Combining frog-leg lateral view may serve as a more sensitive X-ray position in monitoring collapse in osteonecrosis of the femoral head

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ABSTRACT

Load-bearing capacity of the bone structures of anterolateral weight-bearing area plays an important role in the progressive collapse in osteonecrosis of the femoral head (ONFH). The purpose of this study is to assess the efficacy of combined evaluation of anteroposterior (AP) and frog-leg lateral (FLL) view in diagnosing collapse. Between December 2016 and August 2018, a total of 478 hips from 372 patients with ONFH (268 male, 104 female; mean age 37.9 ± 11.4 years) were retrospectively evaluated. All patients received standard AP and FLL views of hip joints. Japanese Investigation Committee (JIC) classification system was used to classify necrotic lesion in AP view. Anterior necrotic lesion was evaluated by FLL view. All patients with pre-collapse ONFH underwent non-operative hip-preserving therapy. The collapse rates were calculated and compared with Kaplan-Meier survival analysis with radiological collapse as endpoints. Forty-four (44/478, 9.2%) hips were classified as type A, 65 (65/478, 13.6%) as type B, 232 (232/478, 48.5%) as type C1 and 137 (137/478, 28.7%) as type C2. Three hundred cases (300/478, (22.5%) were collapsed at the initial time point. Two hundred and twenty six (226/300, 75.3%) hips and 298 (298/300, 99.3%) hips collapse were identified with AP view and FLL view, respectively. An average follow-up of 37.0 ± 32.0 months was conducted to evaluate the occurrence of collapse in 178 pre-collapse hips. Collapses occurred in 89 hips (50.0%). Seventy-seven (77/89, 86.5%) hips were determined with AP view alone and 85 (85/89, 95.5%) hips were determined with the combination of AP and FLL views. The collapse rates at five years were reported as 0% and 0%, 16.2% and 24.3%, 58.3% and 68.1% and 100% and 100% according to AP view alone or combination of AP and FLL views for types A, B, C1 and C2, respectively. The collapse can be diagnosed more accurately by combination of AP and FLL views. Besides, JIC type A and type B ONFH can be treated with conservative hip preservation, but pre-collapse type C2 ONFH should be treated with joint-preserving surgery. Type C1 needs further study to determine which subtype has potential risk of collapse.

INTRODUCTION

Osteonecrosis of the femoral head (ONFH), a devastating disease in young patients, can generally progress to collapse of the femoral head and secondary hip osteoarthritis [1, 2]. Hip pain and loss of function can affect the quality of life resulting in the need of total hip arthroplasty [3]. Hip preservation is the preferred therapy for young patients. Femoral head collapse is the most significant clinical turning point of ONFH. Once collapse occurs, the clinical outcomes of hip preservation can be greatly deteriorated [4]. Therefore, accurate prediction and evaluation of collapse risk in the pre-collapse stages can help to identify patients who might benefit from intervention to prevent collapse [5].

Femoral head collapse is closely linked to the size and location of the necrotic lesion [6-10]. Especially, a large-sized necrotic lesion and lateral lesion of the femoral head are two major causes of femoral head collapse [6]. Although some reports have attempted to evaluate the collapse risk of ONFH focusing on the area or volume of necrotic lesion, these studies have limitations in terms of small sample sizes and lacking of consistency in clinical application [8, 9]. Recently, Cheng et al. indicates that the location of the necrotic lesion is considered as the highest risk factors. Necrotic lesion located on the lateral and anterior femoral head had high collapse risk in the use of magnetic resonance (MR) imaging, computed tomography (CT) or radiographs [11-13]. The anterior and lateral bone structure of the femoral head is a

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critical weight-bearing area and the reduced load-bearing ability is a result of impairment of necrotic bone [14–18]. Therefore, an ideal method for predicting the progression of ONFH should focus on evaluating the bone structure in the weight-bearing area. An appropriate treatment to the weaken anterolateral weightbearing area of the femoral head is essential for those in high risk of collapse [19].

The ability to easily predict femoral head collapse based on plain radiological types would be valuable and preferable for clinical use, especially in developing countries [20]. Although the Japanese Investigation Committee (JIC) classification that is defined according to AP view is proven to be a reliable system to predict the collapse risk [13], the limitation is that JIC may overlook collapse exiting on the anterior femoral head. Some cases with necrotic lesions occupying more than the medial two-thirds of the weight-bearing area behave as an intact articular surface in AP view but a significant collapse was found in FLL view [21]. This situation reminds us a combined evaluation of AP and FLL views might be more accurate to assess the status of a necrotic femoral head.

