Safety and Accuracy of Suction Rectal Biopsy in Preterm Infants

Yanan Zhang  
Capital Medical University Beijing Children's Hospital

Jinshi Huang (✉ jsdr2002@126.com)  
Capital Medical University Beijing Children's Hospital  https://orcid.org/0000-0003-2857-632X

Yongwei Chen  
Capital Medical University Beijing Children's Hospital

Shen Yang  
Capital Medical University Beijing Children's Hospital

Yichao Gu  
Capital Medical University Beijing Children's Hospital

Kaiyun Hua  
Capital Medical University Beijing Children's Hospital

Yong Zhao  
Capital Medical University Beijing Children's Hospital

Research article

Keywords: suction rectal biopsy, Hirschsprung disease, preterm infants, safety, accuracy

Posted Date: October 21st, 2020

DOI: https://doi.org/10.21203/rs.3.rs-93842/v1

License: ☺️ ☑️ This work is licensed under a Creative Commons Attribution 4.0 International License.  
Read Full License
Abstract

**Purpose:** Little attention were paid in the diagnosis of preterm infants with Hirschsprung disease (HD) in most pediatric surgeons. We aimed to explore the safety and accuracy of suction rectal biopsy (SRB) in preterm infants.

**Methods:** A retrospective review was conducted on 45 preterm patients who underwent SRB from 2015 to 2019 in our hospital. We collected the clinical characteristics and pathology results of the patients, and followed up. The sensitivity and specificity of SRB for HD diagnosis were calculated.

**Results:** The median gestational age of the patients was 35 weeks (range: 28.9 to 36.9 weeks) while the median gestational age at biopsy was 38.6 weeks (range: 33.4 to 60.0 weeks), and the median weight was 2790 g (range: 1580 to 4100 g). Fifteen cases (33.3%) were positive for HD which were confirmed after the pull-through surgery. The ganglion cells presented on 30 cases. And the diagnosis of HD was excluded in 29 patients after discharge follow-up. The sensitivity was supposed to be 93.7%-100% and specificity was 100%. Among the patients whose biopsy age is < 37 weeks (10 cases) or biopsy weight is < 2000 g (5 cases), no complications occurred after SRB.

**Conclusion:** SRB is safe and accurate in late preterm infants.

Introduction

Hirschsprung disease (HD) in premature infants has attracted more attentions than past as the preterm infants’ medical condition have been improved in recent years. Delayed passage of meconium is common in premature infants. The incidence appears to be inversely correlated with gestational age [1]. In the past, the occurrence of HD in premature infants was considered uncommon. Symptoms of the delayed passage was attributed to the immature development of its enteric nervous system [2]. One series in 2013 described a cohort of premature newborns with HD treated at a single center shows that HD occurs significantly less often in premature infants than in term infants,hence SRB should be used more selectively in preterm infants [3]. But in recent years, two large-population-based studies show that preterm HD (PHD) are comparable to term infants HD in occurrence. The incidences of both are about 1/5000. And the PHD accounted for about 6% of all HD. Diagnosis of HD is often delayed in premature newborns [4, 5]. In the view of embryonic development, it is currently believed that HD is caused by the destruction of the early embryonic neuroblast migration process. The migration of neuroblasts generally occurs between 5 to 12 weeks of gestation in human embryos [6, 7]. So confirming PHD early is beneficial for clinicians to make the appropriate treatment plans and to shift focus to other diagnoses for those patients excluded of HD. Suction rectal biopsy is considered the gold standard for diagnosis of HD. However, suction rectal biopsy (SRB) is often delayed until the child reaches term adjusted gestational age owing to a belief that SRB is unreliable in preterm infants. The objective of this study was to quantify the sensitivity, specificity, complication and the outcomes of preterm infants by reviewing the data of our preterm infant suspected HD.
Methods

Patients

We performed a retrospective review of the preterm infants (gestational age < 37 weeks) who underwent SRB from December 2015 to June 2019. We selected and documented the following data: sex, gestational age of birth, gestational age of biopsy, weight of birth, weight of biopsy, biopsy results, procedural complications, surgical procedures performed, and clinical outcome.

