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ESTABLISHMENT OF THE 6-SHOGAOL EXTRACTION PROCESS FROM 6-SHOGAOL-ENRICHED GINGER FOR CANCER TREATMENT

Ho Ba Ngoc Minh[1], Nguyen Trong Diep[1], Pham Van Hien[1], Vu Binh Duong[1]*

Abstract

Objectives: To optimally extract the potential phytochemical "6-shogaol" from fermented shogaol-enriched ginger. ***Methods:*** Extraction process parameters were investigated using 6-shogaol as a marker. ***Results:*** The suitable parameters of the 6-shogaol extraction process: Hot extraction using reflux equipment, solvent of 90% ethanol, solvent/herb ratio of 15/1, temperature of 70ºC, duration of 90 minutes, and two-time extraction. The 6-shogaol extraction efficiency was 89.61 ± 0.68%. ***Conclusion:*** The optimal process for 6-shogaol extraction from fermented shogaol-enriched ginger has been studied.

Keywords: 6-shogaol; Extraction process; 6-shogaol-enriched ginger.

INTRODUCTION

Ginger (*Zingiber officinale* Rosc.), a member of the Zingiberaceae family, has been found to possess various biological activities, including antioxidant, anti-inflammatory, antibacterial, anti-diabetic, and anti-cancer effects, etc. [1, 2, 3]. These biological effects relate to some active ingredients such as 6-gingerol, zingiberol, etc. Many studies have shown that 6-shogaol has more potent therapeutic effects in anti-inflammatory, antioxidant, and cancer-preventing than 6-gingerol [4, 5]. Moreover, 6-shogaol inhibited the growth of several cancer lines, such as lung, breast, and cervical cancer etc. [6]. However, the 6-shogaol content in dried ginger (Can Khuong) is very low, while in fresh ginger, it is almost undetectable [7]. To solve the problem, some studies have been conducted to enrich 6-shogaol by dehydrating 6-gingerol. The conversion of 6-gingerol to 6-shogaol is accelerated in high-temperature and acidic environments [4, 8]; this process is referred to as ginger fermentation.

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Currently, we have been studying the enrichment of 6-shogaol in ginger by steaming it in an acidic environment [9, 10] to create raw materials with potential treatment in pharmaceutical production. To effectively incorporate these herbal materials into practical production, it is crucial to thoroughly research and develop a process for extracting 6-shogaol from fermented ginger. Therefore, this study aims to: *Establish the necessary process parameters for extracting 6-shogaol from shogaol-enriched ginger.*

MATERIALS AND METHODS

1. Materials

Including 6-shogaol-enriched ginger (fermented ginger) provided by the Drug Research and Development Center, Vietnam Military Medical University meeting institutional standards.

* *Standard substance:* 6-shogaol 98% from ChemFaces, China; chemicals and solvents: Acetonitrile, glacial acetic acid, methanol, ethanol 98%, etc., qualified analytical purity standards and pharmacopeia.

* *Equipment:* HPLC system e2695 and PDA Detector 2998, Waters, USA; C_{18} column (150 x 4.6mm; 5µm), InertSustain AQ, Japan; Reflux extractor, China; Rotary vacuum evaporator N1200B, EYELA, Japan.

2. Methods

* *Extraction method:* To develop the extraction process, the following investigations were conducted:

Extraction method: Hot extraction using reflux equipment, ultrasonic extraction, microwave extraction;

Solvent: Ethanol (50%, 70%, 90%, 96%);

Ratio of solvent/herb: 5/1, 10/1, 15/1, 20/1, 25/1 (mL/g);

Temperature: 40, 50, 60, 70, and 80ºC;

Duration: 30, 60, 90, 120 minutes per time;

Extraction time: 1 time, 2 times, 3 times.

* *The quantitative method of 6-shogaol by high-performance liquid chromatography (HPLC):*

Standard sample: 50mg of 6-shogaol was accurately weighed and dissolved in methanol in a 25mL volumetric flask to obtain a stock standard solution with a concentration of about 2000 µg/mL. Working concentrations of standards and samples (about 10, 50, 100, 200, and 500 µg/mL) were made by diluting standard stock solutions with methanol. Samples were filtered through a 0.45µm membrane prior to HPLC quantification.

Material sample: About 2g of fermented ginger was accurately weighed into a round-bottom flask, added 30mL of 90% ethanol, and refluxed for 90

minutes at 60ºC. Extracting 3 times. Combine all extracts and volumetrically measure in a 100mL flask. Centrifuge the extract at 5500rpm for 10 minutes. Collecting the clear solution, then diluting it twofold to obtain a suitable concentration. Samples were filtered through a 0.45µm membrane prior to HPLC quantification.

Extract sample: Fermented ginger was extracted according to the investigated conditions. Centrifuge the extract samples at 5500rpm for 10 minutes. Collecting the clear solution, then diluting it to a suitable concentration. Samples were filtered through a 0.45µm membrane prior to HPLC quantification.

Chromatographic conditions*: InertSustain AQ C_{18} column (150 x 4.6mm; 5µm); wavelength: 280nm; mobile phase: Acetonitrile (C) and 0.4% acetic acid (A) (60/40; v/v); flow rate: 1 mL/min; injection volume: 5µL.

The 6-shogaol content was calculated by the following formula:

$$\text{The 6-shogaol content (mg/g)} = \frac{C \times V \times n}{m \times 1000 \times (1 - h/100)}$$

C: Concentration of 6-shogaol in the extract (µg/mL) calculated from the calibration curve; V: Volume of the herb extract (mL); n: Dilution factor; m: Weight of herb (g); h: Moisture content of herb (%).

*This chromatographic condition has been validated using ICH guidelines. The calibration curves (y = 13462x + 190171) showed an acceptable correlation ($r^2 > 0.99$). The 6-shogaol average recoveries (%) for LQC, MQC, and HQC were 107.91%, 107.92%, and 101.50%, respectively, and RSD for each recovery varied from 0.58% to 1.38%. The precision of 6-shogaol on the same day (intra-day precision) and three consecutive days (inter-day precision) had RSD < 2.00%. LOD and LOQ. The LOD was found to be 0.012 µg/mL, and the LOQ was determined to be 0.040 µg/mL under the specified experimental conditions. These results indicate that the method is capable of detecting and quantifying low concentrations of 6-shogaol with a high degree of accuracy.

3. Ethics

The study has been approved by the Ethics Committee according to decision No. 3934/QĐ-HVQY, dated 19/9/2023. The Institute of Pharmaceutical Education, Vietnam Military Medical University granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest.

RESULTS AND DISCUSSION

1. Results of determination of 6-shogaol content in fermented ginger

Table 1. 6-shogaol content in fermented ginger (n = 3).

m (g)	h (%)	V (mL)	n	S peak (µAU.s)	C (µg/mL)	6-shogaol content (mg/g)
2.012	18.16	100	2.5	3372834	235.01	35.68
2.009	18.16	100	2.5	3352110	237.52	36.11
2.013	18.16	100	2.5	3287472	230.49	34.97
Mean ± SD						35.59 ± 0.57

From table 1, the content of 6-shogaol in fermented ginger is 35.59 ± 0.57 mg/g calculated on the dry material. When examining the extraction process parameters, the extraction efficiency was calculated based on the content of 6-shogaol in the fermented ginger.

* *Investigation of the effect of the extraction method:*

The fermented ginger was extracted under the same conditions using 90% ethanol solvent, solvent/herb ratio of 10/1, temperature of 60ºC, duration of 90 minutes, and one-time extraction by different methods: Ultrasonic extraction, hot extraction, and microwave extraction. The results are shown in table 2.

Table 2. The effect of extraction method on 6-shogaol extraction from fermented ginger (n = 3).

Method	6-shogaol content (mg/g)	Extraction efficiency (%)
Ultrasonication	19.01 ± 0.13	53.41 ± 3.56
Hot extraction	22.20 ± 0.12	62.38 ± 3.35
Microwave	21.92 ± 0,14	61.59 ± 3.91

The results in table 2 showed that hot extraction using reflux equipment gave the highest extraction efficiency of 62.38%. The ultrasonic extraction method yielded only 53.41%, and the microwave extraction method yielded 61.59%. During the hot extraction process, the herb was exposed to the solvent at a high

temperature, which increased the diffusion coefficient, thereby increasing the amount of extracted active ingredients, leading to an increase in extraction yield. Therefore, the hot extraction using the reflux method with simple and easy-to-operate equipment was chosen for further research.

** Investigation of the effect of extraction solvent:*

The fermented ginger was extracted through hot extraction using reflux equipment at 60ºC, solvent/herb ratio of 10/1, duration of 90 minutes, and one-time extraction with different solvents, including ethanol (50, 70, 90, 96%). The results are shown in table 3.

Table 3. The effect of extraction solvent on 6-shogaol extraction from fermented ginger (n = 3).

Solvent	6-shogaol content (mg/g)	Extraction efficiency (%)
EtOH 50%	19.07 ± 0.42	53.59 ± 3.37
EtOH 70%	20.48 ± 0.11	57.54 ± 2.96
EtOH 90%	22.20 ± 0.14	62.38 ± 3.90
EtOH 96%	23.38 ± 0.12	65.70 ± 3.24

Table 3 shows that when the EtOH concentration increased from 50% to 96%, the extraction efficiency of 6-shogaol also gradually increased from 53.59% to 65.70%. This is due to the poor solubility in water of 6-shogaol, with a solvent which is low alcohol concentrations and high-water content, the ability to dissolve the active ingredient is lower, so the content and extraction efficiency of 6-shogaol is lower. However, the extraction efficiency of active ingredients when extracted with 90% ethanol (62.38%) was slightly lower than when extracted with 96% ethanol (65.70%). Therefore, 90% ethanol is the most suitable solvent for extracting fermented ginger and conducted for further research.

** Investigation of the effect of the herb/solvent ratio:*

The fermented ginger was extracted through hot extraction using reflux equipment with the solvent of 90% ethanol, temperature of 60ºC, duration of 90 minutes, and one-time extraction with different solvent/herb ratios, including 5/1, 10/1, 15/1, 20/1, 25/1. The results are shown in table 4.

Table 4. The effect of the herb/solvent ratio on 6-shogaol extraction from fermented ginger (n = 3).

Ratio	6-shogaol content (mg/g)	Extraction efficiency (%)
5/1	20.96 ± 0.05	58.90 ± 1.41
10/1	22.20 ± 0.13	62.38 ± 3.68
15/1	24.89 ± 0.11	69.94 ± 2.93
20/1	25.74 ± 0.13	72.33 ± 3.51
25/1	25.68 ± 0.32	72.15 ± 0.90

Table 4 shows that the solvent/herb ratio has an influence on extraction efficiency. When the solvent ratio increased, the content and extraction efficiency of 6-shogaol also increased. At the solvent/herbal material ratio of 15/1, the extraction efficiency nearly reached 70%; however, when the solvent ratio increased to 20/1, the extraction efficiency increased insignificantly. When extracting at a solvent/ herbal material ratio of 20/1, the extraction efficiency reached 72.33%. The extraction efficiency of 6-shogaol increased slightly when the ratio of active ingredients and solvent reached saturation. Therefore, the study chose the solvent/herb ratio of 15/1 to conduct further tests.

* *Investigation of the effect of extraction temperature:*

The fermented ginger was extracted through hot extraction using reflux equipment with the solvent of 90% ethanol, solvent/herb ratio of 15/1, duration of 90 minutes, and one-time extraction with different temperatures of 40, 50, 60, 70, and 80ºC, respectively. The results are shown in table 5.

Table 5. The effect of temperature on 6-shogaol extraction from fermented ginger (n = 3).

Temperature (ºC)	6-shogaol content (mg/g)	Extraction efficiency (%)
40	15.42 ± 0.12	43.32 ± 3.29
50	20.30 ± 0.02	57.03 ± 0.48
60	24.89 ± 0.05	69.94 ± 1.36
70	26.80 ± 0.05	75.31 ± 1.49
80	26.61 ± 0.12	74.77 ± 3.40

Table 5 shows that when increasing the temperature from 40ºC to 70ºC, the extraction efficiency also increased gradually, reaching its highest at 70ºC (75.31%). However, when the extraction temperature increased to 80ºC, the extraction efficiency decreased slightly (from 75.31% to 74.77%), which may be due to the decomposition of 6-shogaol when extracted at high temperature for a long time (90 minutes), leading to a decrease in extraction efficiency. Therefore, the study chose the temperature of 70ºC to conduct the following parameter research.

** Investigation of the effect of extraction duration:*

The fermented ginger was extracted through hot extraction using reflux equipment with a solvent of 90% ethanol, solvent/herb ratio of 15/1, temperature of 70ºC, and one-time extraction with different durations of 30, 60, 90, and 120 minutes, respectively. The results are presented in table 6.

Table 6. The effect of duration on 6-shogaol extraction from fermented ginger (n = 3).

Duration (minute)	6-shogaol content (mg/g)	Extraction efficiency (%)
30	21.02 ± 0.14	59.07 ± 3.96
60	23.80 ± 0.10	66.88 ± 2.88
90	26.80 ± 0.08	75.31 ± 2.29
120	25.17 ± 0.15	70.73 ± 4.03

Table 6 shows that extraction duration also has a significant effect on extraction efficiency. 30-minute extraction gave the lowest efficiency. When increasing the extraction duration to 90 minutes, the efficiency increased significantly (75.31%). However, if the duration is extended, specifically to 120 minutes, the extraction efficiency decreases slightly (from 75.31% to 70.73%). Therefore, the study chose the duration of 90 minutes to continue the following research.

** Investigation of the extraction times:*

The fermented ginger was extracted through hot extraction using reflux equipment with a solvent of 90% ethanol, solvent/herb ratio of 15/1, temperature of 70ºC, and duration of 90 minutes. The samples were extracted three times, and the extract was quantified separately each time. The results are presented in table 7.

Table 7. The effect of the extraction time on 6-shogaol extraction from fermented ginger (n = 3).

Times	6-shogaol content (mg/g)	Extraction efficiency (%)
1	26.80 ± 0.11	75.31 ± 0.57
2	5.09 ± 0.02	14.30 ± 0.11
3	2.45 ± 0.01	6.90 ± 0.04

The results of table 7 show that after one-time extraction, the extraction efficiency of 6-shogaol is quite high (75.31%). After two-time extractions, the extraction efficiency of 6-shogaol is nearly 90%. Therefore, to save time and solvent, the study chose the number of extractions to be 2 times, with the extraction efficiency of 6-shogaol reaching 89.61 ± 0.68%.

With the goal of investigating and selecting the optimal parameters to extract the fermented ginger rich in 6-shogaol for application in the preparation of dry powder and other pharmaceutical products, ethanol - water solvent is the most suitable, although organic solvents may extract 6-shogaol better. Increasing the amount of solvent can also increase the extraction efficiency of 6-shogaol. However, it will waste solvent, and the extract will be diluted when concentrated, so it will waste energy and prolong the concentration duration, so there will be a risk of reducing the active ingredient content. According to Seon Ok and Woo-Sik Jeong, the higher the extraction temperature, the greater the 6-shogaol content obtained. At 60ºC, the extracted 6-shogaol content was about 10 mg/g, but at 80ºC, it reached about 22 mg/g [3]. The difference in the extracted 6-shogaol content between our research and the study of Seon Ok and Woo-Sik Jeong may be related to the herb/solvent ratio as well as the raw herb used, but they all have a common trend of relatively high extraction temperature, about 70 - 80ºC.

CONCLUSION

The process parameters for extracting 6-shogaol from 6-shogaol-enriched ginger were investigated, and the most suitable extraction condition was selected: Hot extraction using reflux equipment, solvent of 90% ethanol, solvent/herb ratio of 15/1, temperature of 70ºC, duration of 90 minutes per extraction, 2-time extractions. The extraction efficiency of 6-shogaol was 89.61 ± 0.68%. With the obtained results, this extraction process can be applied in the study of modernizing 6-shogaol-enriched ginger.

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EVALUATION OF *PTGER4* GENE METHYLATION IN PLASMA FOR LUNG CANCER DETECTION

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Abstract

Objectives: To evaluate the diagnostic value of *PTGER4* (*Prostaglandin E Receptor 4*) gene methylation in plasma for lung cancer detection compared to healthy controls. ***Methods:*** A cross-sectional, case-control study was conducted on 149 non-small cell lung cancer (NSCLC) patients and 100 healthy individuals. Peripheral blood samples were collected for *PTGER4* methylation analysis using real-time methylation-specific PCR (MSP). Data were analyzed using SPSS 26.0, including group comparisons, receiver operating characteristic (ROC) analysis, and logistic regression. ***Results:*** The *PTGER4* methylation positivity rate was significantly higher in lung cancer patients (18.8%) compared to healthy controls (3.0%, $p < 0.001$). *PTGER4* methylation positivity increased the risk of lung cancer by 6.3 times (OR = 6.30; 95%CI: 1.90 - 20.40). The area under the ROC curve (AUC) was 0.58, with a sensitivity of 18.8% and a specificity of 97.0%. ***Conclusion:*** *PTGER4* gene methylation is a promising biomarker for lung cancer detection. Despite its limited sensitivity, the high specificity suggests its potential as a confirmatory diagnostic tool, particularly when combined with imaging modalities such as low-dose computed tomography (LDCT).

Keywords: *PTGER4* methylation; Lung cancer; Biomarker; Non-invasive diagnosis; Methylation-specific PCR.

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INTRODUCTION

Lung cancer is one of the most prevalent cancers and the leading cause of cancer-related deaths worldwide. According to the Global Cancer Observatory (GLOBOCAN), lung cancer ranks first in terms of both new cases and cancer-related deaths reported in 2022 [1]. In Vietnam, lung cancer is also among the most common cancers, particularly in men [2]. Alarmingly, the majority of patients are diagnosed at advanced stages, leading to limited treatment efficacy and reduced survival rates [3].

Early diagnosis of lung cancer plays a crucial role in improving prognosis and increasing the chances of successful treatment. Current diagnostic methods, such as LDCT and tissue biopsy, have shown significant advancements. However, they still present limitations in terms of cost, accuracy, and broad applicability [4]. Therefore, the search for non-invasive biomarkers in peripheral blood for early detection of lung cancer has become a promising research direction and has attracted significant attention within the medical community [5].

Among various biomarkers, DNA methylation, an epigenetic modification that regulates gene expression, has emerged as a potential tool for early cancer detection [6]. DNA methylation changes, especially hypermethylation in the promoter regions of tumor suppressor genes, can lead to gene silencing and contribute to cancer development. The *PTGER4* gene, a member of the prostaglandin E2 receptor family, is involved in inflammatory processes and has been implicated in tumor progression and metastasis, including lung cancer [7]. Recent studies have demonstrated that abnormal methylation of the *PTGER4* gene (*mPTGER4*) can be detected in the plasma of lung cancer patients and holds diagnostic potential [8].

Although several international studies have highlighted the potential of *mPTGER4* for early lung cancer detection, similar research in Vietnam remains limited. Therefore, this study aims to: *Evaluate the diagnostic value of PTGER4 gene DNA methylation in the peripheral blood of lung cancer patients, compared to the healthy control group.* The findings of this study will contribute to clarifying the role of *mPTGER4* as a non-invasive biomarker, supporting early detection and diagnosis of lung cancer in clinical practice.

MATERIALS AND METHODS

1. Subjects

This study included two groups: NSCLC patients and healthy controls.

The NSCLC group consisted of patients with histopathologically confirmed primary NSCLC who had not received chemotherapy, radiotherapy, or surgery before blood sampling. The control group included individuals with no history of cancer and no pulmonary lesions on chest X-rays or LDCT, matched by age and gender to the NSCLC group.

* *Exclusion criteria:* Patients with small-cell lung cancer (SCLC), secondary lung cancer, advanced chronic diseases (COPD, tuberculosis, autoimmune disorders), withdrawal of consent, or incomplete clinical data.

All participants provided written informed consent, ensuring confidentiality and the right to withdraw at any time without affecting their medical care.

* *Sample size calculation:*

The sample size was calculated using the formula for comparing two proportions to detect a significant difference in *mPTGER4* positivity between lung cancer patients and healthy controls.

$$n = Z^2_{(1-\frac{\alpha}{2})} \frac{p(1-p)}{d^2}$$

n: Minimum required sample size per group; Z: 1.96 (for 95% confidence level); p: Expected *mPTGER4* positivity rate (15%, based on exploratory study); d: Margin of error (6%).

The calculation yielded a minimum of 91 participants per group. To enhance statistical power, the final sample sizes were 149 NSCLC patients and 100 healthy controls.

2. Methods

* *Study design:* A cross-sectional, case-control study was conducted from April 2021 to August 2024 at the National Lung Hospital and the Institute of Biomedicine & Pharmacy, Vietnam Military Medical University.

Data collection included demographics, clinical history, and biological samples.

For each participant, 10mL of peripheral blood was collected into EDTA tubes, centrifuged within 6 hours, and plasma was stored at -80°C. DNA was extracted using the QIAamp Circulating Nucleic Acid Kit (Qiagen, Germany) and bisulfite-treated with the EZ DNA Methylation-Gold™ Kit (Zymo Research, USA). *mPTGER4* methylation was analyzed via real-time methylation-specific PCR (qMSP) using primer and probe sequences from Wei et al. (2021) [8]. Results were classified as positive (methylation detected) or negative (no methylation) and used for statistical analysis.

* *Study variables:*

The study included two categories of variables: Demographic variables,

including age, gender, BMI, and smoking history; and primary variables, including *PTGER4* DNA methylation test results (positive/negative). These variables were analyzed to assess the association between *mPTGER4* and lung cancer diagnosis.

* *Statistical analysis:*

Data were analyzed using SPSS 20.0. Categorical variables were summarized as frequencies (%) and compared using the Chi-square test, while continuous variables were presented as Mean ± SD and analyzed using the T-test or Mann-Whitney U test (for non-normal distributions).

The diagnostic performance of *mPTGER4* was assessed via ROC curve analysis. Logistic regression estimated the association between *mPTGER4* positivity and lung cancer risk, expressed as ORs with 95%CIs. $p < 0.05$ was considered statistically significant.

3. Ethics

This study was approved by the Ethics Committee for Biomedical Research at Military Hospital 103, Vietnam Military Medical University, under the official decision No. 182/2021/CNChT-HĐĐĐ, dated August 10, 2021. The approval ensured that all procedures adhered to the Declaration of Helsinki and Vietnamese regulations for biomedical research. The data used in this study were collected and analyzed with the approval of the Military Hospital 103, Vietnam Military Medical University. The institution has granted permission for the use and publication of the research findings in compliance with applicable regulations. The authors declare that there are no conflicts of interest related to this study.

RESULTS

1. Characteristics of study participants

The study included 149 NSCLC patients and 100 healthy controls. Table 1 presents the demographic and clinical characteristics of the study population.

The lung cancer group had a significantly higher mean age than the healthy control group (60.9 ± 8.6 vs. 54.9 ± 10.7 years, $p < 0.001$). The proportion of males was also higher among lung cancer patients (65.8%) compared to healthy controls (46.0%, $p = 0.0026$). Regarding BMI, lung cancer patients were more likely to be underweight (7.4% vs. 3.0%) and less likely to be overweight/obese (23.5% vs. 43.0%, $p = 0.0029$). Smoking history was more prevalent among lung cancer patients (57.0%), nearly 1.8 times higher than in healthy controls (32.0%, $p < 0.001$).

Table 1. Demographic and clinical characteristics of study participants.

Characteristics	Lung cancer patients (n = 149)	Healthy controls (n = 100)	p
Age (Mean ± SD, years)	60.9 ± 8.6	54.9 ± 10.7	< 0.001
Male gender (%)	65.8	46.0	0.0026
BMI (Categories, %)	Underweight: 7.4 Normal: 69.1 Overweight: 23.5	Underweight: 3.0 Normal: 54.0 Overweight: 43.0	0.0029
Smoking history (%)	57.0	32.0	< 0.001

2. *PTGER4* methylation status: Lung cancer patients versus healthy controls

To evaluate the diagnostic potential of *mPTGER4*, the positivity rates were compared between lung cancer patients and healthy controls. The results are presented in table 2.

Table 2. *mPTGER4* results by study groups.

***mPTGER4* results**	**Lung cancer patients (n = 149)**	**Healthy controls (n = 100)**	**p**
Negative	121 (81.2%)	97 (97.0%)	< 0.001
Positive	28 (18.8%)	3 (3.0%)	

mPTGER4 positivity was significantly higher in lung cancer patients (18.8%) compared to healthy controls (3.0%, $p < 0.001$). The OR for *mPTGER4* positivity in lung cancer patients was 6.3 (95%CI: 1.9 - 20.4), indicating that lung cancer patients were over 6 times more likely to test positive for *mPTGER4* than healthy individuals.

3. Diagnostic accuracy of *mPTGER4*: ROC analysis

The diagnostic performance of *mPTGER4* was further evaluated using ROC curve analysis. The AUC, sensitivity, and specificity are presented in table 3.

Table 3. Diagnostic performance of *mPTGER4* for lung cancer detection.

Index	Lung cancer patients vs. healthy controls
AUC	0.58
Sensitivity	18.8%
Specificity	97.0%

The AUC was 0.58, indicating modest diagnostic accuracy. The sensitivity was 18.8%, suggesting that *mPTGER4* alone may not be sufficient as a standalone screening test. However, the specificity was 97.0%, meaning that a positive *mPTGER4* result strongly supports lung cancer presence.

4. Logistic regression: Independent association of *mPTGER4* with lung cancer

To evaluate the independent association between *mPTGER4* positivity and lung cancer diagnosis, a multivariate logistic regression analysis was performed, adjusting for potential confounders, including age, gender, BMI, and smoking history. The results are shown in table 4.

mPTGER4 positivity remained a strong independent predictor of lung cancer, with an OR of 6.30 (95%CI: 1.90 - 20.40, $p < 0.001$). Importantly, other factors, including age, gender, BMI, and smoking history, were not statistically significant ($p > 0.05$).

Table 4. Logistic regression: The association between *mPTGER4* and lung cancer diagnosis.

Variable	Coefficient (Coef.)	OR (95%CI)	p
mPTGER4 positive	2.159	6.30 (1.90 - 20.40)	< 0.001
Age	0.027	1.03 (0.99 - 1.07)	0.208
Male gender	-0.331	0.72 (0.30 - 1.36)	0.458
BMI	-0.518	0.60 (0.30 - 1.22)	0.145
Smoking history	-0.018	0.98 (0.95 - 1.01)	0.268

DISCUSSION

These findings highlight the potential of *mPTGER4* as a non-invasive biomarker for lung cancer detection. The significantly higher positivity rate in lung cancer patients compared to healthy controls aligns with previous studies, including the study by Schotten et al. (2021) reported elevated *mPTGER4* in lung cancer patients compared to those with benign pulmonary nodules and chronic obstructive pulmonary disease [3]. The high specificity (97.0%) observed in this study suggests that *mPTGER4* is useful for confirming lung cancer diagnosis, particularly when imaging findings are inconclusive. However, the modest sensitivity (18.8%)

indicates that *mPTGER4* alone may not be sufficient as a primary screening tool. Instead, it should be considered as part of a multi-marker panel or used alongside imaging modalities like LDCT.

A study by Weiss et al. (2017) further supports this approach, showing that combining *mPTGER4* with other biomarkers such as SHOX2 significantly improved diagnostic accuracy for differentiating malignant from non-malignant lung disease. This approach may enhance early detection while minimizing false-positive results.

Moreover, the significant association between *mPTGER4* positivity and lung cancer (OR = 6.3, $p < 0.001$) reinforces its role as an independent biomarker. This association remained significant even after adjusting for age, gender, BMI, and smoking history, suggesting that *mPTGER4* reflects cancer-specific epigenetic changes rather than general risk factors.

Despite these promising results, the study has certain limitations. The relatively small sample size and cross-sectional design limit the ability to assess longitudinal changes and survival outcomes. Future studies with larger, multi-center cohorts and longitudinal follow-up are needed to validate these findings and explore the utility of *mPTGER4* for monitoring disease progression and treatment response.

CONCLUSION

In conclusion, *mPTGER4* is a promising biomarker for lung cancer detection, particularly for confirming diagnosis in patients with suspicious imaging findings. While its limited sensitivity precludes its use as a standalone screening tool, its high specificity makes it a valuable addition to current diagnostic approaches.

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RESULTS OF ALZHEIMER'S DISEASE ANIMAL MODEL INDUCTION BASED ON THE INTRAHIPPOCAMPAL INJECTION OF AMYLOID B-PEPTIDE (1-42) AT VIETNAM MILITARY MEDICAL UNIVERSITY

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Abstract

Objectives: To evaluate some behaviors and histological results of the hippocampus in a rat model of Alzheimer's disease (AD). ***Methods:*** A longitudinal, descriptive study was conducted on 21 Wistar rats. Intrahippocampal amyloid beta (Aβ) injection was performed, followed by behavioral testing and analysis and hippocampal histological examination. ***Results:*** Rats in the AD model showed learning impairment with a reduced average latency time (52.09 ± 10.67s) and reduced latency time on the 5th day (27.08 ± 6.88s) during the learning phase of the Morris water maze (MWM) test. Memory impairment was indicated by a decreased percentage of spontaneous alternations (34.45 ± 7.03%) in the Y-maze test, reduced time spent (14.84 ± 4.61s), and shorter distance traveled (2.26 ± 0.82m) in the target quadrant during the probe trial of the MWM test. Neurodegeneration in the hippocampus was observed, with an increased degeneration score (1.83 ± 0.31). ***Conclusion:*** Intrahippocampal Aβ injection is an effective method for inducing the AD model in rats, characterized by learning and memory impairments as well as neurodegeneration in experimental animals.

Keywords: Alzheimer's disease model; Hippocampus; Stereotaxic surgery.

INTRODUCTION

Alzheimer's disease is a progressive, age-related degenerative brain disorder with a multifactorial and heterogeneous etiology. Among many hypotheses proposed for the pathogenesis of AD, the amyloid hypothesis is the most widely accepted pathological mechanism. Extracellular amyloid plaques, intracellular neurofibrillary tangles, neuronal degeneration, and consequent memory impairment are hallmark features of AD [1].

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AD models serve as valuable tools for replicating pathological changes, deciphering the disease's pathogenesis, and testing novel therapeutic approaches [1]. Many AD models have been developed on a global scale, with the rat intrahippocampal Aβ injection model being demonstrated as one of the most representative and reliable AD animal models [2]. However, this specific AD model has not yet been reported in Vietnam. Therefore, this study aims to: *Evaluate behaviors and histological outcomes of AD rat model induction via intrahippocampal Aβ injection.*

MATERIALS AND METHODS

1. Subjects

Including 21 adult healthy Wistar rats experimented in strict compliance with The Animal Center Guidelines for Care and Use of Laboratory Animals at Vietnam Military Medical University.

* *Location and time:* The study was conducted at the Practical and Experimental Surgery Department from September 2022 to July 2024.

2. Methods

* *Study design:* A longitudinal, descriptive study.

* *Sample size:* The sample size was determined using the formula:

$$n = DF/k + 1$$

n: Sample size of each group; DF: Degree of freedom with a value of 10 or 20; k: Number of comparison groups. With k = 2, the sample size of each group is: $6 \leq n \leq 11$ [3]

* *Research procedure:*

21 rats were divided into 3 groups: The control group (n = 6), the sham operation (SO) group (n = 8), and the Aβ injection (AB) group (n = 7).

Drug: $A\beta_{1-42}$ (ab120959) was purchased from Abcam and was dissolved in PBS (2 μg/μL).

Surgery: Rats were anesthetized with intraperitoneal ketamine and xylazine, and their heads were shaved and fixed onto the stereotaxic apparatus (Japan). Injection points (coordinates: AP = -4mm, ML = ± 2.2mm) were marked in stereotaxic frame. The hole was drilled into the skull using an electric driller (Komax drill, Germany). The Hamilton needle (Hamilton 10μL pump, USA) was lowered into the hippocampus at a depth of DV = -3.2mm. For the AB group, 2.5μL of Aβ (2 μg/μL) was injected bilaterally. In the SO group, the needle was inserted without injecting any substance. After the injection, the needle was carefully withdrawn, the skull opening sealed with composite solder, and the skin sutured [2].

Behavioral test:

+ Y-maze test: The Y-maze test was performed according to the procedures

described in a previous study (Prieur E et al., 2019). The Y-maze is a three-arm maze with equal angles between all arms of 75cm in length and 15cm in width, with 15cm-high walls. The maze floor and the walls were constructed using black-painted wood. Rats were initially placed within one arm, and the sequence and number of arm entries were recorded over an 8-minute period for each rat and monitored using a video tracking system (ANY-maze, Stoelting, USA) [9].

+ MWM test: The MWM test was performed according to the procedures described in a previous study (Vorhees CV et al., 2006) [4]. The MWM is a black circular pool (150cm in diameter and 60cm in height). The circular pool was filled with water at a temperature of 21°C ± 1°C. The pool was divided into four equal quadrants. A transparent platform (10cm in diameter and 28cm in height) was centered in one of the four quadrants of the pool and submerged 2cm below the water's surface so that it was invisible at the surface. The water maze experiment was performed on 6 days. On the 5th of learning, the rat received swimming training for 120 seconds in the presence of the platform. The rat underwent a daily session of four training trials each, with an inter-trial interval of 2 minutes. In each training trial, rats were placed in water facing the wall of the pool in a randomly selected pool quadrant. Once the rat located the platform, it was allowed to remain on it for 10 seconds. If the mouse failed to locate the platform within 120 seconds, it was placed on it for 10 seconds and then removed. On the 6th day, in the probe trial, the platform was removed, and rats were tested for memory retrieval by swimming for 60 seconds. The trajectory of each rat while swimming was monitored using a video tracking system (ANY-maze, Stoelting, USA).

Histological analysis: At the end of the experimental phase, the hippocampus of 6 rats of each group (AB and SO) was extracted and fixed in 10% formalin, then paraffin block was cast and cut into 3 - 4µm slices and stained with Hematoxylin-Eosin (HE). The specimens were observed under an optical microscope.

** Research variables and indicators:*

Behavioral assessment indicators: Determined by Anymaze software:

+ Number of alternating movements is the number of times the rat successfully enters three consecutive wings (ABC, ACB, BCA, etc.) and percent alternation [9]. % Alternation = (Number of alternating movements)/(Total number of times entering the wings -2) x 100%.

+ The average escape latency (s) on each day of learning. The swimming time (s) and distance (m) at the target quadrant in the probe trial [4, 5].

Pathological indicators:

Neurodegeneration is determined by cell damage in the hippocampus on HE staining, which includes cytoplasm eosinophilic, vacuole, dispersed chromatin, and loss of nuclear membrane integrity. Each damage is given 1 score; the neurodegenerative score is the sum of the scores of the lesions [6].

* *Data collection and processing methods:* Behavioral data were extracted from the Excel files of Anymaze software. Statistical analysis was performed using SPSS software. Quantitative variables were expressed as Mean ± SEM. Alternation (%) in the Y-maze test, the time and distance of swimming in the target quadrant in the MWM probe trial were analyzed using one-way ANOVA, the Tukey's post-hoc test in case of multiple comparisons. Escape latencies and swimming distance in the training trials in the MWM test were analyzed using repeated-measures two-way ANOVA and Bonferroni adjustment for multiple comparisons. Neurodegenerative scores were analyzed using the Independent-Sample T-test. The differences were considered statistically significant with $p < 0.05$.

3. Ethics

The research was conducted according to Decision No. 3424/QĐ-HVQY dated September 19, 2022. The Department of Practical and Experimental Surgery granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the research.

RESULTS

1. Learning performance of AD model in rats

Repeated measure two-way ANOVA test showed that there was a difference in latency time between groups [($F(2, 10) = 14.07$; $p = 0.001$]. Bonferroni adjustment for multiple comparisons showed that the latency time of the AB group (52.09 ± 10.67s) was higher than that of the SO group (22.84 ± 2.62s) and the control group (20.94 ± 1.93s). On the 5th day of the learning phase, the latency time of the AB group (27.08 ± 6.88s) was higher than that of the SO group (7.18 ± 0.88s) and the control group (6.73 ± 0.83s); the difference was statistically significant with $p < 0.05$. There was no difference in latency time between the SO group and the control group during learning days with $p > 0.05$.

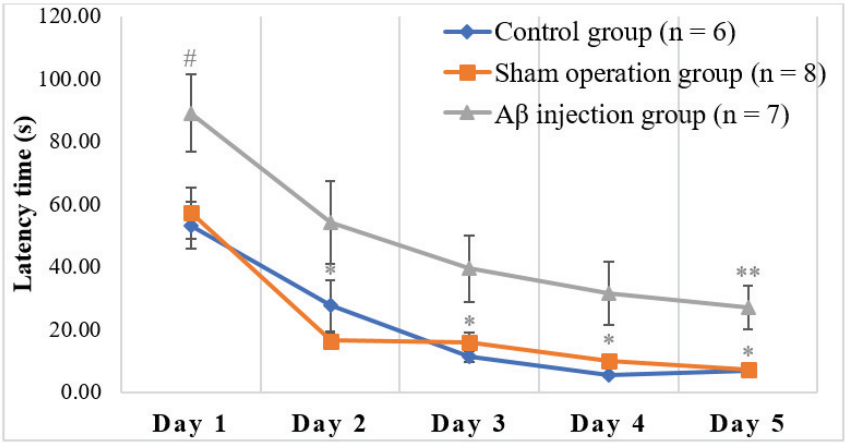

Figure 1. The swimming time to the platform of rats in the MWM.

*(**: Compare between the AB group and SO group, control group (** p < 0.05); *: Compare between the SO and control group (* p > 0.05). #: Compare between three group (# p > 0.05))*

2. Memory impairment of AD model in rats

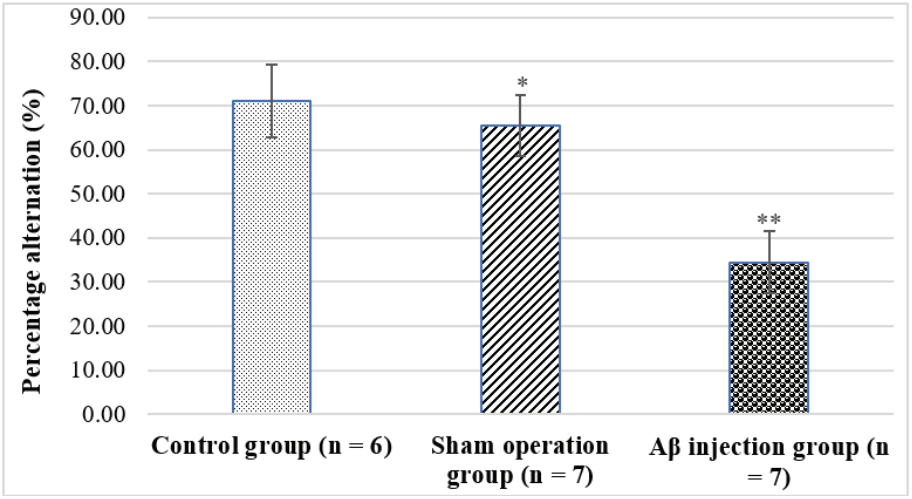

Figure 2. Percentage alternation of rats in Y-maze.

*(**: Compare between the AB group and SO group, control group (** p < 0.05); *: Compares between the SO and control group (* p > 0.05))*

One-way ANOVA test showed that there were significant differences in % alternation between the three groups [F(2, 19) = 7.36; p = 0.005], Tukey's post-hoc test showed that % alternation in the AB group (34.45 ± 7.03%) was lower than that in the SO group (65.44 ± 6.84%) and control group (71.09 ± 8.19%); the

difference was statistically significant with $p < 0.05$. There was no difference in % alternation between the SO group and the control group with $p > 0.05$

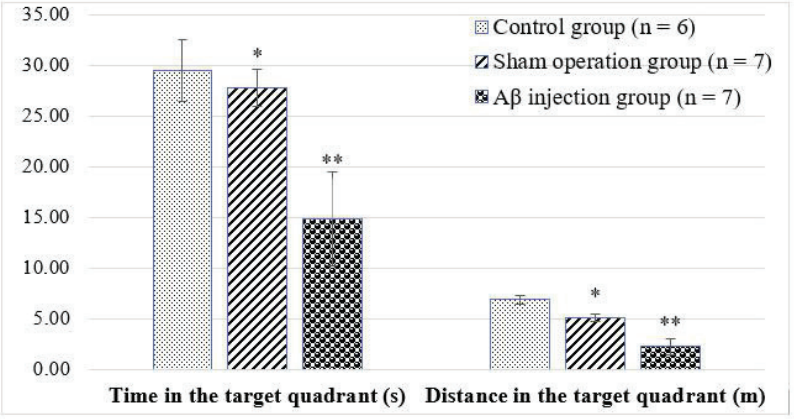

Figure 3. Time and distance of the rats at the target quadrant.

*(**: Compare between the AB group and SO group, control group (** $p < 0.05$); *: Compare between the SO and control group (* $p > 0.05$))*

One-way ANOVA test showed that there were significant differences in time [$F(2, 20) = 5.89$; $p = 0.01$], distance [$F(2, 20) = 15.32$; $p < 0.001$] of swimming in the target quadrant between the three groups. Tukey's post-hoc test showed that the swimming time of the AB group (14.84 ± 4.61s) was shorter than that of the SO group (27.79 ± 1.78s) and control group (29.52 ± 3.06s); the swimming distance of the AB group (2.26 ± 0.82m) was shorter than that of SO group (5.11 ± 0.34m) and control group (6.87 ± 0.48m).

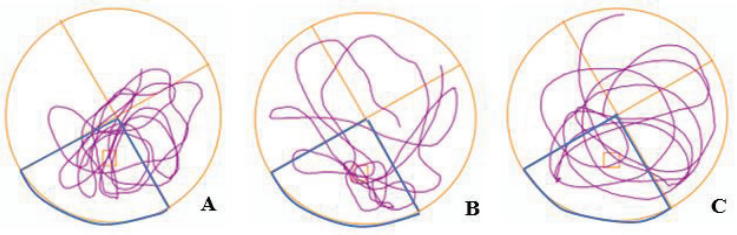

Figure 4. Illustration of the swimming path of rat in the MWM.

(A: The swimming path of rats in the control group; B: The swimming path of rats in the SO group; C: The swimming path of rats in the AB group)

Rats in the AB group spent less time swimming in the target quadrant (blue border) than those in the control group and SO group.

Figure 5. Neurodegenerative scores in the hippocampus of model rats.

*(**: Compare between the AB group and SO group (**p < 0,05))*

Independent Sample T-test showed that the neurodegenerative score in the hippocampus of the AB group (1.83 ± 0.31) was higher than that of the SO group (0.67 ± 0.33); the difference was statistically significant with $p < 0.05$.

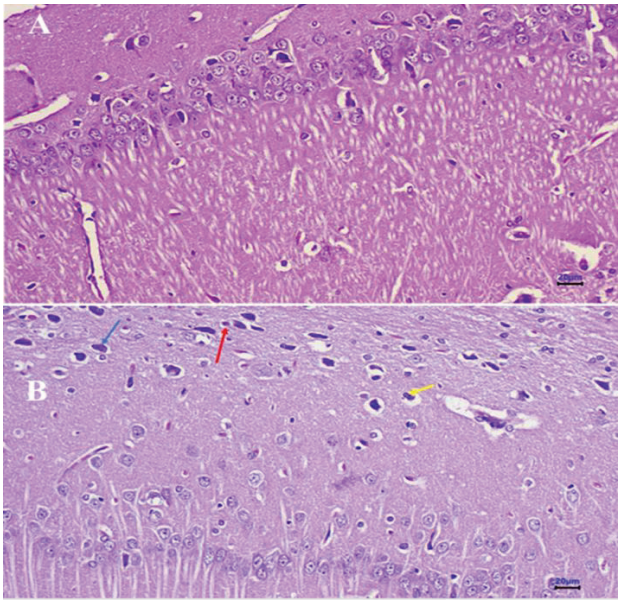

Figure 6. Microscopic image of rats' hippocampus.

(Normal hippocampus in SO rats (A), neurodegeneration in AB rats (B) with dispersed chromatin (green arrow), vacuoles (yellow arrow), eosinophilic bodies (red arrow))

DISCUSSION

1. Intrahippocampal Aβ injection impairs learning and memory performance in AD model rats

Memory is the brain's ability to categorize, encode, store, and retrieve acquired information. Declarative memory is stored in the middle of the temporal lobe and the hippocampus. The hippocampus plays a significant role in spatial learning and memory. The hippocampal subregions support the creation of episodic memory; the CA3 (cornus ammonis: CA) region generates sharp wavelets to consolidate memories. CA1 and CA3 have neurons that represent points in the space of an environment, which Nadel L et al. (1971) called place cells; this finding is called the place cell theory [4]. Neuronal connections in the CA of the hippocampus form a trisynaptic loop that plays a role in converting short-term memory into long-term memory and maintaining by unidirectional progression of synaptic connections through the loop.

Memory impairment is a hallmark symptom of AD, characterized by impaired acquisition of new information and progressive loss of information retrieval over time. Prieur E et al. (2019) showed that rats with a high % alternation had good working memory because they recalled the wings they had visited and tended to visit the recently visited wings less. This requires interaction across several brain regions, such as the hippocampus and prefrontal cortex [9].

In the SO group, 1 rat had a stimulating behavior on the behavioral test day on Y-maze; to avoid data interference, we did not include it in the analysis. The study showed that AB rats had impaired memory compared to SO rats and control rats. The % alternation of the AB group (34.45%) was lower than that of the SO group (65.44%) and control group (71.09%). This result is consistent with the study of Jeong H et al. (2021) on ICR mice (n = 10/group), showing that AB mice had memory impairment with % alternation (50%) lower than the control group (70%) [10].

The MWM test to assess learning ability and spatial memory is based on the survival instinct of animals; when put in water, they have to swim to survive. Learning ability was assessed in the training phase. Othman M et al. (2022) showed that the latency time gradually decreased, reflecting increased learning ability [4]. The results of the study showed that AB rats had reduced learning ability compared to SO and control rats. The average latency time of 5 days of learning in the AB group was higher than that of the SO group and control group; the difference was

on the 5th day of the learning phase. This result is similar to previous studies, such as Rahman S et al. (2020) on the AD model of intrahippocampal Aβ injection in Wistar rats (n = 8/group), which showed that latency time in AB rats was higher than that in the control group [6].

Spatial memory was assessed in the probe test phase (on the 6th day of the MWM test). Time and distance swimming within the target quadrant were the most frequently used parameters. Previous studies have shown that longer time spent in the target quadrant reflects better memory retrieval because they remember the area as safe [4, 5]. The results showed that AB rats had impaired memory compared to SO and control rats. The swimming time in the target quadrant of the AB group (14.84s) was lower than that of the SO group (27.79s) and control group (29.52s). The swimming distance in the target quadrant of the AB group (2.26m) was lower than that of the SO group (5.11m) and control group (6.87m). This result is similar to the study of Rahman S et al. (2020), which shows that the time of the AB group (13s) was smaller than that of the control group (22s). Jeong H et al. (2021) showed that this time in the AB mice (15s) was lower than that of the control group (25s) [10].

2. Aβ injection causes hippocampal neurodegeneration

The pathological features of AD include extracellular amyloid plaques, intracellular neurofibrillary tangles, and degeneration and loss of neurons and synapses [1]. Aβ causes synaptic dysfunction, changes in cell membrane permeability, calcium homeostasis, oxidative stress, inflammation, and neurodegeneration [8]. Faucher P et al. (2016) showed in animal models that Aβ causes molecular and cellular changes in the hippocampus, resulting in cognitive impairment even in the absence of amyloid deposition [7]. The microscopic pathological finding of the hippocampus by HE staining showed that the AB rats group had a higher neurodegenerative score (1.83) than the SO rats group (0.67). The results of the study are consistent with previous studies that intrahippocampal Aβ injection causes neurodegeneration, such as Rahman S et al. (2020) showed that the number of degenerated neurons in the AB rats (70) was higher than the control group (5) [6].

