Regular arrangement of collecting venules (RAC) as an endoscopic marker for exclusion of Helicobacter pylori (H. pylori) infection: A systematic review and meta-analysis

Fan Yu¹, shaoyou qin¹, Song Wang², and Jiangbin Wang¹

¹China-Japan Union Hospital of Jilin University
²The First Hospital of Jilin University

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Abstract

Background: Helicobacter pylori (H. pylori) is the most common cause of gastric cancer. Growing evidence suggests that the regular arrangement of collecting venules (RAC) can be used as an endoscopic marker to diagnose H. pylori infection. However, data on the diagnostic accuracy of RAC for H. pylori infection are conflicting. We performed a systematic review and meta-analysis of relevant studies to determine the diagnostic accuracy and clinical utility of RAC for the diagnosis of H. pylori infection. Methods: We systematically searched PubMed, Embase, Web of Science, and the Cochrane Library between inception and Oct 29, 2020, for studies that assessed the diagnostic accuracy of RAC for H. pylori infection. Results: The literature search yielded 2921 nonduplicated screened titles, of which 58 underwent full-text review. Fifteen studies, representing a total of 6621 patients, met the inclusion criteria. The area under the summary receiver operating characteristic curve was 0.98 (95% CI 0.96 to 0.99). The pooled estimates for RAC were 0.98 (95% CI 0.95 to 0.99) for sensitivity and 0.75 (95% CI 0.54 to 0.88) for specificity. The pooled positive likelihood ratio (PLR) and negative likelihood ratio (NLR) were 3.8 (95% CI 1.9 to 7.7) and 0.03 (95% CI 0.02 to 0.07), respectively. Conclusions: RAC can be used as an endoscopic marker for exclusion of H. pylori infection. However, it cannot be recommended as a single indicator for the confirmation of H. pylori infection. The conclusion of this study should be treated with caution because significant heterogeneity exists between the evaluated studies.

Regular arrangement of collecting venules (RAC) as an endoscopic marker for exclusion of Helicobacter pylori (H. pylori) infection: A systematic review and meta-analysis

Running title: RAC as an endoscopic marker for Hp infection

Fan Yu¹, Shaoyou Qin¹, Song Wang²,* , Jiangbin Wang¹,*

¹ Department of Gastroenterology and Hepatology, China-Japan Union Hospital of Jilin University, Changchun, China.
² Department of Urology, The First Hospital of Jilin University, Changchun, China.

* Correspondence: Jiangbin Wang, Department of Gastroenterology and Hepatology, China-Japan Union Hospital of Jilin University, No. 126 Xiantai Street, Changchun 130033, China. E-mail: jiangbin@jlu.edu.cn . Song Wang, Department of Urology, The First Hospital of Jilin University, No. 71 Xinmin Street, Changchun 130021, China. E-mail: w_s@jlu.edu.cn.

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**KEYWORDS:** regular arrangement of collecting venules, endoscopic marker, Helicobacter pylori

1 | INTRODUCTION

Helicobacter pylori (H. pylori) is a gram-negative bacterium affecting up to 50% of the population worldwide and is the most common cause of chronic gastritis, peptic ulcer, and gastric cancer(1-3). Eradication of chronic H. pylori infection significantly reduces gastric cancer risk(4-6). A systematic review including ten randomized controlled trials provided evidence that H. pylori eradication therapy reduces gastric cancer incidence in healthy individuals and patients with gastric neoplasia(7). The strong association between H. pylori and gastric cancer risk highlights the importance of adequate detection and eradication of H. pylori in clinical practice.

Conventional examinations for the diagnosis of H. pylori infection include noninvasive and gastroscopic biopsy-based tests. These examinations, however, cannot evaluate endoscopic findings associated with H. pylori infection (8-13). Recently, many investigators have attempted to characterize endoscopic features indicative of H. pylori infection. Several endoscopic features in the Kyoto classification of gastritis, such as the regular arrangement of collecting venules (RAC), diffuse redness, or atrophy, have been proposed to evaluate the status of H. pylori infection and the potential risk of developing gastric cancer(14).