Procedure of SRB

All SRBs were performed by the pediatric surgical fellow or attending surgeon. We used the standard techniques described on the RBi2 website [8]. A rectal irrigation was performed before the biopsy. The device with the cartridge was inserted into the anus to approximately 3 cm to 4 cm from the anal verge. At least 2 biopsies were performed posteriorly or laterally (6 o’clock or 4, 8 o’clock). Specimens were delivered to pathology in formalin. Mucosa and submucosa were contained in each biopsy basically.

The biopsies were processed and embedded by routine procedures. At least 6 slides were made from one tissue, could had been more according to the stained results. Hematoxylin and eosin stain and calretinin immunohistochemical stain were performed in each patient’s tissue. All pathology slides were reviewed by a pathologist with subspecialty training in pediatric pathology.

Statistical analysis

We analyzed all the data on SPSS 23.0. Continuous variables were presented as the mean with standard deviation or median and interquartile range if the normality hypothesis test rejected the null hypothesis of normal distribution. Categorical variables were reported as counts and percentages. The sensitivity and specificity of SRB for HD diagnosis were calculated.

Results

Patient characteristics

There were 45 preterm infants underwent SRB during the study period (Table 1). describes the demographic information of patients in the study cohort. Males comprised 48.9% of the cohort while females 51.1%. The median age at birth was 35 weeks (range: 28.9 to 36.9 weeks), the median age at biopsy was 38.6 weeks (range: 33.4 to 60 weeks). The median weight at biopsy was 2790 g (range: 1580 to 4100 g). No biopsy complications were found in our cases.

Pathological features, treatment strategies, and prognosis

As shown in Table 2, histologic examination reports were described as 3 types. One group had normal ganglion cells in the submucosa, calretinin (+). The HD cases were absent of ganglion cells with
hypertrophic nerves while calretinin (-). The third type was described as present of the ganglion cells in the submucosa, but part of them were hypoplasia ganglion cell while calretinin (+). All 15 cases whose biopsy was absent of ganglion cells underwent a surgery: pull through operation (n = 12); colostomy (n = 2); ileostomy (n = 1). The patients with colostomy underwent the pull through operation after 3 months. The patients with ileostomy was confirmed total colonic HD. The patient’s parents abandoned further treatment, and this case had no further follow-up records. The diagnosis of HD was confirmed through demonstration of an aganglionic segment on final pathology in all 15 cases. Among the 30 patients with ganglion cells on initial biopsy, the diagnosis of HD was excluded in 29 patients in after-discharge follow-up. The median follow-up period was 2 years (range: 1 to 4 years). Five of them underwent the ileostomy because of the necrotizing enterocolitis (NEC) or bowel resection for the gut stenosis after NEC. Close surgeries were performed after 3 months. Symptoms resolved with appropriate treatment in each case. One case died at home few days after abandonment of treatment.

Among the 15 PHD patients (10 boys and 5 girls), 8 cases had rectosigmoid disease, 6 cases had long segment disease and 1 had total colonic aganglionosis. The median gestational age of the operation was 38.6 weeks (range: 36.3 to 43.4 weeks), and the median weight was 2940 g (range: 2100 to 4000 g). Among all cases, there were 4 cases whose weight was < 2500 g.

The accuracy of SRB in preterm infants

If we consider the case that died after treatments abandonment, whose biopsy result excluded HD, as a false-negative since no autopsy was performed, the sensitivity of SRB was 93.7% (95% CI 67.7-99.7%) and specificity was 100% (95% CI 85.4-100.0%) in the cohort. If the case was true-negative, the sensitive of SRB was 100% (95% CI 74.7-99.7%) and specific was 100% (95% CI 85.9-100.0%).

Discussion

SRB has proven to be a valuable diagnosing technique of HD since its recording in 1965, especially for its high accuracy and minimal invasion. It has been proved that the sensitivity and specificity of SRB are 96.8% and 99.4% in some systematic reviews in recent years [9]. Most clinicians will suggest preterm patients who are suspected of HD to not perform SRB until reaching term-corrected age or gain more weight. The sensitivity and specificity of SRB on term-corrected infants ranged from 46–100% and 97–100% [10–12]. It has been confirmed that the intestinal wall muscle layer will increase with age, and the intestinal wall of preterm infants were thinner than term infants. Therefore, in theory, the risk of bowel perforation in preterm infants undergoing rectal biopsy is greater than that of full-term or older children. But D.R.Halleran et. review the RSB of PHD in their institute believed that suction rectal biopsy can be performed safely in preterm infants as small as 1590–2000 g with high accuracy. Clinicians should not hesitate to perform a biopsy for a premature infant when clinically appropriate [13]. In our cases, there were 10 patients who were less than 37 gestational weeks at the time of biopsy. The biopsy results suggested HD in 4 of them and was confirmed after the surgery. Among the rest, 5 were cured with appropriate treatment in each case and well developed till now. There were 35 patients who were older
than 37 gestational weeks at the time of biopsy. Eleven of them had HD that were verified in surgery. The rest 24 cases were cured with appropriate treatment and well developed till now. Then the sensitivity of SRB was 93.7–100%, and specificity was 100% in our cohort.