The purpose of this study was to estimate and compare the collapse rate by using the AP view alone and a combination of AP and FLL views and develop personalized therapeutic strategies for patients with pre-collapse ONFH.

MATERIALS AND METHODS

This retrospective cohort study have included 478 hips in 372 patients (268 men, 104 women) diagnosed with ONFH between December 2016 and August 2018. Clinical data and radiographic information were collected. This study was approved by the ethics committee of the Third Affiliated Hospital of Guangzhou University of Chinese Medicine (NO. PJ-KY-20210401-001).

All patients were diagnosed with ONFH and classified into Association Research Circulation Osseous (ARCO) staging system [22] and the JIC classification system [13]. The inclusion criteria were set as follow: (i) above the age of 14, (ii) with AP and FLL views of both hip joints, (iii) agreed to perform conservative treatment and (iv) follow-up of >24 months. The exclusion criteria were as follows: (i) accompanying with serious diseases, such as dysplasia of the hip joint, rheumatoid arthritis, ankylosing spondylitis, joint tuberculosis or pyogenic arthritis, (ii) absence of clinical or radiographic data and (iii) previously underwent hip-preserving surgery. Demographic, radiological and surgical intervention data from Orthopaedics Database System v1.0 were collected, which had obtained the copyright in computer software (2017SR274625) registered by the State Copyright Administration. The demographic data have included age, sex, laterality, etiology (steroid use, alcohol intake, idiopathic disease or traumatic), duration of symptom (asymptomatic, pain duration of hip ≤ 6 months and >6 months) and surgical procedure (if relevant). Radiological data have included the stage of ONFH at initial diagnosis and radiological change(s) during the follow-up.

Radiological evaluation

AP and FLL views of both hip joints were performed at the time of initial diagnosis. All plain radiographs were taken using

standardized techniques. For the AP view, the patient was positioned supine on the X-ray table and bilateral legs abducted in neutral position so that the distance between the two feet is equal to shoulder width. For the FLL view, the patient was positioned supine on the X-ray table and the bilateral hips was flexed at a degree of 30° . The thigh was abducted and externally rotated while ensuring that the feet contacted close to each other at the level of ipsilateral knee. The X-ray beam was directed anterior to posterior and centered on the femoral head, while the plane of the pelvis was parallel to the plane of the table (Fig. 1) [23].

The stage of ONFH was defined according to the ARCO staging system [22]: stage I is identified as a normal radiograph with an abnormal MRI or bone scan; stage II is identified as cystic and sclerotic changes in the femoral head but without any evidence of subchondral fracture or flattening of the femoral head; stage III is identified as a crescent sign and/or flattening of the articular surface, this stage is further divided into IIIA (collapse ≤ 2 mm) and IIIB (collapse >2 mm); and stage IV is identified as the appearance of degenerative changes with accompanying joint space narrowing and/or joint destruction. The degree of the collapse was also measured using concentric circles on both AP and FLL views using the Image J program [12]. This study have included patients in ARCO stages II and III and signs of necrotic zone, sclerosis change and/or collapse could be seen on X-ray clearly. Consequently, the JIC type was determined basing on the AP and FLL views. The JIC classification comprises four types according to necrotic location: A, B, C1 and C2 [13]. Type A represents necrotic lesion is located in the medial onethird of the weight-bearing surface relative to acetabular eyebrow arch; type B involves medial two-thirds of the weight-bearing surface; type C1 lesions are characterized by a necrotic zone that spans more than the medial two-thirds of the weight-bearing surface at the acetabular edge; type C2 involves the necrotic zone that spans more than the lateral one-third of the weight-bearing surface and exceeds the acetabular edge. To establish reliability, all image data measurements were made by two experienced orthopedic surgeons in a blinded manner.



Fig. 1. (**A**, **B**) A typical AP and FLL view of bilateral hip. (**C**, **D**) A standard filming feature of the FLL view: patients were obtained in a supine position with the bilateral hips and knees flexed and the feet contacted close to each other; the thigh was abducted and externally rotated at a degree of 30° while ensuring that the plane of the pelvis was parallel to the plane of the X-ray table. $\alpha = 30^{\circ}$.