CONCLUSION

This study successfully established an AD model using intrahippocampal Aβ injection in rats characterized by learning and memory impairments as well as neurodegeneration in the hippocampus.

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OPTIMIZATION OF THE FLOW CYTOMETRIC METHOD FOR ANALYZING NK CELL CYTOTOXIC ACTIVITY IN BREAST CANCER

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Abstract

Objectives: To establish a protocol for analyzing NK cell cytotoxic activity (NKAc) in breast cancer patients using the flow cytometric technique. ***Methods:*** We first optimized the protocol based on the previously published studies, focusing on choosing fluorescent dye concentration, incubation time length, and Effector:Target cell ratio (E:T ratio). Subsequently, we preliminarily applied the established protocol to characterize NKAc in some breast cancer patients and healthy controls. Lastly, we compared the NKAc versus NK cell secretory activity (NKAs) data obtained from the study. ***Results:*** The fluorescent dye concentration could be used according to the manufacturer's suggestion (CFSE: 2.5 - 10µM; Zombie NIR: 1:1000 - 1:100 dilution). The co-culture period of effector and target cells and the E:T ratio could be 4 hours and 5:1, respectively. NKAc was reproducible for 1 month. 2/7 breast cancer patients had NKAs < 200 pg/mL and NKAc < 10%. ***Conclusion:*** It is feasible to examine NKAc in breast cancer patients via the flow cytometric method established in this study. Further studies are needed to validate the diagnostic and prognostic value of NK cell function tests.

Keywords: NK cell cytotoxic activity; NK cell secretory activity; Flow cytometry; Breast cancer.

INTRODUCTION

NK cells, an important population of the innate immune system, protect the body against viruses and malignant cells. Infected cells or malignant cells often express activating ligands and reduce inhibitory ligands for the counterpart receptors of the NK cells, which stimulates

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NK cells to perform two major NK cell functions: NKAc and NKAs. It has been well documented that NKAc and NKAs were suppressed in a subgroup of cancer patients, especially in prostate, lung, colon, and breast cancer, though the mechanism of these observations has not been clarified yet [1, 2, 3, 4, 5]. The effectiveness of NK cell therapy in cancer could be indicated by clinical and immunological responses. Therefore, it is important to have a good system to quantify the activity of NK before and after NK cell therapy in cancer patients. In terms of NKAs, there is an IVD assay to measure the level of IFN-γ secretion of Promoca (engineered recombinant cytokines)-stimulated pNK via ELISA availably [6]. There is currently no standardized commercial assay to measure NKAc; therefore, each laboratory must set up its own protocol and document the reference range of NKAc [1]. In this study, we aim to: *Establish the protocol for measuring NKAc and apply it to breast cancer samples. Specifically, we tested different conditions of fluorescent dye concentration for labeling the target cells, incubation time, and the E:T ratio. Then, we preliminarily apply the established protocol for measuring NKAc of some breast cancer patients and then discuss the method to analyze the obtained data.*

MATERIALS AND METHODS

1. Materials

Peripheral blood was collected from breast cancer patients (hospitalized in the Military Hospital 103) and healthy controls (the medical staff of the Department of Immunology, Vietnam Military Medical University).

* *Exclusion criteria:* Donors were in status of acute infection or autoimmune diseases.

* *Location and time:* At the Department of Oncology, Military Hospital 103 and the Department of Immunology, Vietnam Military Medical University, from June to July 2024.

2. Methods

* *Study design:* To optimize the protocols, we used 4 blood samples from healthy medical staff. Then, apply the established protocol to the blood samples from 7 breast cancer patients and 5 healthy medical staff.

* *NKAc assay:*

Live fluorescent-CFSE-labelled K562 cells were killed when co-cultured with the NK cells (CFSE - 5-(and 6)-Carboxyfluorescein diacetate succinimidyl ester of CFDA SE, Biolegend, #423801). Dead K562 cells can be subsequently stained by dead-cell fluorescent dye (Zombie NIR, Biolegend, #423105) and counted by flow cytometry with a dual laser (red, blue laser). Heparinized

blood was collected from the donors. Peripheral blood mononuclear cell (PBMC) preparation was performed via the Ficoll-gradient separation method. The percentage of NK cells was analyzed via flow cytometry (described below). K562 cancer cell line was bought from Cell Line Service (CLS, Germany). Each condition was prepared in triplicate wells.

Flow cytometric analysis: Label the E:T suspension with the dead-cell fluorescent dye (Zombie NIR, Biolegend) following the manufacturer's recommendation in 15 minutes before analyzing on the flow cytometry analyzer (Novocyte, ACEA, USA). Analyzing and interpreting the data following Wu et al. (2020) [7]. Percent specific lysis [%] of K562 cells was calculated as the following method based on the percentage of dead K562 cells (CFSE+Zombie NIR+):

$$\text{Specific lysis [\%]} = [(n_1-n_2)/(100-n_2)]*100$$

n_1: Percentage of dead K562 cells in the E:T wells (Average of the triplicates); n_2: Percentage of dead K562 cells in the negative control wells (Average of the triplicates).

* *NKAs assay:*

NKAs or IFN-γ levels in the supernatant are measured via the ELISA method following the standard protocol of the IVD NK-VUE kit (ATGen, Korea).

* *NK cell subset analysis by flow cytometry:* PBMCs after Ficoll gradient separation were washed by PBS-1X and stained with a cocktail of fluorescent conjugating antibodies (Anti-CD45-PercP, anti-CD3-FITC, anti-CD56-PE from Biolegend). NK cells are defined as CD45+CD3-CD56+ by flow cytometry analysis.

* *NK cell purification*: To purify NK cells from peripheral blood, we followed the protocol of Phuc et al. (2023) [3] by using the Miltenyi Biotec human NK cell isolation kit.

* *Statistical analysis:* Data are presented as mean (standard deviation-SD). Statistical analysis was performed on Microsoft Excel (2023). T-test was used for the comparison of the two populations; $p < 0.05$ was considered to be statistically significant. Coefficient correlation (r) is interpreted as follows: 0.00 - 0.199: Very weak; 0.2 - 0.399: Weak; 0.4 - 0.599: Medium; 0.6 - 0.799: Strong; 0.80 - 1.00: Very strong.

3. Ethics

The study was approved by the Institutional Review Board of Vietnam Military Medical University (2352/QĐ-HVQY, June 2024). All donors were explained and consented to the study. The participants did not pay any fee related to the study. The Department of Oncology, Military Hospital 103, Vietnam Military Medical University granted permission for the use and

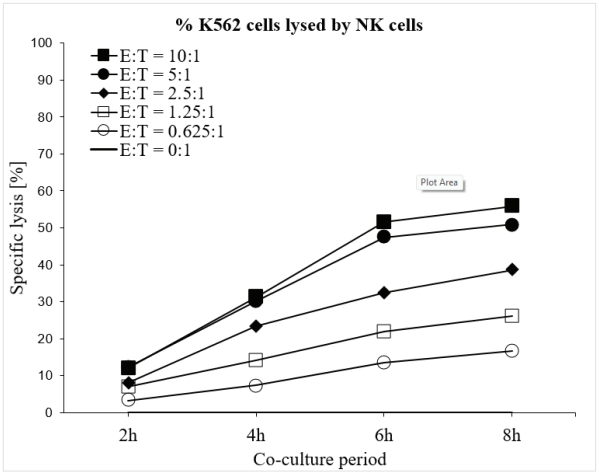
publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS AND DISCUSSION

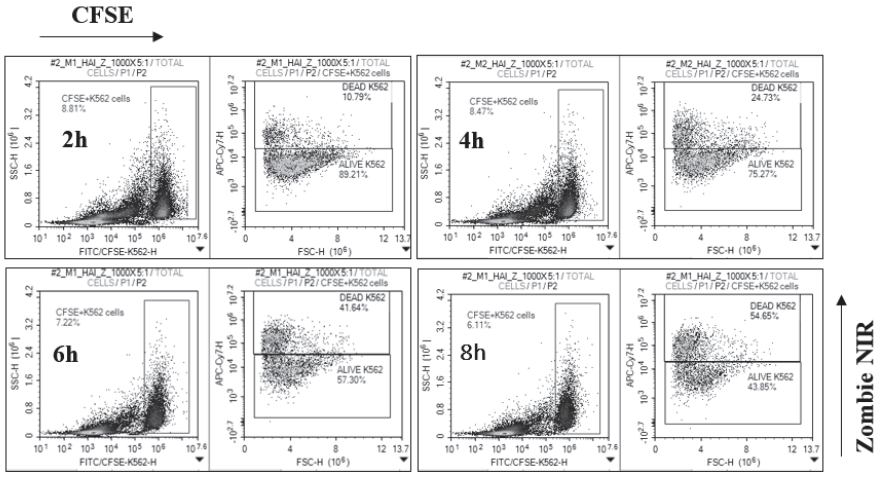
In an attempt to set up our own protocol to analyze NKAc, we have tested the following conditions. Firstly, we chose two fluorescent dyes (CFSE and Zombie NIR) to stain alive vs. dead target cells as they are read by the two distinct fluorescent channels (FITC by blue laser and APC-Cy7 by red laser), which means there is no overlapping signal between live cells (CFSE-FITC positive) and dead cells (Zombie NIR-APC-Cy7). In the procedure before co-culture, K562 cells would be stained by CFSE (conjugating to intra-cellular proteins). K562 cells stained with CFSE concentration > 2.5μM would show a strong and uniform population on flow cytometry (Median fluorescent intensity - MFI exceeded 10^6) (Data not shown). It is important to note that CFSE has been reported to lose its fluorescent signal during long incubation time [9]; thus, we thought it would be optimal to use 5μM of CFSE for pre-staining K562 cells. Next, we performed heat-killed CFSE-labelled K562 cells and stained them with dead-cell fluorescent dye (Zombie NIR - ZB). The dead cell population (CFSE+ZB+) could be clearly separated from the alive cell population (CFSE+ZB-) even when using the modest concentration of ZB (1:1000 dilution) as recommended by the manufacturer's suggestion (Data not shown). Thus we fixed the following condition of 5μM CFSE and 1:1000 diluted ZB concentration for staining alive and dead target cells when analyzing them by flow cytometry.

Most of the published protocols used fresh PBMCs instead of purified NK cells, and the reasons could be: (1) Isolating NK cells requires time & consumable costs and weakens the NK cell activity; (2) NK cells might be the major sole effector cells to kill K562 cells as it was shown that PBMCs without NK cells did not kill K562 cells [9]. However, since NK cells occupy a relative fluctuation frequency (5 - 15%) of PBMCs, fixing the number of PBMCs as the effector cells might be not a good approach. We always checked the frequency of NK cells (CD3-CD56+) on flow cytometry and adjusted the density of PBMCs based on the NK cell: K562 cell ratio. About the E:T ratio in terms of NK cells as the effector cells, we researched the previous publications and found that the E:T ratio has been used around 5:1 and 1:1 [1, 6, 9]. About incubation time, most papers showed a consensus of 4 hours [1, 6, 9]. In our established system, we checked again with a matrix of time (2 - 8 hours) and the E:T ratio (10:1 to 0.625:1). The data in figure 1

suggested that: (1) Incubation time should be 4 hours or > 2 hours to see a properly lysis activity of target cells; (2) There were no clear differences of NKAc between the two conditions of E:T = 10:1 and 5:1. Thus, we fixed the conditions of time and E:T ratio as 4 hours and 5:1, respectively.



(a)



(b)

Figure 1. NKAc varied depending on the parameters of co-culture time and E/T ratio.

(a) The specific lysis of K562 cells in the co-culture system with NK cells/PBMCs at different conditions of time and E:T ratios;

(b) A representative image of flow cytometric analysis of the co-culture system with the E:T ratio = 5:1 and different time incubation. The data are presented as the mean of 4 independent samples from healthy donors. E: Effector cells, T: Target cells.

Subsequently, we checked the stability of the NKAc assay by testing again the NKAc of 4 healthy donors with a time interval of 1 month. The average of NKAc (n = 4) remained unchanged with p > 0.05 (*Table 1*). Thus our protocol of the NKAc assay might be stable.

Table 1. NKAc of the donors at two timepoints (1-month interval).

Sample #	Specific lysis (%)	
	Timepoint #1	Timepoint #2
#1	14.3	20.5
#2	18.4	17.3
#3	24.0	28.4
#4	31.9	31.0
Mean	22.2	24.3
SD	7.6	6.5
p	> 0.05	

We then tried to apply the established assay to preliminarily examine a small number of donors, including some newly-diagnosed breast cancer patients (n = 7) and a healthy control group (n = 5). NKAc of the patients and the healthy control group were 26.0 ± 15.4% and 17.2 ± 8.7%; there was no significant difference with p > 0.05 (*Table 2*).

Table 2. NKAc in the study groups.

Specific lysis, % (Mean, SD)	
Breast cancer patients (n = 7)	26.0 (15.4)
Healthy controls (n = 5)	17.2 (8.7)
p	> 0.05

In order to preliminarily validate the optimized protocol, we tested the NKAc of the NK cells in PBMCs or the purified NK cells in the co-culture system with K562 cells with the defined E:T (NK cells: K562 cells). Data shown in table 3 suggested that the NKAc of NK cells in PBMCs could be similar to that of the purified NK cells.

Table 3. NKAc (% K562 cells) lysed by NK cells in PBMCs or purified NK cells.

E:T ratio (Mean ± SD)	NKAc (specific lysis, %)	
	PBMCs (NK cells)	Purified NK cells
E:T = 10:1	24.4 ± 2.3	35.6 ± 1.8
E:T = 5:1	20.9 ± 1.4	25.0 ± 1.7
E:T = 2.5:1	18.4 ± 0.2	18.9 ± 2.0
E:T = 1.25:1	13.5 ± 1.3	13.1 ± 1.1
E:T = 0.625:1	11.6 ± 0.7	9.7 ± 0.2
p	> 0.05	

Altogether, the study data suggest a favorable protocol that the NKAc assay could be performed by fixing the following criteria: (1) Staining CFSE with 5μM, Zombie NIR with 1:1000 dilution; (2) E:T co-culture in 4 hours with the E:T = 5:1; PBMCs density should be adjusted based on the frequency of NK cell in PBMCs.

Lastly, we discuss the NK cell activity in general when it comes to both NKAs and NKAc. NKAs were also documented in the 12 study participants. Our small-size data show that 3/7 breast cancer patients had NKAc levels < 500 pg/mL and 2/7 breast cancer had NKAs levels < 200 pg/mL; whereas we did not notice any NKAs of 5 healthy controls < 500 or 200 pg/mL. Interestingly, 2 breast cancer patients with NKAs levels < 200 pg/mL also showed the lowest NKAc of less than 10% (*Table 4*). There is a medium strength of correlation between NKAs and NKAc in this study (r = 0.523), though the sample size is small.

Table 4. NKAs and NKAc in the study groups.

Study groups Mean (SD)	NKAs (IFNγ, pg/mL)	NKAc (specific lysis, %)
Patients (n = 7)	1243 (1192)	26.0 (15.4)
Healthy controls (n = 5)	1505 (1042)	17.2 (8.7)
p	> 0.05	> 0.05
	r = 0.523	

In this study, we tried to establish an in-house protocol for examining NKAc of breast cancer patients based on the previously published characterization of non-standardized methods of assessing NKAc. Our study suggests a method that is rather feasible and stable to examine the NKAc of the patients.

However, as a preliminary study, our study has mainly focused on optimizing the protocol and has only been tested in small-size samples, which might hinder the strong conclusion of the study. Besides, we did not validate our established protocol with another gold-standard protocol. Thus, we did not give a reference range of NKAc; this task requires analyzing NKAc in larger-scale samples.

CONCLUSION

It is possible to assess the NKAc in breast cancer patients using the flow cytometric technique developed in this study. Additional research is required to confirm the diagnostic and prognostic significance of NK cell function tests.

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RESEARCH ON CHANGES IN SOME CHARACTERISTICS OF ACTIVATED UMBILICAL CORD BLOOD PLATELET-RICH PLASMA AFTER STORAGE AT NORMAL COLD TEMPERATURE

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Abstract

Objectives: To evaluate sensory changes, infection, epidermal growth factor (EGF), and vascular endothelial growth factor (VEGF) concentrations in activated umbilical cord blood platelet-rich plasma (PRP) after cold storage at 2 - 6 degrees Celsius (°C). ***Methods:*** The study was conducted on 8 umbilical cord blood samples and 8 venous blood samples of adults. ***Results:*** EGF concentrations in PRP from umbilical cord blood and adult blood after activation were 115.72 (51.59 - 217.61) pg/mL and 113.04 (69.71 - 155.73) pg/mL ($p > 0.05$), with VEGF concentration being 193.99 (75.58 - 320.25) pg/mL and 16.12 (11.70 - 49.03) pg/mL, respectively, $p < 0.01$. After cold storage at 2 - 6°C for 7 days, 10 days, and 14 days, the concentrations of EGF and VEGF in PRP from activated umbilical cord blood showed no difference compared to immediately after activation ($p > 0.05$). PRP remained clear and yellow, with no signs of infection. ***Conclusion:*** PRP from umbilical cord blood has high VEGF concentration, no significant change in appearance, EGF and VEGF concentrations, and sterility after cold storage for 7 - 14 days.

Keywords: Platelet-rich plasma; Umbilical cord blood; Change; Cold storage.

INTRODUCTION

Platelet-rich plasma contains many growth factors that stimulate wound healing stages. These factors are much higher than normal when concentrated from plasma [1]. Autologous PRP has many advantages in treating chronic wounds but encounters some difficulties when implementing. The source is umbilical cord blood with a lot of potential [2, 3].

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Stored umbilical cord blood is used to treat many diseases in the form of allogeneic [2, 3]. In Vietnam, umbilical cord blood is mainly used for stem cell products to treat diseases or as a precaution for potential future autologous disease treatment. Blood and blood products are also studied for application in cold storage conditions, usually at 2 - 6°C, which is convenient for the actual treatment process [4]. Umbilical cord blood contains hematopoietic stem cells and mesenchymal stem cells; however, the number of stem cells is quite low, leading to slow graft growth and a high risk of infection [5]. Therefore, the application of activated umbilical cord blood PRP in wound treatment mainly depends on the role of growth factors. EGF and VEGF are two of the growth factors from platelet alpha granules having an important role in initiating and stimulating the four stages of wound healing [6, 7]. This study aims to: *Evaluate the changes in some characteristics of PRP from activated umbilical cord blood after conventional cold storage to support further understanding in the application of umbilical cord blood PRP in clinical wound treatment.*

MATERIALS AND METHODS

1. Subjects

8 samples of cord blood immediately after birth from healthy pregnant women and 8 samples from healthy adult donors.

* *Inclusion criteria:*

Pregnant women aged 20 - 45; no history of obstetric diseases, no concomitant diseases such as congenital diseases, tuberculosis, cancer, autoimmune diseases, mental illness, screened for HIV, HBV, HCV with negative results before giving birth, no obstetric complications; gestational age > 36 weeks; born within 24 hours of rupture of membranes; newborn weight ≥ 2.800g.

Volunteer donors: Agree to give blood samples; no acute or chronic diseases; HIV, HBV, HCV screening tests with negative results; no fever (< 38°C); aged 20 - 45 years old.

* *Location and time:* Umbilical cord blood was collected at the Department of Obstetrics and Gynecology, Military Hospital 103. Blood from healthy adults was collected, and PRP extraction, hematology, biochemistry, and bacterial culture were performed at the Department of Paraclinical Medicine, Le Huu Trac National Burn Hospital. EGF and VEGF were tested at the Military Medical Research Institute, Vietnam Military Medical University. The study was conducted from May 2023 to September 2024.

* *Raw materials and equipment:* PRP separation kit New-PRP Pro kit; Thermo Fisher Scientific's human EGF, VEGF test kit; syringes, blood collection tubes for hematology and biochemistry tests, bacterial culture kits; newborn weight ≥ 2.800g; K4500 machine with 18 hematological parameters by Sysmex of Japan; AU480 blood biochemistry analyzer; ELISA machine; DLAB centrifuge; biological and immunological laboratory equipment; freezer -80°C; refrigerator for storage at 2 - 6°C.

3. Methods

* *Study design:* Umbilical cord blood and venous blood were collected according to aseptic procedures, blood was taken into test tubes in the PRP extraction kit, transferred to the laboratory of the testing department immediately after collection, and PRP was extracted using the double centrifugation method. PRP was activated with $CaCl_2$ (ratio 1:10). Activated PRP was stored at 2 - 6°C, and characteristics and test indexes were collected and evaluated the day after activation, 7 days, 10 days, and 14 days after activation.

* *Method of implementation:*

Umbilical cord blood is aspirated with a sterile 10mL syringe immediately after the aseptic procedure, the blood is collected into the test tube of the kit. A negative pressure needle is used to insert directly into the test tube of the kit to collect peripheral blood from the volunteer donor according to the procedure. PRP was separated according to the procedure of the kit. Whole blood is partially collected in the same process, and a quantity of PRP after activation is tested, evaluated, and compared.

PRP preparation process as described in the instructions of GeneWorld:

- Phase 1 (collection of unactivated PRP): Take venous/umbilical cord blood into test tubes, each tube with a total of 10mL (including 1.5mL of available anticoagulant). Centrifuge the first blood tube to separate plasma, red blood cells, and platelets at 2,000rpm x 10 minutes. Gently aspirate the yellow plasma layer on top, aspirate the white layer next to the red blood cell layer (buffy coat layer), and put it into a centrifuge tube labeled PLASMA (total volume recovered from 3 tubes is about 12 - 16mL). Centrifuge the second time at 3,500rpm x 5 minutes with a PLASMA tube with a counterweight to obtain a solution that separates into 2 layers. Gently aspirate the yellow liquid on top and discard it, leaving 6mL of liquid at the bottom of the tube. This is the unactivated PRP.

- Phase 2 (activate PRP with $CaCl_2$): Aspirate the remaining 6mL of PRP into another test tube that contains a small amount of $CaCl_2$ activator solution. Add and mix well for 2 - 5 minutes until a solid mass appears. Use a sterile pipette to gently rotate the solid mass until it separates from the tube wall. Wait 5 - 15 minutes until the solid mass shrinks completely, then aspirate the solid mass. The final product is a clear yellow activated PRP solution with a volume of about 4 - 5mL.

* *Measurement of EGF and VEGF:* Using the ELISA reaction method. Quantify the presence of antibodies/ antigens by the "sandwich" method. A layer of antibody specific for EGF/VEGF is coated on the well plate. The base sample, test sample, and antibody are added to the well if the biotin of the EGF/VEGF receptor is detected. At the bottom of each well, EGF/VEGF will attach itself to the available antibody at the bottom and to the biotin-linked detection antibody after incubation. Remove all non-specific binders, and add streptavidin - HRP. Wash and add substrate solution to the well. A color reaction occurs in which the intensity of the color matches the concentration of EGF/VEGF. Stop the reaction with another solution. Measure the color intensity, then calculate the concentration of EGF/VEGF in the sample.

* *Bacterial culture:* Take 0.1mL of activated PRP and culture bacteria according to the routine procedure of the microbiology lab, and determine the species and number of bacteria (if any grow).

* *Research indicators*

Color characteristics, clarity-turbidity, quantitative test indexes of EGF and VEGF of activated PRP (T1), after activation, stored cold at 2 - 6°C for 7 days (T7), 10 days (T10) and 14 days (T14); PRP infection testing culture were performed at the above times.

* *Data processing:* The obtained data are calculated as the average in the form of $\overline{X} \pm$ SD compared by T-test or in the form of Q2 (Q1 - Q3) compared by Mann Whitney U-test, Wilcoxon test. Analyze data using SPSS 22.0 software. There are statistically significant differences when $p < 0.05$.

4. Ethics

The research content was approved by the Expert Subcommittee for evaluation according to Decision No. 4692/QD-HDTSSDH dated November 18, 2022, and approved by the Ethics Council in Biomedical Research of

Military Hospital 103 (certification No. 14/CNChT-HDDD dated January 6, 2023). Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest when implementing or publishing the results of this study.

RESULTS

Table 1. Bacterial culture results of PRP after storage period.

Time after storage	PRP from umbilical cord blood (n = 8)	PRP from adult human blood (n = 8)
Immediately after activation (T1)	Negative	Negative
After 7 days of storage (T7)	Negative	Negative
After 10 days of storage (T10)	Negative	Negative
After 14 days of storage (T14)	Negative	Negative

PRP samples stored at 2 - 6°C after 7, 10, and 14 days did not exhibit colonial growth.

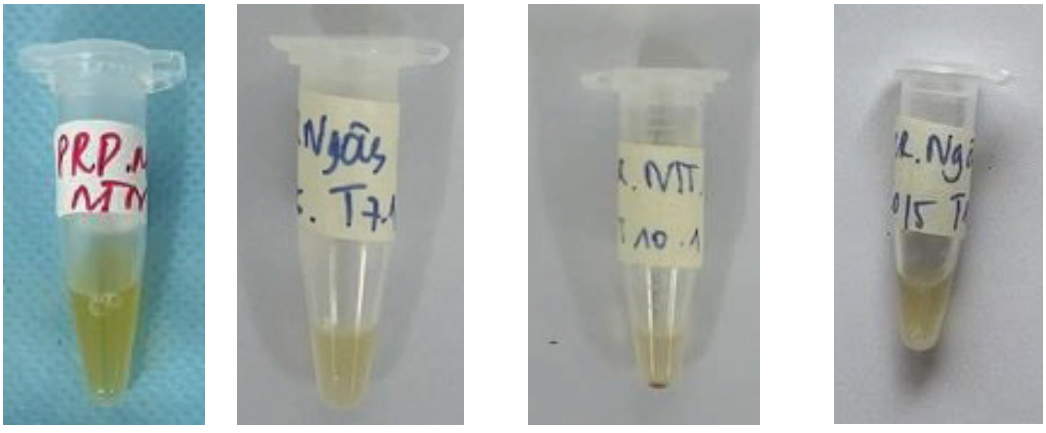

Figure 1. Activated PRP after 14 days of cold storage.

PRP samples stored at 2 - 6°C after 7 days, 10 days, and 14 days showed no change in color (yellow), no turbidity, and no air bubbles (clear).

Table 2. EGF concentrations in activated PRP stored after different storage periods[+].

EGF concentration in PRP Q2 (Q1 - Q3)	Cord blood (n = 8)	Adult human blood (n = 8)	p
Immediately after activation (T1) (1)	115.72 (51.59 - 217.61)	113.04 (69.71 - 155.73)	> 0.05*
After 7 days of storage (T7) (2)	91.82 (47.71 - 258.20)	164.91 (96.11 - 438.32)	> 0.05*
After 10 days of storage (T10) (3)	109.02 (53.79 - 329.35)	149.67 (86.35 - 198.39)	> 0.05*
After 14 days of storage (T14) (4)	109.80 (45.42 - 284.79)	192.77 (99.43 - 459.71)	> 0.05*
p_{21}**	> 0.05	< 0.05	
p_{31}**	> 0.05	> 0.05	
p_{41}**	> 0.05	< 0.05	

*([+]: Storage conditions: Cold storage 2 - 6°C; *: Mann Whitney U test; **: Wilcoxon test)*

During storage time points, the EGF concentration in activated PRP from umbilical cord blood was not statistically different compared to that from adult blood ($p > 0.05$). After 7, 10, and 14 days of cold storage at 2 - 6°C, the EGF concentration in activated PRP from umbilical cord blood and from adult blood was not statistically different compared to the initial time point ($p > 0.05$).

Table 3. VEGF concentrations in activated PRP preserved after different storage periods$^+$.

VEGF concentration in PRP Q2 (Q1 - Q3)	Cord blood (n = 8)	Adult human blood (n = 8)	p
Immediately after activation (T1) (1)	193.99 (75.58 - 320.25)	16.12 (11.70 - 49.03)	< 0.01*
After 7 days of storage (T7) (2)	240.61 (49.68 - 462.65)	11.70 (11.70 - 59.08)	< 0.05*
After 10 days of storage (T10) (3)	211.78 (40.22 - 266.13)	11.70 (11.70 - 49.86)	< 0.05*
After 14 days of storage (T14) (4)	147.44 (50.53 - 254.44)	17.90 (11.70 - 54.16)	< 0.05*
p_{21}**	> 0.05	> 0.05	
p_{31}**	> 0.05	> 0.05	
p_{41}**	> 0.05	> 0.05	

*($^+$: Storage conditions: Cold storage 2 - 6°C; *: Mann Whitney U test; **: Wilcoxon test)*

During storage time, the concentration of VEGF in activated PRP from umbilical cord blood was statistically higher than that from adult blood ($p < 0.05$). After 7, 10, and 14 days of cold storage at 2 - 6°C, the concentration of VEGF in activated PRP from umbilical cord blood and from adult blood was not statistically different compared to the initial time ($p > 0.05$).

DISCUSSION

The umbilical cord consists of 2 smaller arteries spiraling around a larger vein. The umbilical vein supplies oxygen-rich blood and nutrients to the fetus. The arteries return deoxygenated blood to the placenta. The umbilical vein is large and easily visible. When taking umbilical cord blood, blood is taken from the umbilical vein because it is large and easy to see. This ensures easy collection and the quality of blood because this blood contains a lot of nutrients and is transported to the fetus from the placenta through the osmotic pathway in the placenta, not directly

from the mother's blood [5]. The source of umbilical cord blood has a lot of potential as an abundant blood source, ensuring economic and medical ethics, especially being able to bring a good quality product and creating a ready-made PRP product for wound treatment. As this source is very accessible, donors do not face safety risks, infectious diseases have a very low risk of transmission, and umbilical cord blood also produces low immunity [2, 3]. Preserved umbilical cord blood can be used for autologous transfusion for patients with blood diseases. It is also used in cases of patients with neurological diseases, pulmonary diseases, systemic skin diseases, etc., in the form of allogeneic [2, 3]. Studies have shown that autologous PRP has a wide range of clinical applications. However, it may also contain many inflammatory cytokines, and its quality depends on several factors, such as the number and quality of platelets, the age of the patient, and the treatments the patient has received. Studies have shown that umbilical cord PRP contains more anti-inflammatory and growth factors than peripheral blood PRP [8]. Therefore, research on the application of umbilical cord blood PRP will open up a new direction in the treatment of chronic wounds today.

EGF is secreted from platelets and is also produced by macrophages and fibroblasts. It is present throughout the epidermis, especially in the basal layer. EGF plays a major role in re-epithelialization by stimulating the proliferation and migration of keratinocytes, increasing the tensile strength of the new skin layer [6]. VEGF is not only secreted from platelets but also from many other types of cells, such as fibroblasts, endothelial cells, smooth muscle cells, macrophages, and neutrophils. VEGF plays a major role in promoting the process of neovascularization, increasing permeability through capillaries during wound healing [7]. In acute wounds, VEGF begins to increase from day 1, increases significantly from day 3 - 5, and returns to normal from day 7 - 14 after injury. In chronic wounds, VEGF decreases abnormally [7]. Chronic wounds are weakened by excessive vascularization, leading to the lack of oxygen and micronutrients, thereby causing more tissue damage [6]. This phenomenon causes a decrease in many cytokines, chemokines, and growth factors necessary for wound healing, including EGF and VEGF [6]. Many studies have mentioned the use of exogenous EGF and VEGF in the treatment of chronic wounds. EGF and VEGF are found in high concentrations in PRP, so PRP is a good source of EGF and VEGF in the treatment of chronic wounds [6, 7].

Blood and blood products have long been commonly used in treatment. The preservation of blood and blood products is also regulated to be able to practice in accordance with actual conditions. According to Circular 26 of the Ministry of Health in 2013, the shelf life of plasma can be up to 14 days from the time of preparation in a closed system [4]. Thus, the preservation of blood products is easily implemented in clinical practice conditions, especially in grade 1 medical facilities. There are reports that at room temperature, PRP can be preserved for 5 - 8 days [9]. In this study, we preserved activated PRP at 2 - 6°C for 7 days, 10 days, and 14 days, the evaluation criteria were color, turbidity, bacterial and fungal culture, and measurement of EGF and VEGF concentration index. Through evaluation, we found that the color of PRP did not change, maintaining a clear yellow color during storage. PRP was also not infected with bacteria or fungi. The concentration indexes of EGF and VEGF in PRP from adult blood and umbilical cord blood did not fluctuate significantly compared to PRP immediately after activation ($p > 0.05$). Our results are similar to the findings of previous reports [9, 10]. Under the above storage conditions, the EGF concentration was not different between the origin of umbilical cord blood and adults, but VEGF in PRP from activated umbilical cord blood was significantly higher than that from adult blood, consistent with previous reports [8]. Thus, when storing PRP in cold conditions of 2 - 6°C, if PRP is extracted tightly and ensures the process, it can be stored for a long time, possibly 7 - 14 days, as in our evaluation. Application in clinical practice can be deployed to suit the conditions of each medical facility.

CONCLUSION

Platelet-rich plasma from activated umbilical cord blood has higher VEGF concentrations than that from adult blood sources. Cold storage at 2 - 6°C for 7 - 14 days keeps PRP clear, yellow, and free of bacteria, and EGF and VEGF concentrations in PRP do not change significantly.

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A RARE CASE REPORT: LUNG ADENOCARCINOMA WITH DIFFUSE LESIONS IN A YOUNG MALE PATIENT

Dao Ngoc Bang[1], Ta Ba Thang[1], Nguyen Tien Dung[1]*

Abstract

The images of diffuse lung involvement are seen in various respiratory diseases, including lung adenocarcinoma (ADC). After ruling out acute infectious causes, lung biopsy is valuable for a definitive diagnosis, with transbronchial biopsy via flexible bronchoscopy being an effective and safe diagnostic approach. However, the widespread lung damage in these patients poses a challenge for transbronchial biopsy. Chemotherapy is often difficult due to the overall poor health of the patient. Targeted therapies, specifically tyrosine kinase inhibitors (TKIs), have shown efficacy in lung cancer treatment.

Keywords: Adenocarcinoma; Lung cancer; Tyrosine kinase inhibitors (TKIs).

INTRODUCTION

Lung cancer is a malignancy with increasing incidence and mortality rates. Its slow progression and nonspecific clinical symptoms contribute to low early-stage diagnosis rates, often leading to misdiagnosis or confusion with other respiratory diseases, especially in cases with atypical X-ray or computed tomography (CT) findings. ADC with diffuse lesions (named bronchioloalveolar carcinoma - BAC before) is rare in clinical practice in Vietnam, making diagnosis challenging [1, 2, 3]. In patients with epidermal growth factor receptor (EGFR) mutation and low PS [3, 4], TKIs are the first choice for treatment, in which Afatinib is a suitable indication for ADC with a G719x mutation in exon 18. With this case study, we would like to: *Present the clinical characteristics of a rapidly progressed ADC case with diffuse lesions, having G719x mutation in exon 18, and the results of treatment by Afatinib as first-line therapy.*

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CASE REPORT

Male patient, 39 years old, non-smoker, no history of tobacco use, no exposure to cancer risk factors, and no family history of cancer. Occupation: Military officer. The symptoms have been present for about 4 months, including occasional cough with minimal sputum, dull chest pain, mild progressive dyspnea, and mild fever. The patient was admitted to the Internal Department of a regional general hospital, where chest X-rays, chest CT, acid-fast bacillus (AFB) testing, and GeneXpert/MTB testing of negative sputum were conducted. Following consultation, the patient was diagnosed with miliary tuberculosis and received a 2RHZE/6RH regimen. After 3 weeks of tuberculosis treatment, the patient's symptoms were unresponsive, and respiratory distress worsened, leading to admission to the Respiratory Medicine Center, Military Hospital 103. On admission, the patient exhibited signs of respiratory failure (persistent dyspnea, cyanosis, respiratory muscle retractions; SpO_2 80%), no fever, no palpable peripheral lymph nodes, and reduced breath sounds in both lungs without rales. Imaging findings on chest X-ray and CT revealed hazy opacities interspersed with diffuse reticular patterns in both lungs, concentrated in the mid and lower zones of both lungs, with no hilar lymphadenopathy, involvement of the mediastinum, or pleural effusion (*Figure 1*).

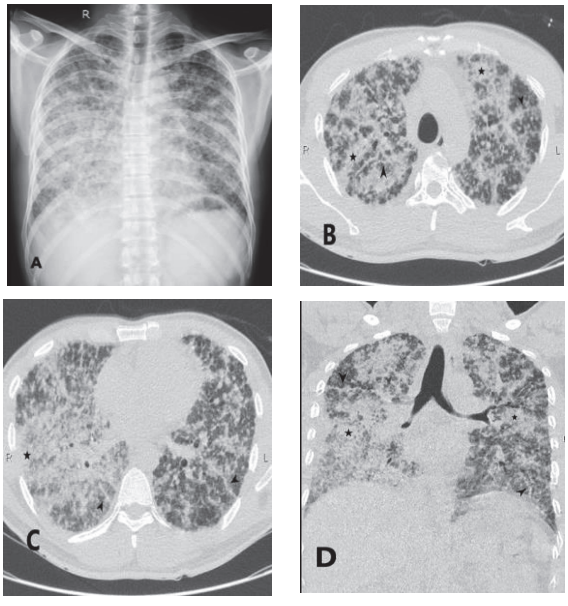
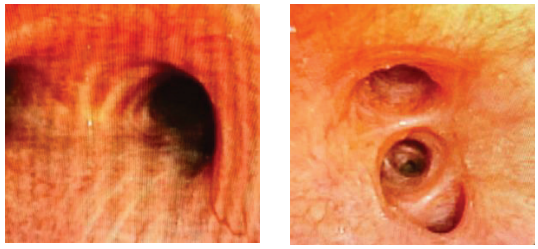


Figure 1. Initial chest X-ray (A) demonstrates bilateral diffuse reticular opacities with consolidation in the right lung predominant. Axial CT scan (B, C) and coronal reconstructed CT image (D) showed bilateral extensive septal thickening and reticular opacities (arrowhead) with consolidation (star).

Arterial blood gas (ABG) results: PaO_2 = 57 mmHg, $PaCO_2$ = 37 mmHg, pH = 7.38. The patient underwent treatment for respiratory failure (high-flow nasal cannula oxygen therapy - HFNC), symptom management, and bronchoscopy. Bronchoscopy images show normal findings with no increased mucus secretion (*Figure 2*). Selective bronchial lavage technique is applied, collecting bronchial lavage fluid for microbiological and cytological examinations, and performing transbronchial biopsy for pathological examination. Pathological examination reveals papillary ADC of the lung; EGFR mutation testing indicates a G719x mutation on exon 18 (*Figure 3*). Microbiological tests, including AFB, GeneXpert/MTB-Rif, bacterial and fungal cultures of bronchial fluid, all yield negative results. The patient undergoes additional diagnostic imaging tests (abdominal ultrasound, cranial MRI) for staging.



A **B**

Figure 2. Bronchoscopy did not detect lesions in the main bronchus (A) and segmental bronchus (B), so a transbronchial biopsy was indicated.

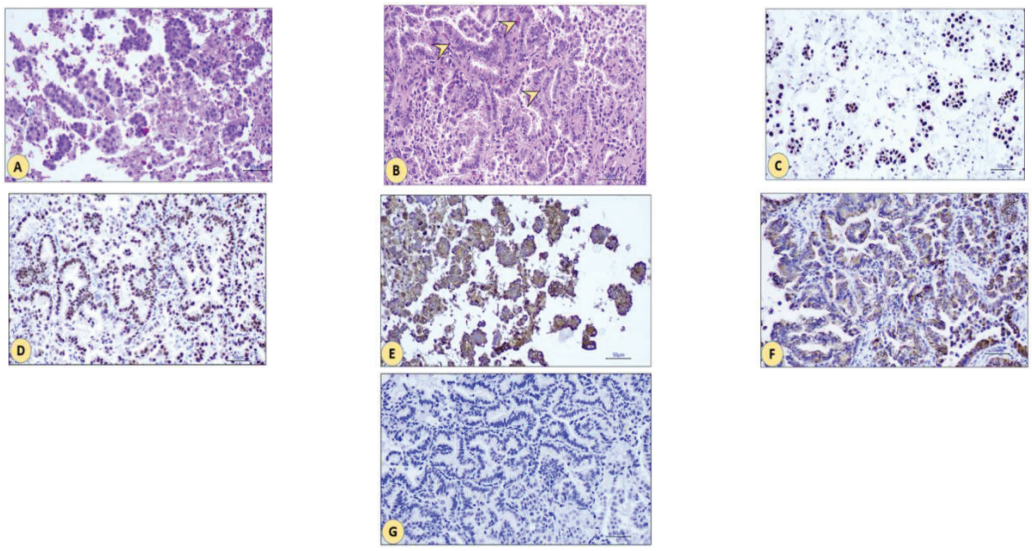


Figure 3. Initial chest X-ray (A) demonstrates bilateral diffuse reticular opacities with consolidation in the right lung predominant. Axial CT scan (B, C) and coronal reconstructed CT image (D) showed bilateral extensive septal thickening and reticular opacities (arrowhead) with consolidation (star).

The patient is diagnosed with non-mucinous ADC of the lung, stage IV ($T_{1mi}N_0M_1$), with EGFR mutation, and respiratory failure as a complication. The patient is reviewed by a multidisciplinary team and opts for treatment with second-generation TKI: Afatinib at a dose of 40 mg/day, taken 1 hour before lunch. Response assessment after 1 month indicates a partial response: Significant reduction in cough, sputum, and chest pain; weight gain (BMI 21.3 kg/m^2), no need for supplemental oxygen, and improved ABG (PaO_2 = 87 mmHg, $PaCO_2$ = 38 mmHg). Chest X-ray images show a partial reduction in lung lesions after 10 days of treatment (*Figure 4*), and CT scans demonstrate a noticeable decrease in the extent of spread compared to the patient's initial presentation (*Figure 5*). Mild adverse effects of TKIs are observed (mild skin rash). After 6 months, the patient has signs of disease progression. Afatinib should be replaced by Osimertinib as a recommendation of NCCN [7].

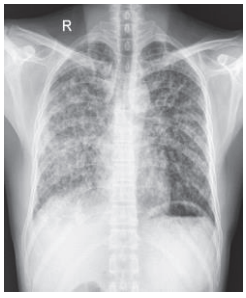


Figure 4. Chest X-ray obtained 10 days after treatment revealed a decrease in consolidation area and reticular opacities after the patient's symptoms improved.

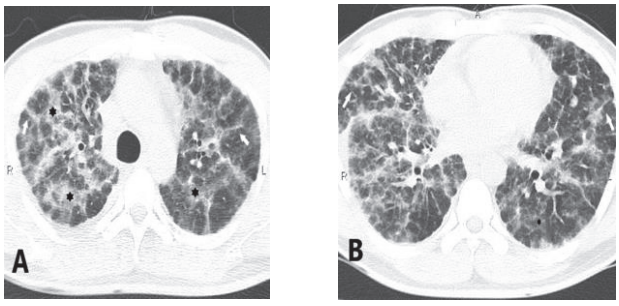


Figure 5. CT scan obtained 15 days after treatment showed a decrease in consolidation, but we still saw bilateral septal thickening, and reticular opacities (arrow) with ground glass opacities (star). (A) Axial section of apex (B) Axial section of basal.

DISCUSSION

1. Diagnosis of diffuse lung ADC

Diffuse lung ADC is a type found at a rate of 3 - 6% of all types of lung cancer. It is more commonly found in young, non-smoking females. However, this type is rare in Vietnam. Key clinical symptoms include chronic cough, progressively increased difficulty in breathing and a significant amount of coughed-up sputum. In patients with localized lesions visible on X-rays, clinical symptoms may not be apparent, and the diagnosis relies primarily on the

technique of invasive specimen collection for pathological examination. Patients with rare cases of bilateral spread of lesions present more significant diagnostic challenges, especially in late-stage patients with complicated respiratory failure, making it challenging to perform diagnostic specimen collection techniques. The choice of biopsy procedures depends on the overall condition of the patient, the characteristics of lung lesions observed on X-rays, and chest CT scans. Treatment options are limited due to the extensive spread of lung damage, and the patient's overall health index is compromised due to respiratory failure [3, 4]. Moreover, when imaging reveals lesion patterns such as nodules or a diffuse mesh spreading across both lungs, differential diagnosis is crucial to distinguish from various respiratory infections (disseminated tuberculosis, mycobacteria infection, fungi), non-infectious diffuse lung diseases (sarcoidosis, hypersensitivity pneumonitis, pneumoconiosis), and malignant conditions (secondary lung cancer, Kaposi's sarcoma) (*Table 1*).

Table 1. Causes of diffuse pulmonary nodules.

Infections	Diffuse lung diseases	Malignancies
Miliary tuberculosis	Hypersensitivity pneumonitis	Metastasis
Atypical mycobacterial infection	Diffuse pulmonary meningotheliomatosis	Diffuse lung ADC
Lung fungal infection	Pneumoconiosis (namely silicosis)	Lymphangitic carcinomatosis
Septic emboli	Amyloidosis	Kaposi sarcoma

In the clinical case of a young patient with no risk factors for lung cancer, presenting minimal clinical symptoms, mainly gradual onset of mild breathlessness and occasional mild fever, the initial differential diagnosis considered is subacute disseminated pulmonary tuberculosis. This is a chronic infectious lung disease with widespread lesions commonly seen in Vietnam. Simultaneously, the second differential diagnosis raises the possibility of a systemic disease, a group of conditions often found in young individuals. Microbiological tests for tuberculosis, including AFB, and

GeneXpert/MTB-Rif from sputum, all yield negative results. Therefore, a lung biopsy via bronchoscopy is recommended for a conclusive diagnosis. However, due to the patient's respiratory distress, a bronchoscopy with a flexible bronchial tube is performed with the support of the ICU. As the bronchoscopy results reveal no significant mucosal lesions in the large bronchi, a transbronchial biopsy is performed at the right lower lobe, guided by chest CT scan images, focusing on areas with multiple consolidated lesions. As a result, 8 biopsy samples are safely obtained for histopathological examination. With an adequate amount of biopsy material, immunohistochemical and molecular biology techniques are employed [6]. The diagnosis confirms ADC with non-mucinous components and a G719x mutation on exon 18. The pathological findings explain the patient's clinical symptoms despite the widespread involvement of ADC in both lungs, with no prominent cough symptoms observed clinically.

2. Regarding the results of TKI treatment

The G719x mutation on exon 18 is classified as a drug-sensitive mutation, responsive to TKI treatment. Clinical studies have demonstrated that second-generation TKIs (Afatinib) yield better outcomes than the first-generation for this mutation [7, 8]. Third-generation TKIs (Osimertinib) are also an effective option in treating the G719x mutation [9]. Additionally, patients may be combined with chemotherapy and immunotherapy to enhance cancer treatment effectiveness [7, 10].

In the clinical case, despite having a confirmed diagnosis, treatment decisions for the patient are challenging due to the patient's severe overall condition, frequent respiratory failure requiring HFNC support, and low overall performance status (PS = 4). Consequently, chemotherapy is no longer recommended. Lung transplantation is a potential solution, but the patient faces a risk of mortality while awaiting a transplant. TKI treatment proves to be the most suitable option for the patient, with Afatinib as the first-line choice. However, a notable concern is the undesirable side effect of TKIs causing peripheral lung damage, which is difficult to detect due to the patient's diffuse lung involvement. Therefore, closely monitoring treatment response and the unintended effects of TKIs requires a synergistic approach between clinical and chest imaging [8].

The outcome is a positive response to both clinical and X-ray imaging treatment. Just 1 week into treatment, the patient no longer requires HFNC, and after 2 weeks, gentle physical activity is resumed.

Visible reductions in lung lesions are observed on X-ray and chest CT after 1 month of treatment. The favorable treatment response is also linked to the patient's ADC subtype with non-mucinous components, indicating a better prognosis compared to the mucinous subtype. However, the response to second-generation TKIs is limited to approximately 12.7 months [7, 8]. In the future, if the patient does not respond to Afatinib, re-biopsy for T790m mutation and immune tests will be essential to determine the next treatment approach [9, 10].