We have 16 preterm infants whose biopsy weights were less than 2500 g, while 5 cases less than 2000 g. The youngest baby was 31.5 weeks whose biopsy age was 33.5 weeks while biopsy weight was 1580 g. The biopsy result of him was normal. All of the cases had no complications after RSB. This study demonstrates that the diagnosis of PHD can be made reliably. Because most premature infants have delayed feces which is similar symptom to the manifestations of HD, it’s difficult to distinguish them. The accuracy of contrast was lower in younger HD while the risk of NEC maybe rose. From our case, the safety and accuracy of RSB in late preterm infants are high. Early diagnosis can provide the best treatment for children in a targeted manner.

But it is worth noting that the premature infants may have poor clinical conditions because of its hypoplasia. In this research, we focus on the gastrointestinal symptoms and RSB and omit other conditions, especially those affecting the time of the performing. This may be one of the reasons why we have a small cohort. But from this study we consider that RSB is safe and accurate on premature babies their biopsy age were more than 37 weeks. Of course, more cases are needed draw a correct conclusion. The performer needs to evaluate the conditions of the preterm. Bedside rectal suction biopsy is not recommended worldwide for premature infants less than 32 weeks old, especially with a body weight of < 1500 g.

In addition, an experienced pathologist is very important. Immature ganglion cells may not be perceived since the biopsy result depends on the entire process from material extraction, sectioning, staining, etc. Even if our clinicians obtain satisfied specimens (at least 3 mm diameter and a minimum of one-third of the sample should include the submucosa according to the International Working Group of the 2009 World Congress of Gastroenterology), biopsies do not have ganglion cells on every slide. The experience from our hospital’s pathologists was that about 13–15 slides were made from each tissue while some needs to be continuously sliced or continuously sliced repeatedly according to the specimens. At the same time, calretinin immunohistochemical stain was performed. When the nucleus and cytoplasm were positive, it was identified as ganglion cells. However, the dysplastic ganglion cells are less typical than normal ganglion cells in staining effect, while there is no uniform definition of development stages of ganglion cell in the world, such as cell size and nucleoplasm morphology. An experienced pathologist has higher accuracy in identifying those cells. According to the follow-ups of our cases, the pathological results are consistent with the clinical outcomes.

These results suggest that RSB is safe and accurate for late preterm infants. As for the premature infants’ various clinical symptoms, the timing of biopsy still needs to be decided by clinicians.

**Declarations**

**Funding:** None
Conflicts of interest: The authors declare that they have no conflict of interest.

Availability of data and material: All data were in the article.

Code availability: None

Authors’ contributions: Yichao Gu, Kaiyun Hua, Yong Zhao and Yanan Zhang participated the clinical work. Yanan Zhang carried out data collection, analysis of data and preparation of the manuscript. Yongwei Chen and Jinshi Huang designed the study. Shen Yang participated in analysis of data and preparation of the manuscript. All authors read and approved the final manuscript.

Ethics approval for retrospective studies:

This study was conducted retrospectively from data obtained for clinical purposes. We consulted extensively with the IRB of Beijing Children’s Hospital who determined that our study did not need ethical approval.