Non-operative management was recommended to patients with ARCO stage II ONFH. These conservative treatments consisted of pain relief, restricted weight bearing and physical therapy. Restricted weight-bearing was maintained with the use of two crutches for the first 3 months after initial diagnosis. Partial weight-bearing using one crutch was recommended in the next following 3 months. Then, full weight-bearing was allowed when the patient was free of hip pain. All patients underwent clinical and radiographic examination at each 3 or 4 month during follow up for the first 2 years and then receive annually examination thereafter. Demographic features, ARCO stage, JIC type and collapse or not on FLL view were recorded at initial diagnosis. Above data information in patients with ARCO stage III ONFH also were collected before treatment.

Statistical analysis

Statistical analyses were performed by using SPSS version 13.0 software (SPSS Inc., USA). Chi-squared test was used to compare the laterality, etiology, JIC type and duration of symptom. Further, Kaplan–Meier survival analysis was performed to calculate the collapse rates of the femoral head and collapse was set as the endpoint. A comparison of the Kaplan–Meier curves for the relevant factors (JIC type, etiology and onset of symptom) was performed with a log-rank test in the pre-collapse cohort. A cox regression model was used to evaluate the independent prognostic factors associated with collapse, including JIC type (AP view only or combination with FLL view), etiology and duration of pain. All statistical analyses were two-sided and evaluated with a *P*-value of less than 0.05 indicating statistical significance.

RESULTS

Demographics

Between December 2016 and August 2018, 478 hips from 372 patients with ONFH were reviewed. At initial diagnosis, 178 hips in 135 patients including 93 male and 42 female with a mean age of 40.4 ± 11.5 years (range, 17–64) were defined as ARCO stage II. A number of 300 hips in 237 patients including 175 male and 62 female with a mean age of 36.21 ± 10.8 years (range, 14–62) were defined as ARCO stage III with stage IIIA 197 hips and IIIB 103 hips, respectively. According to the JIC types, 44 hips were classified as type A, 65 hips as type B, 232 hips as type C1 and 137 hips as type C2. The demographic, clinical and radiographic characteristics are shown in Table I.

General collapse rates

As shown in Table II, there were no significant differences in collapse rate between left and right sides ($X^2 = 0.00$, P = 0.98). There was also no significant difference between patients with and without steroid use ($X^2 = 0.14$, P = 0.71). There were significant differences in terms of the collapse rate among four JIC types ($X^2 = 291.15$, P < 0.01). The collapse rates in types A, B, C1 and C2 were 2.5%, 43.1%, 69.0% and 77.4%, respectively. Moreover, there was also a significant difference in collapse rate at initial diagnosis between asymptomatic and symptomatic hips ($X^2 = 92.08$, P < 0.01). The collapse rates of asymptomatic hips (72.2%).

Table I. Demographic data

Parameters	N
Number of patients/hips	372/478
Mean age at initial diagnosis, years (range)	37.9±11.4 (14–63)
Gender (Males/Females)	268/104
Side (Left/Right)	242/236
Bilateral:unilateral	106:266
Etiology	
Steroid	186
Alcohol	123
Idiopathic	41
Trauma	22
ARCO stage (Baseline)	
II	178
IIIA	197
IIIB	103
Location (Baseline)	
A	44
В	65
C1	232
C2	137
Onset of symptom	
Asymptomatic	79
≤ 6 months	255
>6 months	144

Collapse rates observed at initial diagnosis

In the 300 collapsed hips, the number of the collapsed femoral head in ARCO stage IIIA found by FLL view was significantly higher than that found by AP view ($X^2 = 81.53$, P < 0.01). However, there were no significant differences in the collapse rate between two position radiographs ($X^2 = 2.02$, P = 0.16). In the 197 hips with ARCO stage IIIA, 2 collapsed hips (1.0%) were detected in AP view, but not in FLL view. Seventy-two (72/197, 36.5%) collapsed hips were detected in FLL view but not in AP view (Table III). Further analysis of this 72 hip indicated that 6 hips were JIC type A, 24 hips type B, 41 hips type C1 and 1 hips type C2. Necrotic femoral heads were collected after hip replacement, and hard tissue slicing showed a collapse at the anterolateral portion of the femoral head or anterior portion alone (Fig. 2).