CONCLUSION

Diffuse lung involvement in primary lung cancer is rare and diagnostically challenging, particularly in BAC. The biopsy is crucial for determining the cause after excluding infectious etiologies. Testing for EGFR mutations guides treatment decisions, with second-generation TKIs like Afatinib showing effectiveness in cases with a G719x mutation.

Ethics: The authors have obtained written informed consent from the patient for the publication of this case report. The patient consented to de-identified clinical information and images being used for the purposes of this report. The authors of the manuscript retain this informed consent and can provide it to the journal upon specific request. The data supporting this research are available from the authors upon reasonable request. The Respiratory Medicine Center, Military Hospital 103, Vietnam Military Medical University granted permission for the use and publication of the research data. The authors declare that they have no competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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INITIAL ANALYSIS OF SEVERAL CLINICAL FEATURES AND SPERM CONCENTRATION IN MEN WITH PERICENTRIC INVERSION OF CHROMOSOME 9 FINDING ASSISTED REPRODUCTIVE TREATMENTS

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Dinh Van Sinh[2], Ho Sy Hung[3]

Abstract

Objectives: To evaluate some clinical features and sperm concentration among men with pericentric inversion of chromosome 9, and to find assisted reproductive treatments. ***Methods:*** A retrospective, descriptive study was conducted on 36 men with pericentric inversion of chromosome 9 at the Military Institute of Clinical Embryology and Histology, Vietnam Military Medical University, from January 2020 to December 2023. Age, height, weight, and body mass index (BMI) were recorded. Semen analysis was conducted according to World Health Organization (WHO) 2021 guidelines. ***Results:*** 36 male patients with pericentric inversion of chromosome 9 were observed, of which the majority had variant 46,XY,inv(9)(p11q13) with a rate of 72.22%. Evaluation of clinical characteristics showed that height, weight, and BMI were all within the normal ranges for men. Semen analysis: 24/36 patients (66.67%) had normal sperm concentration; only 12/36 patients (33.33%) were observed to have a slight decrease in sperm concentration; no cases of severe oligozoospermia or azoospermia were recorded. A case of pericentric inversion of chromosome 9 was recorded as inherited in the family after natural reproduction. ***Conclusion:*** In men who found assisted reproductive treatments in our assisted reproductive center and were recorded pericentric inversion of chromosome 9 in the karyotype, age, height, weight, and BMI were all within the normal ranges of Vietnamese people and manifested mild oligozoospermia or normal sperm concentration.

Keywords: Pericentric inversion of chromosome 9; Oligozoospermia; Assisted reproduction.

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INTRODUCTION

Infertility is a common medical problem that affects many couples of reproductive age worldwide, in which genetic factors are important causes. Chromosomal abnormalities, especially structural variations such as inversions, have been noted to contribute to reproductive disorders. In fact, the most common inversion observed in humans is the pericentric inversion of chromosome 9, which is highly susceptible to rearrangements, especially pericentric inversions, in which the p (short) and q (long) arms of chromosome 9 are rotated 180° around the centromere. The pericentric inversions of chromosome 9: inv(9)(p11q13) and inv(9)(p12q13) are common cases, accounting for 1 - 3.57% of the general population [1].

The pericentric inversion of chromosome 9 is generally considered by cytogeneticists to be benign and does not cause serious clinical features [1]. The clinical features associated with this variant in humans vary between developmental stages, and there is no clinical feature specific to a specific developmental stage. However, some studies have shown that the chromosome 9 inversion may be associated with various conditions such as congenital anomalies, growth retardation, idiopathic reproductive failure [2], infertility [3], and recurrent miscarriage [4]. In men, in particular, this inversion may affect spermatogenesis, leading to reduced sperm concentration, motility, and morphological abnormalities. Therefore, we conducted this study: *To evaluate some clinical features and sperm concentration in men who found assisted reproductive treatments in our assisted reproductive center and recorded pericentric inversion of chromosome 9 in the karyotype.*

MATERIALS AND METHODS

1. Subjects

Including 36 men with pericentric inversion of chromosome 9 at the Military Institute of Clinical Embryology and Histology, Vietnam Military Medical University, from January 2020 to December 2023.

* *Inclusion criteria:* Men finding assisted reproductive treatments; patients had karyotype results confirming the pericentric inversion of chromosome 9 (inv(9)).

* *Exclusion criteria:* Obstruction of the vas deferens, varicocele, hypothalamic-pituitary axis disorders, history of scrotal or testicular surgery; other genetic causes of infertility; patients with acute or chronic infectious diseases affecting the reproductive system; did not agree to participate in the study.

* *Time and location:* From January 2020 to December 2023, at the Military Institute of Clinical Embryology and Histology, Vietnam Military Medical University.

2. Methods

* *Study design:* A retrospective, descriptive study.

* *Sample size:* Total sampling: All male patients with chromosome 9 inversion were examined and treated for infertility at the Military Institute of Clinical Embryology and Histology. During the study period, we collected 36 patients who met the criteria to participate in the study.

* *Research process:*

Male patients examined and treated for infertility at the Military Clinical Embryology Institute were recruited and underwent karyotype testing. Chromosomal analysis and karyotype were established using bioinformatics software. More than 20 G-banded metaphase chromosomes were detected for each patient. Chromosomal disorders were described in accordance with the International System for Human Cytogenetic Nomenclature, 2020.

Male patients with pericentric inversion of chromosome 9 were clinically examined to record personal information, including age, medical history, reproductive history, and clinical symptoms.

Semen analysis was conducted according to WHO 2021 standards to assess sperm concentration, then classified according to levels [5]:

- Normal: Sperm concentration ≥ 15 million/mL;

- Mild oligozoospermia: Sperm concentration 5 - 15 million/mL;

- Severe oligozoospermia: Sperm concentration < 5 million/mL;

- Azoospermia: No sperm in semen after three times on testing.

* *Data processing:* Data were processed and analyzed using STATA 14.0 software. Quantitative variables are described as mean ± standard deviation (SD). Qualitative variables are described as percentages (%).

3. Ethics

All procedures performed in the study involving human participants were approved by the Military Institute of Clinical Embryology and Histology. Patients voluntarily participated in this study. Patients' personal information is guaranteed confidential while conducting the study and when publishing the study and used only for scientific purposes. The Military Institute of Clinical Embryology and Histology granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS

1. Characteristics of pericentric inversion of chromosome 9

Table 1. Results of karyotype testing (n = 36).

Karyotypes	Frequency (n)	Rate (%)
46,XY,inv(9)(p11q13)	26	72.22
46,XY,inv(9)(p11q13),Yqh+	8	22.22
46,XY,inv(9)(p11q13),Yqh-	1	2.78
46,XY,inv(9)(p12q13)	1	2.78

Of the 36 infertile male patients participating in the study, 26 patients had karyotype testing results of 46,XY,inv(9)(p11q13), accounting for the highest rate (72.22%). Other variants appeared at lower frequencies.

2. Clinical features

Table 2. Clinical features of study patients (n = 36).

Features	Mean ± SD	Min	Max
Age (years)	30.72 ± 3.06	26	40
Height (cm)	169.83 ± 5.76	160	180
Weight (kg)	68.47 ± 6.53	55	80
BMI (kg/m^2)	23.70 ± 1.43	19.49	26.17

Table 2 showed that age, height, weight, and BMI characteristics of men with pericentric inversion of chromosome 9 were all within the normal range of Vietnamese people.

3. Sperm concentration

Table 3. Sperm concentration results (n = 36).

Sperm concentration	Frequency (n)	Rate (%)
Normal	24	66.67
Mild oligozoospermia	12	33.33
Severe oligozoospermia	0	0
Azoospermia	0	0

In enrolled men with pericentric inversion of chromosome 9, a majority of patients (n = 24) manifested normal sperm concentration, accounting for 66.67%.

There were 12 cases of mild oligozoospermia, accounting for 33.33%. We did not record any cases of azoospermia or severe oligozoospermia.

4. Hereditary to offsprings

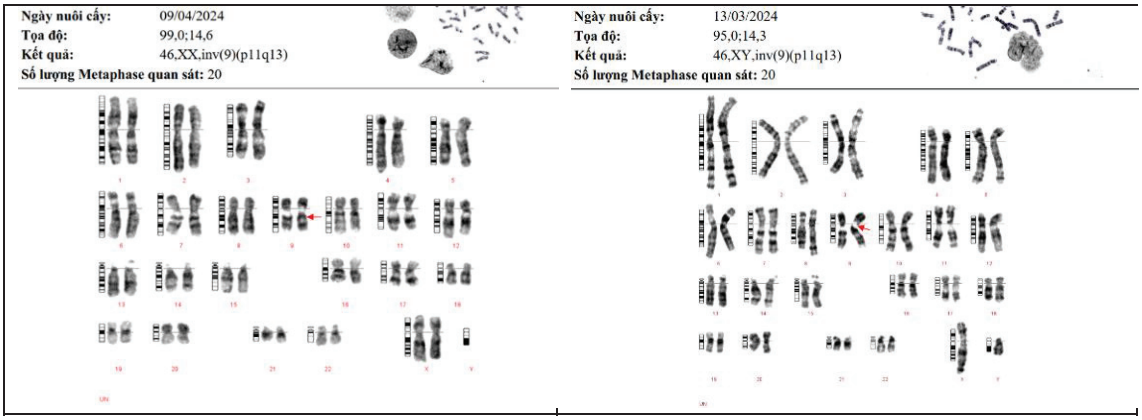

Figure 1. Karyotypes of mother (left) and son (right) both carry the pericentric inversion of chromosome 9.

The results of the karyotype test in figure 1 show that the karyotype of the mother 46,XX,inv(9)(p11q13) (left) and the son 46,XY,inv(9)(p11q13) (right) both carry the pericentric inversion of chromosome 9.

DISCUSSION

Pericentric inversion of chromosome 9 is a frequently observed chromosomal rearrangement in humans. It is relatively common, found in approximately 1 - 3.57% of the general population [1], though rates vary by ethnic group. Its high frequency suggests that it may not have significant deleterious effects on individuals carrying the inversion. Pericentric inversions occur when there are breaks in both p and q arms of a chromosome, and the fragment between the breaks is reinserted in an inverted orientation. For chromosome 9, this often involves breaks at p11 and q13, with re-ligation causing a flip in the orientation of this region. This mechanism involves balanced chromosomal rearrangement without gene loss, generally allowing carriers to have normal phenotypes and natural fertility, while there may be a slight increase in reproductive risks due to potential meiotic missegregation.

Regarding the clinical characteristics of the patients, our study results showed that some clinical characteristics were within normal limits, which is consistent with previous studies that pericentric inversion of chromosome 9 is often considered benign and does not cause

serious clinical features in most cases [1]. This benign nature is a primary reason why the inversion of chromosome 9 is often considered a chromosomal polymorphism rather than a pathogenic mutation. A study by Dana et al. (2012) on 1,800 infertile patients noted that the rate of pericentric inversion of chromosome 9 in both infertile men and women was not significantly higher than that of the normal population [6].

The reduction in sperm concentration in several patients with pericentric inversion of chromosome 9 in our study is also consistent with some previous studies. The study by Xie et al. (2020) noted that in 31 male patients with pericentric inversion of chromosome 9, there were 3 cases of oligospermia and 3 cases of azoospermia [7]. Maeda et al. (1991) pointed out that pericentric inversion of chromosome 9 can be linked to the disruption of specific genes on chromosome 9 associated with spermatogenesis during meiosis, leading to reduced sperm production or production of morphologically abnormal sperm [8]. However, all patients in our study had slightly reduced or normal sperm concentrations, suggesting that the pericentric inversion of chromosome 9 has little effect on sperm concentration. Numerous cases have demonstrated that individuals with pericentric inversion of chromosome 9 are capable of achieving natural conception, and there is a possibility that their child may inherit this inversion as well [9]. This occurs because, despite the structural rearrangement, the genetic content remains balanced, meaning that there is no significant gain or loss of critical genetic material.

The results of this study also highlight the important role of genetic testing, such as karyotype, in the diagnosis of unexplained infertility. The detection of the pericentric inversion of chromosome 9 in infertile patients without obvious clinical symptoms suggests that genetic testing could provide insights into the potential genetic causes of infertility that are not diagnosed by conventional clinical methods and, therefore, assist in family planning, including assisted reproductive technologies if necessary. Although this study has provided important information for men with pericentric inversion of chromosome 9, there are some limitations that need to be considered. First, the small sample size may limit the ability to generalize the results. Second, this is only a descriptive study, has not evaluated the relationship, and has only considered one genetic factor, which is the pericentric inversion of chromosome 9, while other factors such as environment and other genetics that may also contribute to the cause of infertility have not been fully investigated. Therefore, further research with a larger

sample size and more information is needed to clarify the relationship between the pericentric inversion of chromosome 9 and infertility.

CONCLUSION

In men who found assisted reproductive treatments in our assisted reproductive center and were recorded pericentric inversion of chromosome 9 in the karyotype, age, height, weight, and BMI were all within the normal ranges of Vietnamese people and manifested mild oligozoospermia or normal sperm concentration.

Acknowledgments: The authors would like to thank the Board of Directors and employees of the Military Institute of Clinical Embryology and Histology for assisting us in conducting this study and all patients for their cooperation.

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PHASE ANGLE PREDICTOR OF SARCOPENIA IN PATIENTS WITH STABLE ISCHEMIC HEART DISEASE

Nguyen Duy Dong[1], Nguyen Thi Thanh Diem[2]*

Abstract

Objectives: To examine how phase angle (PhA) contributes to sarcopenia and factors influencing sarcopenia in patients with stable ischemic heart disease (SIHD). ***Methods:*** A cross-sectional descriptive study was conducted on 52 SIHD patients who were recruited, and relevant data was gathered. Patients were diagnosed with sarcopenia based on the Asian Sarcopenia Working Group 2019 (AWGS 2019) diagnostic criteria. Differences between groups were compared, and statistically significant factors were included in the logistic regression analysis to screen for independent factors affecting sarcopenia. The receiver operating characteristics (ROC) and the area under the curve (AUC) were used to evaluate the predictive value of PhA in sarcopenia. ***Results:*** The prevalence of sarcopenia was 36.5% in patients with SIHD. Multivariate logistic regression analysis showed that PhA was an independent factor influencing sarcopenia (OR: 0.078; 95%CI: 0.012 - 0.528; p = 0.009). The AUC of PhA predicting sarcopenia was 0.852, p < 0.001; the best PhA cut-off value for sarcopenia was 5.95° for both sexes (sensitivity and specificity were 0.677 and 0.947, respectively); the PhA cut-off points were 6.05° and 5.25° for men and women, respectively (p < 0.05). ***Conclusion:*** PhA is an important determinant of sarcopenia in patients with SIHD. PhA may have an optimistic predictive value for determining sarcopenia in this population.

Keywords: Stable ischemic heart disease; Sarcopenia; Phase angle.

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INTRODUCTION

Sarcopenia is a disease characterized by progressive deterioration of skeletal muscle mass, accompanied by low muscle strength or muscle dysfunction and often exacerbated by chronic comorbidities, including cardiovascular diseases, chronic kidney disease, and cancer [1]. Sarcopenia is associated with a faster progression of cardiovascular disease and a higher risk of falls, fractures, and other adverse consequences, increasing disability and mortality, particularly among older patients. The prevalence of sarcopenia is about 25% in coronary artery disease (CAD) hospitalized and 12.5% in community-dwelling older adults [2]. Sarcopenia may also be a risk factor for CAD. Previous studies have shown that low skeletal muscle mass among asymptomatic community-dwelling older adults is associated with subclinical atherosclerosis, increased coronary artery calcium score, arterial stiffness, and carotid arterial wall thickening [3, 4]. PhA, a key parameter obtained from bioelectrical impedance analysis (BIA), has attracted significant attention. Recent studies have shown that PhA can predict sarcopenia to a certain degree in healthy elderly people or patients with cachexia due to cirrhosis [5, 6]. Patients with cardiovascular disease are at high risk of sarcopenia. If PhA can be used as a simple indicator for early detection of sarcopenia, it could significantly improve quality of life, reduce treatment costs, and increase survival time in cardiovascular disease patients. Therefore, this study aimed to: *Analyze some factors affecting sarcopenia and investigate the association between PhA and sarcopenia in patients with SIHD.*

MATERIALS AND METHODS

1. Subjects

Including 52 patients meeting the criteria who were included in the analysis.

* *Inclusion criteria:* Patients diagnosed with SIHD (by percutaneous coronary angiography, with or without indication for intervention and coronary artery stenting); aged over 18.

* *Exclusion criteria:* Patients at the time of the study were comatose and had surgery, emergency procedures, and limitations to perform the tests needed to evaluate muscle strength and function, as did those with pacemakers, and those who could not stand were excluded from the study sample.

* *Time and location:* From April 2022 to October 2022 at the Department of Cardiovascular Interventions, Military Hospital 103.

2. Methods

* *Study design:* A cross-sectional descriptive study.

Patients were selected for the study according to the convenience sampling method. Collected data includes patients' general information (age, gender, medical history), anthropometric information (weight, height, calf circumference, BMI), information about the laboratory, information about measuring body composition using BIA (Inbody S10, Seoul, Korea) (skeletal muscle mass index, PhA).

Figure 1. Image of bioelectrical impedance meter (Inbody S10) and measurement results.

* *Measuring skeletal muscle mass index (SMMI) and PhA:* An Inbody S10 bioelectrical impedance analyzer (Seoul, Korea) was used to analyze the body composition of SIHD patients. We performed BIA approximately 24 hours after the patient was admitted to the department. Before measuring BIA, patients were asked to fast for 2 hours, empty their bladder, take things out of pockets, remove necklaces, bracelets, rings, and other jewelry, take off their shoes and socks, wear clothing of known weight, and make contact with their hands and feet with an eight-point tactile electrode. We entered the patient's

name, age, gender, height, and weight in the analysis system and then started measuring BIA. The 8-electrode technique of the Inbody body composition analyzer allows for fractional impedance measurements, performed with a current of 100μA at frequencies from 1 to 1000kHz. The device acquires resistance and reactance values at a frequency of 50kHz and provides additional calculation through a proprietary algorithm developed by the company. SMI is calculated according to height (kg/m^2). PhA was calculated with resistance (R) and reactance (Xc; measured at 50kHz) by the following equation: PhA (°) = arctangent(Xc/R) x (180/π).

* *Measuring muscle strength and physical activity ability (muscle function):* Handgrip strength (HGS) was assessed with an electronic dynamometer (Camry, China) after measuring BIA. The dominant hand is used to hold the dynamometer firmly with the elbow straight away from the body. The measurement is taken twice; the highest value is recorded in kilograms (kg). Muscle function is assessed by sit-to-stand test (SST). Patients sitting in chairs without armrests were asked to stand up and sit down five times at their highest ability. The average value was recorded after two consecutive measurements.

* *Diagnosis of sarcopenia:* According to the diagnostic criteria of the Asian Working Group for Sarcopenia 2019 (AWGS) [7], sarcopenia can be diagnosed when muscle mass loss (SMI < 7.0 kg/m^2 and < 5.7 kg/m^2 in men and women, respectively) plus one of the two criteria of reduced muscle strength (HGS < 28kg and < 18kg in men and women, respectively) and reduced muscle function (time last over 12 seconds).

* *Data analysis:* SPSS version 20 (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. A logistic regression model was used to screen influencing factors for sarcopenia. The ROC curve and its corresponding AUC were used to evaluate the predictive value of PhA with sarcopenia. The cut-off point was defined as the maximum value of sensitivity + specificity-1. A two-sided $p < 0.05$ was considered a statistically significant difference.

3. Ethics

This study complies with the ethics of biomedical research at Military Hospital 103. Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS

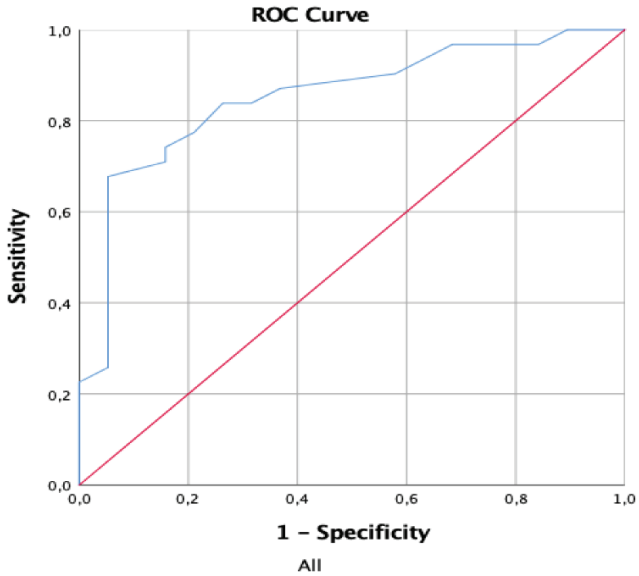
A total of 52 patients were recruited in this study, of which 38 patients (73.10%) were men. The average age was 66.40 ± 10.20 years old; 73.10% of patients were ≥ 60 years old. Among the study subjects, 19 patients (36.5%) were diagnosed with sarcopenia.

Table 1. Multivariate logistic regression analysis of factors influencing sarcopenia (n = 52).

Variables	OR	95%CI	p
Age > 60 (year)	0.81	0.04 - 16.80	0.89
Male	0.96	0.11 - 8.31	0.97
History of hypertension	0.49	0.05 - 4.67	0.54
History of diabetes mellitus	0.60	0.07 - 5.24	0.64
BMI > 25			0.35
BMI: 18.5 - 24.9	8.58	0.15 - 498.90	0.30
BMI < 18.5	7.77	0.48 - 126.30	0.15
Low hemoglobin (g/L)	0.61	0.07 - 5.06	0.64
NLR	1.18	0.93 - 1.49	0.17
High CPR (mg/L)	1.49	0.21 - 10.35	0.68
PhA (°)	0.08	0.01 - 0.53	0.01

(NLR: Neutrophil-to-lymphocyte ratio; BMI: Body mass index; CRP: C-reactive protein)

Table 1 shows a multivariate logistic regression analysis of factors affecting sarcopenia. The results showed that only PhA was an independent factor affecting sarcopenia.



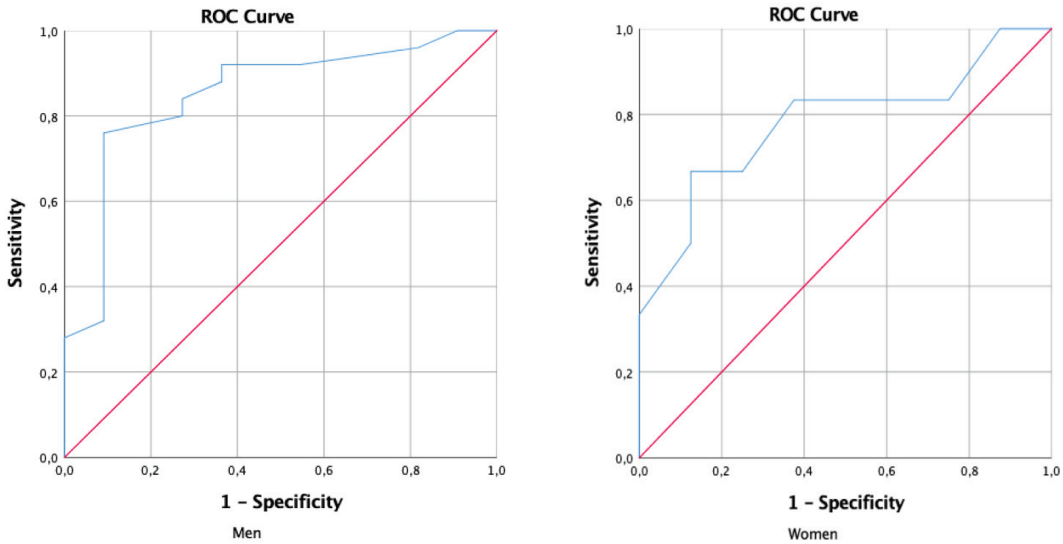
AUC = 0.852; p < 0.001

Figure 2. ROC curve of PhA in the diagnosis of sarcopenia.

Table 2. Cut-off, sensitivity, and specificity of phase angle for diagnosing sarcopenia in the study subjects.

Gender	Cut-off	Sensitivity	Specificity
All (n = 52)	5.95°	0.677	0.947
Men (n = 38)	6.05°	0.760	0.909
Women (n = 14)	5.25°	0.667	0.875

The ROC curve shows the predictability of sarcopenia based on PhA. The AUC was 0.852, and the best cut-off of PhA for sarcopenia was 5.95° for both genders, with a sensitivity of 67.7% and a specificity of 94.7% ($p < 0.001$). The AUC was 0.851, and the best cut-off of PhA for sarcopenia was 6.05° for men, with a sensitivity of 76.0% and a specificity of 90.9% ($p < 0.001$). The AUC was 0.781, and the best cut-off of PhA for sarcopenia was 5.25° for women, with a sensitivity of 66.7% and a specificity of 87.5% ($p < 0.05$). The results determined that PhA has high predictive power in the diagnosis of sarcopenia (*Figure 2, 3*).



AUC = 0.781; p = 0.04 AUC = 0.851; p = 0.001

Figure 3. ROC curve of the ability of PhA to predict sarcopenia in men (left) and women (right).

DISCUSSION

Sarcopenia is a widespread health problem associated with poor physical capacity and poor prognosis in patients with chronic diseases. Current diagnostic criteria for sarcopenia have limitations in their widespread clinical use in large numbers of patients in hospital settings. The present study investigated the role of PhA through BIA (a measure of body composition) as a simple alternative tool for detecting sarcopenia in patients with SIHD and found that it was an independent factor influencing sarcopenia (OR: 0.078; 95%CI: 0.012 - 0.508; p = 0.009). Kilic et al. [8] found that decreased PhA was a risk factor for sarcopenia in elderly patients (OR: 0.59; 95%CI, 0.40 - 0.87; p = 0.008), and the risk of sarcopenia increases 1.69 times for every 1° decrease in PhA. Kosuku, et al. [9] reported that PhA is an important influencing factor of sarcopenia in kidney transplant patients (OR: 0.36; 95%CI: 0.16 - 0.82; p = 0.015). PhA (°) = arctangent (reactance/resistance × 180°/π) using reactance and resistance at 50kHz. Here, the resistance represents the volume of the water reservoir, which is inversely related to the amount of body fluid. Reactance reflects the energy storage capacity of the cell membrane, which is positively related to the number of cells and the integrity of the cell membrane. Literature has reported that PhA is associated with cellular function, inflammation, nutritional status, muscle mass, disease prognosis, and mortality. One possible explanation is that the loss of muscle mass may reduce the amount of water in the cells, so electrical resistance will decrease and PhA will be lower. Second, as a marker of cellular health, PhA is also lower in patients with poor muscle cell function. Third, skeletal muscle mass was also calculated from the resistance and reactance obtained at 50kHz; therefore, part of the association can be explained by this factor.

To further investigate the role of PhA in diagnosing sarcopenia in this chronic disease population, the ROC curve was used to examine the diagnostic role of PhA in sarcopenia. The results showed that the best value for diagnosing sarcopenia in patients with SIHD was 5.95°, which was higher than the results in the study by Hirose et al. [5] on the role of PhA as an indicator of sarcopenia, malnutrition, and cachexia in patients with cardiovascular disease (27% IHD) (4.55° with men and 4.25° with women); Kilic et al. [8] reported an optimal PhA cut-off of 4.55° to detect sarcopenia in 263 community-dwelling and hospitalized older adults (> 65 years). However, our study results were similar to the results

in the study of Reis et al. [10] on kidney transplant subjects, which were 5.80 and 6.20 in men and women, respectively. This may be explained by the differences in population size, mean age, study population, and ethnicity. In addition, it is also possible that the methods of measuring muscle mass and the diagnostic criteria for sarcopenia used in the studies were different, thus influencing the results.

To our knowledge, this is the first study to evaluate the association of PhA with sarcopenia in patients with SIHD. The results of the study suggest that higher PhA is a protective factor for sarcopenia, and therefore, it can be used as a predictor of sarcopenia. When sarcopenia is difficult to diagnose in chronic patients, such as when muscle strength or function cannot be measured or the patient is unwilling to cooperate, we propose to use PhA in clinical practice as an additional assessment method. According to PhA, healthcare professionals can identify patients at risk of sarcopenia in advance and implement appropriate early interventions to prevent the occurrence of sarcopenia and improve the prognosis and quality of life of chronically ill patients. However, this study has some limitations. It is a cross-sectional, single-center study that includes a small number of patients with SIHD, so the generalizability of the results is limited, and it is not possible to conclude a causal relationship between PhA and sarcopenia. In addition, we also excluded patients at the highest risk of sarcopenia, such as bed- or wheelchair-bound patients, critically ill patients, and patients with cognitive impairment.

CONCLUSION

The current study shows that PhA is an independent influencing factor of sarcopenia (according to AWGS 2019) in patients with SIHD. PhA as a nutritional index may have good predictive value to identify sarcopenia in this population.

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LIVER BIOCHEMICAL CHANGES IN PATIENTS AFTER DIFFERENTIATED THYROID CANCER SURGERY AND BEFORE RADIOIODINE THERAPY

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Abstract

Objectives: To investigate liver enzyme changes and assess liver function in patients with differentiated thyroid cancer who have undergone surgery and are preparing for radioiodine therapy (^{131}I therapy). ***Methods:*** A cross-sectional descriptive study on 163 patients with differentiated thyroid cancer 4 - 6 weeks after surgery at the Military Institute of Radiation Medicine and Oncology from April 2023 to April 2024. Evaluating liver enzymes and liver function at the time of preparing for ^{131}I therapy, compared with a number of paraclinical parameters to find factors related to liver damage. ***Results:*** 38.0% of patients had liver damage (increased AST or ALT), of which 35.6% increased AST, and 24.5% increased ALT, mainly mildly increased (40 to < 100 U/L) and a low rate of liver dysfunction (5.5% slight increase in total Bilirubin and 11.0% decrease in Prothrombin ratio). Male gender and decreased FT4 concentration < 1.17 pmol/L were two factors related to liver damage with ORs of 2.56 and 2.74, respectively, $p < 0.01$. ***Conclusion:*** Liver damage is a relatively common phenomenon related to male gender and FT4 levels in patients with thyroid cancer after surgery and being prepared for ^{131}I therapy.

Keywords: Liver biochemical index; Thyroid cancer; ^{131}I therapy.

INTRODUCTION

Differentiated thyroid cancer is the most common endocrine cancer, has a high incidence, and is rising worldwide. It has a good prognosis if detected and treated properly, including total or nearly total thyroidectomy, followed by ^{131}I therapy to destroy remaining normal thyroid tissue to prevent recurrence or destroy local/distant metastases (if any)

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in combination with thyroid stimulating hormone (TSH) suppression therapy [1, 2]. In order to increase the ability of ^{131}I absorbent into the thyroid tissue, post-operation patients are required not to take thyroid hormone replacement therapy and practice a low-iodine diet, which will lead to a state of active hypothyroidism. This condition can result in some consequences, such as myxoedema, constipation, neuropsychiatric disorders, etc. It also affects the liver, causing liver enzyme elevation and liver dysfunction [3]. Liver damage in patients with post-operative hypothyroidism has been demonstrated in many studies worldwide [4, 5], however, research on this issue has not yet been recorded in Vietnam. Therefore, we conducted this study to: *Assess changes in liver enzymes and some liver function indicators in differentiated thyroid cancer patients who have undergone surgery and are preparing for ^{131}I therapy.*

MATERIALS AND METHODS

1. Subjects

Including 163 patients with thyroid cancer after surgery and indicated for ^{131}I therapy at the Military Institute of Radiation Medicine and Oncology from April 2023 to April 2024.

* *Inclusion criteria:* Patients with thyroid cancer diagnosed by histopathology and who had undergone total or nearly total thyroidectomy 4 - 6 weeks ago; indicated and prepared for ^{131}I therapy (not using thyroid hormone replacement with a low-iodine diet); aged over 18; had normal neuropsychiatric status and agreed to participate in the study.

* *Exclusion criteria:* Patients who have been using liver protection medicines during the postoperative period, active hepatitis B/C virus infection, alcohol abuse, comorbidity of another cancer, etc.

2. Methods

* *Study design:* A cross-sectional descriptive study.

Patients with differentiated thyroid cancer after surgery who meet the inclusion and exclusion criteria were asked for their medical history (directly on the patients and through the surgical medical records). Post-operative disease staging was determined according to the American Joint Committee on Cancer 8th, 2017.

Patients did not use thyroid hormones for 4 - 6 weeks after surgery with a low-iodine diet to increase TSH concentration > 30 μIU/mL (causing active hypothyroidism) to increase the ability to absorb ^{131}I into the remaining thyroid tissue and metastatic tissues (if any) to destroy thyroid tissue and destroy any remaining cancer cells [2].

Liver enzyme tests, some indicators to evaluate liver function (Albumin, total Bilirubin, and Prothrombin ratio) and thyroid hormones (TSH, T3, FT4) at 4 - 6 weeks after surgery on the AU680 biochemical system (Beckman, Coulter, USA) and the ACL-TOP500 automatic coagulation machine.

Classify the level of increased liver enzymes AST, ALT according to the criteria for evaluating adverse events version 4.0 of the US National Cancer Institute (CTCAE v4.0) [6], as follows:

+ Normal < 40 U/L (ULN - upper limit normal).

+ Mild increase in liver enzymes: ≥ ULN and < 2.5 ULN.

+ Moderate increase in liver enzymes: ≥ 2.5 ULN and < 5 ULN.

+ High increase in liver enzymes: ≥ 5 ULN.

Assess patients with liver damage when AST and/or ALT increase > 40 U/L.

Change the value of biochemical indexes according to the threshold at the Military Institute of Radiation Medicine and Oncology and physiological parameters of Vietnamese people:

+ Normal albumin 34 - 48 g/L, decreased when < 34 g/L.

+ Total bilirubin: Normal ≤ 17 μmol/L, increased when > 17 μmol/L.

+ Prothrombin ratio: Normal ≥ 70%, decreased when < 70%.

+ Normal T3 1.3 - 3.1 nmol/L, decreased < 1.3 and increased when > 3.1 nmol/L.

+ Normal FT4 13 - 23 pmol/L, decreased < 13 and increased > 23 pmol/L.

+ Normal TSH 0.27 - 4.2 μIU/mL.

* *Data processing:* Using SPSS 22.0 software.

3. Ethics

The study was approved by the Ethics Council of Military Hospital 103 (No. 2030/HDDD) on June 23rd, 2023. The Military Institute of Radiation Medicine and Oncology granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS

Table 1. Some characteristics of the study subjects (n = 163).

Variables	$\overline{X}$ ± SD or n (%)
Average age	45.92 ± 16.06
Gender (male; female)	30 (18.4); 133 (81.6)
Postoperative disease stage (I; II; III; IV)	131 (80.4); 28 (17.2); 4 (13.5); 0 (0.0)
Histopathology (papillary; follicular; mixed)	156 (95.7); 2 (1.2); 5 (3.1)
Nearly total; total thyroidectomy	28 (17.2); 135 (82.8)

The average age was 45.92 ± 16.06; women were the main subjects with 4.43 times higher than men (81.6% vs. 18.4%). The disease stage was mainly stage I (80.4%) with 95.7% papillary.

Table 2. Thyroid hormone characteristics (n = 163).

TSH (μIU/mL)	Median (Q1 - Q3)	91.89 (67.68 - 100)
	≥ 30 (n, %)	163 (100)
	< 30 (n, %)	0 (0,0)
T3 (nmol/L)	Median (Q1 - Q3)	0.36 (0.30 - 0.58)
	Decrease < 1.3 (n, %)	163 (100)
	Normal/Increase	0 (0.0)
FT4 (pmol/L)	Median (Q1 - Q3)	1.17 (0.73 - 2.89)
	Decrease < 13 (n, %)	163 (100)
	Normal/Increase	0 (0.0)

100% of patients in the study had hypothyroidism at the time of ^{131}I therapy, showing increased TSH concentration > 30 μIU/mL and decreased T3 concentration < 1.3 nmol/L and FT4 < 13 pmol/L.

Table 3. Liver enzyme concentration characteristics (n = 163).

AST (U/L)	Median (Q1 - Q3)	31.32 (21.48 - 49.14)
	< 40 (n, %)	105 (64.4)
	40 - < 100 (n, %)	52 (31.9)
	100 - < 200 (n, %)	4 (2.5)
	≥ 200 (n, %)	2 (1.2)
	Min - Max	12.4 - 290.9
ALT (U/L)	Median (Q1 - Q3)	30.05 (23.82 - 39.72)
	< 40 (n, %)	123 (75.5)
	40 - < 100 (n, %)	38 (23.3)
	100 - < 200 (n, %)	1 (0.6)
	≥ 200 (n, %)	1 (0.6)
	Min - Max	13.6 - 276.5
Elevated liver enzyme (AST/ALT)	62 (38.0)	

The median AST enzyme level before ^{131}I therapy was 31.32 U/L; 35.6% of patients had increased AST enzyme, of which 31.9% had a mild increase, 2.5% had a moderate increase, and 1.2% had a high increase. ALT enzyme increased in 24.5% of patients, of which 23.3% had a mild increase, 0.6% had a moderate and high increase. 38.0% of patients had liver damage (increased AST and/or ALT).

Table 4. Characteristics of some liver function indicators (n = 163).

Bilirubin TP (µmol/L)	Median (Q1 - Q3)	9.40 (7.62 - 12.14)
	≤ 17 (n, %)	154 (94.5)
	> 17 (n, %)	9 (5.5)
	Min - Max	4.8 - 42.0
Prothrombin (%)	Median (Q1 - Q3)	85.90 (75.21 - 98.75)
	≥ 70% (n, %)	145 (89.0)
	< 70% (n, %)	18 (11.0)
	Min - Max	53.5 - 133.4
Albumin (g/L)	Median (Q1 - Q3)	46.64 (44.72 - 48.28)
	≥ 34 (n, %)	163 (100)
	< 34 (n, %)	0

Only 9 patients (5.5%) had increased total bilirubin > 17 µmol/L (the highest was 42 µmol/L), and 18 patients (11.0%) had decreased prothrombin ratio < 70% (the lowest was 53.5%). No patient had decreased plasma albumin before using ^{131}I.

Table 5. Univariate regression analysis of factors related to liver damage.

Variables		No liver damage	Liver damage	OR	95%CI	p
Age	≤ 45 (n, %)	41 (68.3)	19 (31.7)	1.55	0.79 - 3.02	0.20
	> 45 (n, %)	60 (58.3)	43 (41.7)			
Gender	Male (n, %)	13 (43.3)	17 (56.7)	2.56	1.11 - 5.86	0.027
	Female (n, %)	88 (66.2)	45 (38.8)			
TSH* (µIU/mL)	< 91,89 (n, %)	50 (61.7)	31 (38.3)	0.81	0.40 - 1.64	0.56
	≥ 91,89 (n, %)	51 (62.2)	31 (37.8)			
T3* (nmol/L)	< 0,36 (n, %)	46 (56.8)	35 (43.2)	0.83	0.40 - 1.71	0.62
	≥ 0,36 (n, %)	55 (67.1)	27 (32.9)			
FT4* (pmol/L)	< 1.17 (n, %)	27 (46.6)	31 (53.4)	2.74	1.39 - 5.37	0.003
	≥ 1.17 (n, %)	74 (70.5)	31 (29.5)			

(: Median)*

Male gender and decreased FT4 concentration < 1.17 pmol/L were predictors of liver damage with ORs of 2.56 and 2.74, p = 0.027 and 0.003, respectively.

DISCUSSION

1. Some characteristics of the study subject

80.4% of the patients in the study group were female (the female/male ratio was 4.43/1), with an average age of 45.92 ± 16.06. This result is consistent with the study by Dang Trung Dung et al. (2023) on 98 patients with differentiated thyroid cancer, recording an average age of 43.0 ± 14.5, with 87.8% women [7].

Regarding the stage and histopathology, we found that 80.4% of differentiated thyroid cancer was in stage I when the tumor was still localized in the thyroid gland, and the major histopathological type was papillary (accounting for 95.7%). The study by Dang Trung Dung et al. (2023) also showed that papillary type accounted for 95.6%, and stage I accounted for 75.6% [7].

2. Changes in liver enzymes and liver function in thyroid cancer patients after surgery who are preparing for ^{131}I therapy and some related factors

Currently, the standard treatment for differentiated thyroid cancer is surgery (total or nearly total thyroidectomy), followed by ^{131}I therapy to destroy the remaining thyroid parenchyma (to avoid local recurrence) and/or destroy metastatic lesions (if any). To prepare well for ^{131}I therapy immediately after surgery, the patient must not use thyroid hormone replacement and have to practice a low-iodine diet to induce

hypothyroidism (increase TSH, decrease T3, FT4 concentrations in the blood), thereby increasing the ability to absorb ^{131}I into the thyroid tissue to increase the destruction efficiency. The results of our study showed that 4 - 6 weeks after surgery, 100% of patients had hypothyroidism (*Table 2*). However, when hypothyroidism occurs, it will affect the function of organs, including the liver. We noted 38.0% of patients with liver damage showing an increase in at least one of the two liver enzymes (AST/ALT), of which mild increases were mainly recorded (31.9% with mild AST increase and 23.3% with mild ALT increase), the proportion of patients with moderate and high increases was quite low < 3%. At the same time, a small proportion of patients had impaired liver function (5.5% had a mild increase in total Bilirubin, and 11.0% had a decreased Prothrombin ratio). The study by Ji Y et al. (2022) on 996 patients with hypothyroidism after thyroid cancer surgery also recorded 31.6% of patients with impaired liver function without clinical symptoms, and the most common abnormality was increased liver enzymes (AST or ALT) accounting for 47.5%, also mainly mild increase in liver enzymes (80%) [5]. Another study by Han Y et al. (2012) on 77 hypothyroid patients (who did not use thyroid hormone replacement after surgery) observed liver damage, showing that AST and ALT levels were significantly higher than before surgery (37.9 ± 16.2 vs. 19.5 ± 4.5 U/L and 39.6 ± 21.7 vs. 19.2 ± 17.3 U/L, respectively) [4]. Thus, liver damage is a fairly common phenomenon in thyroid cancer patients after surgery who are prepared for ^{131}I therapy by inducing active hypothyroidism (without using thyroid hormone), but mainly mild enzyme elevation. The mechanism of increased liver enzymes is thought to be related to reduced lipid metabolism in the liver and fatty liver due to reduced thyroid hormone levels leading to increased lipolysis, accumulation of lipid droplets in liver lysosomes, etc. In addition, increased AST is also related to myopathy due to hypothyroidism [3].

We also identified some factors associated with liver damage through univariate logistic regression analysis. The results in table 5 show that 2 factors that increase the risk of liver damage are male gender (OR: 2.56; 95%CI: 1.11 - 5.86, p = 0.027) and decreased FT4 concentration < 1.17 pmol/L (OR: 2.74; 95%CI: 1.39 - 5.37, p = 0.003), similar to the results of Ji Y et al. (2022) [4]. The results are consistent with the biological role of thyroid hormones, which is to participate in blood lipid metabolism, especially lipid metabolism in the liver. Decreased FT4 will increase the risk of fatty liver and liver damage [3].

CONCLUSION

38.0% of patients had liver damage (increased AST or ALT enzymes), of which 35.6% had increased AST, and 24.5% had increased ALT, mainly mild increase (40 - < 100 U/L) and low rate of liver dysfunction (5.5% mild increase in total Bilirubin and 11.0% decreased Prothrombin ratio). Male gender and decreased FT4 concentration < 1.17 pmol/L were two factors related to liver damage with OR of 2.56 and 2.74, $p < 0.01$, respectively.

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CLINICAL AND PARACLINICAL CHARACTERISTICS OF DENGUE HEMORRHAGIC FEVER DURING PREGNANCY AT MILITARY HOSPITAL 103 IN 2023

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Abstract

Objectives: To describe clinical and paraclinical characteristics of Dengue hemorrhagic fever (DHF) during pregnancy at the Department of Infectious Diseases, Military Hospital 103 in 2023. ***Methods:*** A cross-sectional descriptive study was conducted on 162 pregnant patients diagnosed with DHF and treated at the Department of Infectious Diseases during the 2023 epidemic (from January 2023 to December 2023). ***Results:*** The average age was 28.92 ± 5.64 years old, and the average gestational age was 18.98 ± 16.75 weeks. Clinical characteristics: Fever was 84.4%; headache and eye socket pain was 76.5%; bleeding gums was 8.6% and abnormal vaginal bleeding was 2.5%; pleural or peritoneal effusion was 7.4%. Paraclinical characteristics: Decreased white blood cell (WBC) (< 4 G/L) was 28.4%, decreased platelet (< 150 G/L) was 69.1%. The first-trimester group had the highest average hematocrit (HCT) (%) and WBC ($p < 0.05$), and the second-trimester group had the lowest average platelet ($p < 0.05$) compared to other groups. Blood biochemistry tests showed increased AST (≥ 40 U/L) and ALT (≥ 40 U/L) in 46.7% and 49.3% of cases, respectively. The third-trimester group had the highest average AST and ALT compared to other groups ($p < 0.01$). Disease severity was classified as DHF with warning signs in 24.7% of cases and severe DHF in 1.8% of cases. The third-trimester group had the highest rate of DHF with warning signs and severe DHF ($p < 0.05$). ***Conclusion:*** Pregnant women with DHF exhibit different clinical and paraclinical characteristics depending on the stage of pregnancy. The third-trimester group experienced more severe liver injury and disease severity compared to the first and second-trimester groups.

Keywords: Pregnancy; Dengue hemorrhagic fever; Clinical and paraclinical characteristics.

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INTRODUCTION

Southeast Asia is among the regions most affected by Dengue, and Vietnam is one of the five countries in this region with the highest burden. In 2022, the country recorded 367,729 DHF cases and 140 deaths, and as of December 17, 2023, the country recorded 166,619 infections, including 42 deaths. Dengue fever can occur in all subjects, including pregnant women. Dengue during pregnancy increases the risk of pre-eclampsia, severe DHF, fetal distress, preterm delivery, caesarean delivery, and maternal mortality [1]. In addition, physiological changes during pregnancy, according to the stages of pregnancy, have been shown to have certain effects on the progression and prognosis of the disease [2]. Currently, in Vietnam, the number of studies on Dengue fever in pregnant women published in medical literature is limited. Therefore, we conducted this study to: *Describe the clinical and paraclinical characteristics of DHF in pregnant women. At the same time, we compared some clinical and paraclinical differences between stages of pregnancy.*

MATERIALS AND METHODS

1. Subjects

Including 162 pregnant patients diagnosed with DHF and treated at the Department of Infectious Diseases, Military Hospital 103 in 2023.

* *Inclusion criteria:* Pregnant patients diagnosed with DHF according to the Vietnamese Ministry of Health's standards in 2019 - Decision No. 3705/QD-BYT, with positive Dengue - NS1 or Dengue - IgM test results [3].

* *Exclusion criteria:* Patients who did not agree to participate in research.

* *Time and location:* From January 1, 2023 to December 30, 2023 at the Department of Infectious Diseases, Military Hospital 103.

2. Methods

* *Study design:* A cross-sectional descriptive study.

* *Sample size:* Purposive convenience sampling method, all eligible patients were included in the study.

* *Research content:*

General information: Characteristics of age, gestational age.

Group division by pregnancy stage: First 3 months, middle 3 months, and last 3 months of pregnancy.

Clinical characteristics: Symptoms and signs at the time of admission.

Paraclinical tests: Red blood cells (RBC), Hemoglobin (Hb), HCT, WBC, platelets (PLT), AST, ALT; Abdominal and pleural ultrasound assessed at the time of admission.

Assessment of disease severity: According to the 2019 Ministry of Health guidelines, divided into levels of DHF, DHF with warning signs and severe DHF [4].