RAC is an endoscopic feature defined as numerous red dots regularly distributed over the entire gastric body(15, 16). The presence of RAC at the level of the distal part of the lesser gastric curvature has been regarded as a characteristic endoscopic feature of the H. pylori-negative normal stomach. However, data on the diagnostic accuracy of RAC as an endoscopic marker for the diagnosis of H. pylori infection are conflicting, with reported sensitivity ranging from 86% to 100% and specificity ranging from 7% to 98%(17-31). Variations in the sensitivity and specificity of RAC for H. pylori infection in these studies highlight the need for a comprehensive evaluation of the diagnostic performance of RAC before broader application. Therefore, this study was conducted to determine the diagnostic accuracy and clinical utility of RAC for the diagnosis of H. pylori infection.

2 | METHODS

This study followed the Preferred Reporting Items for a Systematic Review and Meta-Analysis of Diagnostic Test Accuracy Studies (PRISMA-DTA)(32, 33). The study protocol was registered in PROSPERO prior to study selection (Registration number: CRD42020216437).

2.1 | Data sources and search strategy

We systematically searched PubMed, Embase, Web of Science, and the Cochrane Library for studies that
assessed the diagnostic accuracy of RAC for H. pylori infection. Our medical subject heading terms (for PubMed), EMTREE terms (for Embase), and search text (for others) were "("Kyoto classification of gastritis" OR "endoscopic findings" OR (endoscopic features) OR "regular arrangement of collecting venules" OR RAC) AND ("Helicobacter pylori" OR H. pylori)." The detailed search strategy is shown in Supplementary Table 1. We searched the databases between inception and Oct 29, 2020. The reference list of each primary study identified was also searched.

2.2 | Study selection criteria
The inclusion criteria for studies were as follows: (1) Assessed the accuracy of RAC for determining the status of H. pylori infection; (2) Used a rapid urease test, serum H. pylori antibody, and histological examination to define the reference standard for H. pylori infection; and (3) provided sufficient information (true and false positives and negatives) to construct the 2×2 contingency table.

The exclusion criteria were as follows: 1) reviews, letters, and case reports; 2) studies with insufficient data to construct the 2×2 table; and (3) studies not written in English.

2.3 | Data extraction and quality assessment
Two researchers independently examined all potentially relevant papers, extracted the data, and assessed the quality of retrieved studies. Any disagreement was resolved by consensus. The extracted data were characteristics of the study population (adults or children), region, sample size, types of endoscopy devices, gold standard test for H. pylori infection, prevalence of H. pylori infection, and true and false positives and negatives. We used the Quality Assessment of Diagnostic Accuracy Studies 2 (QUADAS-2) checklist to evaluate the quality of the included studies(34).

2.4 | Statistical analysis
We used the extracted True positives (TP), True negatives (TN), False positives (FP), and False negatives (FN) data to calculate the pooled sensitivity, specificity, positive likelihood ratio (PLR), negative likelihood ratio (NLR), and diagnostic odds ratio (DOR) based on bivariate generalized linear mixed modeling(35). We constructed a summary receiver operating characteristic (SROC) curve. The overall diagnostic accuracy of RAC for H. pylori was determined by calculating the area under the SROC curve (AUROC). We calculated the Q-value and I² to assess heterogeneity between studies, which can be quantified as low, moderate, and high, with upper limits of 25%, 50%, and 75% for the I² statistics, respectively(36). Meta-regression and subgroup analysis were performed to identify potential factors that could contribute to heterogeneity between studies.

We used the Fagan nomogram to estimate a patient’s posttest probability of being infected with H. pylori based on a pretest probability. Furthermore, a likelihood ratio scattergram was used to evaluate the clinical utility of RAC for the diagnosis of H. pylori infection. In detail, a PLR>10 suggests that the index test can be used for confirmation of H. pylori infection, and an NLR<0.1 suggests it can be used for the exclusion of H. pylori infection. Publication bias was assessed using the Deeks funnel plot asymmetry test. We used the MIDAs module for STATA (version 12, StataCorp LP in College Station, TX) for the bivariate summary receiver operating curve analysis. Revman software (version 5.3, Cochrane Collaboration) was used for quality assessment.