Collapse rates assessed at 1, 3 and 5 years follow-up

The risk factors of collapsing in 178 pre-collapse hips were analyzed by log-rank test, and the mean follow-up period was 37.0 ± 32.0 months (range 1–141). The collapse rates at five-year follow-up were evaluated and described as 0% in type A, 16.2% in type B, 58.3% in type C1 and 100% in type C2 (P < 0.01; Fig. 3A); With the definition of JIC types according to FLL view, type A have the collapse rate of 0%; type B 24.3%; type C1 68.1%; and type C2 100% (P < 0.01; Fig. 3B). In addition, there was no significant difference in steroid and non-steroid groups (P = 0.19; Fig. 3C). However, a significant difference was indicated between patients with asymptomatic and symptomatic ONFH (P < 0.01; Fig. 3D). The cox regression analysis indicated that JIC type was an independent risk factor associated

Parameter/stage	ARCO stage					
	II	IIIA	IIIB	n	Collapse hips at initial diagnosis, n (%)	P-value
Side (Left/Right) ^a						0.982 ^b
Left	90	101	51	242	152/242(62.8%)	
Right	88	96	52	236	148/236(62.7%)	
Etiology ^a						0.706 ^b
Steroid	91	104	56	251	159/251(63.3%)	
Non-Steroid	87	93	47	227	140/227(61.7%)	
Location (Baseline) ^a						<0.001 ^b
А	38	6	0	44	6/244(2.5%)	
В	37	27	1	65	28/65(43.1%)	
C1	72	118	42	232	160/232(69.0%)	
C2	31	46	60	137	106/137(77.4%)	
Onset of symptom ^a						<0.001 ^b
Asymptomatic	67	9	3	79	12/79(15.2%)	
Symptomatic ≤ 6 months	74	129	52	255	181/255(71.0%)	
Symptomatic > 6 months	37	59	48	144	107/144(74.3%)	

Table II.	Collapse rates	at initial diagnosis	analyzed by d	lemographic data
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^aValues are given as the number of hips.

^bChi-square test.

Table III. Radiographical characteristics of patients with collapsed ONFH

	ARCO stage		
Parameters	IIIA	IIIB	
Number of hips (<i>n</i>)	197	103	
AP view			
Femoral head non-collapse (n)	72	2	
Femoral head collapse (n)	125	101	
Negetive in FLL view (n)	2	0	
Collapse rate (%)	63.5%	98.1%	
FLL veiw			
Femoral head non-collapse (n)	2	0	
Femoral head collapse (n)	195	103	
Negetive in AP view (n)	72	2	
Collapse rate (%)	99.0%	100%	
Combination of AP and FLL views			
Collapse rate (%)	100%	100%	

ONFH = osteonecrosis of femoral head.

with collapse [P < 0.01, hazard ratio (HR) 4.551; 95% confidence interval (CI) 3.263–6.348]. A lower HR value was shown when FLL view was combined with AP view to determine collapse [P < 0.01, HR 4.11; 95% CI 3.04–5.55]. Moreover, the HR value was 4.32 for type C1 compared to type B (P < 0.01) and 3.12 for type C2 compared to C1 (P < 0.01) (Fig. 4). In addition, there was no significant association between collapse and other clinical factors such as etiology (P = 0.68) and pain duration of hip (P = 0.94).

Different treatments at initial diagnosis or during the follow-up period

Surgical treatment at initial **diagnosis** was performed for a total of 76 hips (25.3%) in 300 collapsed hips, namely, total hip



Fig. 2. Five necrotic femoral heads shown on AP and FLL views. (A) A 63 year old man with alcohol induced-ONFH, and (B) a 40 year old man with idiopathic ONFH, both showed non-collapse of the femoral head on AP view and collapse on FLL view. Corresponding the gross specimen photograph showed cartilage defect or folds at the anterior portion of the femoral head. (C) A 32 year old woman with steroid induced-ONFH and (D) a 49 year old man with alcohol induced-ONFH, both showed collapse of the femoral head on AP and FLL views, and corresponding the gross specimen photograph showed cartilage folds at the anterolateral portion of the femoral head. ONFH: osteonecrosis of femoral head; AP: anteroposterior view; FLL: frog-leg lateral view.