* *Data collection and processing:* Data was collected from unified research medical records, entered into Excel 16.0 software, and processed with SPSS 22.0 software.

3. Ethics

The research data does not impact testing rights or expenses and is gathered from patient data that is regularly tested throughout hospital inpatient treatment. All patient data is kept private and confidential. The Department of Infectious Diseases, Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest.

RESULTS

Through a study of 162 pregnant patients diagnosed with DHF, the average age was 28.92 ± 5.64 years old. The age group of 21 - 30 years old accounted for the highest percentage (51.9%), followed by the 31 - 40-year-old group at 29.0%.

The average gestational age was 18.98 ± 16.75 weeks, with the smallest being 6 weeks and the largest being 39 weeks. According to the trimesters, the middle 3 months accounted for the largest proportion with 39.5% (64 pregnancies); the first 3 months and the last 3 months accounted for 32.1% (52 pregnancies) and 28.4% (46 pregnancies), respectively.

1. Clinical characteristics at the time of admission

Table 1. Symptoms and signs of DHF in pregnant women.

Symptoms and signs		Number (n = 162)	Percentage (%)
Systemic	Fever	136	84.0
	Headache, eye pain	124	76.5
	Muscle and joint pain	98	60.5
Digestive	Nausea and vomiting	76	46.9
	Liver pain	12	7.4
	Epigastric pain	32	19.8
	Diarrhea	10	6.2
Hemorrhage	Nose bleeding	8	4.9
	Gum bleeding	14	8.6
	Vaginal bleeding	4	2.5
	Gastrointestinal bleeding	0	0.0
Other	Pleural/peritoneal effusion	12	7.4

Some common symptoms include fever (84.4%); headache and eye pain (76.5%); nausea and vomiting (46.9%); epigastric pain (19.8%); bleeding gums (8.6%) and abnormal vaginal bleeding (2.5%); pleural or peritoneal effusion (7.4%).

2. Paraclinical characteristics at the time of hospitalization

Table 2. Characteristics of some hematology tests.

Results		First 3 months (n = 52)(1)	Middle 3 months (n = 64)(2)	Last 3 months (n = 46)(3)	p	Number (n = 162)	Percentage (%)
RBC (T/L)	$\overline{X} \pm SD$	4.67 ± 0.65	4.53 ± 0.86	4.52 ± 0.92	> 0.05	4.58 ± 0.75	
Hb (g/L)	$\overline{X} \pm SD$	118.94 ± 12.30	118.32 ± 13.68	116.34 ± 10.12	> 0.05	117.21 ± 12.34	
	< 110	10 (19.2)	14 (21.9)	10 (21.7)	> 0.05	34	21.0
	≥ 110	42 (80.8)	50 (78.1)	36 (78.3)		128	79.0
HCT (%)	$\overline{X} \pm SD$	46.23 ± 8.64	43.67 ± 6.06	44.26 ± 5.25	$p_{(1-2.3)} < 0.05$	44.56 ± 6.45	
	< 47	45 (86.5)	53 (82.8)	35 (76.1)	> 0.05	133	82.1
	≥ 47	7 (13.5)	11 (17.2)	11 (23.9)		29	17.9
WBC (G/L)	$\overline{X} \pm SD$	6.38 ± 3.23	6.06 ± 3.38	5.78 ± 4.34	$p_{(1-3)} < 0.05$ $p_{(1-2.\ 2-3)} > 0.05$	6.21 ± 4.03	
	< 4	20 (38.5)	16 (25.0)	10 (21.7)		46	28.4
	4 - 10	29 (55.8)	40 (62.5)	25 (54.4)	< 0.05	94	58.0
	> 10	3 (5.7)	8 (12.5)	11 (23.9)		22	13.6
PLT (G/L)	$\overline{X} \pm SD$	56.78 ± 62.43	42.65 ± 52.65	48.87 ± 56.34	$p_{(2\ -1.3)} < 0.05$	50.02 ± 58.39	
	< 50	8 (15.4)	18 (28.1)	11 (23.9)		38	23.4
	50 - 149	28 (53.8)	36 (56.3)	27 (58.7)	> 0.05	74	45.7
	≥ 150	16 (30.8)	10 (25.6)	8 (17.3)		50	30.9

Pregnant women with DHF had a rate of decreased Hb (< 110 g/L) of 21.0%, increased HCT (≥ 47%) of 17.9%, decreased WBC (< 4 G/L) of 28.4%; decreased PLT with < 50 G/L and 50 - 149 G/L of 23.4% and 45.7%, respectively. The first 3-month group had a higher average HCT (%) and WBC than the other groups ($p_{1-2.3} < 0.05$ and $p_{1-3} < 0.05$). The middle 3-month group had the lowest average number of WBC among the comparison groups ($p < 0.05$).

Table 3. Characteristics of some liver function tests.

Results		First 3 months (n = 49)(1)	Middle 3 months (n = 61)(2)	Last 3 months (n = 40)(3)	p	Number (n = 150)	Percentage (%)
AST (U/L)	$\overline{X} \pm SD$	85.67 ± 125.64	96.76 ± 234.45	126.56 ± 326.76	< 0.01	102.16 ± 267.58	
	< 40	33 (67.3)	27 (44.3)	20 (50.0)	> 0.05	80	53.3
	40 - 399	15 (30.7)	30 (49.2)	15 (37.5)		60	40.0
	400 - 999	1 (2.0)	3 (4.9)	3 (7.5)		7	4.7
	≥ 1000	0 (0.0)	1 (1.6)	2 (5.0)		3	2.0
ALT (U/L)	$\overline{X} \pm SD$	96.54 ± 106.43	93.34 ± 156.76	145.56 ± 289.70	< 0.01	121.13 ± 206.23	
	< 40	30 (61.2)	30 (49.2)	16 (40.0)	> 0.05	76	50.7
	40 - 399	17 (34.7)	26 (42.6)	17 (42.5)		60	40.0
	400 - 999	2 (4.1)	4 (6.6)	5 (12.5)		11	7.3
	≥ 1000	0 (0.0)	1 (1.6)	2 (5.0)		3	2.0

(Exclude pregnant women with a history of chronic liver disease)

The rates of increased AST and ALT (≥ 40 U/L) were 46.7% and 49.3%, respectively. Of these, increased AST ≥ 1000 U/L and ALT ≥ 1000 U/L both accounted for 2.0%. In the last 3 months of pregnancy, the average AST and ALT enzyme activity increased more than the other two groups ($p < 0.01$).

Table 4. Classification of disease level of study subjects.

Disease severity	First 3 months (n = 52)(1)		Middle 3 months (n = 64)(2)		Last 3 months (n = 46)(3)		p	Number (n = 162)	Percentage (%)
	n	%	n	%	n	%			
DHF	46	88.5	47	73.4	26	56.6		119	73.5
DHF with warning signs	6	11.5	16	25.0	18	39.1	< 0.01	40	24.7
Severe DHF	0	0.0	1	1.6	2	4.3		3	1.8

The rates of DHF with warning signs and severe DHF in pregnant women accounted for 24.7% and 1.8%, respectively. The last 3-month group had a higher rate of DHF with warning signs and severe DHF than the other two groups, with $p < 0.05$.

DISCUSSION

1. Clinical and paraclinical characteristics of Dengue fever in pregnant women

** Characteristics of age and gestational age:*

According to our study results, pregnant women were all of childbearing age, with an average age of 28.92 ± 5.64 years old. The results are similar to Nguyen Thi Thu Huyen (2018), with an average age of 27.9 ± 4.8 [4]; and Trinh Tien Dat, with an average age of 24.8 ± 3.7 [5].

The average gestational age in the study was 18.98 ± 16.75 weeks, of which the middle-trimester group accounted for the highest proportion at 39.5%, followed by the first- and last-trimester groups. The studies by Machain-Williams C (2018) in Mexico [6] and Nguyen Thi Thu Huyen (2018) [4] also noted that the middle 3-month group had a higher rate than the other groups.

** Clinical characteristics:*

Some clinical symptoms include fever (84.4%), headache and eye pain (76.5%), nausea and vomiting (46.9%),

and epigastric pain (19.8%). These are common symptoms similar to those of patients with common Dengue fever.

Abnormal vaginal bleeding in our study accounted for 2.5%, lower than the research results of Nguyen Thi Thu Huyen (2018) at 11.8% [4], but similar to Hoang Xuan Cuong (2022) at 4.1% [7]. The difference can be explained by the research subjects, stage characteristics, and severity of the disease. In addition, the progression, severity, and complications of DHF change every year according to the characteristics of each epidemic.

The rate of peritoneal and pleural effusion in our study accounted for a significant proportion of 7.4%. This is one of the signs included in the diagnostic criteria for DHF, indicating hospitalization and close monitoring and treatment for these subjects. Physiological changes in the cardiovascular system of pregnant women have been mentioned in some studies, such as decreased systemic vascular resistance, increased capillary permeability leading to decreased plasma volume, and increased risk of preeclampsia. Placental ischemia in the first 3 months can occur secondary to increased vascular permeability and fluid leakage from interstitial spaces [1].

2. Paraclinical characteristics of Dengue fever in pregnant women

** Complete blood count:*

According to a report by Hoang Xuan Cuong (2022) on 727 subjects with Dengue fever in 2022, the rate of PLT reduction was 96.3%, in which PLT < 50 G/L ranged from 70 - 80%; HCT increase (≥ 47%) accounted for about 20 - 30% [7]. Compared to our study, the rate of HCT increase was lower, accounting for only 17.9%. As mentioned above, the physiological process of pregnancy is often accompanied by vasodilation, so changes in hematological status and blood concentration due to plasma leakage are often not as obvious as in normal subjects.

Comparing the stages of pregnancy, the first-trimester group had a higher average HCT than the other two groups ($p < 0.05$). This can be explained by the physiology of pregnancy. In the first trimester, plasma volume often decreases, then gradually increases and stabilizes in the last 3 months of pregnancy. According to the guidelines for diagnosis and treatment of DHF in pregnant women in Sri Lanka, special attention is required, as pregnant women with Dengue fever in the first 3 months face an increased risk of plasma leakage from the blood vessels, trapped blood pressure, preeclampsia, and acute pulmonary edema [8].

Regarding PLT, the overall PLT reduction rate in the study was 69.9%. Comparing between pregnancy stages, the second-trimester group had a significantly lower PLT reduction than other stages in pregnancy ($p < 0.01$). During pregnancy, the second-trimester period is often accompanied by hypercoagulability, and gestational thrombocytopenia can occur in 4.4 - 11.6% [9]. Therefore, the group of pregnant women in this period with DHF may have a combination of physiological and pathological thrombocytopenia.

Regarding the WBC index, in the guidelines for diagnosing Dengue fever, leukopenia or leukopenia is one of the common signs. However, in pregnant women, leukopenia is rarely recorded. In our study, the overall rate of leukopenia was 28.6%. Comparing the pregnancy groups, the first-trimester group had a higher leukopenia count than the other two groups, with a clear difference recorded when comparing the first and last trimesters of pregnancy ($p < 0.05$). This is explained by the fact that pregnant women often have increased leukocytes, especially in the first trimester of pregnancy. Therefore, the guidelines for Dengue fever in pregnant women also noted that leukopenia may not be recorded, especially in the first trimester of pregnancy [8].

** Liver function test:*

In pregnant women with Dengue fever, Machain-Williams C (2018) reported elevated ALT and AST with the following rates: 92.6% and 46.3% in the DHF group; 93.3% and 66.7% in the DHF with warning signs; and 92.3% and 92.3% in severe DHF [6]. The results of our study were lower, the rate of increased AST and ALT in pregnant women (> 40 U/L) accounted for only 46.7% and 49.3%, respectively. The difference can be explained by the time of test assessment. Our study chose a cross-sectional time point upon admission, the above studies chose a time point when the tests changed most clearly during the hospital follow-up, so the rate was different.

Comparing between gestational age groups, we noted that in the last 3 months of pregnancy, elevated AST and ALT were significantly higher than in the first 3 months and the middle 3 months of pregnancy ($p < 0.05$). Currently, there is no study that addresses changes in liver function during pregnancy. In our opinion, in addition to the mechanisms of liver damage caused by the Dengue virus (the virus directly attacks liver cells and Kupffer cells, cytotoxicity through T lymphocytes, cytokines such as IL-2, IL-6, TNF-α, and IFN-γ, ...); there are

also mechanisms of increased liver enzymes in pregnant women such as cholestasis in the liver, HELP syndrome, fatty liver, etc. Pathological liver enzyme elevation disorders in pregnant women are often concentrated in the last months of pregnancy, so when infected with Dengue virus, it will aggravate liver cell damage.

** Characteristics of disease severity:*

In our study, the rates of DHF with warning signs and severe DHF were 24.7% and 1.8%, respectively. The last 3-month group had higher rates of DHF with warning signs and severe DHF than the other two groups ($p < 0.05$). Our results were similar to those of Machain-Williams C (2018), the rates of DHF with warning signs and severe DHF were 18.3% and 15.9%, respectively. No cases of DHF with warning signs and severe DHF were recorded in the first 3 months of pregnancy. The last 3-month group also had the highest rate of severe Dengue fever compared to the other groups [6]. Thus, the rates of DHF and severe DHF may be slightly different, possibly due to different standards and guidelines for classifying disease severity according to the regulations of each research country. However, our results and some studies mentioned above show that the last 3-month group was at high risk of DHF with warning signs and severe DHF when compared with subjects in other stages of pregnancy.

CONCLUSION

Compared to the other two groups, the average HCT (%) and WBC count of the first-trimester group was higher ($p < 0.05$). On the other hand, the last-trimester group showed an increase in the liver enzymes AST and ALT ($p < 0.05$), whereas the middle-trimester group had the lowest average platelet count ($p < 0.05$). The percentages of DHF with warning signs and severe DHF were 24.7% and 1.8%, respectively, in relation to the disease's severity. When compared to the other two groups, the last-trimester group had the highest percentage of DHF with warning signs and severe DHF ($p < 0.05$).

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CHARACTERISTICS OF MULTIMORBIDITY IN HOSPITALIZED ELDERLY PATIENTS

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Abstract

Objectives: To evaluate the characteristics of multimorbidity in hospitalized elderly patients and identify factors associated with the length of stay and the number of comorbidities. ***Methods:*** A retrospective, cross-sectional study was conducted using 507 medical records of patients aged > 60 years. Descriptive statistics, one-way ANOVA, Pearson correlation analysis, and multiple regression analysis were used to evaluate the associations between age, sex, number of comorbidities, and length of stay. ***Results:*** The median age of patients was 73 years, and the sex distribution was relatively balanced (49.3% male, 50.7% female). The median length of stay was 9 days, and the median number of comorbidities was 3. Hypertension was the most common comorbidity (61.9%), followed by chronic lung disease (30.4%) and diabetes (26.4%). Older age and number of comorbidities were associated with longer length of stay. The number of comorbidities was an independent factor affecting the length of hospital stay (OR = 1.63; 95%CI = 1.41 - 1.89). ***Conclusion:*** Multimorbidity is common in hospitalized elderly patients, and factors such as advanced age and the number of comorbidities increase the length of hospital stay. Management and medical care should be strengthened to meet the needs of this group of patients.

Keywords: Elderly patients; Multimorbidity; Length of hospital stay; Chronic diseases.

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INTRODUCTION

Multimorbidity is the simultaneous occurrence of multiple chronic diseases in the same patient, especially in the elderly [1, 2]. The biological mechanism may be due to aging and the development of different diseases, the impact of which can lead to complex developments [2]. Elderly people often have complex health problems due to the combination of chronic diseases such as hypertension, diabetes, cardiovascular disease, and other disorders [3]. This increases the risk of prolonged hospitalization, affects quality of life, and places a great burden on the health system.

There have been many studies on factors related to multimorbidity in elderly people worldwide. The factors included age, sex, number of comorbidities, living conditions of the elderly individuals, number of visits, number of hospitalizations, length of stay, and treatment costs. These studies have helped to improve the understanding of the need for integrated disease management and the development of effective treatment strategies to reduce hospital stays and improve treatment outcomes [2, 4].

However, in Vietnam, studies related to multimorbidity in the elderly population are still limited. Although there are some small studies on individual chronic diseases, such as hypertension or diabetes, comprehensive studies on combined multimorbidity and factors related to hospital stays have not been widely conducted. This study analyzed the characteristics of combined multimorbidity in elderly patients hospitalized at a large hospital in Vietnam. This paper aims to: *Identify factors associated with length of stay and number of comorbidities, and provide data to support the development of appropriate health strategies.*

MATERIALS AND METHODS

1. Subjects

Including 507 medical records of patients aged > 60 years who were admitted and discharged from the Senior Officer Department, Military Hospital 103, between December 2022 and October 2024.

* *Exclusion criterion:* Medical records of patients who were not admitted or discharged from the research department.

2. Methods

* *Study design:* A retrospective, cross-sectional study based on data collected from inpatient medical records.

* *Protocol:*

Medical records were collected by selecting and coding medical records that met the criteria for the study.

Recording information: Data, including admission and discharge times, year of birth, sex, and disease status, were collected from medical records and coded according to the International Classification of Diseases-10.

Data entry and processing: After the data were collected, the data were entered into Excel software and checked again before analysis with SPSS 26.0.

Diagnostic criteria: (1) Hypertension: According to the Vietnam Heart Association and ESC/ESH 2018: Systolic blood pressure (SBP) ≥ 140 mmHg and/or diastolic blood pressure (DBP) ≥ 90 mmHg measured in a clinical setting; or the patient is on antihypertensive medication. (2) Diabetes mellitus: According to the American Diabetes Association (ADA 2023): Fasting plasma glucose (FPG) ≥ 126 mg/dL (7 mmol/L) after fasting for at least 8 hours; HbA1c ≥ 6.5% (48 mmol/mol); plasma glucose ≥ 200 mg/dL (11.1 mmol/L) 2 hours after a 75-gram oral glucose tolerance test (OGTT); or random plasma glucose ≥ 200 mg/dL (11.1 mmol/L) with symptoms of hyperglycemia; or the patient is on glucose-lowering medication. (3) Arrhythmias: According to the Vietnam Heart Association and ESC 2020: Diagnosed by electrocardiogram (ECG) with the following findings: Atrial fibrillation; premature atrial or ventricular contractions; right or left bundle branch block; atrioventricular block (grade I, II, or III). (4) Heart failure: According to ESC 2021: Clinical symptoms: Dyspnea, fatigue, or leg edema (NYHA class II-IV); echocardiographic findings: Heart failure with reduced ejection fraction (HFrEF): Left ventricular ejection fraction (LVEF) ≤ 40%; heart failure with preserved ejection fraction (HFpEF): LVEF ≥ 50% with signs of heart failure. (5) Dyslipidemia: According to the Vietnam Endocrine Society and ESC/EAS 2019: Total cholesterol (TC) ≥ 5.2 mmol/L (200 mg/dL); LDL cholesterol ≥ 2.6 mmol/L (100 mg/dL); HDL cholesterol < 1.0 mmol/L (40 mg/dL) in men or < 1.2 mmol/L (50 mg/dL) in women; triglycerides (TG) ≥ 1.7 mmol/L (150 mg/dL); or the patient is on lipid-lowering therapy. (6) Chronic kidney disease (CKD): According to KDIGO 2012: Glomerular filtration rate (GFR) < 60 mL/min/1.73m² for ≥ 3 months; or structural or functional kidney abnormalities for ≥ 3 months, evidenced by: Albuminuria ≥ 30 mg/24h; persistent urinary abnormalities (e.g., hematuria, leukocyturia); imaging findings indicating kidney abnormalities. (7) Old stroke: According to the Vietnam Ministry of Health: History of ischemic or hemorrhagic stroke that is no longer in

the acute phase; or imaging evidence (MRI or CT) of previous stroke lesions. (8) Gout: According to ACR/EULAR 2015: Serum uric acid level > 6.8 mg/dL (408 μmol/L); history of recurrent acute arthritis, particularly involving the first metatarsophalangeal joint; identification of urate crystals in synovial fluid or tophi. (9) Musculoskeletal diseases: According to the Vietnam Ministry of Health: Rheumatoid arthritis: Diagnosed according to ACR/EULAR 2010 criteria. Osteoarthritis: Identified by clinical features and X-ray findings (Kellgren-Lawrence criteria). Osteoporosis: Diagnosed by a T-score ≤ -2.5 using a DXA scan. (10) Neurological diseases: According to the Vietnam Ministry of Health and international guidelines; Alzheimer's disease: Diagnosed per DSM-5 or NIA-AA criteria; herniated disc: Confirmed by MRI showing nerve root compression; peripheral neuropathy: Diagnosed through clinical features and electromyography (EMG). (11) Cachexia: According to the International Association of Gerontology: Loss of skeletal muscle mass ≥ 5% within 12 months; body mass index (BMI) < 18.5 kg/m²; reduced muscle strength assessed by clinical tests (e.g., handgrip strength). (12) Chronic lung disease: According to GOLD 2023 and the Vietnam Ministry of Health: COPD: Post-bronchodilator FEV1/FVC < 0.7; asthma: Diagnosed based on clinical history and spirometry results. Chronic bronchitis: Persistent cough and sputum production for ≥ 3 months over 2 consecutive years. (13) Chronic diseases: According to WHO: Non-communicable diseases (NCDs), including cardiovascular diseases, cancer, diabetes, chronic respiratory diseases, musculoskeletal diseases, and neurological disorders; duration of ≥ 6 months, requiring long-term monitoring and management.

** Research variables:*

Independent variable: Number of diseases (calculated according to the ICD-10 diagnosis list). Diseases are coded in binary form (0 - 1), with 1 being the presence of the disease and 0 being the absence of the disease.

Dependent variables: Patients were divided into 4 age groups: 60 - 69, 70 - 79, 80 - 89, ≥ 90; sex (male/female); total number of diseases: The patients were grouped into 1 disease, 2 diseases, 3 diseases, 4 diseases, 5 diseases, and 6 diseases or more; duration of hospital stay (classified by median: Short and medium, or long).

** Data processing:*

Descriptive analysis: Frequencies and proportions were calculated to describe the characteristics of the study sample (age, sex, number of combined diseases, and length of hospital stay).

Comparative analysis: One-way ANOVA and two-way ANOVA were used to compare age group, sex, number of combined diseases, and length of hospital stay. Correlation analysis: The Pearson correlation test was used to evaluate the associations between age, length of hospital stay, and number of combined diseases. Multivariate regression analysis: Factors associated with length of hospital stay were evaluated using odds ratios (ORs) and 95% confidence intervals (CIs).

3. Ethics

The study was conducted in compliance with the Declaration of Helsinki of the World Medical Association. The study data were anonymized without containing specific personal information about the patients to ensure confidentiality and privacy. The Department of Senior Staff, Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS

The study showed a fairly even distribution of male and female patients, with a median age of 73 years. The median length of stay was 9 days, reflecting the severity of the disease. The median number of comorbidities was 3. Patients with 2 or 3 comorbidities predominated (23.7% and 24.3%, respectively). Hypertension was the most common disease (61.9%), followed by chronic lung disease (30.4%) and diabetes mellitus (26.4%) (*Table 1*).

Table 1. Age, sex, number of days in hospital, and disease status of elderly patients (n = 507).

Characteristics	Total, n (%)
Age (years)	
Median (Interquartile Range)	73 (65.80)
Sex	
Male	250 (49.3)
Female	257 (50.7)
Number of days in hospital (days)	
Short and medium length of hospital stay	232 (45.8)
Long length of hospital stay	275 (54.2)

Characteristics	Total, n (%)
Proportion distribution of chronic diseases	
Hypertension	314 (61.9)
Diabetes mellitus	134 (26.4)
Arrhythmias	35 (6.9)
Heart failure	32 (6.1)
Other cardiovascular diseases	15 (3.0)
Dyslipidemia	40 (7.9)
Chronic kidney disease	46 (9.1)
Old stroke	83 (16.4)
Gout	20 (3.9)
Musculoskeletal diseases	18 (3.6)
Neurological diseases	112 (22.1)
Cachexia	52 (10.3)
Chronic lung disease	154 (30.4)

The length of hospital stay and number of comorbidities increased with age ($p < 0.05$) (*Table 2*).

Table 2. The association between age groups and length of hospital stay.

Age group	n	Mean	SD	p
60 - 69	203	3.84	1.477	< 0.05
70 - 79	159	4.45	1.504	
80 - 89	105	5.24	1.929	
≥ 90	40	5.03	1.790	
Total	507	4.41	1.700	

(SD: Standard deviation; n: Number of patients)

The number of comorbidities and length of hospital stay increased with age, particularly in the 80 - 89 and ≥ 90 age groups. (*Figure 1*).

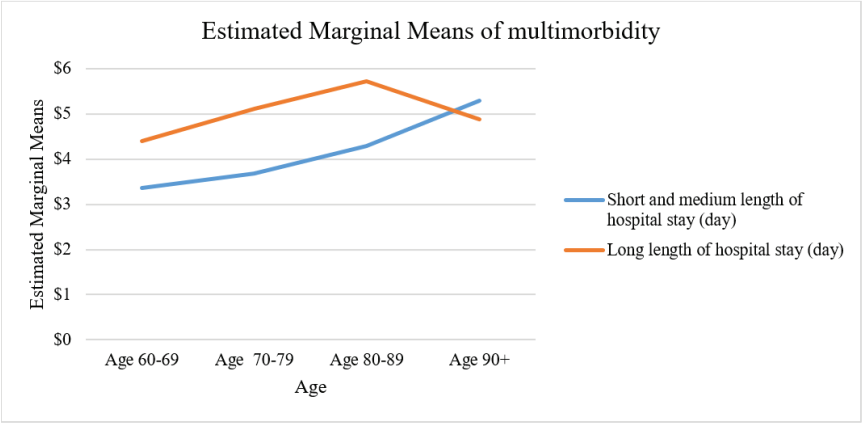

Figure 1. The association between the number of comorbidities and length of hospital stay by age group.

The results showed that age was positively related to the length of hospital stay ($|r| = 0.137$; $p < 0.01$) and the number of comorbidities ($|r| = 0.325$; $p < 0.01$). The greater the number of comorbidities, the longer the hospital stay ($|r| = 0.446$; $p < 0.01$) (*Table 3*).

Table 3. The correlation between age, length of hospital stay, and number of comorbidities (n = 507).

Factors	Pearson correlation	Length of stay	Number of comorbidities
Age	r (p)	0.137 (0.002)	0.325 (0.000)
Length of stay	r (p)		0.446 (0.000)

(r: Pearson correlation)

The number of comorbidities was an independent factor that significantly affected the length of hospital stay (OR = 1.63; 95%CI = 0.000) (*Table 4*).

Table 4. Multivariate regression analysis between length of hospital stay and age, sex, and the number of comorbidities and chronic diseases (n = 507).

Factors	OR	95%CI	p
Age	1.01	0.99 - 1.04	0.18
Gender	0.71	0.49 - 1.05	0.08
Comorbidity	1.63	1.41 - 1.89	0.00*
Chronic diseases	1.41	0.96 - 2.09	0.08

*(OR: Odds ratio; CI: Confidence interval; *: p = 0.000)*

DISCUSSION

The study showed a fairly even distribution of men and women. There was a long hospital stay, a high number of comorbidities, and mainly chronic diseases reflecting the severity of the disease as well as the prevalence of multimorbidity in the elderly patient group, Like other studies showing that multimorbidity is common and a factor that increases the risk of hospitalization, patients with chronic cardiovascular disease and diabetes were identified as complex elderly people [2, 3]. Recognizing age and sex characteristics and the number of comorbidities could identify specific care needs and design personalized treatment intervention programs for elderly people, thereby improving the quality of care and the effectiveness of chronic disease management.

Hypertension was the most common disease, followed by chronic lung disease and diabetes. Jobe, Modou [3] et al. showed that people > 75 years had a prevalence of hypertension of approximately 75%, and that of diabetes was 13%. Xiao Li et al. [5], studying patients aged > 60 years in rural areas in China, reported that the prevalence of hypertension was 50.6%, and that of diabetes, stroke, and COPD were 10.2%, 6.4%, and 5.4%, respectively. Gao S et al. [6], performing an epidemiological study on 210,169 hospitalized elderly patients, showed that hypertension, diabetes, and ischemic heart disease were the leading diseases. Comorbidities are a major problem in hospitalized elderly patients due to an increase in the number of hospitalizations, length of hospital stay, and risk of mortality, which greatly affect the aging process and increase the burden on families and society. Management requires a multidisciplinary approach that includes healthcare staff, geriatric specialists, pharmacists, and the community [6, 7].

The number of comorbidities was an independent factor that increased the length of hospital stay. The older the individual, the higher the risk of multimorbidity and the longer the hospital stay. As in other studies, multimorbidity increased with age from 56% in the 65 - 69 age group to 67% in the 80 - 84 age group [3]. A study by Rodrigues LP et al. [7] showed that multimorbidity was associated with the length of hospital stay, number of hospitalizations, and age. Multimorbidity and long hospital stays are potential risks for polypharmacy and all-cause mortality [8]. Multimorbidity is an independent predictor of long hospital stays and survival [9]. Studies have shown that longer hospital stays are

associated with a number of comorbidities, care services, and caregiver stress. Hospital stays are strongly influenced by health policies and the patient's ability to recover. The extent of the impact of comorbidities may vary from region to region, depending on the quality of health services [7, 8, 9]. Understanding this relationship helps guide appropriate interventions such as increased monitoring and improved hospital care conditions, including the indispensable role of elderly people's healthcare staff. Identifying the relationship between comorbidities and hospital stays is important for health resource planning. Investment in effective chronic disease management programs is needed to reduce hospital stays and improve healthcare quality [10]. The results of this study highlight the importance of effective management and treatment of comorbidities, thereby improving the healthcare quality and reducing its burden. Future studies should extend to multiple hospitals and use a prospective study design to identify effective interventions.

CONCLUSION

The prevalence of comorbidities in hospitalized elderly patients was high, with a median of 3 comorbidities. Hypertension, chronic lung disease, and diabetes were the most prevalent conditions. Older age and the number of comorbidities were associated with the length of hospital stay, with the number of comorbidities identified as an independent factor that increases the length of hospital stay.

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ASSESSMENT OF ANXIETY DISORDER USING THE DASS-21 AND RELATED FACTORS IN MAINTENANCE HEMODIALYSIS PATIENTS

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Abstract

Objectives: To describe the characteristics of anxiety disorders using the DASS-21 (depression - anxiety - stress scale-21) and some related factors in patients with maintenance hemodialysis at Military Hospital 103. ***Methods:*** A cross-sectional descriptive study was conducted on 60 patients with maintenance hemodialysis treated at the Department of Nephrology - Hemodialysis, Military Hospital 103, from August 2023 to May 2024. Patients' anxiety disorders were evaluated using the DASS-21. The Spearman correlation analysis and Mann-Whitney U test were used to analyze the data. ***Results:*** The rate of patients with anxiety disorders was 35.0%. The anxiety disorder score was negatively and significantly correlated with age. The group with the hemodialysis duration of more than 24 months showed significantly higher anxiety scores than the group with the hemodialysis duration of less than 12 months. ***Conclusion:*** The rate of anxiety disorders was high among end-stage renal disease patients with maintenance hemodialysis. Age and hemodialysis duration were factors related to anxiety symptoms in patients with maintenance hemodialysis.

Keywords: Anxiety disorders; DASS-21; Hemodialysis; End-stage renal disease.

INTRODUCTION

Increased anxiety disorders have been reported in patients with chronic kidney disease, especially in patients undergoing maintenance hemodialysis [1, 2]. The prevalence of anxiety disorders in hemodialysis patients ranges from 12% to 52% [1], with higher rates in Europe and Asia [2]. The pathogenesis of anxiety includes many factors in hemodialysis patients, such as comorbidities, chronic pain, chronic inflammation, increased fatigue, uremia, and sleep disturbance, etc. In addition,

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anxiety disorders can lead to decreased quality of life, increased suicidal ideation, sleep disturbances, impaired immune system, and worsening nutritional status. These are all causes of worsening disease and increased mortality in hemodialysis patients. In Vietnam, studies on anxiety disorders in patients undergoing maintenance hemodialysis have been reported [3, 4], but inconsistent results were found. A study by Nguyen Thi Quynh Van et al. reported that the rate of anxiety disorders in patients undergoing hemodialysis at Bach Mai Hospital in 2015 was 40.4% [3]. In contrast, Luong Cong Minh et al. reported that the rate of anxiety disorders in patients undergoing hemodialysis at Nguyen Tri Phuong Hospital was 5.9% [4]. Therefore, we conducted this study to: *Describe the characteristics of anxiety disorders and some related factors in patients undergoing maintenance hemodialysis and treated at Military Hospital 103 from August 2023 to May 2024.*

MATERIALS AND METHODS

1. Subjects

Including 60 patients undergoing maintenance hemodialysis at the Department of Nephrology - Hemodialysis, Military Hospital 103, from August 2023 to May 2024.

* *Inclusion criteria:* Aged ≥ 18 years old and diagnosed as stage 5 chronic kidney disease due to all causes and undergoing maintenance hemodialysis; patients who had started hemodialysis for at least 3 months; patients agreed to participate in this study.

* *Exclusion criteria:* Patients had a history of mental disorders, traumatic brain injury, or use of psychotropic drugs before being diagnosed with chronic kidney disease.

* *Demographic, clinical, and subclinical data:* Age, gender, blood pressure, dialysis duration, body mass index (BMI), medical history, anemia, biochemical test results: Glucose (mmol/L), urea (mmol/L), creatinine (umol/L), uric acid (umol/L), GOT (UI/L), GPT (UI/L), CRP (mg/L), NA+ (mmol/L), K+ (mmol/L), Cl- (mmol/L), Ca++ (mmol/L), albumin (g/L), protein (g/L), cholesterol (mmol/L), triglyceride (mmol/L) of patients were collected before hemodialysis session.

2. Methods

* *Study design*: A cross-sectional descriptive study.

* *Sample size and selection*: Convenient sampling was performed based on the inclusion and exclusion criteria. A total of 60 patients were recruited during the study period.

* *Assessment of anxiety disorder:* The DASS-21 includes 7 questions with a scale from 0 - 3 to assess anxiety

disorders that are widely used in the world to assess patients' anxiety disorders. The patient was asked to answer the anxiety questionnaires on the day of hospital administration. The anxiety score was calculated by the sum of the component scores, then the obtained result was multiplied by 2 with the level of anxiety disorders assessed as follows: Normal (0 - 7 points), mild (8 - 9 points), moderate (10 - 14 points), severe (15 - 19 points), extremely severe (≥ 20 points) [5].

* *Data analysis:* SPSS 21.0 statistical analysis software was used to analyze the data. Descriptive statistics were used to describe patient's characteristics and anxiety disorder scores. Spearman correlation test was used to analyze the correlation between anxiety disorder scores with age and biochemical test values. Mann-Whitney U test was used to compare anxiety disorder scores among patients with different characteristics. The p-value < 0.05 was determined to be statistically significant.

3. Ethics

This study has been approved by the Ethics Council of the Vietnam Military Medical University according to decision No. 2575/QĐ-HYQY dated June 30, 2023. The Department of Nephrology, Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest.

RESULTS

1. Characteristics of the study's participants.

Table 1. Characteristics of the study's subjects.

Characteristics	Median, n	(IQ1, IQ3), [%]
Age (years)	37.0	(31.25, 48.0)
Gender (% male subjects)	25	[41.7]
Height (cm)	160.0	(155.25, 166.0)
Weight (kg)	55.0	(51.0, 58.75)
BMI (kg/m^2)	21.3	(19.3, 22.9)
Smoking status (% yes)	7	[11.7]
Alcohol consumption (% yes)	5	[8.3]
Medical history (% yes)	24	[40.0]
Hemodialysis duration (months)	12.0	(6.0, 36.0)

(n: Number of subjects; SD: Standard deviation; BMI: Body mass index)

Table 1 shows the characteristics of the study subjects. The median (IQ1, IQ2) age was 37.0 (31.25, 48.0) years with 24 (41.7%) subjects of the study were male. The median BMI was 21.2 kg/m^2. 7 (11.7%) and 5 (8.3%) subjects consumed alcohol and smoked cigarettes, respectively. 24 (40.0%) subjects had comorbidity with at least one disease (5 patients with type 2 diabetes, 4 patients with hepatitis B, 7 patients with hepatitis C, 4 patients with coronary artery disease, 1 patient with heart failure, 1 patient with thyroid cancer, and 2 patient with sinusitis). The median duration of hemodialysis was 12.0 months.

2. The rate of anxiety disorders

In a total of 60 patients, there were 21 (35.0%) patients who showed signs of anxiety disorders. Of these, 9 (15.0%) and 7 (11.7%) patients were at mild and moderate levels, respectively, while 1 (1.7%) patient showed severe levels and 4 (6.7%) patients showed extreme levels. The number of subjects without symptoms of anxiety disorders was 39 (65.0%) of the total subjects.

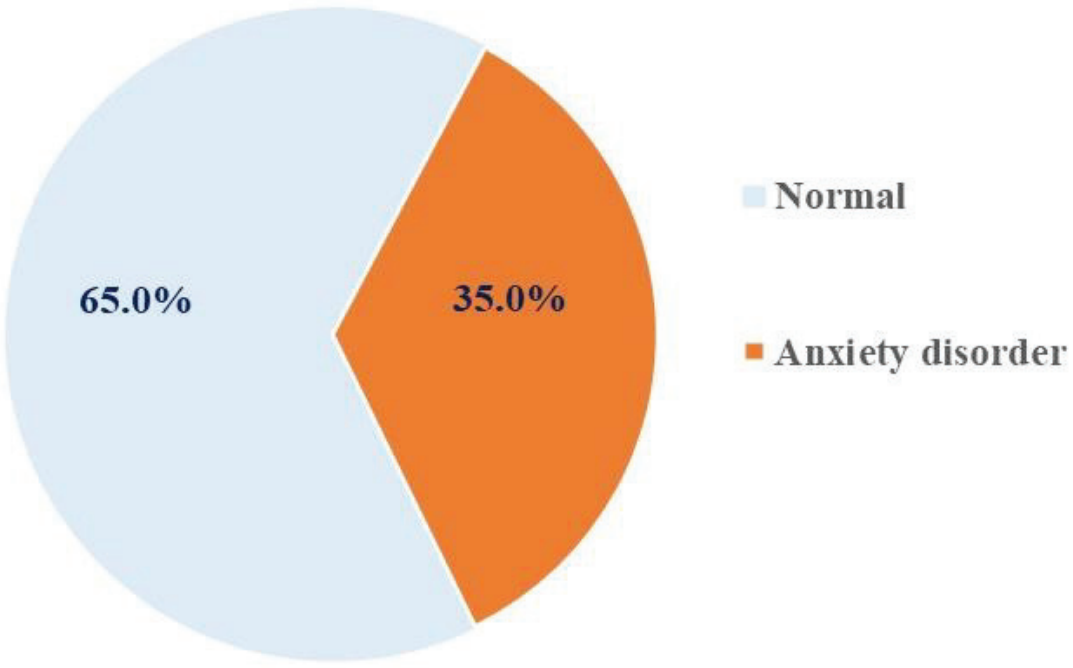

Figure 1. The rate of patients with anxiety disorders.

3. The relationship between anxiety disorders and some clinical characteristics

Table 2. The relationship between anxiety disorder assessment scores and some clinical characteristics.

Characteristics	Units	n	Anxiety score Median (IQ1, IQ3)	p
Gender	Male	25	2.0 (0.5, 5.0)	0.855
	Female	35	2.0 (1.0, 5.0)	
Medical history	Yes	36	2.0 (0.0, 4.75)	0.153
	No	24	2.0 (1.0, 5.75)	
BMI	< 18.5 kg/m^2	7	1.0 (0.0, 5.0)	
	18.5 - 24.9 kg/m^2	49	2.0 (1.0, 5.0)	0.305#
	> 24.9 kg/m^2	4	0.5 (0.0, 2.5)	0.527#
Causes of chronic kidney disease	Chronic glomerulonephritis	37	2.0 (0.0, 7.0)	0.969
	Others	23	2.0 (1.0, 5.0)	
Duration of hemodialysis	< 12 months	32	2.0 (0.25, 5.0)	
	12 - 24 months	9	3.0 (0.5, 5.5)	0.385*
	> 24 months	19	5.0 (1.0, 10.0)	0.048*

*(n: Number of subjects; SD: Standard deviation; *: As compared with group of patients with duration of hemodialysis < 12 months; #: As compared with group of patients with BMI < 18.5 kg/m^2)*

Table 2 shows the relationship between the anxiety disorder score using the DASS-21 and some clinical characteristics of the patients. The results showed that the group with hemodialysis duration over 24 months had a statistically significant increase in the anxiety disorder score as compared with the group with hemodialysis duration under 12 months ($p < 0.05$). No significant difference was observed in the anxiety disorder score between the groups with differences in gender, comorbidities, BMI, cause of renal failure (*Table 2*) or hypertension,

anemia (unpublished data).

4. The correlation between anxiety disorder assessment scores with age and some paraclinical indicators

Table 3. The correlation between anxiety disorder scores with age and some paraclinical indicators.

Indices	Anxiety score	
	r	p
Age	-0.346	0.045
Glucose (mmol/L)	-0.196	0.133
Ure (mmol/L)	0.124	0.345
Creatinine (umol/L)	0.006	0.961
Acid uric (umol/L)	-0.033	0.804
GOT (UI/L)	-0.070	0.594
GPT (UI/L)	-0.128	0.331
NA + (mmol/L)	0.196	0.134
K + (mmol/L)	0.009	0.946
Cl- (mmol/L)	0.112	0.393
Ca ++ (mmol/L)	0.101	0.441
Albumin (g/L)	0.059	0.654
Protein (g/L)	-0.033	0.800
Cholesterol (mmol/L)	-0.018	0.894
Triglyceride (mmol/L)	-0.094	0.474

Table 3 shows the correlation between anxiety disorder assessment scores with age and biochemical indices. The results showed that age had a negative and significant correlation with anxiety disorder scores (r = -0.346, p = 0.045). There was no significant correlation between biochemical indices with anxiety disorder

assessment scores ($p > 0.05$).

DISCUSSION

The present study was carried out on 60 patients undergoing maintenance hemodialysis from August 2023 to May 2024 and used the DASS-21 to assess anxiety disorders. The rate of anxiety disorders in 60 hemodialysis patients was 35.0%. In a previous study, Nguyen Thi Quynh Van et al. reported that the rate of hemodialysis patients with anxiety disorders at Bach Mai Hospital was 40.4% [3]. The results of the study by Nguyen Thi Quynh Van and our study are higher than those in the study reported by Luong Cong Minh et al. The author reported the rate of anxiety disorders in hemodialysis patients at Nguyen Tri Phuong Hospital was 5.9% [4]. However, our study result is similar to reports from previous studies worldwide. Lee et al. collected 208 patients with chronic kidney disease and reported that the rate of patients with anxiety disorders in patients with stage 5 chronic kidney disease was 34.3% [6]. In a meta-analysis of 6 studies with a total of 578 patients with maintenance hemodialysis, Murtagh et al. reported that the rate of anxiety disorders ranged from 12% to 52%, with an average rate of 38% [1]. Moreover, in a meta-analysis of 87 studies from 44 countries worldwide, Baxter et al. reported that the rate of anxiety disorders in the general population was 7.3% [7].

These findings indicate that the rate of anxiety disorders in hemodialysis patients in our study is about 5 times higher than those in the general population, suggesting, increased neuropsychiatric disorders, especially anxiety disorders might be found in hemodialysis patients.

Previous studies have also shown the important role of the “brain-renal axis” in an increase in anxiety disorders, which are thought to be related to secondary inflammation due to increased urea levels, oxidative stress due to increased cytokine production, and microvascular damage in the brain [8]. In addition, patients with hemodialysis also face stressors such as treatment costs, adherence to complex medication regimens, diet/fluid intake, and management of related complications. This increases the risks of increasing anxiety disorders or other psychiatric disorders such as depression.

Investigating factors related to anxiety disorders, we found that age was negatively and statistically significantly correlated with anxiety disorder scores. This result indicates an increased risk of anxiety disorders in young patients with

hemodialysis. According to a study by the American Psychiatric Association, stress about work, finances, and family are the most common causes of increased anxiety rates in young people compared to middle-aged and elderly people [9]. However, these factors were not measured in our study. Therefore, further studies, including social factors, are necessary to investigate anxiety disorders in hemodialysis patients.

In addition, prolonged dialysis time associated with increased anxiety disorders was also observed in our study. This result is similar to the study by Qawaqzeh et al. [10]. Central nervous system complications due to hyperuremia are thought to be related to the accumulation of urea metabolites in the blood. The longer the dialysis time, the more urea metabolites accumulate in the blood and the central nervous system, leading to effects on the patient's neuropsychiatric condition. Therefore, early control, detection, and treatment of increased blood urea concentration play a very important role in the management of hemodialysis patients.

However, as our study was conducted with a small sample size, some other factors, such as family income or fatigue level, which have an impact on anxiety disorders in patients with maintenance hemodialysis, were reported in previous studies [10] but were not evaluated in our study.

CONCLUSION

The rate of anxiety disorders in patients with maintenance hemodialysis is 35.0%. Age and duration of hemodialysis are factors affecting the expression of anxiety disorders in patients with maintenance hemodialysis.

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DEPRESSIVE SYMPTOMS AND GERIATRIC CHARACTERISTICS IN DEMENTIA PATIENTS AT NATIONAL GERIATRIC HOSPITAL

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Abstract:

Objectives: To determine the prevalence of depression symptoms and geriatric characteristics of dementia patients. ***Methods:*** A cross-sectional study was conducted on 87 dementia patients ≥ 60 years old at the Outpatient Department, National Geriatric Hospital. Data were collected using designed tools, including general information, Health-related Quality of Life (HRQoL), Instrumental Activities of Daily Living (IADLs), Activities of Daily Living (ADLs), Pittsburgh Sleep Quality Index (PSQI), Mini Nutrition Assessment Short Form (MNA-SF). Depressive symptoms were diagnosed by the Patient Health Questionnaire 9 (PHQ-9). ***Results:*** A total number of 87 patients were recruited for the study. The mean age was 76.84 years old. Moderate dementia had the highest rate with 43.7%. The remaining levels of mild dementia and severe dementia were 34.5% and 20.7%, respectively. The prevalence of having depressive symptoms was 43.7%. The symptom that occurred the most on all days was "Trouble falling or staying asleep or sleeping too much" at 32.2%. Low quality of life accounted for the highest number of 38 people (43.7%). ***Conclusion:*** The prevalence of having depressive symptoms in dementia patients was respectively high at 43.7%. Early screening for depressive symptoms of dementia patients is essential and should be considered for application in comprehensive geriatric assessment.

Keywords: Depressive symptom; Dementia patient; Geriatric characteristic.

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INTRODUCTION

In the current era of economic growth and improved healthcare, life expectancy is increasing, leading to an increasing proportion of elderly people in many countries around the world, including Vietnam. However, the elderly often suffer from many chronic diseases such as cardiovascular disease, diabetes, bone and joint diseases, etc [1]. Dementia is one of the most severe and common mental disorders among them. The prevalence of the disease increases with age. The prevalence of dementia increases sharply with age from just > 1% between the ages of 65 - 69 years old up to > 30% beyond the age of 90 years old [2]. While dementia is associated with cognitive changes, behavioral changes such as depression also frequently occur, with up to 20% of individuals reporting some degree of clinically significant depressive symptoms [3, 4]. Depressive symptoms can have adverse consequences for patients and their caregivers [5]; thus, a clear understanding of the prevalence of depression in dementia is warranted. It is important to investigate dementia at an early stage to identify any curable conditions, deploy the proper medical treatment, and provide appropriate support and assistance to patients with dementia and their relatives. In Vietnam, so far, there have been a number of studies on dementia; however, the clinical features of depression in dementia patients have not been fully explored. The goal of our study was to conduct a systematic review and meta-analysis to determine the prevalence of depression in dementia among older adults in studies that used validated criteria for the diagnosis of both depression and dementia. This information may help us better understand the overall burden of depression in dementia and the factors associated with depression in dementia. Studying this issue may help doctors diagnose and treat depression in people with dementia, improving the quality of their lives as well as the lives of their caregivers. Therefore, the study is conducted to: *Determine the prevalence of depression symptoms in dementia and the geriatric characteristics of dementia patients.*

MATERIALS AND METHODS

1. Subjects

Including 87 participants ≥ 60 years old who were diagnosed with dementia at the National Geriatrics Hospital.

