain headshots for all authors available at the filming location.

REQUIRED:

- 1.1. **Ziyan Li:** Our research scope is the detection technology of *Helicobacter pylori*. The question we want to answer is the principle, procedure, method and clinical application of *Helicobacter pylori* fecal antigen detection.
 - 1.1.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 2.4.2*

What technologies are currently used to advance research in your field?

- 1.2. **Ziyan Li:** Emerging technologies that promote the development of the field, such as real-time fluorescence quantitative PCR, CRISPR-Cas detection and biosensor and nanotechnology.
 - 1.2.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 2.6.1*

What advantage does your protocol offer compared to other techniques?

- 1.3. **Ziyan Li:** This technology has the advantages of being non-invasive, low-cost cost and easy operation. The advantage of our scheme is that it is suitable for screening in backward and underdeveloped areas. The rapid detection of *H. pylori* antigen in feces is a potential and promising tool for rapid and reliable diagnosis of *H. pylori* infection in remote and backward areas. **NOTE: This statement was changed during the shoot.**
 - 1.3.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 2.8.1*

Videographer: Obtain headshots for all authors available at the filming location.

Testimonial Questions:

How do you think publishing with JoVE will enhance the visibility and impact of your research?

- 1.4. **Ziyan Li:** I think the video format has attracted a large audience. Video documents minimize ambiguity and increase the adoption and citation of other laboratories. JoVE combines traditional peer review with video verification to ensure the robustness of science and technology.

- 1.4.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera. *Suggested B-roll: 2.8.1*

Ethics Title Card

This research has been approved by the Ethics Committee of Guangdong Provincial People's Hospital

Protocol

2. Procedure for fecal *H. pylori* infection antigen detection

Demonstrator: Xiuxian Chen

- 2.1. Place the test tube on the test bench [1], take the fecal sample collection container, and retrieve the sampling rod [2]. **NOTE: The VO has been edited.**
 - 2.4.1. Talent placing the tube on the bench at room temperature. **NOTE: This shot is moved here.**
 - 2.1.1. WIDE: Talent unscrewing the lid of a stool collection container and retrieving the sampling rod.
- 2.2. Insert the sampling rod into five different areas of the stool, ensuring that the threaded end is fully submerged in each position [1]. After sampling is complete, insert the sampling rod back into the reagent tube, ensuring that the total sample volume collected is approximately 5 to 50 milligrams [2].
 - 2.2.1. Talent holding the rod and inserting it into the stool.
 - 2.2.2. Talent carefully returning the sampling rod into the reagent tube and sealing it.
- 2.3. Shake the reagent tube side to side for approximately 10 seconds to thoroughly mix the stool sample in the diluent [1-TXT].
 - 2.3.1. Talent holding and shaking the reagent tube horizontally for 10 seconds, ensuring visible mixing. **TXT: Diluent: EDTA tetrasodium hydrate: 0.018 g/mL; NaCl: 0.01 g/mL**
- ~~2.4. Test the sample within 6 hours if kept at room temperature after sampling [1]. If immediate testing is not possible, store the sample at 2 to 8 degrees Celsius for up to 72 hours [2-TXT].~~
 - ~~2.4.1. Talent placing the tube in a room temperature on the bench. **NOTE: This shot is moved to 2.1.**~~
 - ~~2.4.2. Talent placing other tubes in a refrigerator. **TXT: Store at -25 to -15 °C for 6 months** **NOTE: This shot is moved to 2.8.**~~
- 2.5. Before testing, bring both refrigerated and frozen samples to room temperature [1-

TXT]. NOTE: Please move this step **before 2.1** (this will be the first step of the protocol section).

2.5.1. Talent placing samples from refrigerator and freezer onto a bench. **TXT: Samples may be frozen and thawed up to three times**

2.6. Open the white cover on the reagent tube and hold the tube upright [1]. Press the cap down completely to release the sample onto the test card [2]. Allow the sample to flow across the detection area labeled **T** and the quality control area labeled **C**, both of which are coated with mouse anti-*Helicobacter pylori* antibody [3].