3 | RESULTS

3.1 | Literature search and characteristics of the included studies
This meta-analysis was organized according to the PRISMA statement. Figure 1 summarizes the results of the literature search and study selection. Our database search retrieved 4232 articles. After removing duplicated records, we excluded 1311. After reviewing the titles and abstracts, we excluded 2863. After a full-text review, we initially identified 19 potentially eligible studies. Subsequently, we excluded four studies for reasons reported in the PRISMA diagram. Supplementary Table 2 summarizes the main characteristics
of these four studies excluded at the eligibility stage along with the exact reasons for exclusion. The search of the reference lists of the identified articles did not identify any additional relevant articles.

Table 1 summarizes the baseline characteristics of the eligible studies. All eligible studies were published between 2002 and 2020. A total of 15 studies with 6621 participants were included in the meta-analysis. Of these, one was conducted in America, 2 in Europe, and the remaining 12 in Asia. Sample sizes ranged from 52 to 1875. H. pylori infection was assessed using a rapid urease test in 8 studies, serum H. pylori antibody in 6 studies, and histological examination in 13 studies, suggesting that each individual study might have used more than one test for H. pylori infection at the same time. The RAC pattern was evaluated by standard endoscopy in 10 studies and by high-resolution magnified endoscopy in 7 studies, suggesting that each individual study might have used both standard endoscopy and high-resolution endoscopy at the same time. The prevalence of H. pylori among the included studies ranged between 15.3% and 76% (mean 46.1%).

3.2 | Quality of the included studies

We used QUADAS-2 criteria to evaluate each of the included studies in 4 domains: patient selection, index test, reference standard, and test timing. For the included studies, “Patient Selection” and “Flow and Timing” revealed slight shortcomings (13.3% and 33.3%, respectively), which may indicate bias regarding inclusion. Overall, the study quality was satisfactory (Supplementary Figure 1).

3.3 | Pooled results

The pooled sensitivity and specificity of RAC for the endoscopic diagnosis of H. pylori infection were 0.98 (95% CI 0.95 to 0.99) and 0.75 (95% CI 0.54 to 0.88), respectively (Figure 2). The pooled PLR and NLR were 3.8 (95% CI 1.9 to 7.7) and 0.03 (95% CI 0.02 to 0.07), respectively (Supplementary Figure 2). The DOR was 115 (95% CI 36 to 365) (Supplementary Figure 3). The area under the summary receiver operating characteristic curve was 0.98 (95% CI 0.96 to 0.99), suggesting a high overall diagnostic accuracy for H. pylori infection (Figure 3).

3.4 | Evaluation of clinical utility

The clinical utility of RAC was assessed by utilizing like-hood ratios to simulate a Fagan nomogram. With a 46% pretest probability of H. pylori infection, the posttest probabilities of H. pylori infection were 77% (with a positive test result) and 3% (with a negative test result). The Fagan nomogram revealed that the posttest probability increased by 31% in patients with a positive test but decreased by 43% in patients with a negative test, suggesting that RAC was useful in clinical practice (Figure 4). Furthermore, the likelihood ratio scattergram showed a PLR of <10 and an NLR of <0.1, suggesting that RAC can be used as an endoscopic marker for exclusion rather than confirmation of H. pylori infection (Figure 5).

3.5 | Publication bias and heterogeneity

We used the Deeks plot asymmetry test to examine publication bias. The funnel plot did not reveal significant publication bias (p=0.37) (Supplementary Figure 4). Heterogeneity among the included studies was measured using the Cochran-Q method and $I^2$. Substantial heterogeneity existed among the studies (overall $I^2$ for bivariate model 99.95%, 95% CI 94 to 100). The proportion of heterogeneity likely due to the threshold effect was small ($p<0.05$).