arthroplasty (21 hips), surgical hip dislocation approach combined with anterior iliac bone grafting (31 hips), free vascularized fibular grafting (20 hips) and intra-articular injection of platelet-rich plasma (4 hips). In 178 pre-collapse hips, 89 hips (50.0%) were detected collapse during the follow-up period. Surgical treatments was performed for a total of 38 hips (42.7%), namely, total hip arthroplasty (29 hips), surgical hip dislocation



Fig. 3. Kaplan–Meier survival curves of patients with pre-collapse ONFH. (**A**) the cumulative five-year collapse rates were indicated as follows: using the JIC type on AP view alone, type A, 0%; type B, 16.2%; type C1, 58.3%; and type C2, 100% (P < 0.001, log-rank test). (**B**) collapse rates were evaluated on AP and FLL views as follows: type A, 0%; type B, 24.3%; type C1, 68.1%; and type C2, 100% (P < 0.001, log-rank test). (**C**) collapse rate according to steroid use; there was no difference in terms of time to collapse (P = 0.186, log-rank test). (**D**) collapse rate according to symptomatic; there was a significant difference in terms of time to collapse (P < 0.001, log-rank test). (**D**) collapse rate according to steroid use; there was a significant difference in terms of time to collapse (P < 0.001, log-rank test). (**D**) collapse rate according to symptomatic; there was a significant difference in terms of time to collapse (P < 0.001, log-rank test). (**D**) collapse rate

approach combined with anterior iliac bone grafting (4 hips) and free vascularized fibular grafting (5 hips). No surgical intervention was need in all 75 hips with JIC type A and type B during follow-up.

DISCUSSION

This study reported that the collapse during the progress of ONFH can be determined more accurately by combined evaluation of AP and FLL views. JIC types based on combination of AP and FLL views have a predictable value for collapse according to the survival analysis. The cox regression analysis indicated that JIC types based on the AP view was an independent risk factor. When JIC types were defined by combination of AP and FLL views, the collapse risk was higher. The results emphasized that the necrotic lesion in anterolateral weight-bearing area of the femoral head should be fully evaluated by not only AP view but also combined with FLL view. This method is simple, low-cost and easy to perform and repeat reliably. It is appropriate predicting the occurrence of collapse at initial diagnosis and follow-up.

Lacking of reliable data of the collapse rates make the ideal therapy for asymptomatic remain controversial [24]. Considering the high collapse rates, some studies proposed early surgical hip-preserving intervention in asymptomatic patients, but

there are also studies recommending surgical treatment should be considered only when the disease is symptomatic [25–28]. Accordingly, there are different methods based on the threedimensional images to assess extent and location of the necrotic lesion and, further, predict collapse, such as MRI and CT [8, 29, 30]. It is hard to reach a consensus especially in which layer should be taken to determine for predicting collapse in MRI or CT scan. In addition, CT scan involves higher doses of radiation and MRI is a relatively higher cost examination. Therefore, it is generally believed that the ability to easily and accurate predict femoral head collapse based on plain radiological would be valuable and preferable for clinical used, especially in developing countries [31].

JIC classification system is a classical method to determine the collapse risk and widely used in worldwide because it is based on lesion extent and location involving the lateral weightbearing surface of femoral head [10, 13]. The advantages of JIC type are its accuracy, prognostic value as well as simplicity. Several studies have reported different collapse rates of ONFH using the JIC classification system; however, there are great differences with the clinical results (Table IV) [10, 13, 32]. Compared to Kuroda's study, we have further divided type C to C1 and C2 for different collapse rates in these two subtypes. Recently, a study with large sample data reported that



Fig. 4. Five-year collapse rates of each JIC type as evaluated by the cox proportional hazards regression model. JIC types on AP view identify that the greater the lateral involvement of the necrotic lesion, the higher the risk of collapse. A higher collapse rate can be seen on AP and FLL views, which simultaneously identified the collapse of anterior and lateral location of femoral head. yr: year; AP: anteroposterior view; FLL: frog-leg lateral view.