2.6.1. Talent flipping open the small white cap while keeping the tube in vertical position.

2.6.2. Talent pressing the cap down to release the sample.

2.6.3. Close-up of the test card as liquid flows past areas marked **T** and **C** with visible reaction.

2.7. If the antigen is present, wait for 10 to 20 minutes to allow for chromogenic reaction formation in the detection area [1].

2.7.1. Talent setting a Timer.

2.8. Observe the presence or absence of colored lines in the test and control areas [1]. Compare the result with the color card to determine the concentration of *Helicobacter pylori* antigen [2-TXT]. After the experiment, store the sample at 2 to 8 degrees Celsius for up to 72 hours [3-TXT]. NOTE: The VO has been edited.

2.8.1. Talent examining the test card.

2.8.2. Shot of the test card being held next to a printed color interpretation card, with finger pointing to matching color intensity. **TXT: Confirm the results after performing DNA extraction and qPCR**

2.4.2 Talent placing other tubes in a refrigerator. **TXT: Store at -25 to -15 °C for 6 months** NOTE: This shot is moved here.

Results

3. Results

- 3.1. Among the fecal *Helicobacter pylori* antigen results of 261 subjects, 52 were positive [1] and 209 were negative [2].
 - 3.1.1. LAB MEDIA: Table 3 [Video editor: Highlight 52](#)
 - 3.1.2. LAB MEDIA: Table 3 [Video editor: Highlight 209](#)

- 3.2. Among the results of qPCR with the feces of 261 subjects, 83 were positive [1] and 178 were negative [2].
 - 3.2.1. LAB MEDIA: Table 2 [Video editor: Highlight 83](#)
 - 3.2.2. LAB MEDIA: Table 2 [Video editor: Highlight 178](#)

- 3.3. The positive rate in Shitan village was 19.92% [1], which was slightly lower than the national positive rate of 42.8% [2].
 - 3.3.1. LAB MEDIA: Table 3 [Video editor: Highlight 19.92%](#)
 - 3.3.2. LAB MEDIA: Table 3 [Video editor: Highlight 42.8%](#)

Pronunciation Guides:

1. Fecal

Pronunciation link:

<https://www.merriam-webster.com/dictionary/fecal>

IPA: /'fi:kəl/

Phonetic Spelling: fee-kuhl

2. Reagent

Pronunciation link:

<https://www.merriam-webster.com/dictionary/reagent>

IPA: /ri'eɪdʒənt/

Phonetic Spelling: ree-ay-juhnt

3. EDTA (Ethylenediaminetetraacetic acid)

Pronunciation link:

<https://www.howtopronounce.com/edta>

IPA: /,i:,di:ti:'eɪ/ (acronym)

Phonetic Spelling: ee-dee-tee-ay

4. Tetrasodium

Pronunciation link:

<https://www.howtopronounce.com/tetrasodium>

IPA: /,tetrə'soʊdiəm/

Phonetic Spelling: teh-truh-soh-dee-uhm

5. Helicobacter pylori

Pronunciation link:

<https://www.merriam-webster.com/medical/Helicobacter%20pylori>

IPA: /,helɪkəʊ'bæktər paɪ'lo:raɪ/

Phonetic Spelling: heh-li-koh-bak-ter pie-lor-eye

6. Chromogenic

Pronunciation link:

<https://www.merriam-webster.com/dictionary/chromogenic>

IPA: /,kroʊmə'dʒɛnɪk/

Phonetic Spelling: kroh-muh-jeh-nik

7. qPCR (quantitative Polymerase Chain Reaction)

Pronunciation link:

<https://www.howtopronounce.com/qpcr>

IPA: /,kju:,pi:si:'ɑr/

Phonetic Spelling: kyoo-pee-see-ar

8. Antigen

Pronunciation link:

<https://www.merriam-webster.com/dictionary/antigen>

IPA: /'æn.tə.dʒən/

Phonetic Spelling: an-tuh-jen

9. Stool

Pronunciation link:

<https://www.merriam-webster.com/dictionary/stool>

IPA: /stu:l/

Phonetic Spelling: stool