3.6 | Univariable meta-regression and subgroup analysis

To identify the source of heterogeneity, we performed meta-regression analyses. Three study characteristics were used as covariates for univariable meta-regression analysis, including whether patients with a history of H. pylori infection were excluded, whether enrollment was consecutive, and whether a single reference standard for H. pylori infection was used (Supplementary Figure 5). The results showed that whether patients with a history of H. pylori infection were excluded significantly affected the sensitivity of RAC for the diagnosis of H. pylori infection ($p<0.01$). Likewise, whether enrollment was consecutive significantly affected the sensitivity of RAC ($p<0.05$). Thus, the heterogeneity could partly be explained by meta-regression analysis.
DISCUSSION

Several endoscopic features of the gastric mucosa have been proposed for the diagnosis of H. pylori infection, such as RAC, diffuse or spotty redness, mucosal swelling, and nodular changes(1). Our study included 15 relevant studies(17-31) assessing the diagnostic accuracy of RAC for H. pylori, and the results suggest that RAC has a high diagnostic accuracy for H. pylori infection, with a pooled estimate of 0.98 (95% CI 0.95 to 0.99) for sensitivity and 0.75 (95% CI 0.54 to 0.88) for specificity.

Many studies have evaluated the diagnostic accuracy of RAC as a single endoscopic feature for H. pylori, with inconclusive results. A prospective study in a European population enrolling 140 adults found that the presence of RAC in the lesser curvature evaluated with high-definition endoscopy can help identify patients without H. pylori, with a sensitivity of 100% and a specificity of 48.96%(20). A prospective study in Korea involving 617 individuals concluded that the RAC pattern observed using standard endoscopy could predict H. pylori infection status, with a sensitivity of 93.3% and a specificity of 89.1%(19). Again, in a Brazilian cohort of 99 individuals, Machado et al. investigated the diagnostic accuracy of RAC for H. pylori, suggesting that RAC can provide a sensitivity of 96.9% and a specificity of 88.1% for diagnosing H. pylori infection(26).

In a meta-analysis from 2020 including studies published between 2002 and 2019, Li and colleagues concluded that RAC is a valuable endoscopic finding for predicting patients without H. pylori infection(37). However, this meta-analysis has substantial shortcomings in its quantitative data analysis for heterogeneity, with a reported $I^2$ of 0 (95% CI: 0-100%). In contrast, our study found significant heterogeneity between studies, with an $I^2$ of 99.95% (95% CI: 99-100%). Furthermore, we performed a univariable meta-regression and subgroup analysis to identify the potential source of heterogeneity between the studies. The results showed that whether patients with a history of H. pylori infection were excluded and whether consecutive enrollment was used in each study significantly affected the sensitivity of RAC, indicating that these two factors might partly contribute to the heterogeneity between studies.

Likelihood ratios and posttest probabilities are also of importance because they provide information regarding the likelihood that a patient with a positive or negative test has H. pylori infection or not. In our study, a positive likelihood ratio of 3.8 implies that a person with H. pylori infection is 3.8 times more likely to have a positive test result than is a healthy person. Likewise, a negative likelihood ratio of 0.03 indicates that a person without H. pylori infection is 33 times more likely to have a negative test result than is a person with H. pylori infection. Therefore, RAC can serve as a reliable marker for exclusion rather than confirmation of H. pylori infection.

As our results show, RAC is not a perfect endoscopic marker for the diagnosis of H. pylori. Although RAC has a high overall diagnostic accuracy for H. pylori infection, it is not a good predictor for confirming H. pylori infection because of its low positive likelihood ratio. The pathophysiological process of H. pylori infection is complex and can be affected by many factors, which probably contributes to the variations in endoscopic findings(38-41). Therefore, a single endoscopic feature of RAC might be insufficient for confirmation of H. pylori infection because of the relatively low positive likelihood ratio, thus highlighting the need for a combination of multiple endoscopic features to confirm H. pylori infection.

This study has limitations. First, we detected substantial heterogeneity between the studies and found that two of the study characteristics contributed to the observed heterogeneity. However, there are probably additional study characteristics that have impacted study heterogeneity but have not been addressed, such as sample size, geographic area, variation in the methods used for the diagnosis of H. pylori between studies, and variation in the anatomic location where endoscopic physicians observed the RAC, because these factors were challenging to quantitatively analyze. Second, we only included studies written in English, which might have led to selection bias, although the funnel plot did not reveal significant publication bias (p=0.37). Third, this systematic review did not include studies that assessed endoscopic features other than RAC that are possibly associated with H. pylori infection. Despite these limitations, this meta-analysis investigating the diagnostic accuracy of RAC for H. pylori infection is the largest and most comprehensive assessment to date.