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Table IV. Flevious reports on th	le conapse rate evaluated	u by the japanese mve	sugation Commutee (J1	C) classification system

			Collapse rate, % (by Kaplan–Meier survival analysis)			
Authors (years)	Necrotic hips, n	Mean follow-up, years	Type A	Туре В	Type C1	Type C2
Min et al. (2008) [32]	81	8.3	0	0	13	86
Takashima <i>et al</i> . [10]	86	10.0	0	6	68	82
Kuroda <i>et al.</i> (2019) [13]	212	5.0	0	8	37	85
Present study (2021)	178	5.0	0	24	68	100

five-year collapse rates of 267 necrotic femoral heads in JIC type A, B, C1 and C2 were 0%, 7.9%, 36.6% and 84.8%, respectively [13]. Nevertheless, the present study, a total of 178 hips were followed-up for five-year and reported collapse rates of 0% in type A, 24.3% in type B, 68.1% in type C1 and 100% in type C2, respectively. The reasons for this difference might result from the usage of combined evaluation of AP and FLL views to classify JIC types. Anterior involvement of the femoral head is also an important factor of collapse [12, 33]. Previous studies in hip biomechanics have indicated that the mechanical stress is loaded on the anterolateral area of femoral head which supporting most of the body weight in daily activities [34, 35]. Kubo et al. demonstrated that a completed involvement of the anterior femoral head could increase the risk of collapse. Anterior area affected might collapse eventually even the necrotic lesion did not extend beyond the lateral column [33]. Nam et al. evaluated the fate of untreated asymptomatic ONFH

with a measurement of the size of the anterolateral lesion in AP and FLL views [36]. Both necrotic angle and a modified Kerboul method were put forward to assess collapse of the femoral head in the anterolateral weight-bearing surface of femoral head [37, 38].

This study found that the collapse rate increases as the necrotic lesion on the anterolateral weight-bearing surface of femoral head becomes larger. It can provide useful clues for determining optimal treatment approaches. Conservative treatments are recommended for Type A in the combination of AP and FLL views because of the lowest collapse rate. Type C2 with the highest collapse rate is suggested to accept early hip-preserving surgical treatments. Although type B has a certain collapse rate, conservative treatments are still recommended for most of the patients. The high collapse rate of type C1 in the combination of AP and FLL views suggests that early intervention should be estimated according to the integrity of anterolateral weight-bearing surface of femoral head, but the precise evaluation method is yet to be further studied.

In the present study, a significant difference is noted that the five-year collapse rates are closely related to the existence of clinical symptoms at initial diagnosis. It is well coincided with the previous studies reporting that hip pain was strongly associated with bone marrow edema, subchondral bone fracture and subsequent collapse in pre-collapse ONFH [31, 39–41]. Therefore, we suggest that hip pain may be considered as a sign for progression to advanced ONFH, even prior to collapse. Furthermore, we demonstrated another useful finding that combined evaluation of AP and FLL views at diagnosis has clinical value to predict collapse in patients with pre-collapse ONFH.

There are some limitations in this study. First, this is a retrospective study in nature with limited level of evidence (level 3). Second, the clinical outcomes of patient undergone different treatments were not observed. If collapsing only detected in FLL view but not in AP view could affect the following treatment is yet to be studied. Third, even though the intraobserver and interobserver reliabilities of JIC types are reported with high reliability, further analysis is still need to confirm more indexes with other statistical analysis. Additionally, without the CT scan results of each case, we cannot ensure combining AP and FLL views can detect any collapses in any situation. A further comparing study between the results of CT scan and combination of AP and FLL views may need.

In conclusion, combined evaluation of AP and FLL views for anterolateral necrotic lesion has clinical value to predict collapse in patients with pre-collapse ONFH. Together by using JIC classification, it can provide an optimal choice for therapeutic strategies. Specifically, prediction of collapse and early intervention are the fundamental principles of hip-preserving treatment for both systematic and asymptomatic ONFH.

ETHICAL APPROVAL

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration ant its later amendments or comparable ethical standards.

CONSENT TO PARTICIPATE

Written informed consent was obtained from individual or guardian participants.

CONSENT TO PUBLISH

Written and signed consent to publish the case was obtained from the patient.

AUTHOR CONTRIBUTIONS

Q.-S.W. carried out the experiments and data analysis and drafted and critically revised the manuscript. M.-C.H. critically revised the manuscript. T.-Y.L. revised the manuscript and provided technical assistance in preparing the manuscript. P.Y. carried out the statistical analysis. Z.-Q.C. and Q.-W.Z. supervised the project and critically revised the manuscript for important intellectual content. W.H. is the corresponding author, contributed to the study design, clinical studies and critically revised the manuscript for important intellectual content.

DATA AVAIILABILITY

All data or clinical images used during the study are available from the corresponding author by reasonable request.

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CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest.

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