* *Inclusion criteria:* Older people have a diagnosis of dementia according to DSM V criteria; patients have physical and cognitive abilities to do a face-to-face interview; patients and their families agree to participate.

* *Exclusion criteria:* Acute and malignant diseases (advanced cancers, end-stage chronic diseases, acute myocardial infarction, stroke); symptomatic cardiovascular disease, coronary revascularization within 1 year; clinical evidence of schizophrenia, severe depression, psychiatric or bipolar disorder (according to DSM-IV TR criteria); alcoholism or substance dependence (according to DSM-5 criteria) currently, or within the past 2 years; severe loss of vision, hearing or communicative ability (according to the interRAI Community Health Assessment); participant or family unwilling to participate in the study.

* *Setting and time:* In the Outpatient Department at National Geriatric Hospital, from July to November 2021.

2. Methods

* *Study design:* A cross-sectional study.

* *Sample size and sampling:*

Sampling: Convenience sampling.

Sample size: The proposed sample size was 87 participants.

* *Tools and data collection method:* Data were collected using designed tools, including general information, PHQ-9 for assessing depressive symptoms, HRQoL, IADLs, ADLs, PSQI, and MNA-SF.

- Mini Mental State Evaluation (MMSE);

- Depression: PHQ-9:

+ Performing: PHQ-9 is one of the tools used to screen for the presence and severity of depression and to monitor response to treatment [6].

+ Evaluating: Interpretation of Total Score Depression Severity: PHQ-9 is a 27-point questionnaire with a cut-off point of 10 (having depressive symptoms).

1 - 4: Minimal depression;

5 - 9: Mild depression;

10 - 14: Moderate depression;

15 - 19: Moderately severe depression;

20 - 27: Severe depression.

* *Data processing and analysis:* The process of data coding, entry into Redcap, and analysis was done using SPSS.22. Descriptive statistics were adopted to examine characteristic data: Frequency, percentage, and mean. Inferential statistics was done to perform comparisons between groups: Chi-square andmultivariable regression. Statistical significance was accepted at the 95% confidence level ($p < 0.05$).

3. Ethics

The study tools did not involve sensitive or intimate problems and did not affect the subjects' emotions. National Geriatric Hospital granted permission for the use and publication of the research data. The results of the study were proposed to improve the health of the community, not for other purposes. The authors declare to have no conflicts of interest in the study.

RESULTS

A total number of 87 patients were recruited for the study from July to November 2021. Demographic characteristics are presented in detail below.

Table 1. Demographic characteristics (n = 87).

Demographic characteristics	Frequency (n)	Percentage (%)
Aged group		
60 - 69	18	20.7
70 - 79	36	41.4
≥ 80	33	37.9
Gender		
Female	57	65.5
Male	30	34.5
Marital status		
Married	66	75.9
Single/widowed/divorced	21	24.1
Educational level		
Have not graduated from high school	52	59.8
Graduated from high school	28	32.2
Graduated from university and above	7	8

Patients were ≥ 60 years old, with a mean age of 76.84 years old, in which the most significant distribution was generated by patients aged between 70 - 79 (41.4%). Secondly, patients ≥ 80 years old accounted for 32.1%. Patients from 60 - 69 represented only 20.7%. Females accounted for 65.5% (n = 57), which was higher than male patients (34.5%, n = 30). The female/male ratio was 1.9. Patients who have not graduated from high school accounted for the highest proportion (59.8%), followed by those who graduated from high school (32.2%). Patients who graduated from university and above accounted for the lowest percentage (8%).

Table 2. Characteristic of dementia (n = 87).

Characteristics of dementia	Classification	Frequency (n)	Percentage (%)
The severity of dementia	Mild	30	34.5
	Moderate	38	43.7
	Severe	18	20.7
Time of appearance memory loss	≤ 1 year	33	37.9
	> 1 year	54	62.1
Previously diagnosed with dementia	Yes	34	39.5
	No	52	60.5

Moderate dementia has the highest rate (43.7%). The remaining levels of mild dementia and severe dementia were 34.5% and 20.7%, respectively. 54 patients appeared to have memory loss > 1 year (62.1%), and the remaining 32 patients had dementia ≤ 1 year (37.9%). But only 34 patients (39.5%) were previously diagnosed, the rest (60.5%) were undiagnosed. The depressive symptoms prevalence in mild dementia was 29.7%, in moderate dementia was 43.3%, and in severe dementia was 10%.

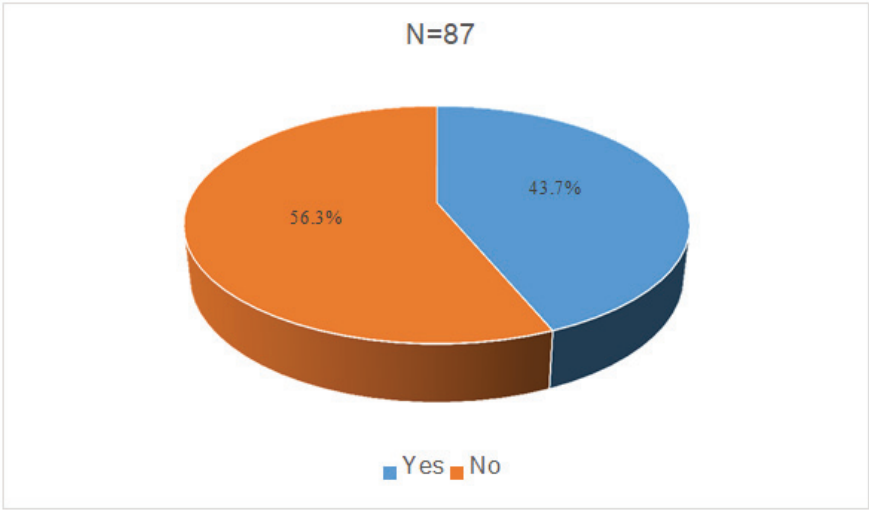

Figure 1. The prevalence of depressive symptoms by PHQ-9 (n = 87).

According to the PHQ-9 questionnaire, the prevalence of having depressive symptoms was 43.7% (*Figure 1*).

Table 3. PHQ-9 elements distribution (n = 87).

Signs and symptoms	Frequency, n (%)			
	Not at all	Several days	More than half the days	Nearly everyday
Little interest or pleasure in doing things	14 (16.1)	37 (42.5)	27 (31.0)	9 (10.3)
Feeling down, depressed, or hopeless	23 (26.4)	41 (47.1)	15 (17.2)	8 (9.2)
Trouble falling or staying asleep or sleeping too much	18 (20.7)	26 (29.9)	15 (17.2)	28 (32.2)
Feeling tired or having little energy	9 (10.3)	34 (39.1)	27 (31.0)	17 (19.5)
Poor appetite or overeating	37 (42.5)	25 (28.7)	16 (18.4)	9 (10.3)
Feeling bad about yourself or that you are a failure or have let yourself or your family down	52 (59.8)	23 (26.4)	10 (11.5)	2 (2.3)
Trouble concentrating on things, such as reading the newspaper or watching television	18 (20.7)	28 (32.2)	16 (18.4)	25 (28.7)
Moving or speaking so slowly that other people could have noticed. Or the opposite, being so fidgety or restless that you have been moving around a lot more than usual	43 (49.4)	25 (28.7)	13 (14.9)	6 (6.9)
Thoughts that you would be better off dead or hurting yourself	79 (90.8)	7 (8.0)	0 (0.0)	1 (1.1)

The symptom that occurred the most on all days was "Trouble falling or staying asleep or sleeping too much" (32.2%), followed by "Trouble diligently on things, such as reading the newspaper or watching television" (28.7%). Symptoms such as "Feeling down, depression, or hopeless" and "Feeling down, depressed, or hopeless" accounted for a high proportion with the frequency of “several days” in the last 2 weeks. The least common symptom in patients is "Thoughts that you would be better off dead, or of hurting yourself" with a very low frequency.

Table 4. Geriatric characteristics of older patients (n = 87).

Geriatric characteristics	Classification	Frequency (n)	Percentage (%)
HRQoL	High/very high	24	27.6
	Very low to moderate	63	72.4
IADLs	Dependent	84	96.6
	Independent	3	3.4
Physical ADLs	Severe dependency	29	33.3
	Moderate dependency	25	28.7
	Slight dependency	33	37.9
	Mean of Barthel Index ± SD: 73.85 ± 26.34		
PSQI	Good sleep	19	21.8
	Poor sleep	68	78.2
Nutritional status	Malnourished-risk of malnutrition	61	60.1
	Normal	26	29.9

Low quality of life accounted for the highest number (43.7%, n = 38). 39 patients (3.4%) were independent in IADLs. The majority (96.6%, n = 84) was dependent on one or more IADLs domains. 29 patients (33.3%) had a severe dependency on physical ADLs, 25 patients (28.7%) had a moderate dependency, and 33 patients (37.9%) had a slight dependency. Most of the patients did not have good sleep (78.2%, n = 68). The MNA-SF measured nutritional status: 26 patients (29.9%) had normal nutrition, 47 patients (54%) were identified as "at risk of malnutrition", and 14 patients (16.1%) were malnourished according to MNA-SF scores.

DISCUSSION

There were 87 participants in the study. The mean age was 76.84 ± 8.38 years, ranging from 60 - 96 years old. The 70 - 79 age group accounted for the highest proportion (41.1%), and aged ≥ 80 and 60 - 68 age groups accounted for 37.9% and 20.7%, respectively. This result was higher than the mean age in the study of CY Liu et al. in China [7], the mean age was 72.72 ± 8.66, ranging from 52 - 92 years. This distribution was similar to the study of Paula Andreasen et al. [8] in low- and middle-income countries, female and male participants accounted for 62.1% and 37.6%, respectively. Globally, the percentage of females with dementia

was higher than males, no matter which country, so the gender ratio in our study is similar to domestic and foreign studies.

The mean score of PHQ-9 was 9.64 ± 4.78, ranging from 1 - 20, and the prevalence of depression in elderly patients was 43.7%. In particular, mild depression was 34.5%, moderate depression accounted for 43.7%, and severe depression was 20.7%. There were 56.3% of participants without depression. This result was higher than other reported rates of 17 - 31% for major depression among Alzheimer's disease patients in the study of Liu et al. (16% of the patients had a depressive disorder); Weiner et al. [9] (1.5% major depression). But this ratio is lower than the study of Migliorelli et al. (1995), the prevalence of depression among individuals with Alzheimer's disease was 51% [10].

With 87 participants, we found that 35 patients (40.2%) had mild depression, followed by moderate depression (27.6%, n = 24) and moderate to severe depression (13.8%, n = 12). And the lowest was severe depression (2.3%, n = 2). The most frequent manifestations of depression were "Being sadness or depressed" (94.5%, n = 35) and "Putting him/herself down or saying like a failure" (32.4%, n = 12). Depression manifestation with the least frequency was "Saying acting as sad or low spirit" (0%, n = 0).

There were several limitations in the research. First, the results could be generalized only to the study area due to the power of the sample size calculation (due to the COVID-19 epidemic, the number of patients was limited). Second, our research was a cross-sectional study; therefore, we were not able to clarify the causal relationship between depression in dementia and some factors.

CONCLUSION

The prevalence of having depressive symptoms in dementia patients was respectively high (43.7%). Early screening for depressive symptoms in dementia patients is essential and should be considered for application in comprehensive geriatric assessment.

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QUALITY OF LIFE AMONG ELDERLY PATIENTS WITH HEART FAILURE AT THONG NHAT HOSPITAL IN 2023

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Abstract

Objectives: To evaluate the quality of life (QoL) among elderly patients with heart failure (HF) treated at Thong Nhat Hospital in 2023. ***Methods:*** A cross-sectional descriptive study was conducted to assess the QoL of 300 elderly inpatients (≥ 60 years old) diagnosed with HF from January 1, 2023 to December 31, 2023. Data were collected using the SF-36 and Kansas City Cardiomyopathy Questionnaire (KCCQ) tools. ***Results:*** 55.33% of patients reported moderate-to-poor overall health, with a mean physical health score of 23.51 ± 19.61 (62% poor, 27.33% moderate-to-poor). Mental health scores were comparatively higher, averaging 57.84 ± 10.84, with 67.67% reporting moderate-to-good outcomes. Marital status, education level, disease duration, and comorbidities (atrial fibrillation) significantly influenced the maintenance of a good QoL. ***Conclusion:*** Maintaining a good QoL among elderly HF patients was predominantly moderate-to-poor, especially concerning physical health. Factors such as marital status, educational attainment, duration of illness, and comorbidities were significant determinants of maintaining a good QoL.

Keywords: Quality of life; Elderly; Heart failure.

INTRODUCTION

Maintaining a good QoL is critical for patients with chronic progressive illnesses, particularly HF. QoL in HF patients is markedly reduced compared to healthy individuals and those with other chronic conditions [1]. QoL reflects the impact of clinical symptoms and treatment modalities on patients' daily lives. HF symptoms such as dyspnea,

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chest pain, fatigue, edema, and insomnia often restrict physical and social activities, leading to diminished QoL [2]. Poor QoL is associated with increased hospitalization and mortality rates [3]. QoL is also a sensitive measure for evaluating intervention effectiveness and an independent predictor of survival in cardiovascular disease patients. Therefore, comprehensive QoL assessments are essential for tailoring appropriate interventions for HF patients. However, research on QoL among Vietnamese HF patients, especially the elderly, remains limited. This study aims to: *Address this gap by evaluating the QoL of elderly HF patients at Thong Nhat Hospital in Ho Chi Minh City in 2023.*

MATERIALS AND METHODS

1. Subjects

Including 300 elderly patients (≥ 60 years) diagnosed with HF and admitted to Thong Nhat Hospital from January 1, 2023 to December 31, 2023.

* *Inclusion criteria:* Inpatients ≥ 60 years old; diagnosed with HF; voluntarily participated in the study.

* *Exclusion criteria:* Acute HF diagnosis; declined participation.

2. Methods

* *Study design:* A cross-sectional descriptive study.

* *Sample size:*

The sample size formula for the descriptive study was used:

$$n = Z^2_{1-a/2} \times \frac{(1-p)}{p \times \varepsilon 2} \times DE$$

n: Sample size; $Z_{1-\alpha/2}$: Confidence coefficient with statistical significance level $\alpha = 0.05$, corresponding to 95% confidence level then $Z_{1-\alpha/2} = 1.96$; DE (Design effect): Design coefficient, choose DE = 2.0; p: Estimate the percentage of elderly people with HF receiving inpatient treatment at Thong Nhat Hospital, choose p = 0.198 according to the study of Tran Song Giang [4]; ε: Relative error, ε = 0.1.

The minimum sample size calculated was n = 189; in fact, the study was conducted on 300 elderly patients with HF who were hospitalized at Thong Nhat Hospital in 2023.

* *Data collection tools:*

SF-36: A widely used health-related QoL scale consisting of 36 questions evaluating physical health (physical functioning, role limitations due to physical health, pain), mental health (vitality, social functioning, role limitations due to emotional problems, mental health), and overall health [4]. Scores range from 0 to 100, with higher scores indicating better QoL. The level of assessment was defined as follows:

Poor (from 0 - 25); medium - poor (from 26 - 50); medium - good (from 51 - 75); good - very good (from 76 - 100).

KCCQ: A 15-item questionnaire assessing physical and psychological health, social relationships, and living environment. Scores are standardized from 0 (poorest) to 100 (best) [5].

* *Data analysis:* Data were entered and analyzed using SPSS 22.0.

3. Ethics

The research was conducted according to Decision No. 196/QĐ-BKHCN dated February 2, 2021, issued by the Ministry of Science and Technology. Thong Nhat Hospital granted permission for the use and publication of the research data. The authors hereby declare that there are no conflicts of interest in this research.

RESULTS

Table 1. Demographic and clinical characteristics of elderly HF patients (n = 300).

General characteristics		Mean
Age ($\overline{X}$ ± SD)		75.95 ± 9.33
Gender, n (%)	Female	138 (46.0%)
	Male	162 (54.0%)
Marital status, n (%)	Currently married	198 (66.0%)
	Single/widowed	102 (34.0%)
Duration of illness ($\overline{X}$ ± SD) (month)		70.08 ± 127.65
Educational attainment n (%)	High school or above	97 (32.33%)
	Below high school	203 (67.67%)
NYHA HF classification, n (%)	I	92 (30.67%)
	II	49 (16.33%)
	III	128 (42.67%)
	IV	31 (10.33%)
Atrial fibrillation n (%)	Yes	108 (36.0%)
	No	192 (64.0%)

(NYHA: New York Heart Association)

The mean age of the study population was 75.95 ± 9.33 years, with a female-to-male ratio of approximately 1:1.17. The mean duration of illness was 70.08 ±

127.65 months. Among the 300 patients, the proportions of the New York Heart Association (NYHA) class I, II, III, and IV were 30.67%, 16.33%, 42.67%, and 10.33%, respectively. Atrial fibrillation was observed in 36% of HF patients.

Table 2. Mean QoL scores (SF-36) of elderly patients with HF.

Domains	$\overline{X} \pm SD$	Min	Max
Physical health	23.51 ± 19.61	0.0	74.05
Mental health	57.84 ± 10.84	29.29	78.57
Overall health	36.90 ± 14.60	17.36	75.28

The mean physical health score of chronic HF patients was 23.51 ± 19.61 points, ranging from 0 - 74.05 points. For mental health, the mean score was 57.84 ± 10.84 points, ranging from 29.29 to 78.57 points. The mean overall health score of the study population, calculated based on physical and mental health scores, was 36.90 ± 14.60 points.

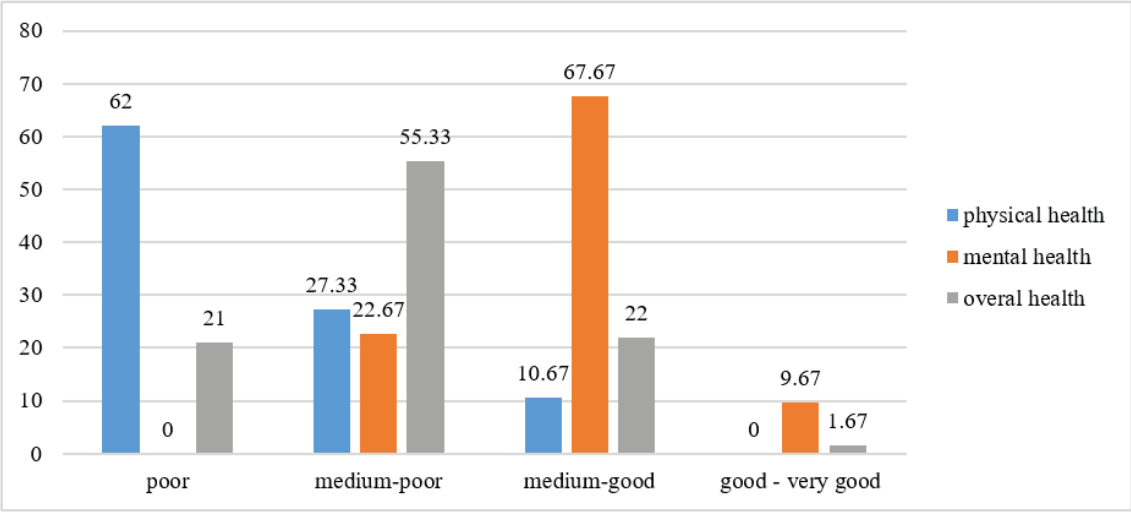

Figure 1. Classification of QoL scores among elderly patients with HF.

Regarding physical health scores, the majority of patients in the study had poor scores (62%) and moderate-to-poor scores (27.33%). None of the patients achieved scores classified as good or very good in physical health. Mental health scores

were higher; specifically, no patients were classified as having poor mental health scores. The proportions of patients with moderate-to-poor, moderate-to-good, and good-to-very-good mental health scores were 22.67%, 67.67%, and 9.67%, respectively. As for overall health scores, 55.33% of patients were in the moderate-to-poor category, 22% in the moderate-to-good category, and 21% were classified as poor.

Table 3. The correlation between mean QoL scores (SF-36) and demographic characteristics of elderly patients with HF (n = 300).

Variables		Physical health		Mental health		Overall health	
		$\bar{X} \pm SD$	p	$\bar{X} \pm SD$	p	$\bar{X} \pm SD$	p
Gender	Male (162)	22.37 ± 18.62	> 0.05	57.28 ± 10.51	> 0.05	36.00 ± 14.07	> 0.05
	Female (138)	24.86 ± 20.70		58.50 ± 11.22		37.95 ± 15.19	
Age	60 - 69 (86)	21.08 ± 18.45	> 0.05	57.93 ± 9.98	> 0.05	35.44 ± 13.92	> 0.05
	70 - 79 (92)	23.80 ± 19.89		57.80 ± 11.87		37.07 ± 14.99	
	≥ 80 (122)	25.02 ± 20.19		57.81 ± 10.70		37.80 ± 14.81	
Marital status	Currently married (198)	30.80 ± 19.63	< 0.05	59.02 ± 11.92	< 0.05	41.82 ± 15.06	< 0.05
	Single/widowed (102)	9.37 ± 9.01		55.56 ± 7.93		27.35 ± 7.02	
Educational attainment	High school or above (197)	29.08 ± 18.63	< 0.05	58.54 ± 10.48	> 0.05	40.64 ± 14.30	< 0.05
	Below high school (203)	20.85 ± 19.56		57.51 ± 11.02		35.11 ± 14.44	

The physical health and overall health scores of elderly HF patients living with a spouse and having an educational attainment of high school or above were significantly higher compared to those who were single/widowed and had an educational attainment below high school ($p < 0.05$). Regarding mental health scores, HF patients living with a spouse also exhibited significantly higher QoL scores compared to single/widowed patients ($p < 0.05$).

Table 4. The correlation between mean QoL scores (SF-36) and clinical characteristics of elderly patients with HF (n = 300).

Variables		n	Physical health		Mental health		Overall health	
			$\bar{X} \pm SD$	p	$\bar{X} \pm SD$	p	$\bar{X} \pm SD$	p
NYHA HF classification	I	92	22.93 ± 19.51	> 0.05	57.19 ± 8.88	> 0.05	36.30 ± 14.01	> 0.05
	II	49	22.81 ± 20.24		54.93 ± 11.25		35.20 ± 14.89	
	III	128	22.84 ± 19.77		58.47 ± 11.36		36.75 ± 14.84	
	IV	31	29.11 ± 18.25		61.80 ± 12.26		41.95 ± 14.51	
Duration of illness (year)	< 1	159	28.50 ± 21.82	< 0.05	60.05 ± 12.14	< 0.05	40.78 ± 16.34	< 0.05
	1 - < 5	47	18.56 ± 15.63		57.50 ± 6.95		33.87 ± 10.44	
	≥ 5	94	17.56 ± 14.72		54.27 ± 9.08		31.85 ± 11.05	
Atrial fibrillation	Yes	108	30.06 ± 20.61	< 0.05	59.03 ± 9.53	> 0.05	41.32 ± 15.56	< 0.05
	No	192	19.83 ± 18.07		57.17 ± 11.49		34.41 ± 13.45	

Elderly patients with a longer duration of HF exhibited progressively lower scores in physical health, mental health, and overall health. Patients with a disease duration of less than 1 year had significantly higher overall health scores compared to those with a duration of 1 to < 5 years and ≥ 5 years, with mean scores of 40.78 ± 16.34, 33.87 ± 10.44, and 31.85 ± 11.05, respectively ($p < 0.05$). HF patients with atrial fibrillation had significantly lower physical health and overall health scores compared to those without atrial fibrillation ($p < 0.05$).

Table 5. QoL of HF patients classified by general characteristics.

Variables		n	KCCQ scale		SF-36 scale	
			Score of KCCQ (X ± SD)	p	Score of SF-36 (X ± SD)	p
Age	60 - 69	86	38.78 ± 21.14	> 0.05	35.44 ± 13.92	> 0.05
	70 - 79	92	40.58 ± 20.32		37.07 ± 14.99	
	≥ 80	122	41.84 ± 20.20		37.80 ± 14.81	
Gender	Male	162	39.83 ± 20.22	> 0.05	36.00 ± 14.07	> 0.05
	Female	138	41.46 ± 20.82		37.95 ± 15.19	
Marital status	No	102	28.51 ± 14.73	< 0.01	27.35 ± 7.02	< 0.01
	Yes	198	46.79 ± 20.27		41.82 ± 15.06	
Educational attainment	High school or above	203	37.46 ± 19.19	< 0.01	35.11 ± 14.44	< 0.01
	Below high school	97	47.10 ± 21.62		40.64 ± 14.30	
NYHA HF classification	I - II	141	38.55 ± 19.63	> 0.05	35.92 ± 14.28	> 0.05
	III - IV	159	42.38 ± 21.10		37.77 ± 14.88	

Using both the KCCQ and SF-36 scoring systems, QoL was significantly higher among patients living with a spouse (KCCQ: 46.79 ± 20.27; SF-36: 41.82 ± 15.06) and those with an educational attainment of high school or above (KCCQ: 47.10 ± 21.62; SF-36: 40.64 ± 14.30) compared to single/widowed patients (KCCQ: 28.51 ± 14.73; SF-36: 27.35 ± 7.02) and those with educational attainment below high school (KCCQ: 37.46 ± 19.19; SF-36: 35.11 ± 14.44) ($p < 0.05$). There were no statistically significant differences in QoL scores between age groups, genders, or NYHA HF classifications on either the KCCQ or SF-36 scales.

DISCUSSION

Our study results indicate that the mean overall health score of 300 chronic HF patients, as measured by the SF-36 questionnaire, was 36.90 ± 14.60. The mean physical health score was 23.51 ± 19.61, lower than the mean mental health score of 57.84 ± 10.84. Regarding QoL classification, the majority of HF patients exhibited poor (62%) or moderate-to-poor (27.33%) physical health. Mental health scores were higher, with no patients classified as poor; most were classified as moderate-to-good (67.67%). As for overall health scores, 55.33% of patients were categorized as moderate-to-poor. These results highlight the significant impact of HF on QoL, with generally low scores across domains. This finding aligns with the study by Ghuloom and Sanad (2022), which reported that among 250 HF patients, 74.8% had poor QoL, 21.6% had moderate QoL, and 3.6% had good QoL [6].

The QoL scores in our study are lower compared to some other national and international studies. For example, Truong Phi Hung (2023) reported reduced QoL across all physical and mental health domains, with median scores of 44.3 (30.5 - 52.1) and 46.9 (32.1 - 58.8), respectively [7]. The COACH study (2012) demonstrated reduced QoL among HF patients across most domains, except for pain perception [8]. Similarly, Chatzinikolaou et al. (2021), using the SF-36 scale, found a mean overall health score of 45.69 ± 21.75, a physical functioning score of 64.75 ± 33.72, and an emotional health score of 50.95 ± 18.67 [9]. Our findings indicate more severe physical health impairment compared to these studies, though mental health scores were comparatively higher, possibly reflecting patient sample characteristics or cultural and social factors.

We observed that elderly HF patients living with a spouse (41.82 ± 15.06) and those with an educational attainment of high school or above (40.64 ± 14.30) had significantly higher QoL scores compared to those who were single/ widowed (27.35 ± 7.02) or had an educational attainment below high school (35.11 ± 14.44) ($p < 0.05$). No statistically significant differences in QoL were found between age groups, genders, or NYHA classifications across both the KCCQ and SF-36 scales. The SF-36 questionnaire is a very popular instrument for evaluating health-related QoL [4]. The KCCQ has been widely recognized for its sensitivity and specificity in measuring the health status of HF patients [5]. These findings underscore the critical role of familial support and care in improving the QoL of HF patients. Higher educational attainment may be associated with better disease understanding, treatment adherence, and access to healthcare services [6].

CONCLUSION

The study conducted on 300 elderly HF patients hospitalized at Thong Nhat Hospital in 2023 revealed that 55.33% of patients had moderate-to-poor overall health scores. Physical health scores were notably low (23.51 ± 19.61), with the majority classified as poor (62%) or moderate-to-poor (27.33%). Mental health scores were relatively better (57.84 ± 10.84), with 67.67% classified as moderate-to-good. Marital status, educational attainment, duration of illness, and comorbidities such as atrial fibrillation were significant factors influencing the QoL of elderly HF patients.

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A CASE REPORT: HER2-TARGETED THERAPY IN COLORECTAL CANCER

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Abstracts

Colorectal cancer (CRC) is a malignant tumor arising from the inner lining of the colon or rectum and is the third most common cancer and the third leading cause of cancer-related deaths in the United States. Human epidermal growth factor receptor 2 (HER2) gene overexpressed or amplified CRC has shown treatment responses with HER2-targeted therapies. This article reports two cases of heavily pretreated metastatic CRC (mCRC) with HER2 overexpression who achieved a remarkable clinical response to trastuzumab plus pertuzumab.

Keywords: Human epidermal growth factor receptor 2 (HER2); Anti-HER2 therapy; Trastuzumab plus pertuzumab; Colorectal cancer.

INTRODUCTION

Colorectal cancer constitutes 10% of global cancer diagnoses and cancer-related deaths annually. At the time of diagnosis, 20% of patients have mCRC, with a 5-year survival rate of less than 20%. Recent advancements in genomic technology with large-scale molecular profiling of tumors have led to new treatment opportunities for these patients.

In approximately 2 - 5% of patients with CRC, overexpression or amplification of HER2 is observed, with a higher incidence in left-sided colon and primary rectal RAS/BRAF-wild-type (RAS/BRAFwt) tumors [1, 2, 3]. Currently, patients with left-sided RAS/BRAFwt tumors are treated with anti-EGFR therapy (i.e., panitumumab or cetuximab) with or without combination chemotherapy and/or anti-VEGF.

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However, accumulating evidence suggests that the presence of HER2 amplification or overexpression is associated with resistance to anti-EGFR therapy [4]. ERBB2 is a well-known oncogene that is successfully targeted in breast, gastric, and esophageal cancers. Recent reports indicate that patients with HER2-positive mCRC (HER2+mCRC) also benefit from anti-HER2 therapy. Despite promising data, targeted therapy for patients with HER2+mCRC is not available in Vietnam. This is mainly due to the challenges of obtaining regulatory approval based on single-arm studies in small subgroups of patients, such as those with HER2+mCRC. However, there is a clear unmet need for the availability of anti-HER2 agents as a standard treatment option for these patients. Therefore, this study aims to: *Report two HER2 overexpression CRC cases responding significantly to trastuzumab plus pertuzumab.*

CASE REPORT

1. Case report 1

We present the case of a 59-year-old man with a history of rectal adenocarcinoma in March 2017. He underwent low anterior resection, with final pathology revealing stage IIIB (T3, N2a, M0 American Joint Committee on Cancer 7th edition). He finished 5 months of adjuvant chemotherapy with mFOLFOX6 regimen and stopped due to anaphylaxis with Oxaliplatin. In July 2019, peritoneal metastasis was found in imaging. The molecular profile revealed RAS and BRAF wide-type, HER2 amplification (3+) in immunohistochemistry (IHC), and there was no mutation found by next-generation sequencing on the tumor specimen. FOLFIRI plus bevacizumab was recommended. He received 6 months of FOLFIRI plus bevacizumab, followed by irinotecan plus bevacizumab. Two years later, in September 2019, his disease progressed to the lungs. Because of positive HER2 amplification, treatment with trastuzumab combined with pertuzumab was initiated because this is the only anti-HER2 drug available in Vietnam. The patient tolerated the treatment well. A repeated CT scan after 12 weeks (about three months) of treatment showed a significant response (*Figure 2*). Treatment was continued and re-evaluation was done every three months. His disease remained responsive until August 2022, when the lung lesion progressed again (*Figure 4*). Subsequence therapy has continued until now. Dual anti-HER2 therapy improved his progression-free survival by 11 months, which was a remarkable benefit for second-line stage IV rectal cancer.

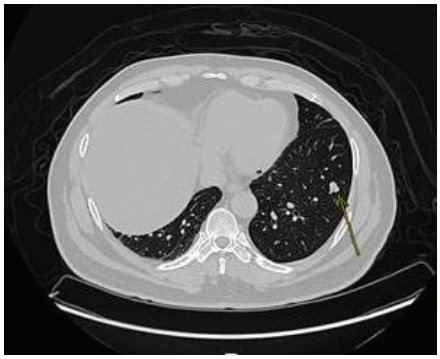

Figure 1. Lung metastasis.

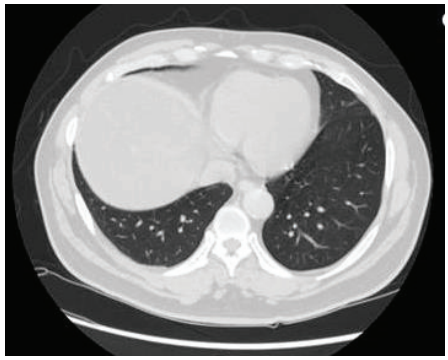

Figure 2. Lung lesion responded after 12 weeks of treatment.

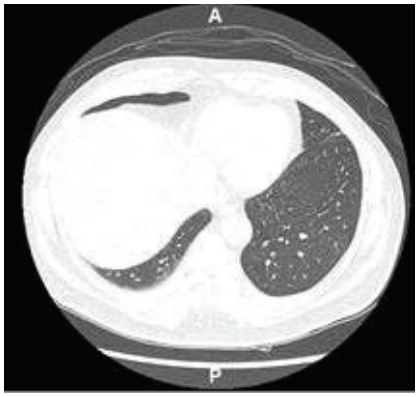

Figure 3. Response remained for 11 months.

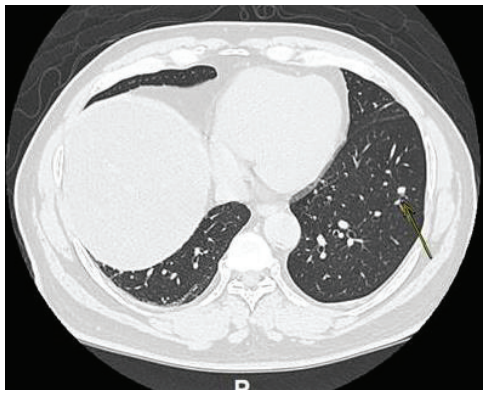

Figure 4. Lung lesion developed again.

2. Case report 2

A 65-year-old male presented in July 2023 with a bowel obstruction due to a mass in the hepatic flexure of the colon. The patient underwent an extended right hemicolectomy with lymph node dissection. Histology revealed moderately differentiated adenocarcinoma, with negative node metastasis, prominent lymphovascular, and perineural invasion. During the operation, multiple lesions in the liver were found. Finally, he was diagnosed with stage IV colon cancer. IHC revealed intact (proficient) DNA mismatch repair proteins (pMMR). The molecular profile revealed KRAS and BRAF wide-type, HER2 amplification (3+) in IHC. Bevacizumab plus mFOLFOX6 was initiated. After 6 cycles of chemotherapy, the patient achieved a partial response. However, he did not tolerate well with chemotherapy (nausea, loss of appetite, weight loss, etc.); the treatment was changed to 5-Fluorouracil (5-FU) combined with Bevacizumab. Five months later, there

was a progression of the disease as CT imaging showed growing hepatic lesions. Based on the tumor genetic profile and the fact that the patient had experienced poor tolerance to chemotherapy, dual HER2-targeted therapy with trastuzumab and pertuzumab was administered. The patient tolerated the treatment well. A repeated CT scan after 12 weeks (about three months) of treatment showed a partial response (*Figure 6*), and the CEA level decreased dramatically from 617 ng/mL to 60 ng/mL. The treatment was continued and re-evaluation was done every three months. His disease remained responsive for 7.5 months when hepatic lesions progressed again (*Figure 7*), and the CEA level increased to 1022 ng/mL. Dual anti-HER2 therapy improved his progression-free survival by 7.5 months, which was a remarkable benefit for second-line stage IV rectal cancer and especially for those who could not tolerate well with chemotherapy.

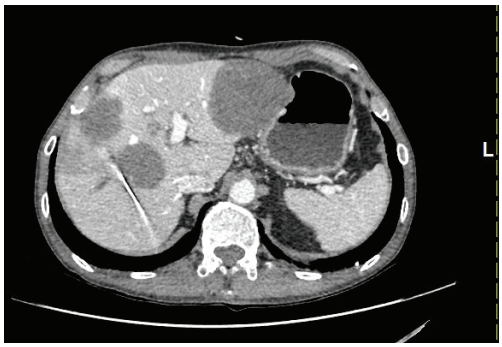

Figure 5. Multiple liver metastases.

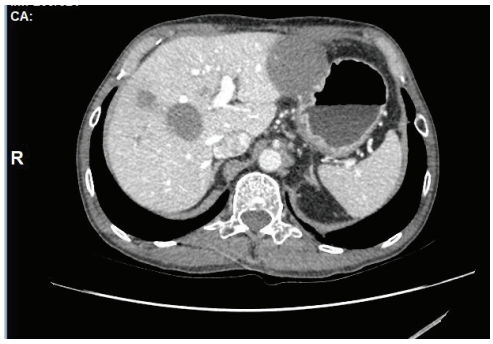

Figure 6. Partial response was achieved after 3 months.

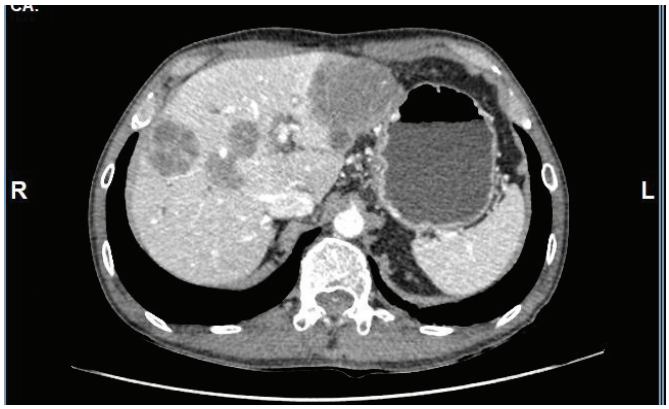

Figure 7. Progression was seen after 7.5 months.

DISCUSSION

HER2 amplification represents approximately 2% of all stage IV CRCs and is associated with resistance to EGFR-based treatment [5, 6]. HER2 overexpression is defined as ≥ 50% staining by IHC or ≥ 10% staining by IHC and positive amplification by fluorescent in situ hybridization according to HERACLES diagnostic criteria [7]. Amplification or overexpression of HER2 oncogene causes hyperactivation of mitogenic signals, even without ligand binding, thereby leading to uncontrolled cell proliferation and tumorigenesis [8].

While clinical studies on the combination of trastuzumab and pertuzumab in metastatic colon cancer are still limited, there is a growing body of evidence supporting the efficacy of HER2-targeted therapies in this context. Early-stage studies and retrospective analyses have demonstrated that a subset of mCRC patients with HER2 amplification or overexpression can benefit from treatment with HER2 inhibitors. However, the response rates in mCRC are often lower compared to breast cancer, underscoring the need for a more precise selection of patients who are likely to benefit.

The combination therapy of trastuzumab plus pertuzumab showed promising results for patients with treatment-refractory HER2-Amp mCRC in the single-arm, phase IIa MyPathway multibasket study and is listed in the National Comprehensive Cancer Network guidelines for HER2-Amp mCRC along with trastuzumab plus lapatinib or fam-trastuzumab deruxtecan-nxki as a category 2A recommendation. Trastuzumab plus pertuzumab demonstrated an objective response rate of 32% with a median OS of 11.5 months and a median progression-free survival of 2.9 months in MyPathway [9]. Similar efficacy was observed with trastuzumab plus pertuzumab in the multicenter phase II TRIUMPH study [10]. However, the clinical response is heterogeneous, and not all HER2-positive CRCs respond to these therapies. Factors such as tumor heterogeneity, mutation status, and co-expression of other biomarkers, such as HER3, EGFR, or PI3K, may influence treatment outcomes. This highlights the need for further studies to refine patient selection criteria and identify those who are most likely to benefit from dual HER2 blockade.

In our case, we present a patient diagnosed with HER2-positive metastatic colon cancer who was treated with a combination of trastuzumab and pertuzumab. The patient had previously failed standard chemotherapy regimens and was found to have HER2

overexpression, which guided the decision to pursue targeted therapy. Our patient experienced stabilization of the disease for several months, indicating that dual HER2 blockade may slow the progression of metastatic disease in HER2-positive colon cancer. This is consistent with findings from clinical trials such as the HERACLES trial, which reported disease stabilization in some patients treated with HER2-targeted agents.

While the patient showed clinical benefit, there were several challenges encountered during treatment. One significant concern was the potential for cardiotoxicity, a well-documented side effect of trastuzumab. Our patient underwent regular cardiac monitoring, and fortunately, no significant changes in left ventricular ejection fraction were observed. Nonetheless, the risk of cardiotoxicity remains a concern in HER2-targeted therapies, and careful cardiac surveillance is essential, particularly in patients with existing comorbidities. Another challenge in this case was treatment resistance. Despite the partial response and stabilization, HER2-targeted therapies often face issues with acquired resistance over time. In our patient, there was concern that the disease may eventually progress due to changes in the tumor microenvironment or the emergence of HER2-negative clones. Monitoring disease progression and considering subsequent lines of therapy, such as combination with other targeted agents or immune checkpoint inhibitors, will be crucial for the long-term management of this patient.

CONCLUSION

These cases highlight the potential benefits of trastuzumab and pertuzumab as a treatment options for patients with HER2-positive metastatic colon cancer. The patient showed a promising partial response and disease stabilization, suggesting that this combination therapy can provide clinical benefit in a subset of mCRC patients. However, the challenges of side effects, resistance, and the need for careful patient selection remain important considerations. Future research and clinical trials will be essential to validate the efficacy of dual HER2 blockade in metastatic colon cancer, refine treatment strategies, and explore the role of combination therapies to further improve patient outcomes.

Ethics: The study was conducted with transparent, honest information, data, and methods. The research results were evaluated objectively and accurately. Vinmec Times City International Hospital granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

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PREVALENCE AND ASSOCIATED FACTORS OF SARCOPENIA IN LUNG CANCER PATIENTS UNDERGOING CHEMOTHERAPY: A CROSS-SECTIONAL STUDY AT A TERTIARY HOSPITAL IN VIETNAM

Pham Duc Minh[1], Bui Do Quynh Huong[1]*

Abstract

Objectives: To evaluate the prevalence of sarcopenia and its associated factors in lung cancer patients undergoing chemotherapy at a tertiary hospital in Vietnam. ***Methods:*** A cross-sectional descriptive study was conducted on 89 lung cancer patients. Sarcopenia was assessed using the Asian Working Group for Sarcopenia (AWGS) 2019 criteria, including measurements of handgrip strength (HGS), appendicular skeletal muscle mass (ASM), and physical performance. Logistic regression analysis identified factors associated with sarcopenia. ***Results:*** The mean age of participants was 62.6 ± 9.8 years; 78.7% were male. The prevalence of sarcopenia was 39.3%, with 40.4% of patients having reduced ASM. Malnutrition (Body mass index (BMI) < 18.5) was found in 15.7% of patients. Logistic regression analysis revealed significant associations between sarcopenia and male gender (OR = 8.19), lower hemoglobin levels (OR = 0.96), and increased lymphocyte counts (OR = 1.05). Patients with normal BMI (18.5 - 23) and high BMI (≥ 23) had lower odds of sarcopenia compared to those with BMI < 18.5. ***Conclusion:*** Sarcopenia is prevalent among lung cancer patients undergoing chemotherapy. Male gender, low BMI, low HGB, and high lymphocyte counts are associated with increased sarcopenia risk in lung cancer patients, whereas normal to high BMI offers protective effects.

Keywords: Malnutrition; Sarcopenia; Lung cancer; Chemotherapy.

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INTRODUCTION

Sarcopenia is a syndrome characterized by the loss of muscle mass, strength, and function, commonly seen in older adults and patients with chronic diseases [1]. In cancer patients, it results from chronic inflammation, metabolic dysregulation, and side effects of cancer treatments. Sarcopenia often coexists with cachexia, a syndrome of chronic inflammation and increased protein breakdown, leading to muscle wasting and functional decline [2].

The prevalence of sarcopenia in cancer patients can reach 80 - 90% in advanced stages, doubling the mortality risk [3]. It is particularly common in elderly cancer patients, increasing chemotherapy toxicity, reducing treatment efficacy, raising postoperative complications, and worsening quality of life, ultimately leading to higher mortality [4].

Lung cancer is one of the most common cancers worldwide. In Vietnam, lung cancer incidence has risen significantly, with an age-standardized rate of 33.2 per 100,000, ranking second after liver cancer [5]. Although chemotherapy is effective, it also exacerbates muscle loss and sarcopenia in lung cancer patients [6]. Nutritional status plays a crucial role in lung cancer management. Some studies on sarcopenia have been published in Vietnam [7, 8]; however, there were no publications on the application of bioelectrical impedance analysis in cancer patients. At Military Hospital 103, standardized multimodal cancer treatment is provided, but further research on malnutrition and sarcopenia prevalence and risk factors is needed to enhance clinical care. Therefore, this study aims to: *Determine the prevalence and risk factors of sarcopenia in lung cancer patients at Military Hospital 103 and evaluate the role of bioelectrical impedance analysis in diagnosing sarcopenia. The findings will provide scientific evidence to improve screening, diagnosis, and nutritional interventions for lung cancer patients, contributing to better treatment outcomes.*

MATERIALS AND METHODS

1. Subjects

Including 89 lung cancer inpatients diagnosed and undergoing chemotherapy at the hospital.

* *Inclusion criteria:* Adults aged ≥ 18 years; undergoing chemotherapy; capable of responding to survey questions; voluntary participation.

* *Exclusion criteria:* Incomplete medical records; conditions affecting bioelectrical impedance (e.g., pacemakers); neurological or communication impairments.

* *Study location and duration:* The research was conducted at the Cancer Center, Military Hospital 103, from October 2023 to December 2024.

2. Methods

* *Study design:* A cross-sectional descriptive study.

* *Sampling method:* All lung cancer patients admitted during the study period and meeting inclusion criteria were selected.

* *Study variables:*

Sarcopenia is diagnosed according to the AWGS 2019 criteria, based on three main criteria: Low muscle mass (mandatory criterion), low muscle strength, and low physical performance. Sarcopenia was diagnosed when low muscle mass presented along with either low muscle strength or low physical performance [1]. Measurement of muscle mass using multi-frequency bioelectrical impedance analysis (MF-BIA) with the InBody S10 (InBody Co., Ltd, Seoul, Korea), SMI = ASM (kg)*((height(m))$^{-2}$). Ensuring the subject fasts for at least 2 hours before the test. Avoiding intensive exercise, alcohol, and excessive water intake before testing. Removing metal accessories and ensuring clean, dry skin for electrode placement. The subject lies down in a supine position for at least 5 minutes before measurement. Reduced ASM was defined as SMI < 7.0 kg/m² for men and SMI < 5.7 kg/m² for women.

Muscle strength measurement by HGS using Camry EH101 (Camry, China). Reduced HGS was defined as < 28kg for men and < 18kg for women.

Physical performance: The 6-meter walk test (Gait Speed Measurement) was done on a flat, non-slip surface with a 6-meter distance clearly marked. A time of ≥ 6 seconds was considered as reduced physical performance.

* *Data collection:*

The assessed variables included patient demographics (age, sex, education), treatment method, and Eastern Cooperative Oncology group (ECOG) performance status.