References

1. Arnoldi R, Leva E, Macchini F, et al. Delayed meconium passage in very low birth weight infants. Eur J Pediatr Surg. 2011; 21(6):395-8.
2. Zhou Y, Yang J, Watkins DJ, Boomer LA, et al. Enteric nervous system abnormalities are present in human necrotizing enterocolitis: potential neurotransplantation therapy. Stem Cell Res Ther. 2013; 4(6):157
3. Sharp NE, Pettiford-Cunningham J, Shah SR, et al. The prevalence of Hirschsprung disease in premature infants after suction rectal biopsy. J Surg Res. 2013; 184:374-7.
4. Earl C. Downey, Elizabeth Hughes, et al. Hirschsprung disease in the premature newborn: A population based study and 40-year single center experience. J Pediatr Surg. 2015; 50123-125
5. Johannes W. Duess, Alejandro D. Hofmann, Prem Puri. Prevalence of Hirschsprung’s disease in premature infants: a systematic review. Pediatr Surg Int. 2014; 30:791-795
6. Haricharan RN, Georgeson KE. Hirschsprung disease. Semin Pediatr Surg. 2008; 17(4):266-275
7. Tam PK, Garcia-Barcelo M. Genetic basis of Hirschsprung’s disease. Pediatr Surg Int. 2009; 25(7):543-558
8. Rbi2 suction rectal biopsy system data sheet. Specialty Surgical Products Inc. https://ssp-inc.com/wordpress/wp-content/uploads/2015/07/rbi2-brochure-electronic-24Aug2015.pdf. Accessed 16 July 2020
9. Friedmacher F, Puri P. Rectal suction biopsy for the diagnosis of Hirschsprung's disease: a systematic review of diagnostic accuracy and complications. Pediatr Surg Int. 2015; 31(9):821-30.
10. Allen AR, Putnam AR, Presson AP, et al. Accuracy of suction rectal biopsy for diagnosis of Hirschsprung's disease in neonates. Eur J Pediatr Surg. 2019; 29(5):425-30.
11. Keyzer-Dekker CM, Sloots CE, Schokker-van Linschoten IK, et al. Effectiveness of rectal suction biopsy in diagnosing Hirschsprung disease. Eur J Pediatr Surg. 2016; 26(1):100-5.

12. Meinds RJ, Kuiper GA, Parry K, et al. Infant's age influences the accuracy of rectal suction biopsies for diagnosing of Hirschsprung's disease. Clin Gastroenterol Hepatol. 2015; 13(10):1801-7.

13. R. Halleran, H. Ahmad, H. Lehmkuhl, et al. Suction Rectal Biopsy Is Accurate in Late Preterm Infants with Suspected Hirschsprung Disease. J Pediatr Surg. 2019; https://doi.org/10.1016/j.jpedsurg.2019.09.055

Tables
Table 1  
Patient characteristics

| Clinical characteristics | n (%)  |
|--------------------------|--------|
| Sex                      | 22 (48.9) |
| Male                     | 23 (51.1) |
| Female                   |        |
| Clinical characteristics | n (%) |
|--------------------------|-------|
| Age of birth             |       |
| < 33 weeks               | 10 (22.2) |
| 33–34 weeks              | 6 (13.3) |
| 34–35 weeks              | 3 (6.7) |
| 35–36 weeks              | 13 (28.9) |
| 36–37 weeks              | 0 |
| Age of biopsy            |       |
| < 33 weeks               | 2 (4.4) |
| 33–34 weeks              | 1 (2.2) |
| 34–35 weeks              | 3 (6.7) |
| 35–36 weeks              | 4 (8.9) |
| 36–37 weeks              | 18 (40.0) |
| 37–40 weeks              | 17 (37.8) |
| > 40 weeks               | 2 (4.4) |
| Weight at biopsy         |       |
| < 1750 g                 | 6 (13.3) |
| 1750–1999 g              | 5 (11.1) |
| 2000–2249 g              | 11 (24.4) |
| 2250–2499 g              | 18 (40.0) |
| 2500–2999 g              | 0 |
| > 3000 g                 | 0 |
| Complications            |       |
| Perforation              |       |
| Hemorrhage               |       |
| Stricture                |       |
Table 2
Pathological features, treatment strategies, and prognosis of the patients

| Pathological diagnosis                             | Treatment strategies                                      | Outcome                              |
|---------------------------------------------------|----------------------------------------------------------|--------------------------------------|
| HD (n = 15)                                        | Pull through surgery (n = 12); Colostomy 2; Ileostomy (total colonic, n = 1) | Normal<sup>1</sup>                    |
| Normal (n = 22)                                    | No surgery (n = 17); Surgery (NEC or gut stenosis after NEC, n = 5) | Normal                              |
| Hypoplasia ganglion cell partly (else normal, n = 8) | No surgery (n = 8)                                      | One died<sup>2</sup>, and the rest were normal |

1: Well-developed, no gastrointestinal symptoms.

2: The baby's parents abandoned the treatment and went home.