Study Protocol and Statistical Analysis Plan

Study on the Relationship Between Pathological Features of Achalasia and Prognosis of Per-oral Endoscopic Myotomy

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Directory

Background

Achalasia is the most common motility disorder of esophagus, characterized by disorders of the lower esophageal sphincter (LES). Normal peristalsis of the esophagus is eliminated and replaced by synchronous or ineffective contraction. Based on high-resolution manometry (HRM), the patients with achalasia were categorized into 3 subtypes, type I: achalasia with minimum esophageal pressurization, type II: achalasia with esophageal compression and type III: achalasia with spasm.

Previous studies have found that the pathological features of the esophageal muscular layers in patients with achalasia are degeneration of nerve plexus, reduction of interstitial cells of Cajal (ICCs), muscular atrophy, fibrosis and infiltration of different inflammatory cells. Different subtypes of achalasia have different clinical characteristics and esophageal motility. Studies have found that ganglion cells in type I patients are significantly reduced compared with type II patients, but the pattern is similar, which may indicate that type I achalasia represents the progression of type II achalasia. Studies have shown that ICCs are preserved in type III achalasia compared with type I and II achalasia. In addition, Achalasia is related to viral infection and autoimmune disease.

Nowadays, per-oral endoscopic myotomy (POEM) is a main therapy for patients with achalasia. Most studies have focused on the relationship

between pathological features and motility characteristics of achalasia, but there are few studies on the relationship between pathological features and therapeutic effect of POEM.

This study will evaluate the pathological characteristics of full-thickness esophageal muscle in patients with achalasia in order to discover the relationship between different pathological characteristics and prognosis of POEM.

Research objective

To determine whether there is difference in prognosis of POEM among patients with achalasia of different pathological characteristics.

Research content

This study prospectively collects the information of 50 participants undergoing POEM for achalasia in Beijing Friendship Hospital, Capital Medical University. During POEM, all patients are required to obtain esophageal muscle biopsy for pathological examination. Participants will be followed up by telephone at 1, 3, 6, and 12 months after POEM for improvement in clinical symptoms.

Research proposal

1. Research design

This is a prospective study based on Beijing Friendship Hospital affiliated to Capital Medical University. The data of 50 patients undergoing POEM due to achalasia were prospectively collected,

including demographic data, drug and surgical treatment data during hospitalization.At POEM, all patients are required to obtain esophageal muscle biopsy and mucosal tissue samples for pathological examination.The patients were followed up by telephone at 1, 3, 6, and 12 months for improvement in clinical symptoms.

2. The research object

2.1 Study Population

The subjects of this study were patients undergoing concurrent POEM surgery for achalasia in Beijing Friendship Hospital affiliated to Capital Medical University.

2.2 Number of planned enrollment cases

50 cases.

2.3 Into the discharge standard

Inclusion criteria:

1) Patients with achalasia treated in Beijing Friendship Hospital, Capital Medical University

2) Age 18-80, no gender limitation

3) Fit and agree to receive POEM therapy

4) Signed informed consent

Exclusion criteria:

1) the previously received treatment of achalasia, Barrett's esophagus lesions, esophageal stricture, liver cirrhosis, and/or

esophageal varices, the digestive tract tumor, active esophagitis, connective tissue disease, allergic disease, blood or blood coagulation patients and hiatal hernia.

2) Use of nonsteroidal anti-inflammatory drugs, corticosteroids, or other immunosuppressive drugs 6 months prior to examination.

3. Research methods

3.1 Diagnosis and classification of achalasia

According to clinical manifestations, endoscopy and high-resolution esophageal manometry, achalasia was diagnosed.

3.1.1 Clinical symptom score

The Eckardt score was used to score clinical symptoms based on preoperative dysphagia, regurgitation, retrosternal pain, and reduced body mass. Doctors need to collect information about the patient's history of viral infections and autoimmunity, such as herpes simplex virus (HSV), varicella zoster, measles, and human papillomavirus (HPV). Autoimmune diseases include urticaria, eczema, ankylosing spondylitis, Hashimoto's thyroiditis, rheumatoid arthritis and sjogren's syndrome.

3.1.2 Endoscopy

All patients underwent endoscopy to exclude tumors, inflammatory reactions or large ulcers in the esophagus and stomach, especially in the lower esophagus or the gastroesophageal junction.

3.1.3 High-resolution manometry

The pressure measurement included resting pressure data, 10 times of swallowing with 5ml water in supine position, and fast swallowing data. The data includes UES, LES, and esophageal body pressure measurement data. Achalasia is divided into three subtypes.

3.2 POEM and muscular pathology

Patients received routine POEM surgery, and full-thickness esophageal muscle biopsy was performed from LES, distal esophagus (about 5cm above EGJ) and middle esophagus (about 10-20cm above EGJ) during the operation. Intraoperative biopsy to take pathological tissue depends on the patient's condition, and the collection method is to use biopsy forceps to take tissue, and take 1 piece at one place.

3.3 Pathological staining and evaluation

Tissue samples were fixed with formalin and embedded in paraffin before sectioning and staining.

3.3.1 Specific dyeing items

1) HE staining is used to evaluate the atrophy degree of smooth muscle and the number of inflammatory cells.

2) Azan-Mallory staining was used to assess the degree of fibrosis.

3) C-KIT immunohistochemistry: C-KIT was positive, and trypsin-like IHC staining was negative to evaluate ICC cells.

4) S100: Evaluate the degree of ganglion loss

5) The number of eosinophil cells was evaluated by HE staining, MBP

and EDN

6) Mast cells: C-kit positive, trypsin-like IHC staining positive.

7) Lymphocyte inflammation.

3.4 Follow-up contents

Follow-up data were collected, including symptom relief (Eckardt score), reflux symptom score (GerdQ score), gastroscopic manifestations, reflux esophagitis, esophagography and esophageal manometry. Follow-up collection time is 1 month, 3 months, 6 months and 12 months after POEM. Patients' symptom improvement and postoperative reflux will be inquired.

4. Clinical outcomes

4.1 Main clinical outcomes

The primary end point event of patients is Eckardt score in 1 month, 3 months, 6 months and 12 months after POEM. Any Eckardt score >3 points are defined as poor prognosis after POEM.

4.2 Secondary clinical outcomes

Secondary end points were the presence of reflux symptoms (GerdQ score >7) or evidence of reflux esophagitis under endoscopy during follow-up. In addition, we will evaluate pathological differences between different sites to provide evidence for selecting the best biopsy site.

Data processing and analysis

Measurement data are expressed as x±s or median, as appropriate,

and count data rate and percentage are expressed. The chi-square test or Fisher's exact test was used to analyze the count data, and the Student's T test or Mann-Whitney test was used to analyze the qualitative and quantitative differences among the subgroups. All P values were bilateral, and P <0.05 was considered as statistically significant difference.

Quality control

In this study, two doctors will participate in patient enrollment, input of baseline data and follow-up data, and jointly check the accuracy and authenticity of data.

References

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