Hematology and biochemical data collection: Blood samples were collected in the morning after overnight fasting. Complete blood count (CBC) analysis was performed using the UniCel DxH 600 hematology analyzer based on flow cytometry and morphological analysis utilizing laser technology. Biochemical parameters, including albumin, lymphocytes, potassium, sodium, chloride, creatinine, and hemoglobin, were analyzed using the AU5800 - Beckman Coulter, which employs the turbidimetric immunoassay method analyzed using standard laboratory techniques. All laboratory tests were

conducted in the hospital's central laboratory to ensure consistency and reliability.

The ECOG performance status scale was designed to assess the level of functioning of patients with cancer in terms of their ability to care for themselves, daily activity, and physical ability. Data were collected through surveys and processed using statistical software. Bias was minimized through investigator training and clear inclusion/ exclusion criteria.

* *Data analysis:* Data were entered and analyzed using SPSS 26.0. A p-value < 0.05 was considered statistically significant.

3. Ethics

The study was approved according to the Decision of the Research Project of the Military Medical University (Decision No. 2404/QĐ-HVQY, 25/6/2024). Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in this study.

RESULTS

Table 1. Characteristics of study participants (n = 89).

Characteristics	Mean ± SD or n (%)
Age (years)	62.6 ± 9.8
Age ≥ 60 (n, %)	60 (67.4)
Male (n, %)	70 (78.7)
Education above high school (n, %)	57 (64)
Stable income (n, %)	39 (43.8)
Advanced-stage cancer (n, %)	75 (84.3)
Treatment method (n, %)	
Surgery - Chemotherapy	23 (25.8)
Surgery - Radiotherapy - Chemotherapy	2 (2.2)
Chemotherapy	31 (34.8)
Radiotherapy - Chemotherapy	33 (37.1)
BMI (kg/m^2)	21.5 ± 2.8
BMI < 18.5 (n, %)	14 (15.7)
BMI 18.5 - 23 (n, %)	48 (53.9)
BMI ≥ 23 (n, %)	27 (30.3)

Characteristics	Mean ± SD or n (%)
ECOG performance status (points)	0.9 ± 0.9
ECOG 0 (n, %)	32 (36.0)
ECOG 1 (n, %)	44 (49.4)
ECOG 2 (n, %)	4 (4.5)
ECOG 3 (n, %)	9 (10.1)
Hemoglobin (g/L)	122.8 ± 19.9
Lymphocytes (G/L) #	12.7 (1.5 - 26.2)
Albumin (g/L) #	40.3 (38.2 - 43.1)
Potassium (mmol/L)	3.9 ± 0.3
Sodium (mmol/L) #	139.0 (136.4 - 141.1)
Chloride (mmol/L) #	101.7 (100.1 - 104.8)
Creatinine (µmol/L) #	73.7 (87.9 - 97.4)

(#: Presented with interquartile ranges (25^{th} - 75^{th} percentile))

Characteristics of the study population are shown in table 1. The mean age was 62.6 years, with a predominance of males (78.7%). Most participants had advanced-stage cancer (84.3%) and a BMI in the range of 18.5 - 23 (53.9%), while 15.7% were malnourished (BMI < 18.5). Radiotherapy - Chemotherapy is the most common treatment method (37.1%).

Table 2. Sarcopenia characteristics in patients (n = 89).

AWGS 2019 criteria	n	%
Reduced HGS	81	91.0
Reduced physical performance	83	93.3
Reduced ASM	36	40.4
Sarcopenia	35	39.3

Data on sarcopenia characteristics in table 2 showed that 39.3% of patients were diagnosed with sarcopenia according to AWGS 2019 criteria, and 40.4% had reduced ASM. The percentages of reduced HGS (91.0%) and physical performance (93.3%) were notably high.

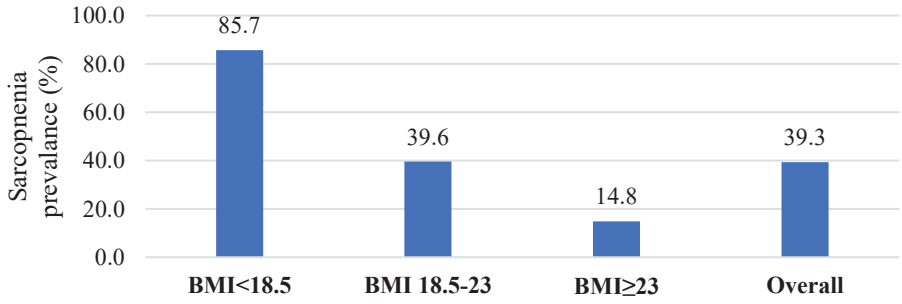

Chart 1. The prevalence of sarcopenia according to BMI categories (n = 89).

Chart 1 shows the prevalence of sarcopenia according to BMI categories. The data presented that with BMI < 18.5, sarcopenia accounted for the highest prevalence at 85.7%. As for the BMI 18.5 - 23 group, the prevalence drops to 39.6%. Finally, when BMI ≥ 23, the lowest prevalence is witnessed at 14.8%.

Table 3. Logistic regression model: Sarcopenia and associated factors (n = 89).

Characteristics	Sig.	OR	95%CI
Model 1			
Age (years)	0.96	1.00	0.92 - 1.09
Male gender	0.074	7.77	0.82 - 73.51
Education above high school	0.902	1.10	0.24 - 4.95
Stable income	0.718	0.77	0.18 - 3.23
Advanced-stage cancer	0.09	9.76	0.70 - 135.92
Surgery - Chemotherapy (reference)		1.00	
Surgery - Radiotherapy - Chemotherapy	0.91	1.27	0.02 - 81.14
Chemotherapy	0.235	0.30	0.04 - 2.18
Radiotherapy - Chemotherapy	0.351	0.39	0.05 - 2.84
BMI < 18.5 (reference)		1.00	
BMI 18.5 - 23	0.00	0.03	0.00 - 0.27
BMI ≥ 23	0.00	0.01	0.00 - 0.16
ECOG score (points)	0.07	2.00	0.95 - 4.20
Albumin (g/L)	0.37	1.05	0.94 - 1.17
Creatinine (mcrmol/L)	0.38	1.02	0.98 - 1.05

Characteristics	Sig.	OR	95%CI
Hemoglobin (g/L)	0.02	0.95	0.92 - 0.99
Lymphocyte (G/L)	0.04	1.05	1.00 - 1.10
Kali (mmol/L)	0.51	2.03	0.25 - 16.58
Natri (mmol/L)	0.44	0.91	0.71 - 1.16
Clo (mmol/L)	0.833	1.026	0.81 - 1.31
Constant	0.489	8442.76	
Model 2			
Male gender	0.023	8.19	1.34 - 50.17
BMI 18.5 - 23	0.002	0.06	0.01 - 0.35
BMI ≥ 23	0	0.02	0.00 - 0.17
ECOG score (points)	0.083	1.75	0.93 - 3.28
Hemoglobin (g/L)	0.013	0.96	0.93 - 0.99
Lymphocyte (G/L)	0.024	1.05	1.01 - 1.10
Constant	0.038	82.063	

(Model 1: Full logistic multivariate model; Model 2: Adjusted logistic multivariate model)

Table 3 analyzes factors associated with sarcopenia. The significant predictors of sarcopenia include male gender (OR = 8.19), BMI (BMI 18.5 - 23: OR = 0.06; BMI ≥ 23: OR = 0.02), Hemoglobin levels (OR = 0.96), and Lymphocyte count (OR = 1.05).

DISCUSSION

1. The prevalence of sarcopenia

This study found that 39.3% of patients met the AWGS 2019 criteria for sarcopenia, consistent with global estimates of 30 - 60% in lung cancer patients, especially in advanced stages. Reduced muscle mass (40.4%) and impaired functional performance (93.3%) highlight the impact of chronic inflammation, tumor burden, and cancer treatments [2, 6]. Sarcopenia was most prevalent (85.7%) in undernourished patients (BMI < 18.5), confirming low BMI as a major risk factor. Notably, 39.6% of patients with normal BMI (18.5 - 23) also had sarcopenia, indicating muscle loss can occur despite normal weight. In overweight/obese

patients (BMI ≥ 23), sarcopenia was the lowest (14.8%), suggesting a protective effect of higher BMI.

2. Risk factors for sarcopenia

Significant factors associated with sarcopenia included male gender, low hemoglobin levels, and increased lymphocyte count. For instance, a study published in the Journal of Parenteral and Enteral Nutrition found that the prevalence of sarcopenia in lung cancer patients varies between 35% and 50%, with a higher prevalence observed in males [9]. This may result from a combination of biological factors, lifestyle choices, and disease-related influences. However, it is important to note that the prevalence and risk factors can vary across populations, and further studies are needed to understand the gender-specific mechanisms underlying sarcopenia.

Anemia, as reflected by reduced hemoglobin, compromises oxygen delivery to muscles, further impairing their function [10]. Elevated lymphocyte counts may reflect an underlying inflammatory response, a hallmark of cancer-associated sarcopenia [2]. Interestingly, BMI is a protective factor, higher BMI significantly reduced the risk of sarcopenia. This supports the importance of maintaining or improving nutritional status to mitigate muscle loss. The association between sarcopenia and poor clinical outcomes, such as higher treatment toxicity, reduced treatment tolerance, and increased mortality, is well-documented [5]. The high prevalence of sarcopenia in this study underscores the urgent need for early identification and interventions targeting nutritional and physical rehabilitation to improve patient outcomes. This study emphasizes the importance of integrating sarcopenia assessments, such as HGS and muscle mass measurements, into routine oncology care. Early nutritional support, tailored physical activity programs, and anti-inflammatory strategies may help mitigate sarcopenia's progression and improve the quality of life for lung cancer patients [10].

However, this study has some limitations. Its cross-sectional design prevents causal conclusions, highlighting the need for longitudinal research on sarcopenia in lung cancer patients. The small sample size may limit generalizability. Additionally, the impact of specific treatments and co-morbidities on sarcopenia was not explored. Future studies should address these gaps for better understanding.

CONCLUSION

Sarcopenia is a prevalent and serious condition among lung cancer patients. The study identified key predictors of sarcopenia, including male gender, low hemoglobin levels, and increased lymphocyte count, reinforcing the role of systemic inflammation and anemia in muscle deterioration. Notably, BMI was found to be a protective factor, highlighting the importance of maintaining adequate nutritional status.

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THE EFFECTIVENESS OF LASER ACUPUNCTURE COMBINED WITH ACUPRESSURE MASSAGE IN PATIENTS SUFFERING FROM LOW BACK PAIN DUE TO SPONDYLOSIS

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Abstract

Objectives: To evaluate the effectiveness of laser acupuncture (LA) combined with acupressure massage in treating low back pain, focusing on pain intensity, functional disability, and lumbar range of motion. ***Methods:*** An uncontrolled, randomized, clinical trial was conducted to compare the effectiveness before and after treatment in 30 volunteered patients aged ≥ 38 diagnosed with low back pain due to spondylosis, regardless of gender or occupation, at the Traditional Medicine Department, Military Hospital 103. ***Results:*** Combining the LA method with acupressure massage showed good effectiveness, with a success rate of 96.7%. The pain level, according to the Visual Analogue Scale (VAS), improved significantly after 14 days of treatment, decreasing from an average of 5.4 ± 1.886 to 1.17 ± 1.234 ($p < 0.05$). The lumbar spine expansion and daily activity limitation measured by the Oswestry Disability Index (ODI) significantly improved. ***Conclusion:*** The treating method using LA combined with acupressure massage has been proven to be an efficient adjunct therapy in treating low back pain due to spondylosis.

Keywords: Laser acupuncture; Acupressure massage; Low back pain; Spondylosis; Lumbar spine degeneration.

INTRODUCTION

Low back pain is a pain syndrome characterized by the compression or irritation of nerve roots in the lumbar region of the spine, with an average prevalence in the general population ranging from 5 - 10%. According to Christopher E Alexander (2025), this syndrome is the leading cause of disability among individuals aged 45 and older in developed countries [1].

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The symptoms include lower back pain that radiates down the lower extremities along the compressed nerve roots, accompanied by numbness and burning sensations along the sciatic nerve pathway [2].

From a traditional medicine perspective, low back pain due to spondylosis is described within the scope of "Bi syndrome" with the disease name "Yao Tong". Treatment in traditional medicine includes both pharmacological and non-pharmacological methods such as acupuncture, electroacupuncture, hydro-acupuncture, acupressure massage, moxibustion, and qigong. In recent years, the integration of traditional and modern medicine has garnered increasing attention, with more combined techniques being applied, including LA. LA is becoming a reliable treatment method by leveraging the biological stimulation effects of laser light combined with acupuncture point theory according to traditional medical theory, which unblocks meridians, harmonizes Qi, and relieves pain. However, there is still insufficient evidence to evaluate the efficacy of this method, so we conducted the research to: *Assess the effectiveness of the laser method combined with acupressure massage to treat low back pain due to spondylosis.*

MATERIALS AND METHODS

1. Subjects

Including 30 patients aged ≥ 38 years, regardless of gender or occupation, diagnosed with low back pain due to spondylosis came for examination and treatment at the Traditional Medicine Department, Military Hospital 103. In traditional medicine, patients were diagnosed with "Yao Tong" due to liver and kidney Ying deficiency. Patients voluntarily participated and complied with the treatment regimen and did not apply other treatments during the period, which comprised data related to the 6-month period from June 2024 to December 2024.

* *Exclusion criteria:* Patients with a history of allergies; pregnant women; spinal tuberculosis; cancer; surgical indications or severe chronic diseases (liver, kidney, heart failure, severe hypertension, diabetes, etc.).

* *Criteria for treatment discontinuation:* Patients who refused to continue; did not adhere to treatment, or experienced side effects (increased pain, skin redness, itching, burning, etc.).

* *Research materials:* Laserneedle touch (manufactured and developed by the Laserneedle GmbH Company of the Federal Republic of Germany, consisting of 10 laser emitters, including 7 red light emitters with a wavelength of 658nm and 3 blue light emitters with a

wavelength of 405nm); measuring tape; VAS; ODI questionnaire.

2. Methods

* *Research design:* An uncontrolled, randomized, clinical trial comparing effectiveness before and after the treatment.

* *Research sample size:* 30 patients.

* *Procedure:*

- Clinical examination and imaging (X-ray of the lumbar spine).

- Patient selection.

- Treatment protocol:

+ LA: 1 time a day, continuous for 15 minutes in 14 days (except Saturday and Sunday). Using eye protection glasses for both patients and medical staff. Acupuncture formula: Ashi point, Jiaji L4-L5, Jiaji L5-S1, Weizhong (bilateral). Using 7 red light emitters with a wavelength of 658nm at a frequency of 935.5Hz and a power density of 4.07 W/cm^2 for 15 minutes.

+ Acupressure massage: According to the Traditional Medicine Technical Process of The Ministry of Health in 2013, these movements include rubbing, squeezing, rolling, pressing, acupressure, distribution, and lumbar spine exercise. 1 time a day for 15 consecutive minutes in 14 days (except Saturday and Sunday).

- The indicators are monitored and evaluated before treatment and at D7 and D14.

- Observing any side effects.

* *Research criteria:*

General characteristics of the research subjects include age, gender, occupation, and duration of pain.

Clinical criteria: Conducting evaluation at the time D0, D7, and D14.

- Evaluating the level of pain according to the VAS. The VAS score from 1 - 10 according to the level of pain: The patient was selected with $3 \leq VAS \leq 8$.

- Measurement of lumbar spine expansion (Schober's test) is classified into 4 levels: Good, fair, moderate, and poor. Normally, the daily index ranges from 14 - 16cm, a measurement of less than 14cm is considered abnormal.

- Assessment of daily activity limitation using the ODI questionnaire: This consists of 10 questions evaluating the extent of daily activity limitation in patients diagnosed with low back pain. The assessment focuses on 8 out of 10 activities: Pain intensity, self-care, lifting, walking, sitting, standing, sleeping, and social activities. Each activity is scored from 0 - 5, with a total score ranging from 0 - 40. A higher score indicates greater impairment in daily functioning. The functional limitation index is calculated based on the Oswestry Disability:

ODI% = (Actual score / Theoretical score) * 100%

- Clinical symptoms in traditional medicine: Irritability, hot flashes, wiry pulse, red tongue, and yellow coating.

- The overall effectiveness of the treatment was based on the comparison of the total scores (VAS, Schober index, and ODI) before and after treatment.

** Statistical analysis:*

The data analysis in this study was conducted using SPSS version 27.0. The results are shown in the form of average value ± standard deviation and percentage. The Nonparametric Wilcoxon matched-pairs signed-rank Test and Friedman Test were used at each time point compared to D0. The research results are considered statistically significant when $p < 0.05$.

3. Ethics

The research complied with all regulations and has been approved for use and publication according to the instructions of the Traditional Medicine Department, Military Hospital 103. Patient personal information is used exclusively for research purposes. The authors declare to have no conflicts of interest in the research.

RESULTS

1. General characteristics of the research subjects

Table 1. General characteristics of the research subjects.

Criteria		Frequency (n)	Percentage (%)
Sex	Female	17	39.4
	Male	13	51.5
Age (year)	< 40	4	13.3
	40 - 59	18	66.7
	≥ 60	6	20
	Average age (year, $\overline{X} \pm SD$)	60.37 ± 11.19	
Duration of pain (month)	≤ 3	5	16.7
	3 - 6	7	23.3
	≥ 6	18	60
Occupation	Manual labor	19	63.3
	Intellectual labor	11	36.7

The study included 30 patients, consisting of 17 females and 13 males. The average age was 60.37 ± 11.19, ranging from 36 - 75 years old. The highest disease prevalence was observed in the 40 - 59 age group, accounting for 66.7%. The majority of affected individuals were manual laborers. 60% of patients had been suffering from the disease for more than 6 months.

2. Evaluation of treatment effectiveness

Table 2. Classification of pain intensity.

Day	Pain levels (%)				p
	Severe pain	Moderate pain	Mild pain	No pain	
D0	36.7	43.3	20	0	
D7	0	33.3	53.4	13.3	< 0.001
D14	0	3.3	46.7	50	< 0.001

The changes in pain intensity occurred early at D7 compared to D0 ($p < 0.05$). After 14 days of treatment, the pain intensity decreased markedly, with a statistically significant difference ($p < 0.05$).

Table 3. Changes in average VAS, Schober index, and ODI % before and after treatment.

Scale	D0 ($\bar{X} \pm SD$)	D7 ($\bar{X} \pm SD$)	D14 ($\bar{X} \pm SD$)	p
VAS	5.4 ± 1.886	2.8 ± 1.919	1.17 ± 1.234	< 0.001
Schober index (cm)	13.15 ± 0.464	13.75 ± 0.548	14.38 ± 0.741	< 0.001
ODI (%)	66.4 ± 18.226	42.75 ± 19.79	26.75 ± 11.27	< 0.001

The average VAS score and ODI % showed a decreasing trend over the follow-up period, whereas the average Schober index gradually rose. The differences between the pre-treatment time point (D0) and the post-treatment time points (D7, D14) were statistically significant ($p < 0.05$).

Table 4. Changes in Schober Index before and after treatment.

Schober index (cm)	D0		D7		D14		p
	n	%	n	%	n	%	
≥ 14/10 - 16/10	0	0	1	3.3	17	56.6	< 0.001
≥ 13/10 - 14/10	8	26.7	17	56.7	11	36.7	< 0.001
≥ 12/10 - 13/10	13	43.3	8	26.7	2	6.7	< 0.001
< 12/10	9	30	4	13.3	0	0	< 0.001

Before treatment, lumbar spine expansion assessed by the Schober test was classified as moderate and poor in 73.3% of patients. After 7 days, this rate decreased to 40%, and by the end of the treatment period, no patients had poor lumbar spine expansion.

Table 5. Changes in clinical symptoms before and after treatment.

Symptoms	D0		D7		D14	
	n	%	n	%	n	%
Irritability	22	73.3	9	30	0	0
Hot flashes	17	56.7	11	36.7	3	10
Wiry pulse	25	83.3	16	53.3	7	23.3
Red tongue, yellow coating	20	66.7	13	43.3	5	16.7

Before treatment, many patients exhibited the four characteristic symptoms of liver and kidney Yin deficiency. The symptoms improved significantly after 14 days, with no patient continuing to show irritability.

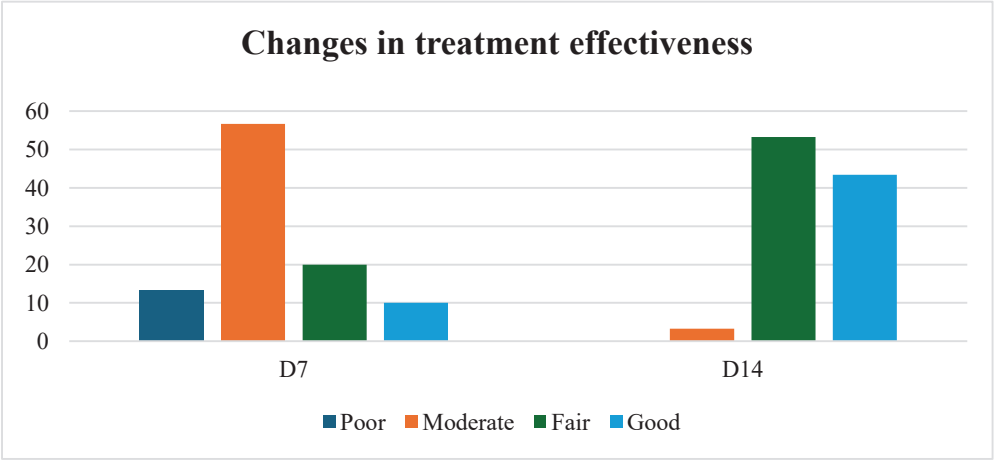

Chart 1. Overall treatment effectiveness.

Overall, treatment effectiveness after 7 days demonstrated a positive change, with 30% of the total patients achieving outcomes of fair and better. This rate increased to 96.7% after 14 days of treatment, and no patients had poor treatment outcomes.

DISCUSSION

Our study recorded a female proportion of 51.5%, which is 1.3 times higher than the male proportion (39.4%). Many studies indicate that women have a higher rate of spondylosis than men due to the effects of pregnancy and hormonal changes after menopause, leading to an estrogen deficiency that reduces calcium absorption - an important component in the structure of the spine. This finding is consistent with Davide Bizzoca's study (2023), which showed that the higher disease incidence in women is due to hormonal changes (estrogen and progesterone) during menstruation and pregnancy or to anatomical differences that may cause biological changes in the spine and pelvis, thereby increasing the likelihood of lower back pain [3].

The average age of the research patients was 60.37 ± 11.19, with 80% being under 60. This suggests that spondylosis is commonly observed among middle-aged workers. According to traditional medicine, when individuals over 40 years begin to enter a stage of senescence, the body's vital energy starts to decline, which results in Kidney Qi deficiency, accompanied by a gradual reduction of original Qi and weakened function of the internal organs (including the spleen, stomach, and kidneys). This decline leads to insufficient nourishment of the musculoskeletal system, which is a major contributor to the aging process of cartilage, joints, and ligament tissues.

All occupations can be affected by lumbar spine degeneration. According to our study, manual laborers exhibited a higher disease prevalence compared to intellectual workers, at 63.3% and 36.7%, respectively. This result is consistent with Jan Hartvigsen's study (2018) [4]. Manual laborers often engage in prolonged, heavy physical work and adopt improper postures, which lead to structural changes in the spine. These changes accelerate musculoskeletal degeneration, resulting in pain, muscle stiffness, and spinal deformity.

For patients with low back pain due to lumbar spine degeneration, pain is the most prominent and bothersome symptom, affecting daily activities, work, and quality of life, prompting them to seek medical care. Lumbar pain triggers a reflex contraction of the lumbar muscles, and as these muscles contract, the pain intensifies. When pain

is accompanied by stiffness in the paraspinal muscles, it further restricts lumbar spine mobility. Therefore, pain relief is the primary goal of treatment. Before treatment, we recorded that 43.3% of patients experienced moderate pain, and 36.7% experienced severe pain. After treatment, 50% of patients were pain-free, and none experienced severe pain. The average VAS score was 5.4 ± 1.886 before treatment and decreased to 1.17 ± 1.234 after 14 days - a statistically significant reduction ($p < 0.05$). These findings are consistent with the study by Do Thi Kim Ngan (2021), with the average VAS score after 20 days of treatment was 0.8 ± 0.6 [5]. Thus, the combination of LA and acupressure massage demonstrated clinically significant pain reduction. The analgesic mechanism of LA has been demonstrated through the biostimulatory effect of the photobiological reaction. LA is believed to influence the function of connective tissue cells (fibroblasts), accelerate connective tissue repair, and act as an anti-inflammatory agent by reducing prostaglandin synthesis. As the body absorbs the laser energy, cellular response processes are reorganized, resulting in several positive effects, including peripheral nerve blockade, inhibition of central synaptic nerve activity, modulation of central neurotransmitters, reduction of muscle spasm and edema, and enhancement of anti-inflammatory action. This aligns with the findings of Yousefi-Nooraie et al. (2008), which demonstrated that low-level laser therapy (LLLT) is beneficial in reducing pain and improving disability in patients with subacute or chronic nonspecific low back pain [6]. Moreover, acupressure massage, a form of physical stimulation applied directly to the skin, muscles, and sensory receptors, produces positive effects on multiple systems such as skin, muscles, tendons, joints, nervous system, circulatory system, digestive system, respiratory system, and metabolic processes. It enhances blood vessel and nerve activity in the skin, improves muscle nutrition, reduces muscle spasms, promotes muscle relaxation, increases the elasticity of tendons and ligaments, dilates peripheral blood vessels, improves local blood supply and oxygenation, alleviates tissue inflammation and edema, and promotes the secretion and circulation of joint synovial fluid. All of which are beneficial for managing musculoskeletal disorders.

Pain significantly affects daily life, with up to 80% of research subjects being assessed as having a moderate and poor quality of life. Consequently, rapidly and effectively addressing pain symptoms is the foremost priority for doctors.

CONCLUSION

A combination of LA and acupressure massage to treat low back pain due to spondylosis demonstrated good treatment effectiveness in terms of pain level, Schober index, and daily living function after 14 days of treatment. No adverse side effects were reported during the study.

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PREVALENCE AND ASSOCIATED FACTORS OF VERTEBRAL COMPRESSION FRACTURE AMONG OLDER WOMEN WITH OSTEOPOROSIS

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Nguyen Van Tan[1,3], Nguyen Duc Cong[3,4]

Abstract

Objectives: To investigate the prevalence and associated factors of vertebral compression fracture (VCF) in older women with osteoporosis. ***Methods:*** A cross-sectional study was conducted on 279 older women with osteoporosis at the Rheumatology and Neurosurgery Department and Rheumatology Clinic, University Medical Center, from August 2022 to May 2023. ***Results:*** 102/279 older women (36.6%) had at least one vertebral fracture, and more than 50% of participants were symptomatic. In the adjusted logistic regression, physical activity (OR: 0.44; 95%CI: 0.20 - 0.94; p = 0.038), osteoarthritis (OR: 0.24; 95%CI: 0.12 - 0.48; p < 0.001), frailty (OR: 7.41; 95%CI: 3.45 - 16.73; p < 0.001), falls (OR: 3.86; 95%CI: 1.68 - 9.32; p = 0.002), T-score at femoral neck (OR: 0.63; 95%CI: 0.41 - 0.92; p = 0.002) were associated with vertebral fracture. ***Conclusion:*** The prevalence of VCF was quite high among older women with osteoporosis, highlighting a disease burden in this population. Physical activity, osteoarthritis, and higher T-score at the femoral neck decreased the odds of VCF, while frailty and falls increased the odds of VCF.

Keywords: Vertebral fracture; Osteoporosis; Aged; Women.

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INTRODUCTION

A VCF is a break in the vertebral body, mostly due to osteoporosis in older women. VCF affects around 20 - 25% of older people across the globe and increases with age up to 40% at the age of 80 [1]. VCF predicts morbidity comprising back pain, kyphotic deformity, and loss of height, resulting in subsequent vertebral fracture and mortality eventually [2, 3]. Unfortunately, VCF is under-recognized in clinical settings [4]. In Vietnam, the increasing number of older women has put a heavy burden of osteoporosis and VCF on this population. Although there was some research about osteoporotic VCF, the participants were less responded to conservative management and had indications of surgical intervention. These articles also did not examine the prevalence of VCF in older women with osteoporosis and did not focus on the geriatric female population, which can have different clinical and radiologic characteristics because of the aging process, frailty, and multimorbidity. The study aims to: *Investigate the prevalence and several factors associated with VCF among older females at University Medical Center, Ho Chi Minh City.*

MATERIALS AND METHODS

1. Subjects

Including 279 older women with osteoporosis at the Rheumatology and Neurosurgery Department and Rheumatology Clinic, University Medical Center, from August 2022 to May 2023.

* *Inclusion criteria:* Females aged ≥ 60 whose bone density scan had a T-score ≤ -2.5, which is the World Health Organization's definition of osteoporosis.

* *Exclusion criteria:* We excluded any cases suspected of having non-fragility fracture or secondary osteoporosis to focus on post-menopausal osteoporosis.

2. Methods

* *Study design*: A cross-sectional study.

* *Study procedure:*

A geriatrician asked the participants about background information and previous height - the highest one they remembered at the age of 30. The participants' height and weight were measured. Their bone mineral density was measured using dual-energy X-ray absorptiometry (DEXA) at the lumbar

spine and femoral neck, and thoracolumbar X-ray results were recorded. The participants' inquiry information, along with X-ray and DEXA results, which were retrieved from electronic health records, was then recorded on data collection sheets.

** Definition of variables:*

VCF is diagnosed using the morphology of vertebrae based on Genant's method. We also examined back pain and kyphosis using clinical judgment. Height loss was defined if the previous height minus the current height was at least 4cm.

Comorbidities were collected using electronic medical records, and multimorbidity was determined if there were at least two diseases. Activities of daily living (ADL) were assessed using the Katz index, and instrumental activities of daily living (IADL) were assessed using the Lawton index. Frailty was diagnosed using the Clinical Frailty Scale (CFS) and categorized as non-frailty (CFS $\leq$ 3), pre-frailty (CFS = 4), and frailty (CFS $\geq$ 5).

** Statistical analysis:*

Data were analyzed using R (R Foundation for Statistical Computing, Vienna, Austria). Categorical variables were expressed as frequencies and percentages, and quantitative variables were expressed as means and medians for normally distributed variables or medians and interquartile ranges for non-normally distributed variables. Variables between groups were compared using the Chi-square test or Fisher's exact test for categorical variables and the T-test for normally distributed quantitative variables. Logistic regression assessed associations of related factors and VCF. Statistical significance was defined as a p-value < 0.05.

3. Ethics

This study was approved by the Ethics Committee of the University of Medicine and Pharmacy in Ho Chi Minh City (approval number 639/HDDD dated 1st August 2022). The University Medical Center granted permission for the use and publication of the research data. All participants were informed of the objectives and obtained informed consent. The authors declare to have no conflicts of interest in this research.

RESULTS

279 patients were included in the study.

Table 1. Baseline characteristics of older women with T-score ≤ -2.5.

Characteristics	Overall (n = 279)
Age (year)[b]	72.0 ± 7.3
Age groups, n (%)	
60 - 69	114 (40.9)
70 - 79	115(41.2)
≥ 80	50 (17.9)
Previous VCF, n (%)	15 (5.4)
Bisphosphonate usage, n (%)	77 (27.6)
Physical activity, n (%)	95 (34.1)
Current height (cm)[b]	152 ± 4.9
Weight (kg)[b]	52.4 ± 8.8
BMI (kg/m^2)[b]	22.6 ± 3.5
Comorbidities, n (%)	
Hypertension	114 (40.9)
Diabetes mellitus	52 (18.6)
Osteoarthritis	142 (50.9)
Stroke	11 (3.9)
Chronic kidney disease	20 (7.2)
Multimorbidity, n (%)	119 (42.7)

(BMI: Body mass index; [a]VCF group vs. non-VCF group; [b]Mean ± Standard deviation)

The majority of our participants were between 60 - 79. The VCF group also had less physical activity, less osteoarthritis occurrence, and lower T-scores at all three sites (femoral neck, total hip, and lumbar spine) than the non-VCF group.

Table 2. Clinical characteristics and X-ray images of VCF.

Characteristics	VCF		p*
	Yes (n = 102)	No (n = 177)	
VCF, n (%)	102 (36.6)		
Back pain, n (%)	74 (72.5)	20 (11.3)	< 0.001
Kyphosis, n (%)	69 (67.6)	5 (2.8)	< 0.001
Height loss, n (%)	64 (62.7)	4 (2.3)	< 0.001
T-score[a]			
Femoral neck	-2.8 ± 1.0	-2.4 ± 0.8	< 0.001
Total hip	-2.4 ± 0.9	-1.9 ± 0.9	0.001
Lumbar spine	-3.1 ± 1.0	-2.8 ± 0.8	0.009
Number of fractures[b]	1.5 (1.0 - 2.0)		
One fracture	51 (50.0)		
Two fractures	27 (26.5)		
Three or more fractures	24 (23.5)		
Severity, n (%)			
Mild	4 (3.9)		
Moderate	14 (13.7)		
Severe	84 (82.4)		

*([a]Mean ± Standard deviation; [b]Median (Interquartile range); *VCF group vs. non-VCF group)*

The prevalence of VCF among the participants was 36.6% (102/279). Overall T-scores at the femoral neck, total hip, and lumbar spine were -2.5 ± 0.9, -2.2 ± 0.9, and -2.9 ± 0.9, respectively. More participants with VCF had back pain, kyphosis, and height loss than those without VCF. Most participants with VCF had one fracture and severe deformity.

Table 3. Geriatric characteristics of older women with T-score ≤ -2.5.

Characteristics	Overall (n = 279)	VCF		p[a]
		Yes (n = 102)	No (n = 177)	
ADL dependence, n (%)	49 (17.6)	33 (32.4)	16 (9.0)	< 0.001
IADL dependence, n (%)	101 (36.2)	66 (64.7)	35 (19.8)	< 0.001
Frailty, n (%)				
Non-frailty	132 (47.3)	22 (21.6)	110 (62.1)	< 0.001
Pre-frailty	43 (15.4)	12 (11.8)	31 (17.5)	
Frailty	104 (37.3)	68 (66.7)	36 (20.3)	
Falls, n (%)	51 (18.3)	36 (35.3)	15 (8.5)	< 0.001

([a]VCF group vs. non-VCF group)

The VCF group had more frailty, falls, and ADL and IADL dependence than the non-VCF group.

Table 4. Logistic regression model of factors associated with VCF (n = 279).

Factors	Univariate regression		Multivariate regression	
	OR (95%CI)	p	Adjusted OR (95%CI)	p
Age	1.10 (1.06 - 1.14)	< 0.001	1.02 (0.96 - 1.08)	0.521
Physical activity	0.25 (0.14 - 0.45)	< 0.001	0.44 (0.20 - 0.94)	0.038
BMI	0.99 (0.93 - 1.07)	0.939		
Diabetes mellitus	1.98 (1.08 - 3.67)	0.027	1.45 (0.65 - 3.25)	0.359
Osteoarthritis	0.42 (0.25 - 0.68)	< 0.001	0.24 (0.12 - 0.48)	< 0.001
Frailty				
Non-frailty	Reference		Reference	
Pre-frailty	1.94 (0.84 - 4.30)	0.109	1.55 (0.59 - 3.96)	0.363
Frailty	9.44 (5.21 - 17.71)	< 0.001	7.41 (3.45 - 16.73)	< 0.001
Falls	5.89 (3.08 - 11.77)	< 0.001	3.86 (1.68 - 9.32)	0.002
T-score				
Femoral neck	0.53 (0.37 - 0.73)	< 0.001	0.63 (0.41- 0.92)	0.002
Lumbar spine	0.65 (0.47 - 0.88)	0.007	0.70 (0.48 - 1.00)	0.055

(OR: Odds ratio; CI: Confidence interval)

Variables with p-value < 0.2 in the univariate regression model were included in the multivariate regression model. The T-score at the total hip was omitted because the T-score at the femoral neck could affect the T-score at the total hip, leading to multicollinearity. Age was included in the two models as a quantitative variable.

In multivariate logistic regression (*Table 5*), physical activity, osteoarthritis, and high T-score at the femoral neck decreased the odds of VCF, while frailty and falls increased the odds of VCF.

DISCUSSION

Our study found that the proportion of VCF among older females with osteoporosis was 36,6% (*Table 2*). For baseline characteristics, the participants in this study were older, had lower T-scores, and were more frail compared to previous studies [5].

The most striking finding was the substantial prevalence of VCF in older women with osteoporosis. Xia et al. used Genant's semiquantitative method and found that the prevalence of VCF was 21.6% among women aged > 65 years. Waterloo et al. reported that the prevalence of VCF was 12.6% in Sweden. Our results were higher than those of published studies worldwide for several reasons as follows: (1) Our participants were older than those in previous studies; (2) over 80% of our participants experienced severe deformity of the VCF; (3) a third of participants were ADL- and IADL-dependent, and 40% were frail. Frailty is an issue that requires further research in older women because of the increased risk of VCF, multimorbidity, and mortality burden.

Physical activity has been proven to benefit skeletal health, but whether physical activity helps to prevent vertebral fractures is still debatable. A previous study showed no significant association between total physical activity and clinical vertebral fracture [6]. On the other hand, Ling et al. demonstrated that women doing heavy physical work were less likely to have VCF (hazard ratio: 0.87; 95%CI: 0.78 - 0.96; p < 0.006). The protective effect of physical activity on bone health can be explained by attenuating age-related bone mineral loss, improving balance and muscle strength, and thus reducing fall risk and fall-related fractures [7].

Our data showed an inverse association between osteoarthritis and vertebral fracture (*Table 4*). Osteoarthritis becomes more prevalent with age; therefore, it could have affected the age variable in the multivariate logistic regression model. Nevertheless, longitudinal research is needed to discover the association

between osteoarthritis at various sites and vertebral fractures among older females with osteoporosis.

Frailty is a common syndrome in older adults with osteoporosis, increasing morbidity and worsening quality of life. Our study was consistent with previous research. Interestingly, fracture also increased the risk of subsequent frailty [8], and the more severe frailty was, the more the hazard ratio increased after adjusting for age, sex, and socioeconomic status [9]. This bidirectional interaction between frailty and VCF demonstrated how the aging musculoskeletal system plays a vital role in the development of frailty [8]. Therefore, it is critical that clinicians should pay more attention to frailty, prescribe walking aids, and fall education, and encourage older women to exercise to minimize the risk of vertebral fracture.

Falls interact with VCF due to age-related alteration in spinal structure. Hyperkyphotic posture at thoracic spine level could jeopardize patients by increasing fall risk (OR = 2.13, 95%CI: 1.10 - 4.51) [10]. The VCF group in our study had more kyphosis than the non-VCF group (*Table 2*). Our study implies that physicians should employ strategies to prevent falls in older women with osteoporosis.

The limitations of our study include several factors that were self-reported, and data on vitamin D status, comprehensive medication history, and chronic inflammatory disease were absent, which may lead to overestimation or underestimation of VCF prevalence. Second, osteoarthritis becomes more prevalent with age; therefore, it could have affected the age variable in the multivariate logistic regression model. Finally, our study conducted quite distant from 2025 (from August 2022 to May 2023) could affect the evolution of factors associated with osteoporosis and vertebral fracture.

CONCLUSION

The prevalence of VCF among older women is high. Physical activity and osteoarthritis decreased the odds of VCF, whereas frailty, lower T-score, and falls increased the odds of VCF.

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PREVALENCE OF DEPRESSION IN ELDERLY PATIENTS FOLLOWING ACUTE CORONARY SYNDROME AT DISCHARGE FROM THONG NHAT HOSPITAL

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Abstract

Objectives: To estimate the frequency of depression among older adults with acute coronary syndrome (ACS) and identify relevant factors that are associated with depression at the time of discharge. ***Methods:*** A cross-sectional descriptive study was conducted on 117 elderly patients with ACS who were discharged from Thong Nhat Hospital between March 2024 and June 2024. Depression was assessed using the 30-item Geriatric Depression Scale (GDS-30), with a total score of ≥ 10 indicating the presence of depression. ***Results:*** The prevalence of depression at discharge among elderly ACS patients was 15.4% (95%CI: 8.7% - 22.0%). In the multivariate regression analysis, female gender, illiteracy, high-risk CCI, experiencing two or more stressful life events, and low perceived social support were significantly associated with a higher prevalence of depression. ***Conclusion:*** The prevalence of depressive symptoms at the time of discharge among elderly patients recovering from ACS was 15.4%. Early detection of depression is crucial, particularly in patients who are female, have illiteracy, present with multimorbidity, particularly type 2 diabetes mellitus, experience stressful life events, and have low perceived social support.

Keywords: Depression; Elderly; Acute coronary syndrome (ACS); Discharge; Geriatric Depression Scale (GDS-30).

INTRODUCTION

Vietnam is currently one of the fastest-growing aging populations in the world and is predicted to enter the aging population period by 2035. Depression is a mood disorder characterized by persistent sadness, loss of interest or pleasure, sleep and appetite disturbance, psychomotor agitation or retardation, and thoughts of self-harm.

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ACS is a severe medical emergency that includes three clinical forms: ST-elevation myocardial infarction (STEMI), non-ST-elevation myocardial infarction (NSTEMI), and unstable angina (UA). It has been documented that after an ACS event, patients often experience psychological stress, with depression and anxiety being the most prevalent conditions. The bidirectional relationship between ACS and depression has been extensively studied [1, 2]. Given the limited data on the depression status among post-ACS elderly patients in Vietnam, this study aimed to: *Investigate the prevalence of depression in this population and analyze its associations with sociodemographic factors and clinical features.*

MATERIALS AND METHODS

1. Subjects

A sample size of 117 patients was estimated to be adequate for detecting differences in primary outcomes, utilizing convenient sampling methods from March 2024 to June 2024.

* *Inclusion criteria:* Aged ≥ 60 years; diagnosed with acute myocardial infarction (AMI) or UA based on the 2023 ESC criteria [3]; approved for discharge based on clinical stability, successful revascularization or optimal medical therapy, and adherence to guideline-recommended discharge criteria for ACS.

* *Exclusion criteria:* Patients who were unable to communicate effectively for interview completion, including those with impaired consciousness, dementia, or a history of psychiatric disorders that could affect the accuracy of the information provided.

2. Methods

* *Study design:* A cross-sectional descriptive study.

* *Variable definition:* Depressive status was assessed using the Vietnamese version of GDS-30. Participants were instructed to respond with "Yes" or "No" based on their experiences over the past 2 weeks. Depression is classified as none to minimal (0 - 9), mild (10 - 19), and severe (20 - 30). A cut-off score of ≥ 10 was chosen to maximize sensitivity without compromising specificity. The dependent variable was depression at discharge [4].

Comorbidity severity was assessed using the Charlson Comorbidity Index (CCI), with a score ≥ 3 indicating a high one-year mortality risk. Functional impairment in activities of daily living (ADL) was evaluated using the Katz ADL scale, with ≤ 4 points denoting significant impairment. Sleep disturbances were identified using the Pittsburgh Sleep Quality Index (PSQI), with a score ≥ 5 indicating poor sleep quality. Psychological stress was defined as experiencing two or more stressful

events within the past 12 months or significant life events. Perceived social support was measured with the MSPSS, with an average score < 5.1 indicating low support.

* *Statistical analyses:* Data were analyzed using SPSS version 27.0. Modified Poisson regression with robust standard errors was used to estimate prevalence ratios. Multivariable Poisson regression was performed to identify independent risk factors for post-ACS depression.

3. Ethics

The study was approved by the Ethics Committee of Pham Ngoc Thach University of Medicine according to Decision No. 733/QĐ-TĐHYKPNT dated March 12th, 2024, and the Ethics Committee of Thong Nhat Hospital according to Decision No. 14/BB-BVTN dated April 19th, 2024. Thong Nhat Hospital granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in this study.

RESULTS

1. Baseline characteristics of the patient population

Table 1. Sociodemographic and clinical characteristics of the study population.

Variable	Total (n = 117)
Mean age, M ± SD	71.2 ± 7.4
Age group (year), n (%)	
60 - 69	53 (45.3)
70 - 79	46 (39.3)
≥ 80	18 (15.4)
Gender, n (%)	
Female	47 (40.2)
Male	70 (59.8)
Diagnosis of ACS, n (%)	
STEMI	28 (23.9)
NSTEMI	65 (55.6)
UA	24 (20.5)

Variable	Total (n = 117)
Living area, n (%)	
Rural	42 (35.9)
Urban	75 (64.1)
Education level, n (%)	
Illiteracy	9 (7.7)
Literate/basic education	71 (60.7)
Secondary education	25 (21.4)
Tertiary education	12 (10.3)
Marriage status, n (%)	
Married	83 (70.9)
Single	6 (5.1)
Separation/divorce	2 (1.7)
Widow	26 (22.2)
Living situation, n (%)	
Living with others	111 (94.4)
Living alone	6 (5.1)
Employment status, n (%)	
Employed	38 (32.5)
Unemployed/retired	79 (67.5)

The study included 117 patients with most being aged 60 - 69 years and a slightly higher proportion of males than females. The predominant clinical presentation of ACS was NSTEMI. Most patients came from urban areas, currently lived with family members, lived with a spouse, and were no longer employed. The widowhood rate was relatively high at 22.2%, while the illiteracy rate was low (7.7%).

2. Prevalence of depression following ACS events

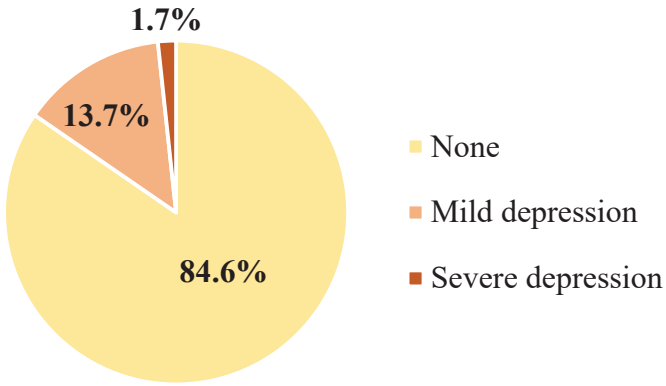

Figure 1. Prevalence of depression in post-ACS elderly patients at discharge.

Depression at discharge was observed in 15.4% of patients, mostly presenting as mild, while severe cases were rare.

Table 2. Univariate analysis of depression with sociodemographic characteristics of post-ACS elderly patients at discharge.

Variable	Depression		Unadjusted PR (95%CI)	p†
	Yes* (n = 18)	No (n = 99)		
Age group (year), n (%)				
60 - 69‡	5 (9.4)	48 (90.6)	1.00	-
70 - 79	8 (17.4)	38 (82.6)	1.84 (0.65 - 5.24)	0.251
≥ 80	5 (27.8)	18 (72.2)	2.94 (0.96 - 9.01)	0.058
Gender, n (%)				
Female	12 (25.5)	35 (74.5)	2.98 (1.20 - 7.38)	0.018
Male‡	6 (8.6)	64 (91.4)	1.00	-
Living area, n (%)				
Rural	7 (16.7)	35 (83.3)	1.14 (0.48 - 2.71)	0.773
Urban‡	11 (14.7)	64 (85.3)	1.00	-

Variable	Depression		Unadjusted PR (95%CI)	p†
	Yes* (n = 18)	No (n = 99)		
Religious practice, n (%)				
Yes	10 (16.1)	52 (83.9)	1.11 (0.47 - 2.61)	0.813
No‡	8 (14.5)	47 (85.5)	1.00	-
Illiteracy, n (%)				
Yes	4 (44.4)	5 (55.6)	3.43 (1.42 - 8.26)	0.006
No‡	14 (13.0)	94 (87.0)	1.00	-
Married status, n (%)				
Others[a]	9 (26.5)	25 (73.5)	2.44 (1.06 - 5.62)	0.036
Married‡	9 (10.8)	74 (89.2)	1.00	-
Living situation, n (%)				
Living alone	2 (33.3)	4 (66.7)	2.31 (0.68 - 7.83)	0.178
Living with others‡	16 (14.4)	95 (85.6)	1.00	-
Employment status, n (%)				
Unemployed/retired	14 (17.7)	65 (82.3)	1.68 (0.59 - 4.77)	0.327
Employed‡	4 (10.5)	34 (89.5)	1.00	-

*(PR: Prevalence ratio; CI: Confidence interval; *: Based on GDS-30 ≥ 10; †: Univariate Poisson regression with robust variance; ‡: Reference variable; [a]: Including single, divorced, separated, and widow status)*

As shown in table 2, the rate of depression was significantly higher among the female patients (p = 0.018), those who were illiterate (p = 0.006), and those who were currently living with their spouse (p = 0.036). Although we noted a greater prevalence of depression in patients aged 80 and older, as well as those living alone, these differences were not statistically significant.