CONCLUSION

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In conclusion, RAC can be used as an endoscopic marker for the exclusion of H. pylori infection. However, it cannot be recommended as a single indicator for the confirmation of H. pylori infection. The conclusion of this study should be treated with caution because significant heterogeneity exists between the included studies.

Conflict of interest
The authors have declared no conflict of interest.

Author contributions
Fan Yu, Shaoyou Qin, and Jiangbin Wang designed the study. Fan Yu wrote the manuscript. Fan Yu, Shaoyou Qin, and Song Wang searched the databases, selected the studies, assessed the quality of the included studies, and performed the statistical analysis. Jiangbin Wang and Song Wang approved the last version and should be regarded as co-corresponding authors. Fan Yu and Shaoyou Qin contributed equally to the manuscript and should be regarded as co-first authors.

Data availability statement
Research data could be shared upon reasonable request to the corresponding author.

References:


Table 1. Characteristics of the studies included in the meta-analysis

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>Region</th>
<th>Population</th>
<th>Simple size(n)</th>
<th>Endoscopy Devices</th>
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<td>Anagnostopoulos GK[18]</td>
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<td>Cho JH[19]</td>
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<td>Korea</td>
<td>Adults</td>
<td>617</td>
<td>Standard Endosco</td>
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<td>2019</td>
<td>Spain</td>
<td>Adults</td>
<td>140</td>
<td>HD Endoscopy</td>
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<tr>
<td>Gonen C[21]</td>
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<td>Turkey</td>
<td>Adults</td>
<td>129</td>
<td>HD Magnifying En</td>
</tr>
<tr>
<td>Author</td>
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<td>Region</td>
<td>Population</td>
<td>Simple size(n)</td>
<td>Endoscopy Devices for index test (RAC)</td>
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<td>--------------------------</td>
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<td>Adults</td>
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<td>Standard Endoscopy</td>
</tr>
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<td>Children and young adult (8-29)</td>
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<td>Japan</td>
<td>14-86 yrs</td>
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RAC: Regular arrangement of collecting venules; H.pylori: Helicobacter pylori; TP: True positives; TN: True negatives; FP: False positives; FN: False negatives; HD: High Definition; IHC: Immunohistochemistry; CLO: Campylobacter-like organism; RUT: rapid urease test; UBT: urea breath test; SAT: stool antigen test; HE: hematoxylin-eosin.

**Fig. 1** The PRISMA flow diagram of the systematic review and meta-analysis.

**Fig. 2** Sensitivity and specificity of RAC for the diagnosis of H. pylori infection.

**Fig. 3** Summary receiver operating characteristic curve of the accuracy of RAC for the diagnosis of H. pylori infection.

**Fig. 4** Fagan nomogram of RAC for the diagnosis of H. pylori infection.

**Fig. 5** Likelihood ratio scattergram of RAC for the diagnosis of H. pylori infection.

**Fig. S1** Quality assessment of the included studies.

**Fig. S2** Likelihood ratios of RAC for the diagnosis of H. pylori infection.

**Fig. S3** Diagnostic odds ratio of RAC for the diagnosis of H. pylori infection.

**Fig. S4** Deeks’ funnel plot for assessing the publication bias of RAC for the diagnosis of H. pylori infection.

**Fig. S5** Univariable meta-regression and subgroup analyses for RAC in the diagnosis of H. pylori infection. (Variable 1, Exclusion of past Hp infection; Variable 2, A single reference standard for H.pylori infection; Variable 3, Consecutive enrollment.)

**Table S1** Detailed search strategies used in this meta-analysis

**Table S2** Studies excluded in this meta-analysis according to the PRISMA flow diagram

**Hosted file**

Records identified through database searching: PubMed, Embase, Cochrane library and Web of science from January 1975 to 25 October 2020 (n=4232)

Records after duplicated removed (n=2921)

Records screened (n=58)

Records excluded after reading the abstracts (n=2863)

Full-text articles and congress abstracts assessed for eligibility (n=19)

Records excluded after reading full-text (n=39)

Full-text articles excluded for reasons specified in table S1 (n=4)

Studies included in quantitative synthesis (meta-analysis) (n=15)

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Q=0.00, df=14.00, p=0.00
Q=190.48, df=14.00, p=0.00
I² = 95.17 [23.66 - 96.00]