Table 1. Univariate analysis of depression with clinical characteristics of post-ACS elderly patients at discharge.

Variable	Depression		Unadjusted PR (95% CI)	p[†]
	Yes[*] (n = 18)	No (n = 99)		
Diagnosis of ACS, n (%)				
STEMI	7 (25.0)	21 (75.0)	1.66 (0.39 - 7.15)	0.144
NSTEMI	9 (13.8)	56 (86.2)	3.00 (0.69 - 13.10)	0.495
UA[‡]	2 (8.3)	22 (91.7)	1.00	-
Complications of ACS, n (%)				
Yes	7 (20.0)	28 (80.0)	1.49 (0.63 - 3.53)	0.363
No[‡]	11 (13.4)	71 (86.6)	1.00	-
PCI treatment, n (%)				
No	7 (22.6)	24 (77.4)	1.77 (0.75 - 4.12)	0.192
Yes[‡]	11 (12.8)	75 (87.2)	1.00	-
Comorbidities[!], n (%)				
Hypertension	14 (14.9)	80 (85.1)	0.86 (0.31 -2.36)	0.764
History of heart failure	5 (20.8)	19 (79.2)	1.49 (0.59 - 3.77)	0.400
History of IHD	4 (28.6)	10 (71.4)	2.10 (0.80 - 5.49)	0.130
Type 2 diabetes mellitus	11 (27.5)	29 (72.5)	3.03 (1.27 - 7.20)	0.012
Chronic kidney disease	4 (26.7)	11 (73.3)	1.94 (0.74 - 5.13)	0.180
Dyslipidemia	12 (16.2)	62 (83.8)	1.16 (0.47 - 2.87)	0.745
GERD	7 (17.1)	34 (82.9)	1.18 (0.50 - 2.81)	0.709
History of CVA	2 (18.2)	9 (81.8)	1.21 (0.32 - 4.57)	0.784
CCI risk, n (%)				
High-risk	12 (33.3)	24 (66.7)	4.50 (1.83 - 11.05)	0.001
Low-risk[‡]	6 (7.4)	75 (92.6)	1.00	-

Variable	Depression		Unadjusted PR (95% CI)	p†
	Yes* (n = 18)	No (n = 99)		
Health behaviors!, n (%)				
Smoking	7 (11.3)	55 (88.7)	0.57 (0.24 - 1.36)	0.200
Alcohol consumption	8 (13.8)	50 (86.2)	0.81 (0.35 - 1.92)	0.637
Physical activity	9 (14.1)	55 (85.9)	0.83 (0.35 - 1.94)	0.663
Geriatric conditions!, n (%)				
ADL impairment	8 (57.1)	6 (42.9)	5.89 (2.80 - 12.38)	< 0.001
Urinary incontinence	6 (46.2)	7 (53.8)	4.00 (1.82 - 8.84)	< 0.001
Sleep disorders	14 (25.0)	42 (75.0)	3.81 (1.34 - 10.90)	0.013
Stressful life events, n (%)				
≥ 2 events	12 (23.5)	39 (76.5)	2.59 (1.04 - 6.43)	0.040
< 2 events‡	6 (9.1)	60 (90.9)	1.00	-
Perceived social support, n (%)				
Low	4 (5.1)	74 (94.9)	7.00 (2.47 - 19.86)	< 0.001
High‡	14 (35.9)	25 (64.1)	1.00	-

*(PR: Prevalence ratio; CI: Confidence interval; GERD: Gastroesophageal reflux disease; CVA: Cerebrovascular accident; CCI: Charlson comorbidity index; IHD: Ischemic heart disease; *: Based on GDS-30 ≥ 10; †: Univariate Poisson regression with robust variance; ‡: Reference variable; !: Reference category for each condition is "No")*

Depression was predominantly observed in patients with AMI, particularly those with STEMI, and was more common than in patients with UA, but this difference was not statistically significant. Depression was more common among patients with type 2 diabetes, high multimorbidity risk, ADL impairment, urinary incontinence, and sleep disturbances. Those experiencing multiple stressful events were also at higher risk. Conversely, higher perceived social support (MSPSS scores 5.1 - 7) was associated with a lower prevalence of depression.

3. Factors associated with outcomes

Table 4. Multivariate modified Poisson regression of predictors of depression in post-ACS elderly patients at discharge.

Factors	Adjusted PR (95%CI)†	p
Female gender	4.36 (1.36 - 13.97)	0.013
Illiteracy	3.76 (1.61 - 8.76)	0.002
Unmarried status	0.64 (0.25 - 1.65)	0.355
Type 2 diabetes mellitus	0.92 (0.3 - 2.86)	0.891
High-risk CCI	3.60 (1.07 - 12.11)	0.038
ADL impairment	1.60 (0.58 - 4.43)	0.367
Urinary incontinence	2.42 (0.7 - 8.33)	0.160
Sleep disorders	1.48 (0.56 - 3.9)	0.426
Experienced ≥ 2 stressful life events	3.69 (1.58 - 8.59)	0.003
Low Perceived Social Support	3.67 (1.73 - 7.82)	< 0.001

(†: Multivariate Poisson regression with robust variance)

We selected 10 statistically significant variables for the multivariate regression model. After controlling for the influence of other covariates, we identified the following factors as significantly associated with post-ACS depression: Female gender, illiteracy, high-risk CCI, experiencing ≥ 2 stressful life events, and low perceived social support.

DISCUSSION

1. Prevalence of depression

The study found that the prevalence of depression at the time of discharge among elderly patients recovering from ACS was 15.4% (95%CI: 8.7% - 22.0%) as measured by the GDS-30 (*Figure 1*). A systematic review by Dong Z et al. (2024), derived from 28 studies, reported a pooled depression rate of 28.5% among ACS patients [5]. When analyzing studies with similar elderly populations, we observed a gradual decline in depression rates over time. Specifically, Hayajneh et al. (2021) reported that 65.7% of elderly patients with AMI exhibited depressive symptoms at the time of emergency admission [6]. In

contrast, Romanelli (2002) and Nguyen Van Tan (2021) found depression rates of 22.9% and 26.4%, respectively [7]. Our study revealed a significantly lower rate of depression at discharge, possibly due to patients feeling more reassured about their medical condition. Additional research is warranted to substantiate these psychological improvements relating to the admission phase.

2. Factors associated with depression in elderly patients post-AMI

Female patients had a 2.98 times higher rate of depression compared to male patients (*Table 2*). It is believed that older females may have a higher risk of depression, possibly due to hormonal imbalances after menopause and increased psychological stress from family and societal responsibilities, which may contribute to a greater likelihood of depression following ACS [8].

Depression was more common among individuals with lower educational attainment, particularly among illiterate patients, who had a 3.43 times higher prevalence rate than other groups (*Table 2*). Illiteracy may create barriers to social communication, leading to feelings of inferiority and difficulty integrating into daily life [9]. Additionally, patients without spouses reported a statistically higher prevalence of depression (PR = 2.44; p = 0.036). However, marital status lost its significance in the multivariate regression model. This finding aligns with numerous surveys on depression in older adults [9].

Although AMI is often associated with greater psychological stress due to its severity and risk of complications, the difference in depression prevalence between AMI and UA was not statistically significant (*Table 2*). This suggests that factors beyond disease severity, such as persistent health concerns and anxiety about recurrent events, may contribute to depression in both groups despite the better short-term prognosis of UA.

Type 2 diabetes mellitus was the only chronic condition showing a statistically significant association with depression (PR = 3.03; 95%CI: 1.27 - 7.20). Furthermore, we found no association between a history of heart failure and depression, which differs from Nguyen Van Tan's findings [7]. This discrepancy may be attributed to population characteristics and the timing of the research.

Chronic multimorbidity has a strong correlation with depression, which is consistent with previous studies. Patients with ≥ 2 chronic conditions are 1.59 times more likely to have depression compared to those without any chronic diseases (adjusted OR = 1.595; 95%CI: 1.01 - 2.52; p = 0.045)

[9]. In our study, high-risk patients had significantly higher depression rates than low-risk individuals (33.3% vs. 7.4%, PR = 4.50; p < 0.001) (*Table 3*). Multivariate analyses confirmed that multimorbidity is an independently associated factor, with a 3.6-fold increased risk of depression in the high-risk group (95%CI: 1.07 - 12.11; p = 0.038) (*Table 4*). Chronic health conditions may lead to dissatisfaction with quality of life, contributing to psychological stress and increasing the risk of depression.

The multivariate analyses suggest that geriatric conditions are not statistically significant in predicting depression. These findings align with the study by Nguyen Van Tan et al. (2021), which reported significant associations with ADL impairment and urinary incontinence but found that these conditions did not remain risk factors after conducting multivariate logistic regression [7].

Patients who reported experiencing two or more stressful life events were 3.69 times (95%CI: 1.58 - 8.59; p = 0.003) more likely to suffer from depression (*Table 4*). According to Do Van Dieu et al. (2018), individuals who have experienced at least one "major life event in the past 12 months" or "major life event in their lifetime" are independent risk factors that lead to a higher likelihood of developing depression, with adjusted OR of 1.9 (p < 0.001) and 2.4 (p < 0.001), respectively [10]. Those identified as having low perceived social support are 3.67 times more likely to have depression (95%CI: 1.73 - 7.82; p < 0.001). This is understandable since social support is an important protective factor in maintaining mental health among elderly patients after an ACS event.

CONCLUSION

This study highlights the significant prevalence of depression among elderly patients recovering from ACS, with a rate of 15.4% at the time of discharge. The findings suggest that patients who are female, have illiteracy, present with multimorbidity, particularly type 2 diabetes mellitus, experience stressful life events, and have low perceived social support are more likely to suffer from depression.

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PREVALENCE AND CHARACTERISTICS OF NON-ALCOHOLIC FATTY LIVER DISEASE IN PATIENTS WITH FATTY LIVER DIAGNOSED BY ULTRASOUND

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Abstract

Objectives: To determine the prevalence and characteristics of non-alcoholic fatty liver disease (NAFLD), including steatosis and fibrosis, in patients with fatty liver detected by ultrasound. ***Methods:*** A cross-sectional study was conducted on 303 patients diagnosed with fatty liver by ultrasound at People's Hospital 115 from August 2019 to October 2020. Steatosis and fibrosis were assessed using FibroScan, employing the controlled attenuation parameter (CAP) and liver stiffness measurements (LSM). Statistical analysis was performed using SPSS version 22.0. ***Results:*** The prevalence of NAFLD in patients with fatty liver detected by ultrasound and assessed by FibroScan using CAP probe was 66%. Among patients with fatty liver on ultrasound who have NAFLD, the distribution of liver fat levels was as follows: S1 = 20.5%; S2 = 27%; S3 = 52.5%. The stages of liver fibrosis were: F0 - F1 at 74.5%; significant fibrosis at 25.5%, advanced fibrosis at 11%, and cirrhosis at 6%. NAFLD patients exhibited higher body mass index (BMI), waist circumference, cholesterol, triglyceride, type 2 diabetes mellitus (T2DM), and obesity rates compared to those without NAFLD. ***Conclusion:*** The study underscores the high prevalence of NAFLD in patients with fatty liver detected by ultrasound, with the highest proportion of patients in stage S3, while 25.5% of the cases are classified as significant fibrosis, which indicates a considerable level of liver damage.

Keywords: Fatty liver; Non-alcoholic fatty liver disease; Ultrasound; FibroScan; Steatohepatitis.

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INTRODUCTION

Non-alcoholic fatty liver disease has continued to rise as a predominant cause of chronic liver disease worldwide, particularly in light of the ongoing global obesity and T2DM pandemics [1]. Recent studies have shown an alarming increase in the prevalence of NAFLD, with an estimated 25 - 30% of the global population affected as of 2024 [2]. The increasing burden of NAFLD is particularly pronounced in Asia, where urbanization and lifestyle changes have driven higher rates of metabolic syndrome, a key risk factor for NAFLD [3]. Although abdominal ultrasound is a common method for detecting fatty liver, no studies have yet evaluated the prevalence of NAFLD in patients with fatty liver identified by ultrasound.

Fatty liver is increasingly detected by abdominal ultrasound, yet it often receives insufficient attention from both patients and healthcare providers. The most common cause of this condition is NAFLD. This disease can lead to serious complications such as liver fibrosis, cirrhosis, and hepatocellular carcinoma [4]. Additionally, NAFLD is recognized as an independent cardiovascular risk factor, leading to cardiovascular problems even in the absence of traditional risk factors like hypertension or dyslipidemia [5].

One of the new and effective non-invasive methods for assessing liver fat and fibrosis levels is FibroScan with the CAP probe. This technique, similar to conventional ultrasound, is simple and quick to perform, but it provides additional crucial information about the patient's liver condition without causing any complications. As a result, it has been recommended by major liver disease research associations worldwide, including the American Association for the Study of Liver Diseases and the European Association for the Study of the Liver, as a useful screening tool in the diagnosis of NAFLD [6]. Therefore, we conducted this study to: *Determine the prevalence and characteristics of NAFLD, including steatosis and fibrosis, in patients with fatty liver detected by ultrasound.*

MATERIALS AND METHODS

1. Subjects

Including 303 patients aged ≥ 18 who were diagnosed with fatty liver by ultrasound and visited People's Hospital 115 from August 2019 to October 2020.

The sample size is calculated using the following formula:

$$n = Z^2_{1-\frac{\alpha}{2}} \frac{p(1-p)}{d^2}$$

p is the proportion of the study variable, with d as the desired margin of error set at 5%. Since no existing

studies determine the prevalence of NAFLD in patients with ultrasound-detected fatty liver, p is assumed to be 0.5 → n > = 196.

* *Inclusion criteria:* All patients diagnosed with fatty liver by abdominal ultrasound within the last 6 months (Increased Echogenicity: Liver appears brighter than right, Vascular Blurring, Ultrasound beam weakens, making deeper liver areas harder to see [7]); aged ≥ 18 years; able to read and sign the consent form to participate in the study.

* *Exclusion criteria:* Unreliable FibroScan results: Patients with FibroScan results showing an interquartile range (IQR) > 30% or a success rate < 60%; elevated liver enzymes > 100 U/L; cholestasis, hepatic congestion, or presence of ascites; SCD (Exceed testing capacity) > 25mm (probe M) to rule out cases of excess abdominal fat in obese patients, an XL probe is required [8]; pregnancy; poor health status or inability to provide blood samples.

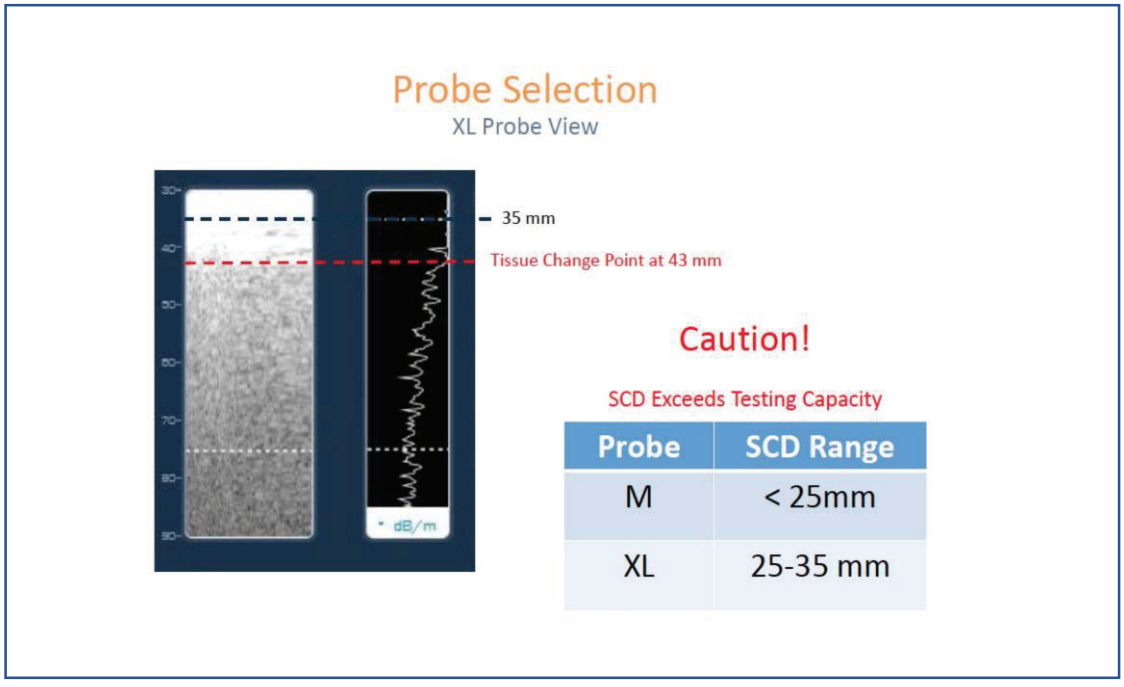

Figure 1. Probe selectition.

* *Location and time*: The study was conducted at People's Hospital 115. Patients who were diagnosed with fatty liver by ultrasound were recruited from August 2019 to October 2020.

2. Methods

* *Study design:* A cross-sectional study.

The FibroScan machine is manufactured by Echosens, a French medical technology company.

The patient fasted for at least 3 hours prior to the measurement. LSM was performed by a hepatology and gastroenterology specialist who has been trained and is proficient in the FibroScan, having conducted over 500 cases up to the time of the study. All patients underwent the procedure with a standard M probe.

Liver fibrosis stages based on kPa values [4]:

- < 7 kPa: Stage F0 - F1;
- ≥ 7 kPa: ≥ F2 (Significant liver fibrosis);
- ≥ 8.7 kPa: ≥ F3 (Advanced liver fibrosis);
- ≥ 11.5 kPa: F4 (Cirrhosis).

Table 1. Liver Steatosis grade [5].

Steatosis grade	Percentage of liver cells with Steatosis	CAP (dB/m)	Remarks
S0	0 - 4%	100 - 233	Normal
S1	5 - 33%	234 - 269	Mild steatosis
S2	34 - 66%	270 - 300	Moderate steatosis
S3	67 - 100%	≥ 301	Severe steatosis

Patients are diagnosed with NAFLD if the CAP score exceeds 233 dB/m (and other causes of steatosis have been excluded [5]:

- Malnutrition: BMI < 18.5 kg/m².
- Parenteral nutrition.
- Hepatitis B or C virus infection (HBsAg (+), Anti-HCV (+)).
- Medications (e.g., amiodarone, methotrexate, tamoxifen, corticosteroids, valproate, antiretroviral drugs).
- Pregnancy.
- Heavy alcohol consumption: ≥ 30 g/day or ≥ 210 g/week for men and ≥ 20 g/day or ≥ 140 g/week for women for at least two consecutive years.

* *Data analysis:* Statistical analysis was performed using SPSS version 22.0.

3. Ethics

The study was approved by the Ethics Committee of People's Hospital 115, Number 108/BV-NCKH, dated October 25, 2019. The authors commit to using licensed data and take full responsibility for it. The authors declare to have no conflicts of interest in the study.

RESULTS

A total of 303 patients were enrolled in this study, with a mean age of 45 ± 12 years. The male-to-female ratio was 1.2:1. Among these patients, 200 (66%) were diagnosed with NAFLD based on the CAP scores obtained via FibroScan.

Table 2. Comparison of characteristics between NAFLD and non-NAFLD patients.

Characteristics	NAFLD (n = 200)	Non-NAFLD (n = 103)	p
BMI (kg/m²) Mean ± SD	24.7 ± 3.6	23.6 ± 3.9	0.002
Waist circumference (cm) Mean ± SD	92.2 ± 10.6	89.8 ± 8.4	0.032
Obesity (%)	44	26.2	0.007
Central obesity (%)	77	71.8	0.325
T2DM (%)	25	13.6	0.021
Dyslipidemia (%)	98	97.1	0.616
Metabolic syndrome (%)	70	59.2	0.06
Glucose (mmol/L) Mean ± SD	6.7 ± 3.6	6.1 ± 2.4	0.084
Cholesterol (mmol/L) Mean ± SD	5.3 ± 1.8	4.8 ± 1.5	0.009
Triglycerides (mmol/L) Median (min - max)	2.6 (1.8 - 3.8)	2.1 (1.6 - 3.1)	0.026
HDL - C (mmol/L) Mean ± SD	1.0 ± 0.3	1.0 ± 0.3	0.655
LDL - C (mmol/L) Mean ± SD	3.3 ± 1.1	3.3 ± 1.2	0.939
ALT (U/L) Median (min - max)	45.2 (27.1 - 77.5)	44.8 (28.1 - 78.5)	0.843
AST (U/L) Median (min - max)	31.5 (23.3 - 50.6)	30.7 (24.5 - 52.8)	0.764

Patients with NAFLD have a higher BMI, larger waist circumference, and higher obesity rates compared to those with fatty liver on abdominal ultrasound without NAFLD. These patients also have higher levels of cholesterol and triglycerides.

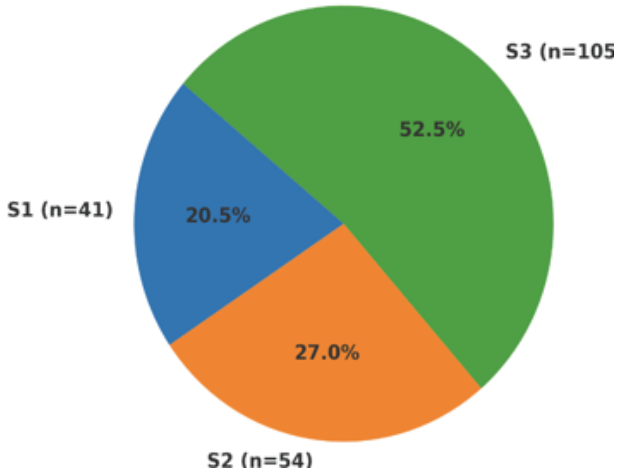

Figure 2. The degree of steatosis in patients with NAFLD.

Table 3. Stages of fibrosis in patients with NAFLD.

Liver fibrosis	Number (n)	Percentage (%)
F0, F1	149	74.5
F2	29	14.5
F3	10	5
F4	12	6
Significant fibrosis (≥ F2)	51	25.5
Advanced fibrosis (≥ F3)	22	11.0
Cirrhosis (F4)	12	6.0

It highlights that 25.5% of the cases are classified as significant fibrosis (≥ F2), which indicates a considerable level of liver damage.

DISCUSSION

The demographic profile of our study population, with a mean age of 45 years and a male-to-female ratio of 1.2:1, reflects the typical demographic distribution of NAFLD, which tends to affect middle-aged individuals with a slight male predominance. This is consistent with other studies showing that NAFLD is more prevalent in middle-aged adults, particularly those with metabolic risk factors such as obesity and T2DM [3]. The high prevalence of NAFLD among patients with these risk factors underscores the importance of early screening and intervention in high-risk

populations [4]. The present study highlights the high prevalence of NAFLD among patients with fatty liver detected by ultrasound.

The findings show that NAFLD patients have significantly higher BMI, waist circumference, and obesity rates. These results strongly support the link between NAFLD and metabolic syndrome, including obesity and T2DM.

The use of FibroScan, particularly the CAP score, proved effective in non-invasively assessing the degree of steatosis and fibrosis [6]. The high prevalence of severe steatosis (S3) in 52.5% of our patients is consistent with findings from other studies utilizing FibroScan, which have reported similar rates of advanced steatosis in Asian populations [2]. Additionally, the prevalence of significant fibrosis (≥ F2) in 25.5% of our patients and cirrhosis in 6% aligns with findings from a recent meta-analysis that reported fibrosis prevalence ranging from 5 - 25% in NAFLD patients [8].

The findings of this study align with recent literature that underscores the high prevalence of NAFLD in patients with metabolic risk factors. Recent studies from 2022 - 2024 have further emphasized the role of metabolic syndrome in accelerating the progression of NAFLD to non-alcoholic steatohepatitis (NASH) and cirrhosis [7]. A comprehensive meta-analysis in 2023 reported that nearly 20% of NAFLD patients progress to NASH, with significant fibrosis present in 10 - 15% of cases [9]. These findings are consistent with the results of our study, which identified a high prevalence of significant fibrosis (25.5%) and cirrhosis (6%) among NAFLD patients.

Emerging evidence also suggests that the burden of NAFLD-related liver disease is likely to increase in the coming years due to the aging population and the rising prevalence of metabolic syndrome [10]. Moreover, the integration of advanced imaging techniques, such as FibroScan has enabled more accurate quantification of hepatic steatosis and fibrosis, offering new avenues for early diagnosis and monitoring of disease progression.

The strength of this study lies in its comprehensive assessment of steatosis and fibrosis using FibroScan, a non-invasive and widely accessible tool. However, the cross-sectional design limits our ability to draw causal inferences or assess the natural progression of NAFLD. Furthermore, the study population was predominantly from an urban setting, which may limit the generalizability of the findings to rural populations.

CONCLUSION

The study found the prevalence of NAFLD in patients with fatty liver detected by ultrasound to be 66%. The distribution of liver fat levels indicated the highest proportion of patients in stage S3 (52.5%), while 25.5% of the cases are classified as significant fibrosis (≥ F2), which indicates a considerable level of liver damage. NAFLD patients exhibited higher BMI, waist circumference, and obesity rates compared to those without NAFLD.

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COMPARISON OF FUNCTIONAL OUTCOMES BETWEEN ADJUSTABLE- AND FIXED-LOOP DEVICES FOR FEMORAL FIXATION IN ARTHROSCOPIC ANTERIOR CRUCIATE LIGAMENT RECONSTRUCTION

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Nguyen Thanh Thao[3], Vu Nhat Dinh[1]

Abstract

Objectives: To compare outcomes of anterior cruciate ligament reconstruction (ACLR) with adjustable- and fixed-loop devices. ***Methods:*** A retrospective, observational study was conducted on 92 patients who underwent ACLR with the fixation of a hamstring graft with the fixed- and adjustable-loop suspensory devices on the femoral side from December 2021 to December 2023. Knee function was evaluated using the Lysholm score, Lachman test, and Pivot-shift test, both preoperatively and at the one-year postoperative follow-up. ***Results:*** One year postoperatively, the Lysholm score averaged 90.62 ± 4.167 in the adjustable-loop group, with 83.1% of cases achieving good grades. In comparison, the fixed-loop group had a mean score of 90.15 ± 4.704, with 77.8% of cases obtaining good grades. However, no significant statistical difference was found between the two groups ($p > 0.05$). A negative pivot shift test was confirmed in 60 cases (92.3%) from the adjustable-loop group and 24 cases (88.9%) from the fixed-loop group ($p = 0.5$). No cases of infection, graft failure, or flexion limitation were recorded. ***Conclusion:*** There were no notable differences in graft laxity and functional outcomes between the fixed- and adjustable-loop devices for femoral fixation in arthroscopic ACLR.

Keywords: Anterior cruciate ligament; Adjustable-loop; Fixed-loop; Suspensory fixation.

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INTRODUCTION

Injuries to the anterior cruciate ligament (ACL) are common, with an annual incidence of approximately 5 per 10,000 individuals, especially among high-impact athletes [1]. Arthroscopic ACLR is considered a standard treatment, with the objective of restoring knee stability and mechanics. Well-defined tunnel placement and reliable graft fixation are important for optimal outcomes. Suspensory fixation techniques, including fixed-loop and adjustable-loop systems, allow the use of longer grafts for femoral tunnels. Fixed-loop devices diminish slippage and ensure graft strength via external cortical bone support and ribbon fixation. However, their capabilities may be limited in short graft tunnels or by improper graft positioning. Adjustable-loop systems, with a finger-trap mechanism, are preferable in short tunnels, supporting more graft material in the tunnel without over-drilling. These systems allow intraoperative tightening to optimize graft placement and minimize complications such as femoral attic formation or the "bungee cord effect" [2]. While fixed-loop devices secure fixation and maintain graft strength, the impact of loop type and tunnel length on clinical outcomes remains unclear. A limited number of studies have compared fixed-loop and adjustable-loop systems in ACLR with hamstring grafts [3, 4]. This study aims to: *Compare two suspensory systems for femoral tunnel on graft laxity and functional outcomes, including Lysholm knee scores, in arthroscopic ACLR.*

MATERIALS AND METHODS

1. Subjects

Including 92 cases suffering from ACL injuries who underwent arthroscopic ACLR at the Department of Joint Surgery, Military Hospital 103 from December 2021 to December 2023.

* *Inclusion criteria:* Aged ≥ 18 years; primary ACL surgery; unilateral ACL tear without additional ligament injuries; no prior knee surgeries; and clinically and MRI-confirmed ACL rupture.

* *Exclusion criteria:* Multiple ligament injuries or significant cartilage damage; alternative femoral fixation techniques; severe osteoarthritis (Kellgren-Lawrence grade 3 - 4) or advanced osteoporosis; and bilateral ACL injuries.

Patient demographics, diagnostic findings, surgical data, and follow-up details were collected from the hospital database, with additional data obtained through phone-based follow-up scheduling during 1 year. All cases in both groups were managed with a similar surgical technique and postoperative protocol.

2. Methods

* *Study design:* A retrospective, observational study.

* *Surgical procedure:* Following arthroscopic confirmation of an ACL tear, a tripled hamstring tendon graft was harvested and pre-tensioned. The femoral tunnel, drilled through the anteromedial portal with the knee in hyper-flexion, was sized to match the graft. The graft was arthroscopically passed through the tunnels with fixed- or adjustable-loop devices. Arthroscopic reassessment confirmed graft tension, with re-tensioning carried out to ensure firm positioning in the femoral socket using alternating traction on the white strands. Knee range of motion was evaluated to verify graft stability and rule out notch impingement. All procedures were performed by senior surgeons, with perioperative antibiotics administered according to institutional guidelines.

* *Rehabilitation procedure:* From the second postoperative day, patients were instructed to bear weight as tolerated using crutches and a knee brace locked in extension. During the first two weeks, rehabilitation focused on patellar mobilization, reaching flexion up to 90° and full passive extension. By 6 weeks, progression to full knee flexion was encouraged, while active terminal extension was restricted until this point. Patients were gradually weaned off the brace and crutches upon demonstrating adequate quadriceps control. The knee brace was recommended for the first 4 weeks and discontinued based on patient comfort. Return to sports was considered after 6 - 8 months.

* *Study variable:* Outcomes were assessed through the Lysholm score, Lachman test, and pivot shift test [2]. The Lysholm score classified results as excellent (from 95 - 100), good (from 84 - 94), fair (from 65 - 83), or poor (< 65). Both groups show comparable demographics and similar preoperative and intraoperative variables. Follow-up evaluations occurred at one year post-surgery.

* *Statistical analysis:* Data analysis was conducted utilizing SPSS software (version 20.0, IBM Corp., USA). Categorical variables were assessed via the Chi-square test, while continuous variables were analyzed with the paired T-test. Statistical significance was determined at a p-value threshold of less than 0.05.

3. Ethics

The research was approved by the Institutional Ethics Committee (No. 192/HĐĐĐ, June 15th 2022). The Department of Joint Surgery, Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS

This study included 92 cases, comprising 79 males (85.9%) and 13 females (14.1%), with an average age of 32.15 ± 10.38 years (range: 19 - 60). Sports-related accidents accounted for 60 cases (65.2%) of ACL injuries, followed by traffic accidents (28 cases, 28.3%) and routine activities (7 cases). The mean interval between injury and surgery was 13.26 ± 14.75 weeks (range: 1 - 72), and meniscal injuries were present in 28 cases (30.4%). The mean hospital stay was 14.27 ± 4.91 days (range: 6 - 31). No significant differences were detected between the groups in demographic characteristics or injury profiles, including age, gender, injury mechanism, affected side, meniscal injury rate, or timing of surgery and hospitalization (*Table 1*).

Table 1. Demographic data and group characteristics (n = 92).

Parameter		Adjustable-loop	Fixed-loop	p
Mean age		31.46 ± 9.757	33.81 ± 11.793	0.325
Gender	Male	57 (87.7%)	22 (81.5%)	0.436
	Female	8 (12.3%)	5 (18.5%)	
Injury causes	Sports injuries	45 (69.2%)	15 (55.6%)	0.442
	Routine activity injuries	16 (24.6%)	10 (37.0%)	
	Traffic accidents	4 (6.2%)	2 (7.4%)	
Time from injury to surgery (weeks)		11.91 ± 12.237	16.52 ± 3.740	0.174
Meniscus tear	Yes	43 (66.2%)	21 (77.8%)	0.270
	No	22 (33.8%)	6 (22.2%)	
Side involved	Right	34 (52.3%)	19 (70.4%)	0.110
	Left	31 (47.7%)	8 (29.6%)	
Length of hospital stay (days)		14.31 ± 4.776	14.19 ± 5.321	0.914

Preoperative Lysholm scores revealed poor knee function in 95.7% of cases, with a mean score of 54.48 ± 5.49 (range: 42 - 66). One year postoperatively, substantial improvements were noted, with 81.5% of cases achieving good outcomes and 18.5% fair outcomes based on the Lysholm score. Preoperative evaluations, including the Lachman test, pivot-shift test, and Lysholm score, displayed no significant differences between the two groups ($p > 0.05$).

Table 2. Preoperative clinical assessment.

Parameter		Adjustable-loop	Fixed-loop	p
Lachman test	Grade 1	4 (6.2%)	1 (3.7%)	0.570
	Grade 2	13 (20%)	8 (29.6%)	
	Grade 3	48 (73.8%)	18 (66.7%)	
Pivot-shift test	Grade 1	6 (9.2%)	3 (11.1%)	0.367
	Grade 2	32 (49.2%)	17 (63.0%)	
	Grade 3	27 (41.5%)	7 (25.9%)	
Lysholm score		54.25 ± 5.640	55.04 ± 5.185	0.533
Poor		61 (93.8%)	27 (100%)	0.188
Fair		4 (6.2%)	0	

One-year postoperative evaluations indicated notable improvements in both groups. The mean Lysholm score increased by 36.00 ± 3.419 (range: 28 - 42). Most cases achieved grade 0 in the Lachman and pivot-shift tests, with no significant intergroup differences ($p > 0.05$). Although both groups showed significant postoperative Lysholm score improvements, the extent of improvement was not statistically different ($p = 0.108$) (*Table 3*). No complications, such as infections, graft failures, or flexion restrictions, were observed in either group.

Table 3. Postoperative clinical assessment at 1 year.

Parameter		Adjustable-loop	Fixed-loop	p
Lachman test	Grade 0	54 (83.1%)	21 (77.8%)	0.551
	Grade 1	11 (16.9%)	6 (22.2%)	
Pivot-shift test	Grade 0	60 (92.3%)	24 (88.9%)	0.596
	Grade 1	5 (7.7%)	3 (11.1%)	
Lysholm score		90.62 ± 4.167	90.15 ± 4.704	0.467
Fair		11 (16.9%)	6 (22.2%)	0.551
Good		54 (83.1%)	21 (77.8%)	
Change in Lysholm score		36.37 ± 3.773	35.11 ± 2.172	0.108

DISCUSSION

Our study evaluated the functional results of arthroscopic ACLR with fixed-loop and adjustable-loop systems through graft laxity and Lysholm knee scores. The average ages in the fixed- and adjustable-loop groups were 34.5 ± 11 and 34.1 ± 9.1 years, respectively, comparable to the findings of Chandru et al. [3], who reported mean ages of 33.81 ± 11.7 and 31.46 ± 9.7 years. Meniscal injuries were recorded in 77.8% of fixed-loop cases and 66.2% of adjustable-loop cases in this study, consistent with Schützenberger et al., who reported a 64% prevalence of meniscal tears in ACL injuries [4]. Adjustable-loop devices, as advanced femoral cortical suspension systems, offer distinct advantages, including eliminating the need to pre-calculate loop length or over-drill the femoral tunnel. They also permit maximal graft insertion in short femoral tunnels, reducing the need for multiple fixed-loop sizes in inventory. In this study, most cases required reconstruction due to grade 2 or 3 instability, as assessed by the Pivot-shift and Lachman tests preoperatively. Postoperatively, knee stability was regained in most cases, with 81.5% testing negative for the Lachman test and 91.3% negative for the Pivot-shift test. Chandru et al. [3] reported that 92.3% of cases in both the fixed- and adjustable-loop groups showed negative graft laxity tests after one year. Asif et al. [6] demonstrated that 87% of cases with a variable-loop device were negative for the Lachman test, and 95.7% were negative for the Pivot-shift test. Shahpari et al. [7] reported negative Lachman and Pivot-shift tests in 81.8% and 87% of fixed-loop cases, respectively. Regarding functional outcomes, Lysholm knee scores in our study were 90.15 and 90.62 in the fixed- and adjustable-loop groups, respectively, at one year, comparable to Chandru et al. [3] (91.54 and 91.69) and Asif et al. [6] (91.4 and 91.0). While no significant difference in scores was observed at one year, our study noted a statistically significant difference at six weeks postoperatively. To the best of our knowledge, this is the only study to periodically compare Lysholm knee scores between fixed- and adjustable-loop groups over one year post-surgery, permitting for an assessment of functional improvement over time. The Lysholm score, a generally used knee-specific scoring system, has revealed acceptable test-retest reliability and internal consistency in a study by Briggs et al. including over 1000 ACLR cases [8]. Its concise and informative nature justified its use as the sole

measure for patient-reported outcomes in this study. Graft laxity was assessed clinically using Lachman and anterior drawer tests by comparing the affected and normal knees, as arthrometer use was not feasible. Despite a longer one-year follow-up period, the study reported no statistically significant difference in graft laxity between the two groups. Some in vitro studies suggest that adjustable-loop devices may be inferior to fixed-loop devices due to elongation under cyclical loads, potentially compromising graft function during the critical 8 - 12 weeks of early recovery. However, other studies report no notable discrepancies between the two devices in terms of functional outcomes [9]. In our study, Lysholm scores and knee laxity assessments (Lachman and pivot shift tests) at the last follow-up were consistent with prior findings on fixed-loop cortical-suspension devices. The gap between biomechanical and clinical studies may arise from limitations in laboratory simulations, which fail to mimic the complex in vivo biomechanical and physiological environment. Graft healing plays a crucial role in the success of ACLR and is influenced by factors including graft type, tunnel length and orientation, graft length within the tunnel, tunnel-graft diameter mismatch, graft tension, motion within the tunnel, and fixation type. Our study focused only on comparing two fixation methods, not mentioning other contributing factors. Although biomechanical studies often suggest increased graft slippage in variable-loop designs compared to fixed-loop designs, clinical studies, including ours, have not illustrated this difference, indicating that biomechanical findings may not fully represent clinical outcomes.

Our study had several limitations. Knee stability was assessed subjectively through the Lysholm score, Lachman test, and pivot shift test rather than instrument-assisted methods like an arthrometer. Nevertheless, all evaluations were performed by a senior professor with over 30 years of clinical experience who was blinded to the surgical method, reducing subjective bias.

CONCLUSION

Our study showed no statistically significant difference in laxity of the graft or functional outcomes of arthroscopic ACLR with fixed- and variable-loop devices at one year of follow-up. These findings offer important evidence to assist surgeons in choosing between the two devices, considering factors like surgical technique preference, cost, or patient-specific needs, without worrying about variations in clinical outcomes.

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CLINICAL OUTCOMES OF TRANSURETHRAL THULIUM LASER ENUCLEATION OF THE PROSTATE IN THE MANAGEMENT OF BENIGN PROSTATIC HYPERPLASIA

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Abstract

Objectives: To evaluate the safety and efficacy of Thulium laser enucleation of the prostate (ThuLEP) as a treatment for benign prostatic hyperplasia (BPH), with a focus on clinical, functional, and anatomical outcomes. ***Methods:*** A prospective, cross-sectional study was conducted on 92 patients diagnosed with BPH at Military Hospital 175 from September 2023 to December 2024. Data were collected on preoperative and postoperative clinical symptoms (IPSS score), urinary function (Qmax and PVR), anatomical outcomes, and quality of life (QoL). Comparative analyses were performed, and results were benchmarked against international literature. ***Results:*** A total of 92 patients diagnosed with BPH, with an average prostate volume of 72.48mL, were included in the study. The average surgery duration was 84.5 minutes, and hemoglobin dropped by 1.1 g/dL postoperatively. The mean IPSS improved significantly from 28.7 preoperatively to 1.3 at six months postoperatively. Qmax increased from 2.7 mL/s to 23.1 mL/s, and prostate volume decreased by more than 50% on average. At six months, 94.2% of patients achieved excellent treatment outcomes, with no severe complications observed. Additionally, patients reported substantial improvements in QoL, with mean QoL scores dropping from 5.8 preoperatively to 0.1. ***Conclusion:*** ThuLEP demonstrates safety and efficacy in managing BPH, offering significant symptom relief, functional improvements, and favorable anatomical outcomes.

Keywords: Lower urinary tract symptom; Benign prostatic hyperplasia; Thulium laser enucleation of the prostate.

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INTRODUCTION

Benign prostatic hyperplasia is a prevalent condition among aging men, significantly affecting their QoL due to lower urinary tract symptoms (LUTS). Cornu JN et al. (2015) highlighted that traditional surgical techniques such as monopolar transurethral resection of the prostate (TURP) and open prostatectomy (OP) have been longstanding standards for managing BPH. However, these methods are associated with considerable perioperative risks, prolonged hospital stays, and higher complication rates [1]. Chen CH et al. (2020) demonstrated that anatomical endoscopic enucleation of the prostate (AEEP) represents a transformative advancement in BPH management. Techniques such as holmium laser enucleation of the prostate (HoLEP) and ThuLEP offer minimally invasive alternatives with efficacy and safety profiles surpassing traditional TURP and OP. These approaches are particularly effective for managing larger prostates [2]. ThuLEP, as highlighted by Herrmann TR et al. (2010) and Bozzini G et al. (2021), combines precise tissue dissection with exceptional hemostatic capabilities. Advantages such as reduced operative blood loss, shorter catheterization times, and reduced hospitalization durations position ThuLEP as a strong alternative to conventional procedures [3, 4]. Moreover, You C et al. (2021) emphasized that the en-bloc enucleation technique further enhances surgical efficiency and precision, particularly for large prostates [5]. Despite its advantages, ThuLEP's adoption remains limited in some regions due to its steep learning curve and the variability of available equipment. Zhang Y et al. (2019) noted that in Vietnam, the application of ThuLEP is still in its early stages, with few centers performing the procedure or studying its outcomes [6]. This study aims to: *Evaluate treatment results of BPH by ThuLEP at Military Hospital 175.*

MATERIALS AND METHODS

1. Subjects

Including 92 patients diagnosed with BPH.

* *Inclusion criteria:* Indication for surgical intervention due to complications such as recurrent urinary retention, bladder stones, or failed medical therapy; ability to tolerate spinal or general anesthesia; histopathological confirmation of BPH postoperatively.

* *Exclusion criteria:* Patients with neurogenic bladder, prostate cancer, or prior prostate surgeries.

* *Location and time:* At the Department of Urology, Military Hospital 175, from September 2023 to December 2024.

The study focused on evaluating clinical outcomes, urinary function, and safety of Transurethral ThuLEP in the management of BPH.

2. Methods

* *Study design:* A prospective, cross-sectional study.

* *Surgical procedure:* All patients underwent ThuLEP using a continuous-wave Thulium laser system and morcellator. The enucleation technique involved three key steps: Creating an initial incision, enucleating the prostate lobes en bloc, and morcellating the extracted tissue for removal. Saline was used as the irrigation fluid to minimize electrolyte disturbances.

* *Data collection:*

Preoperative assessments: International Prostate Symptom Score (IPSS), maximum urinary flow rate (Qmax), and QoL scores; prostate volume measured via transabdominal ultrasound; hematological and biochemical parameters.

Intraoperative data encompassed operative time, enucleation speed, blood loss (measured by hemoglobin reduction), and complications. Postoperative outcomes, including IPSS, Qmax, QoL, and residual prostate volume, were recorded at 1, 3, and 6 months follow-up. The treatment efficacy recorded postoperatively is shown in table 1.

Table 1. Estimate criteria for the efficacy of BPH treatment.

Efficacy	Symptoms (Post/Pre IPSS)	Function (Post - Pre Qmax)	Anatomy (Post/Pre PV)	QoL (Post - Pre QoL)
Excellent	≤ 0.25	≥ 10	≤ 0.5	≥ 4
Good	≤ 0.5	≥ 5	≤ 0.75	3
Fair	≤ 0.75	≥ 2.5	≤ 0.9	2 or 1
Poor	> 0.75	< 2.5	> 0.9	≤ 0

(Overall outcomes are determined by the average results of three primary criteria: Symptoms, function, and QoL)

Source: Homma Y et al. (1996) [7].

* *Statistical analysis:* Data were analyzed using SPSS software. Continuous variables were expressed as Mean ± Standard Deviation (SD) and compared using paired T-tests. Categorical variables were expressed as percentages and compared using Chi-square tests. A p-value of < 0.05 was considered statistically significant.

3. Ethics

The study was approved by the Ethics Council in Biomedical Research of the Vietnam Military Medical University with approval number 07/2022/CNChT-HĐĐĐ on December 12, 2022, and the Ethics Council in Biomedical Research of the Military Hospital 175 with approval number 3377/GCN-HĐĐĐ on September 6, 2023. Military Hospital 175 granted permission for the use and publication of the research data, in full compliance with relevant legal regulations. The authors declare to have no conflicts of interest related to this study.

RESULTS

This study was conducted on 92 patients with BPH treated using the ThuLEP technique at Military Hospital 175, from September 2023 to December 2024. The findings are summarized in the tables below.

Table 2. Baseline characteristics and preoperative clinical data.

Parameter	Mean ± SD	Range
Age (years)	69.38 ± 7.09	50 - 87
Prostate volume (mL)	72.48 ± 33.79	31 - 183
IPSS (score)	28.65 ± 4.23	12 - 35
Qmax (mL/s)	2.66 ± 3.10	0 - 9.6
QoL (score)	5.79 ± 0.41	5 - 6
Surgical time (minutes)	84.46 ± 35.63	30 - 180
Reduction in red blood cells (T/L)	0.34 ± 0.39	0.01 - 1.60
Reduction in hemoglobin (g/dL)	1.06 ± 1.12	0.02 - 5.10
Sodium reduction (mmol/L)	0.06 ± 2.83	-8 - 9
Catheterization duration (days)	2.45 ± 1.53	1 - 13
Hospital stay (days)	3.51 ± 1.72	1 - 14

The majority of patients were aged 60 - 79 years. Most presented with severe LUTS, poor QoL, and impaired urinary function preoperatively.

Table 3. Surgical outcomes and symptom improvement postoperatively.

Parameter	IPSS (score)	Qmax (mL/s)	QoL (score)	Prostate volume (mL)
Preoperative (n = 92)	28.65 ± 4.23	2.66 ± 3.10	5.79 ± 0.41	72.48 ± 33.79
1 month (n = 82)	3.18 ± 2.03	19.06 ± 6.52	0.62 ± 0.54	25.47 ± 12.64
3 months (n = 58)	1.98 ± 1.69	19.62 ± 6.46	0.34 ± 0.55	-
6 months (n = 52)	1.25 ± 1.01	23.09 ± 9.17	0.13 ± 0.35	-

Significant improvements were observed across all postoperative timepoints in LUTS, urinary function, and QoL, with progressive enhancement noted up to 6 months. Prostate volume reduction was sustained postoperatively.

Table 4. Overall treatment effectiveness rates.

Timepoint	Excellent (%)	Good (%)	Fair (%)	Poor (%)
1 month (n = 82)	93.1	5.7	0.4	0.8
3 months (n = 58)	92.6	6.3	1.1	0
6 months (n = 52)	94.2	5.1	0.6	0

Excellent outcomes were maintained across all time points, reflecting sustained treatment effectiveness. No poor outcomes were observed after 3 or 6 months of follow-up.

DISCUSSION

1. Patient characteristics

Our study population had a mean age of 69.38 ± 7.09 years, primarily within the 60 - 79 age range, consistent with the demographic most affected by BPH. Preoperative assessments revealed severe LUTS, with a mean IPSS of 28.65 ± 4.23, and a markedly low mean Qmax of 2.66 ± 3.10 mL/s. These findings indicate significant urinary obstruction and reduced QoL preoperatively.

When compared to other studies, Zhang Y et al. (2019) and Huang SW et al. (2019) similarly reported severe LUTS in BPH patients within comparable age groups. Their work highlights the consistent burden of BPH across populations, validating the clinical profiles observed in our cohort [6].

Additionally, Pang KH et al. (2022) emphasized the substantial urinary obstruction, comparable to our findings, underscoring the urgent need for effective surgical intervention in these patients [8].

2. Intraoperative outcomes

The operative metrics in our study demonstrated the efficacy of the ThuLEP procedure. The mean operative time of 84.46 ± 35.63 minutes and minimal blood loss (mean reductions in red blood cell count: 0.34 ± 0.39 T/L; hemoglobin: 1.06 ± 1.12 g/dL) highlight the precision and hemostatic advantages of this technique. The negligible sodium reduction (0.06 ± 2.83 mmol/L) further underscores the safety of saline as the irrigation fluid. These findings suggest that ThuLEP not only ensures surgical efficiency but also minimizes perioperative complications.

Comparatively, Kim YJ et al. (2015) reported similar operative times for ThuLEP, confirming the reproducibility of efficient surgical workflows [9]. Herrmann TR et al. (2010) highlighted the hemostatic benefits of the Thulium laser, which align with the minimal blood loss observed in our study [3]. Cornu JN et al. (2015) corroborated the safety of saline irrigation, emphasizing its role in maintaining electrolyte stability during enucleation [1].

3. Overall treatment effectiveness

Our findings demonstrated sustained and significant improvement in LUTS, urinary function, and QoL across all follow-up intervals. At six months, the mean IPSS reduced from 28.65 ± 4.23 to 1.25 ± 1.01, while the mean Qmax increased from 2.66 ± 3.10 mL/s to 23.09 ± 9.17 mL/s. Notably, 94.2% of patients achieved excellent treatment outcomes, with no poor outcomes observed beyond 3 months. This underscores the durability and consistency of ThuLEP in addressing both functional and anatomical aspects of BPH.

In comparison, Chen CH et al. (2020) and Huang SW et al. (2019) reported similarly high rates of symptom relief and functional improvement with laser enucleation techniques [2, 10]. Anatomical outcomes in our study, characterized by a reduction in prostate volume exceeding 50%, align closely with the criteria established by Homma Y et al. (1996) for excellent anatomical improvement. The findings of Zhang Y et al. (2019) further reinforce the consistency of our results, highlighting the broad applicability of ThuLEP in diverse patient populations [6].

4. Comparison with other techniques

Compared to other enucleation techniques, such as HoLEP, ThuLEP

demonstrated superior hemostatic control and faster recovery times. You C et al. (2021) noted that ThuLEP had lower complication rates than TURP and HoLEP, particularly in patients with large prostates exceeding 80mL [5]. The "All-in-One" ThuLEP technique described by Kim YJ et al. (2015) further optimized operative efficiency and success rates, consistent with our findings [9]. Additionally, Bozzini G et al. (2021) emphasized the suitability of ThuLEP for treating large prostates, underscoring its versatility [4].

CONCLUSION

This study demonstrates the safety and efficacy of ThuLEP in treating BPH, involving 92 patients with an average prostate volume of 72.48mL. The procedure resulted in over 90% of patients experiencing excellent outcomes in terms of symptom relief, urinary function, and anatomical improvements. These results reinforce ThuLEP as a reliable treatment option for BPH with proven outcomes. Further research with larger cohorts and longer follow-ups is necessary to evaluate its long-term results.

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A CASE REPORT: CARCINOMA AFTER TRAUMATIC BRAIN INJURY CAUSED BY INCENDIARY WEAPONS

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Abstract

Carcinoma in patients with traumatic brain injury having retained metal foreign bodies does not commonly exist. Complications of local cancer by traumatic brain injury, having retained metal foreign bodies can occasionally occur. Changes mainly appear at the wound due to chronic inflammation and cell hyperplasia, which leads to cancer. The clinical case the authors introduce is a patient participating in the Resistance War for National Salvation who was shot by artillery pieces in the right temporal. The patient was admitted to the hospital and had experienced surgery to remove a tumor from the right temporal. The histopathology of the tumor was carcinoma. After surgery, the patient was awake, the incision recovered well, and the patient could live normally.

Keywords: Carcinoma; Traumatic brain injury; Surgery.

INTRODUCTION

Cancer in chronic wounds was first described by Jean-Nicolas Marjolin in 1828. Especially wounds with retained foreign bodies in the head and neck area can result in carcinoma, making up to 80%[1]. Early surgery to remove foreign bodies is needed to prevent carcinoma at the place. The authors present a rare case with the aim to: *Evaluate the status of in situ cancer in a patient with a metal foreign body from a resistance war wound many years ago.*

CASE REPORT

Patient Pham Van P, male, 72 years old, was presented to the hospital due to pain and swelling in his right temple and a mild fever of 37.4°C. The patient participated in the resistance war in 1972 and was hit by shell fragments in the right temple area.

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Since then, the patient has been living with a foreign metal fragment under the scalp in the right temple area. The wound is swelling, recurring many times, and gradually getting bigger. The patient had sleep disorders, dizziness, and weight loss (lost 5kg/2 months). He was admitted to the hospital with a Glasgow score of 15 points, felt pain in the right temporal region, had no paralysis, no epilepsy, and no meningeal syndrome. In the right temple area, there is a shiny mass under the scalp, about 8 x 10cm in size. A computed tomography scan of the brain shows the parenchymal window with oval heterogeneous hyperdense mass including the subcutaneous area, skull, and the subscapular area adjacent to dura mater, bone window with the image of skull bone destruction in the tumor area, metal foreign body image under the skin in the tumor area. The patient underwent surgery to remove the tumor, skull bone, and invaded dura mater. Dural reconstruction with artificial dura mater and cranioplasty with titanium brain mesh. After surgery, the patient had no complications, and the incision recovered well. The patient was recharged from the hospital and lives normally. Histopathology results: Basal cell carcinoma.

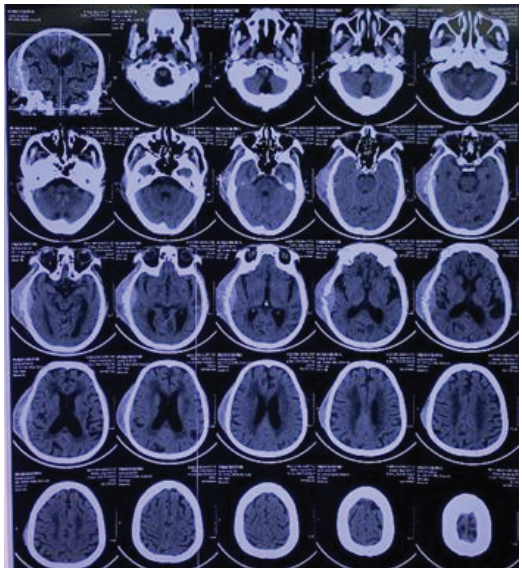

Figure 1. Preoperative computed tomography image of the brain - metal foreign body under the skin of the right temple.

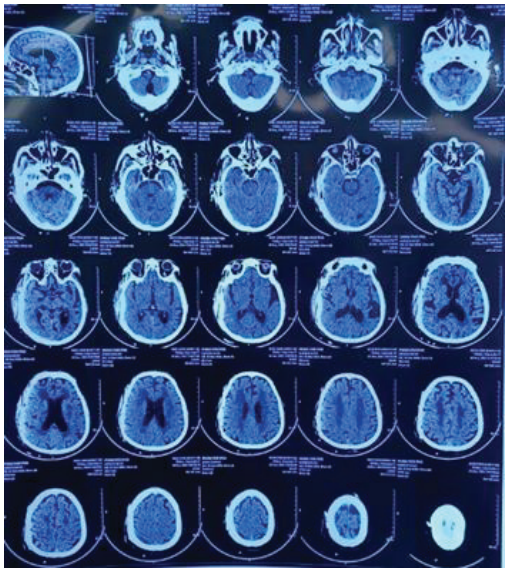

Figure 2. Post-operative computed tomography image of the brain - the tumor in the right temporal region was completely removed.

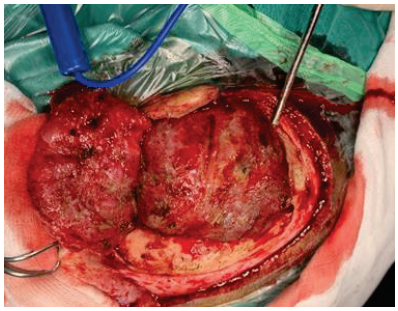

Figure 3. Exposing tumor.

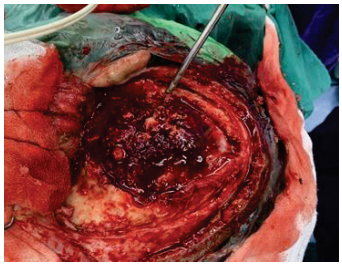

Figure 4. Tumor invades the cerebral dura mater.

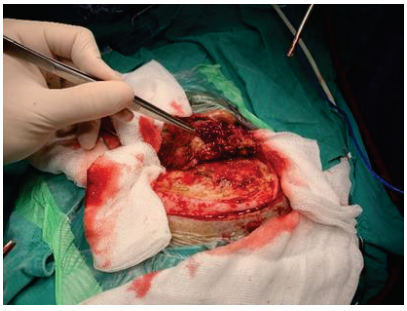

Figure 5. Tumor removal.

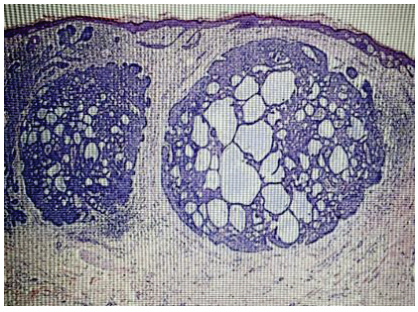

Figure 6. Histopathology.

DISCUSSION

Wounds resulting from retained foreign objects are common in Vietnam. Foreign bodies left on the body after war can be anywhere-under the skin or in the brain parenchyma or lung parenchyma. Metal foreign bodies in the head area can be in the brain parenchyma, skull, or under the scalp. We can meet patients who still have metal foreign bodies in their bodies and live peacefully with these. However, authors around the world have mentioned that cancer is due to lingering foreign objects in the wound. Verifying the relationship between cancer and lasting foreign body injuries is still difficult. Whether the retained metal foreign body is a direct cause of carcinoma remains controversial. However, some hypotheses have been proposed that the poor vascularity of the scar tissue facilitates the development of ulceration in the thin epidermis, making it more susceptible to changes and infection. The retained foreign body will result in changes in epithelial cells during the regeneration process. Dysplasia will occur when the repair process is repeated in chronic wounds, leading to carcinoma in local tissue [2, 3, 4].

The location of the wound by a foreign body is also a crucial factor resulting in carcinoma. The authors believe that location exposure to sunlight will increase the risk of the disease [4, 5]. Ozyazgan I [6] studied 92 patients with basal cell carcinoma, which is common in the elderly, poor wound healing, and exposed skin locations. These are all related to ultraviolet rays and become factors increasing the risk of cancer. Our patient suffered a metal foreign body wound under the scalp in the right temple area. As this is a location that is less covered and usually exposed to sunlight, it will raise the risk of cancer at the wound site.

Wozniak SE describes a patient with a war-related foreign body wound in his hand that had complications causing basal cell carcinoma. The authors believe that these wounds are related to the Marjolin degeneration process of epithelial cells and cause carcinoma in the long term [2]. Ebrahimzadeh (2013) described a case of a 44-year-old male patient with a metal fragment in the lower end of the femur while participating in the Iran-Iraq war, which, after 22 years, caused local bone cancer without any special clinical manifestations. The patient was examined periodically, and found a bone-destroying lesion and the biopsy results showed local bone cancer. The patient underwent surgery to remove the tumor and replace the joint. The authors believe that the cause may be metal or lead poisoning which resulted in changes in the cells around the foreign body [6]. Our patient suffered from a fire injury for 50 years - long enough for the wound to change, causing local carcinoma.

CONCLUSION

Wounds retaining foreign objects can be found anywhere on the body. Foreign metal objects under the skin can cause carcinoma. Surgery to remove foreign bodies is necessary, especially those in skin areas exposed to sunlight. Surgery to remove foreign bodies prevents cancer and prolongs the patient's life.

Ethics: Research strictly complies with the regulations on research ethics of the Ministry of Health. Patients were on operations following the surgical procedures of Military Hospital 103 and the Ministry of Health. The patient's database has been approved for use and publication by Military Hospital 103. The authors commit that the research is carried out objectively and without conflicts of interest.

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PROSEAL LARYNGEAL MASK AIRWAY VERSUS CLASSIC LARYNGEAL MASK AIRWAY FOR AIRWAY MANAGEMENT IN BURN NECROSIS AND SKIN GRAFTING SURGERY

Vo Van Hien[1,2], Le Ngoc Anh[1]*

Abstract

Objectives: To compare the efficiency of the classic laryngeal mask airway (c-LMA) and the proseal laryngeal mask airway (p-LMA) in patients with indications for burn necrosis and skin grafting surgery. ***Methods:*** A clinical descriptive, randomized comparison study was conducted on 60 patients divided into two groups: p-LMA (30 patients) and c-LMA (30 patients) were anesthetized with p-LMA and c-LMA, respectively. Accessing and comparing the ease of placement, respiratory function, and hemodynamic response, and the degree of airway injury after surgery between two groups. ***Results:*** 100% patients were successfully inserted laryngeal mask airway (LMA) with insertion time of 22.14 seconds and 20.56 seconds, respectively ($p > 0.05$); oropharyngeal leak pressure (OLP) was 28.56 cmH_2O (p-LMA) and 19.66 cmH_2O (c-LMA) ($p < 0.05$); hemodynamic and respiratory function during surgery were maintained in normal range and there was no difference between the two groups ($p > 0.05$), and no airway complications related to insertion technique (sore throat, hoarseness, or blood staining) were found in either group. ***Conclusion:*** Both LMAs are useful for managing airways in burn necrosis and skin grafting procedures. Both kinds of masks are simple to put on, do not significantly alter hemodynamic response or respiratory function, and patients recover safely from LMA placement without any problems.

Keywords: Laryngeal mask airway; Proseal laryngeal mask airway; Classic laryngeal mask airway; Burn necrosis and skin grafting.

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INTRODUCTION

Since introducting into clinical use in 1988, the LMA has quickly gained popularity as a convenient tool for airway management in anesthesia and resuscitation. The advantages of utilizing the LMA are its simplicity of use, quick airway control, and minimal postoperative airway injuries. Nevertheless, because LMAs are supraglottic airway devices, they have drawbacks such as malposition during surgery, the possibility of gastroesophageal regurgitation, or air leakage when using positive pressure ventilation [1]. The two most widely used laryngeal masks today are c-LMA and p-LMA, of which p-LMA is a later generation with a silicon cuff and drainage tube for higher mask sealing pressure and can drain gastric juice. On the other hand, c-LMA is easier to manage when inserting because of its smaller size and simpler structure [1, 2, 3]. Burn necrosis and skin grafting surgery often do not require a long surgical time, not requiring deep muscle relaxation, so airway management perioperative with LMA is a reasonable choice. We conduct this study to: *Compare the efficiency of c-LMA and p-LMA in terms of ease of placement, respiratory function and hemodynamic response, and the degree of airway injury after LMA anesthesia in patients with indications for burn necrosis and skin grafting surgery. This will provide evidence for selecting safe equipment for surgery.*

MATERIALS AND METHODS

1. Subjects

Including active sampling of 60 patients diagnosed with burns and indications for necrosis and skin grafting surgery under LMA anesthesia at the Department of Anesthesia, Le Huu Trac National Burn Hospital, from January 2024 to June 2024.

* *Inclusion criteria*: Patients aged 18 - 60 years old; ASA I - III; agreed to participate in the study.

* *Exclusion criteria*: Patients had one of the following factors: Chronic lung disease, airway injury due to burns, and suspected cases of full stomach; patients diagnosed with difficult intubation, obese (BMI > 30); or cases with complications during surgery requiring a change in anesthesia method.

2. Methods

* *Study design:* A clinical descriptive, randomized comparison study.

* *Study grouping*: Patients participating in the study were randomly divided into 2 groups:

The p-LMA group (30 patients): Anesthesia with p-LMA for airway management;

The c-LMA group (30 patients): Anesthesia with c-LMA for airway management.

* *Anesthesia procedure:* 1 day before surgery, patients had preoperative examinations and received instructions on anesthesia and surgical techniques. A consent form must be signed by each patient or relative who decides to take part in the research.

After entering the surgery room, patients received a peripheral intravenous, standard monitoring with non-invasive blood pressure (NIBP), SpO_2, and $EtCO_2$, and an ECG DII. Oxygen was inhaled at 3 L/minute through a face mask. Anesthesia protocol: Slow intravenous injection of midazolam 0.05 mg/kg, fentanyl 3 mg/kg, and propofol 3 mg/kg. A face mask was applied for ventilation support when the patient lost consciousness and stopped breathing. The LMA (Teleflex Medical, Dublin, Ireland) (size 4 for male and size 3 for female) was inserted when muscles were relaxed and the jaw was down. The LMA cuff was inflated with a pressure of 60 mmHg (checked by pressure manometer); check the correct LMA position using $EtCO_2$ waveform and auscultation. The number of insertion attempts was recorded. A failed attempt was defined as the removal of the device from the mouth. Three attempts were allowed before device use was considered a failure.

The airway sealing pressure or "leak" test was measured using the "audible noise" method that was first described by Keller et al. [4]. We set a continuous 100% oxygen flow of 3 L/min with the circuit connected to the reservoir bag and the adjustable pressure limiting valve closed. Then trachea was continuously auscultated in the anterior neck for the audible leak. OLP (cmH_2O) was defined as the airway pressure plateau at which an audible leak occurs.

Ventilate with VC mode (Vt = 5 - 6 L/kg; f = 12 - 14 cycles/minute) and adjust to ensure $EtCO_2$ in the range of 35 - 40 mmHg. Anesthesia was maintained with propofol via an electric syringe at a rate of 10 - 15 mg/kg/hour. When beginning a skin incision, add 100mcg of fentanyl and then another 100mcg every hour. Give 20mg of nefopam combined with 100mL of 0.9% sodium chloride solution 30 minutes before the procedure ends to reach the maximum analgesic concentration when the patient starts to wake up. Anesthesia was stopped completely while the wound was being bandaged. When the patient was completely conscious, able to raise their limbs, and breathing on their own (SpO_2 = 95 - 100% breathing air), LMA was removed. Postoperative blood staining of the LMA, sore throat, and hoarseness were recorded after surgery.

* *Data collection:*

Patients' general characteristics.

Ease of LMA insertion (the number of attempts and insertion time).

Changes in heart rate, NIBP, end-expiratory CO_2 pressure ($EtCO_2$), oxygen saturation (SpO_2), and $EtCO_2$ at the following times: T_0 (before LMA insertion); T_1 (1 minute after LMA insertion); T_2 (5 minutes after LMA insertion); T_3 (skin incision); T_4 (wound bandage); T_5 (after removing LMA).

OLP, peak airway pressure (P-peak), and adverse effects during the procedure. The percentage of patients who suffer from airway injury postoperative.

* *Statistical calculation*: The Statistical Package for the Social Sciences 22.0 (SPSS 22.0) software was used for statistical calculation. Data were expressed as either mean and standard deviation or numbers and percentages. A p-value less than 0.05 is believed to be statistically significant.

3. Ethics

The protocol of LMA anesthesia for burn surgery used in the study referred to the procedure that received approval from the Director of Le Huu Trac National Burn Hospital, Vietnam Military Medical University, according to the decision No. 324/QD-BVB dated April 1, 2020, on promulgating the Guidelines for Procedures for Medical Examination and Treatment at Le Huu Trac National Burn Hospital. All patients' data was secure throughout the study to protect their anonymity. Le Huu Trac National Burn Hospital granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest.

RESULTS

Table 1. Patient general characteristics.

Variables	p-LMA	c-LMA	p
No. of patients/males/females (n)	30/25/5	30/27/3	
Age (years)	32.68 ± 7.92	36.74 ± 7.42	
Height (cm)	165.67 ± 12.55	167.23 ± 8.27	
Weight (kg)	55.55 ± 9.12	57.80 ± 6.35	> 0.05
BMI	20.40 ± 3.75	20.72 ± 5.33	
Duration of surgical procedure (minute)	47.26 ± 6.76	46.73 ± 9.75	
Duration of anesthesia (minute)	62.65 ± 8.55	60.17 ± 6.45	

There is no significant difference in patient general characteristics between the two groups ($p > 0.05$).

Table 2. Comparison between the p-LMA and c-LMA.

	p-LMA	c-LMA	p
Attempts at insertion (n) 1/2/3	23/7/0	27/3/0	> 0.05
Insertion time (seconds)	22.14 ± 1.55	20.56 ± 2.42	> 0.05
Oropharyngeal leak pressure (cmH_2O)	28.56 ± 4.26	19.66 ± 2.88	< 0.05
LMA malposition perioperative, n (%)	0 (0)	2 (6.66)	> 0.05
LMA needs to be positioned or reinserted, n (%)	0 (0)	2 (6.66)	> 0.05
Hoarseness or sore throat, n (%)	0 (0)	0 (0)	> 0.05
Blood staining at removal, n (%)	2 (6.33)	0 (0)	> 0.05

Oropharyngeal leak pressure in patients of the p-LMA group is higher than that of the c-LMA group (p > 0,05).

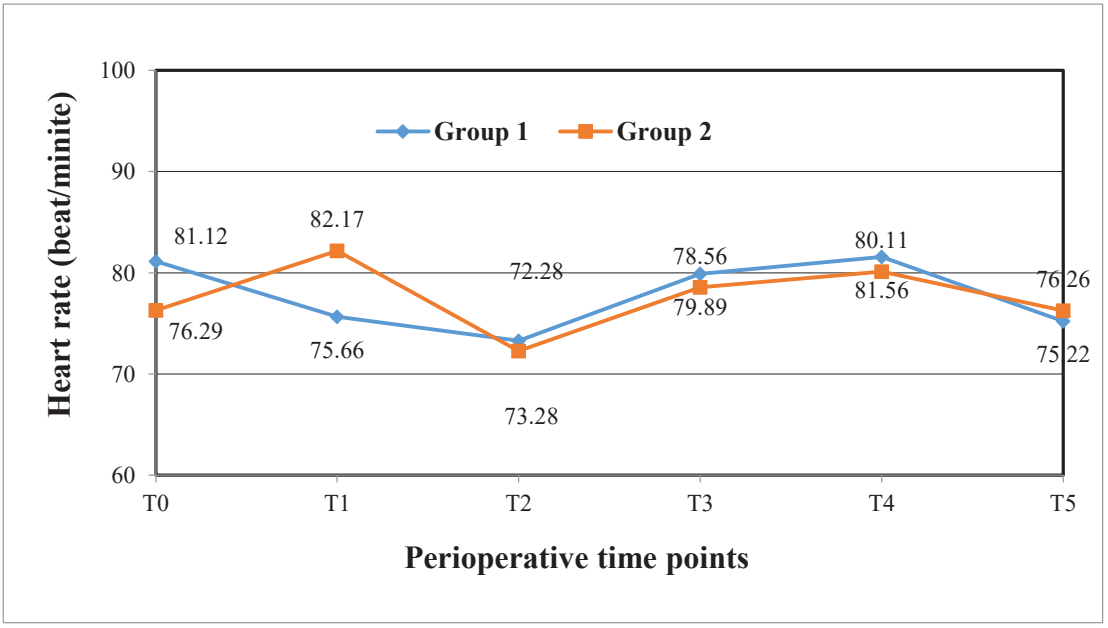

Chart 1. Changes in heart rate during anesthesia.

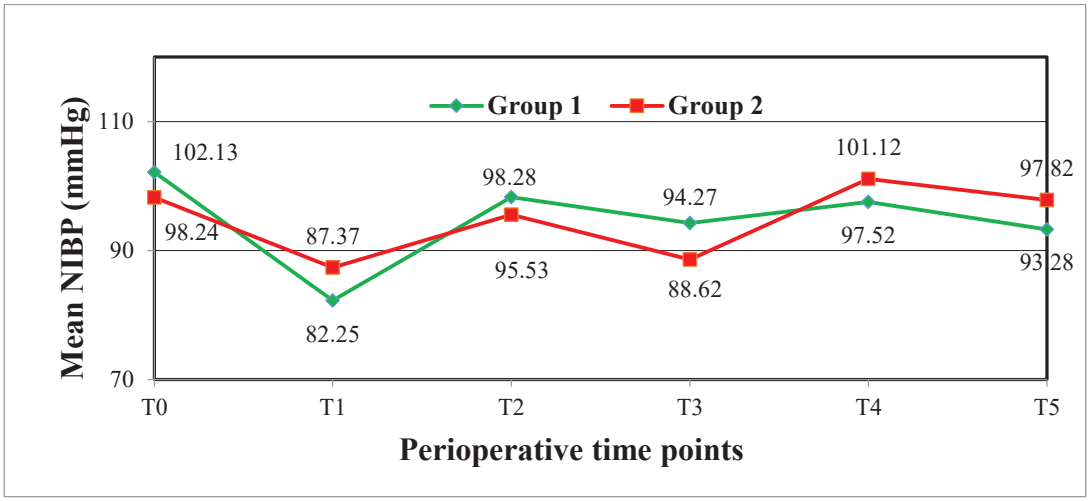

Chart 2. Changes in mean NIBP during anesthesia.

There is no significant difference in hemodynamic changes between the two groups ($p > 0.05$).

Table 3. Changes in SpO_2, $EtCO_2$ and P-peak during anesthesia.

	SpO_2 (%) ($\overline{X} \pm SD$)		$EtCO_2$ (mmHg) ($\overline{X} \pm SD$)		P-peak (cmH2O) ($\overline{X} \pm SD$)	
	p-LMA	c-LMA	p-LMA	c-LMA	p-LMA	c-LMA
T_0	99.87 ± 0.11	99.83 ± 0.15	-	-	-	-
T_1	99.63 ± 0.36	99.05 ± 0.28	38.13 ± 6.34	38.60 ± 5.22	15.26 ± 2.65	13.50 ± 2.60
T_2	99.25 ± 0.54	99.08 ± 0.50	36.13 ± 6.45	37.30 ± 7.98	13.33 ± 4.53	14.28 ± 3.60
T_3	98.97 ± 0.78	98.93 ± 0.97	39.15 ± 7.83	40.10 ± 7.50	15.12 ± 6.51	14.05 ± 4.47
T_4	99.25 ± 0.66	99.42 ± 0.50	40.28 ± 6.34	41.20 ± 5.38	14.62 ± 2.35	13.55 ± 2.40
T_5	99.80 ± 0.26	99.85 ± 0.32	-	-	-	-

There is no significant difference in respiratory parameters between the two groups ($p > 0.05$).

DISCUSSION

Our study results clearly show that using both types of LMA ensures safety and good airway management during anesthesia for patients undergoing burn necrosis and skin grafting. This is reflected in the changes in hemodynamics and respiration of both study groups being within the allowable limits, and there was no significant difference between the two groups ($p > 0.05$) (*Chart 1, Chart 2, Table 3*). In terms of LMA

insertion technique, although p-LMA has a larger structure than c-LMA, the ease of mask placement shown in the number of times and time of successful LMA placement between the two study groups were not different (*Table 2*). This is different from the study by Joseph Brimacombe et al. [5] when comparing the effectiveness of LMA on 384 patients divided into 2 groups using p-LMA and c-LMA. The authors discovered that c-LMA succeeded in 159/192 (83%), whereas p-LMA was successful after just one try in 174/192 (91%). Additionally, p-LMA placement time is longer than c-LMA placement time (22 and 38 seconds, respectively, $p < 0.05$). According to the authors, p-LMA insertion had a lower success rate caused by the mask's larger size and the absence of a backplate, which increased the likelihood that the cuff would fold over at the back of the mouth. The differences with our study may be due to the fact that our operators are experienced anesthesiologists who have performed p-LMA mask insertion on thousands of cases. Furthermore, the reason for this could be that our study had a significantly smaller patient population than the aforementioned study.

The safety of both airway management is also reflected in the results of postoperative adverse effects related to LMA perioperative. According to the study's findings, neither study group experienced any adverse effects like laryngeal laryngospasm or symptoms associated with airway damage, such as hoarseness or sore throat (*Table 2*). This is the main benefit of LMA over other airway control devices or endotracheal tubes. Furthermore, our study's laryngeal mask placement was carried out by experienced anesthesiologists; the patient was sufficiently sedated to eradicate all pharyngeal reflexes, making the procedure simpler and more convenient. According to Pham Quang Minh et al.' study [6], the patients who underwent general anesthesia with endotracheal tube insertion had a 40% and 3.3% higher rate of sore throat and hoarseness symptoms, respectively, than the LMA group, which had a 0% rate ($p < 0.01$). Our findings were in line with those of Belena JM et al. [7], who discovered that patients under LMA anesthesia did not experience hoarseness or sore throat complications.

Our study's most significant finding was that patients in the p-LMA group had higher OLP than those in the c-LMA group (*Table 2*). The results of our study are similar to those of the authors Qamarul Hoda M, Joseph Brimacombe, PP Lu, A Coulson et al. [2, 5, 8, 9]. This is due to the structure

of p-LMA being different from that of c-LMA. According to author Shin et al., a potential risk of LMA use is incomplete airway sealing, which may cause gastric insufflation; inflation of airways at pressures above 20 cmH_2O can induce the opening of the esophageal sphincter [10]. As a supraglottic airway management device, the LMA may move from its intended position, particularly during laparoscopic surgery involving abdominal inflation or when the patient needs to change positions during the procedure. The more OLP there is, the less air leakage there is and the greater the ventilation safety. Furthermore, the LMA is held more securely and is less likely to malposition during surgery when the OLP is higher. Because the patients in our study did not require positional changes during surgery and had short surgical times, this benefit of p-LMA is not immediately apparent.

Our study's limitations included its single-center design and small sample size. Another benefit of p-LMA is that it has a gastric drainage route. However, since this feature was not utilized during anesthesia in our study, there was no way to compare how well these two mask types performed in terms of the aforementioned feature. Therefore, we recommend that multicenter studies with larger sample sizes and in-depth patient selection should use the characteristics of p-LMA to more clearly see the differences between the two study groups.

CONCLUSION

By comparing the effects of p-LMA and c-LMA, we discovered that both LMAs are useful for managing airways in burn necrosis and skin grafting procedures. Both kinds of masks are simple to put on, do not significantly alter hemodynamic response or respiratory function, and patients recover safely from LMA placement without any problems.

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EVALUATION OF GASTRIC CONDUIT PERFUSION RESULTS BASED ON REAL-TIME INDOCYANINE GREEN FLOW SIGNAL IN THORACOSCOPIC SURGERY FOR ESOPHAGEAL CANCER

Nguyen Van Tiep[1], Le Thanh Son[1], Nguyen Anh Tuan[2]*

Abstract

Objectives*:** To evaluate gastric conduit perfusion (GCP) images based on indocyanine green (ICG) flow signal timing in thoracoscopic surgery for esophageal cancer (EsC). ***Methods: A cross-sectional descriptive study was conducted on 70 patients who were applied ICG to evaluate GCP during thoracoscopic surgery to treat EsC at 108 Military Central Hospital and Military Hospital 103 from June 2022 to June 2024. ***Results:*** The mean age was 59.0 ± 7.9 (32 - 71) years old; 100% were male. The anastomotic leak rate was 7.1%, with a mean gastric conduit (GC) width of 5.1 ± 0.2cm. Through ICG imaging, 17 patients with missing GC were detected. The average ischemic GC length was 2.7 ± 0.6cm. The time of appearance of the ICG signal in segments (B-C) and segments (A-D) of the anastomotic leak group was longer than that of the group without anastomotic leak ($p < 0.05$). Multivariable logistic regression analysis found that the greater the rate of ischemic GC, the higher the anastomotic leak rate (OR = 59.27; 95%CI= 1.25 - 2802.03; p = 0.04). ***Conclusion:*** Evaluation of GCP images based on ICG flow signal timing is feasible, safe, and objective. ICG current signal timing helps detect the location of the poorly perfused GC and select the appropriate location to create the anastomosis with a low anastomotic leak rate.

Keywords: Esophageal cancer; Gastric conduit; Indocyanine green.

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INTRODUCTION

Anastomotic leak is a common complication after esophagectomy for EsC, with a rate of about 5 - 25% [1, 2]. Factors related to anastomotic leak include patients' nutritional status, the tunnel where the GC is placed, the location or technique of anastomosis, and the tension of the anastomosis. Among them, ischemia of the GC is an important factor causing anastomotic leak and anastomotic stenosis [3]. Around the world, the near-infrared fluorescence method using ICG has been applied as a valuable, objective tool in surveying and evaluating the perfusion status of the GC. To evaluate GCP, the authors used one of the following three methods: ICG signal duration, flow rate, and ICG current intensity in the GC. Many authors rely on ICG flow signal duration as an objective index to evaluate GCP. Noma et al. reported that if the GC blood supply is enhanced with ICG within 20 seconds, the GC is said to be well perfused at that location, and anastomosis performed in this area will ensure good [4]. Kumagai et al. proposed the "90-second rule" to confirm good perfusion of the GC. All anastomoses performed in the area of the ICG-enhanced GC within 90 seconds of the ICG signal appearing at the origin of the right gastric omental artery [5]. EM de Groot demonstrated that in patients with anastomotic leakage, the mean time to reach maximum ICG intensity at the tip of the GC was 56 seconds (ranging from 30 - 83 seconds) compared with 34 seconds (ranging from 12 - 66 seconds) in patients without anastomotic leakage [6]. In Vietnam, thoracoscopic esophagectomy is performed routinely in many central hospitals, but there has not been any work or research reported on the use of ICG to evaluate GCP. Therefore, we conducted this study to: *Evaluate the results of GC blood supply based on ICG flow signal timing in thoracoscopic surgery for EsC.*

MATERIALS AND METHODS

1. Subjects

Including 70 EsC patients who underwent thoracoscopic esophagectomy and ICG imaging to evaluate the blood supply of the replacement GC for EsC from June 2022 to June 2024.

* *Inclusion criteria:* Diagnosis of carcinoma in the thoracic esophagus and undergoing thoracoscopic radical esophagectomy for the preoperative stage: cT1b-cT2, N0 or cT1b-cT3, N+ after preoperative chemotherapy; undergoing ICG imaging for evaluation of the blood supply of the replacement GC; ASA-PS ≤ 3.

Exclusion criteria: Patients undergoing thoracoscopic esophagectomy for non-cancerous causes or those with tumor invasion at the T4b level, according to the American Joint Committee on Cancer classification.

2. Methods

Study design: A cross-sectional descriptive study.

Surgical procedure using ICG to assess GCP:

Thoracoscopic radical esophagectomy and 2-region lymph node dissection were applied to all patients.

Thoracic step: Thoracoscopic surgery.

Abdominal surgery: Laparoscopic or open surgery was possible in cases with a history of previous abdominal surgery.

Technique to create a large GC:

The reconstructed GC was approximately 5cm wide, retaining the right and left gastroepiploic arteries, some of the first branches of the right gastric artery, and the arcade between the right and left gastroepiploic arteries.

The GC was created using an open stapler with open surgery, assisted laparoscopic surgery, and an endoscopic stapler with laparoscopic surgery.

Fluoroscopy uses ICG to evaluate GCP:

Measure the dimensions of the GC with a tape measure:

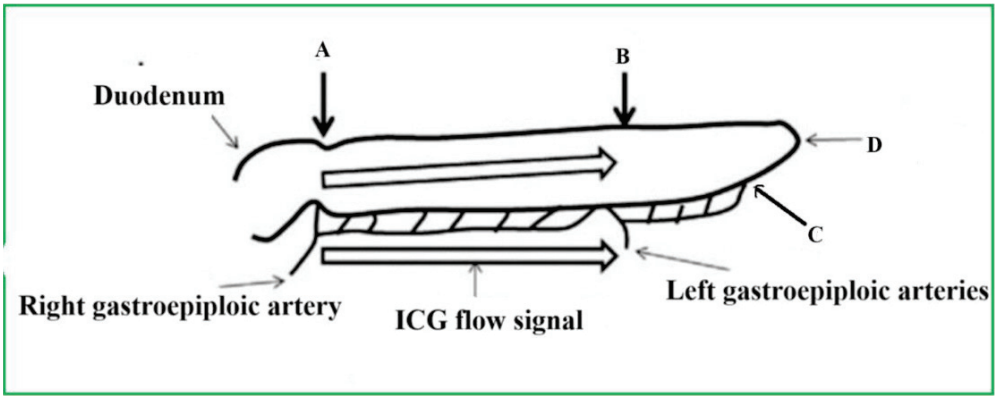

Figure 1. Image of marking positions to measure GC dimensions.
Source: According to Kazuo Koyanagi in 2016 [7].

Point (A) is the pylorus;

Point (B) is the location of the connection between the right and left gastroepiploic arteries;

Point (C) is the last point of the pulsatile GC to the final tip of the stomach;

Point (D) is the final point of the GC.

The camera had a near-infrared light source placed in front of the GC at a 3 - 5cm distance to ensure clear recorded images.

The injection technique in the study was applied according to author Rao-Jun Luo (2020) [5] as follows:

Injection dose: 2.5mg ICG injection/ 1 time.

Injection site: Can be injected into a central vein (right external jugular vein) or peripheral vein (right cephalic vein).

Injection time: Rapid bolus injection of ICG in about 3 - 5 seconds.

Number of injections: 1st ICG injection: After completing the shaping of the GC.

The purpose is to evaluate the GCP status when the shaping is completed. Second ICG injection: Recheck GCP after the GC is inserted through the posterior mediastinal tunnel to the neck.

The time to appear ICG was calculated when the blue signal begins to appear at point a (origin of the right gastroepiploic artery).

Based on the time of ICG appearance calculated from point a to the distal end of the GC in the arterial phase at the 1st injection.

The GC segment was considered to be well perfused when the time to appear ICG is < 60 seconds from point A. The GC segment was considered poorly perfused when the time to appear ICG is ≥ 60 seconds from point A.

* *Data analysis:* All statistical analyses were performed using SPSS software (version 26.0, 64-bit from IBM Corporation, NY, USA).

3. Ethics

This study was conducted in accordance with the declaration of Helsinki, approved by the Ethics Council of Military Hospital 103, on December 9th, 2022, approval number 193/CNChT - HĐĐĐ. Participants fully agreed and voluntarily participated in the study. Written informed consents were obtained with full signatures. Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

RESULTS

70 patients participated in the study from June 2022 to June 2024. The average age was 59.0 ± 7.9 (32 - 71) years old; 100% were male. ASA = 2, accounted for 64.3%. ASA = 3, accounted for 35.7%. Patients received preoperative chemotherapy and radiotherapy, accounting for 77.1%. The anastomotic leak rate was 7.1% (5 patients)

* *Characteristics of GC esophageal replacement used in research:*

GC width: 5.1 ± 0.2cm; GC length (AD): 31.4 ± 1.3cm; length of GC segment (AB): 18.9 ± 1.3cm; length of GC segment (AC): 29.1 ± 1.2cm; length of GC segment (BC): 10.2 ± 1.0cm.

Table 1. Real-time ICG flow signals in the GC (arterial phase).

Real-time ICG flow signals in the GC (arterial phase)	N^0 of patients	Min	Max	Mean ± SD/ Median (Q1-Q3)
First injection				
Segment (A-B) (sec)	70	3	14	6.2 ± 2.0
Segment (A-C) (sec)	70	9	32	18.4 ± 5.1
Segment (A-D) (sec)	70	14	120	24.0 (21.0 - 59.3)
Segment (B-C) (sec)	70	5	24	12.2 ± 4.2
Segment (C-D) (sec)	70	2	105	6.0 (4.0 - 40.3)
Second injection				
Segment (A-C) (sec)	70	12	32	18.9 ± 4.6
Segment (A-D) (sec)	70	17	120	25.0 (22.0 - 48.0)
Segment (C-D) (sec)	70	1	105	7.0 (5.0 - 31.3)

When injecting ICG for the first time, the GC immediately after shaping, the time to appear ICG in the entire GC (A-D) was 24.0 (21.0 - 59.3) seconds. The appearance time of ICG segments (A-C) and (B-C) were 18.4 ± 5.1 seconds and 12.2 ± 4.2 seconds, respectively. When the GC was placed in the posterior mediastinal tunnel, the ICG appearance time of the entire GC (A-D) and segment (A-C) was 25.0 (22.0 - 48.0) seconds and 18.9 ± 4.6 seconds, respectively.

Table 2. Comparison of determining the GC with poor blood supply between visual observation and ICG fluorescence imaging.

Perfusion of the GC		ICG imaging		Total
		No	Yes	n (%)
Visual observation	No	53	8	61 (87.1)
(The first observation)	Yes	0	9	9 (12.9)
Visual observation	No	53	8	61 (87.1)
(The second observation)	Yes	0	9	9 (12.9)
Total		53	17	70 (100)
Management of the ischemic portion of the GC observed by ICG imaging				
Excision of the ischemic portion of GC				17 (24.3)
Suture burying the ischemic part of GC				0
The average length of the GC that was removed (cm): 2.7 ± 0.6 (2 - 4)				

Clinical observation by 2 experienced surgeons found 9 patients with poorly perfused GCs. Re-examination with ICG imaging detected 17 patients with poorly perfused GCs, including 9 patients who were visually observed and the remaining 8 patients who were not visually observed. All poorly perfused GCs had the ischemic part removed; the average conduit length of the ischemic GC was 2.7 ± 0.6cm.

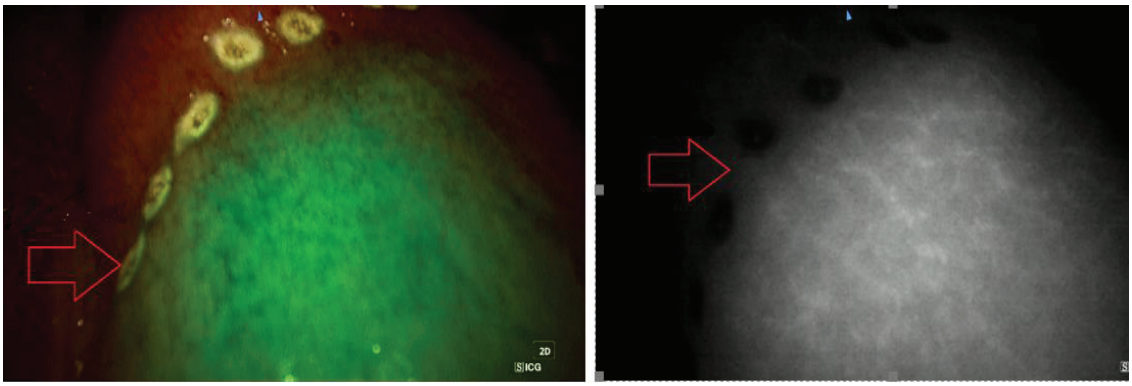

Figure 2. Boundary ICG image identifies an ischemic GC.
Source: Patient Le K, file number 22836724.

Table 3. The relationship between ICG imaging characteristics and injection.

ICG images	Central vein	Peripheral veins	p
First injection			
ICG appearance time of segment (A-B) (sec)	6.1 ± 2.0	6.4 ± 1.9	0.61
ICG appearance time of segment (A-C) (sec)	17.9 ± 4.9	18.9 ± 5.3	0.45
ICG appearance time of segment (A-D) (sec)	24 (21 - 54)	24 (20.8 - 63.5)	0.81
ICG appearance time of segment (B-C) (sec)	11.8 ± 4.1	12.5 ± 4.3	0.49
Second injection			
ICG appearance time of segment (A-C) (sec)	18.6 ± 4.3	19.3 ± 5.0	0.58
ICG appearance time of segment (A-D) (sec)	26 (22 - 42.3)	25 (21 - 63.3)	0.54

There was no relationship between ICG imaging characteristics and the injection route.

Table 4. The relation of ICG flow signal time to the vascular connection between the right and left gastroepiploic arteries.

ICG images	The arcade between the right and left gastroepiploic arteries		p
	No	Yes	
First injection			
ICG appearance time of segment (A-B) (sec)	6.5 ± 2.4	6.1 ± 1.6	0.39
ICG appearance time of segment (A-C) (sec)	18.9 ± 4.6	18.1 ± 5.4	0.56
ICG appearance time of segment (A-D) (sec)	25 (22 - 71)	24 (21 - 37)	0.13
ICG appearance time of segment (B-C) (sec)	12.4 ± 3.3	12.1 ± 4.7	0.76
Second injection			
ICG appearance time of segment (A-C) (sec)	19.1 ± 4.4	18.8 ± 4.8	0.81
ICG appearance time of segment (A-D) (sec)	26 (22 - 72)	25 (22 - 39)	0.36

The real-time appearance of ICG flow signals in the GC at marked locations was not related to the vascular connection between the right and left gastroepiploic arteries.

Table 5. Real-time correlation of ICG flow signal appearance with anastomotic leakage.

ICG image	Anastomotic leakage		p
	No (n = 65)	Yes (n = 5)	
First injection			
ICG appearance time of segment (A-B) (sec)	6.3 ± 2.0	5.6 ± 0.9	0.46
ICG appearance time of segment (A-C) (sec)	18.1 ± 5.1	22.0 ± 2.7	0.10
ICG appearance time of segment (A-D) (sec)	24 (21 - 41.5)	65 (43.5 - 73.5)	0.04
ICG appearance time of segment (B-C) (sec)	11.9 ± 4.1	16.4 ± 2.5	0.02
Second injection			
ICG appearance time of segment (A-C) (sec)	18.7 ± 4.7	22.4 ± 2.5	0.08
ICG appearance time of segment (A-D) (sec)	24 (21.5 - 39.5)	65 (45.5 - 73.5)	0.02

The real-time appearance of ICG flow signals in segments (B-C) and segments (A-D) at the 1st injection of the anastomotic leak group was longer than that of the group without an anastomotic leak. This difference was statistically significant,

with p = 0.02 and p = 0.04. The above results were similar in the second injection when comparing the ICG flow signal time in segments (A-D) in the 2 groups of anastomotic leak and without an anastomotic leak.

Table 6. Multivariable logistic regression for factors predicting anastomotic leak.

Variables	OR	95%CI	p
Age	0.83	0.66 - 1.05	0.12
Smoke	4.19	0.16 - 113.41	0.40
BMI	1.02	0.55 - 1.89	0.94
Protein	1.10	0.82 - 1.46	0.53
Albumin	0.69	0.35 - 1.36	0.29
Surgery time	1.03	0.98 - 1.07	0.28
Blood loss during surgery (mL)	1.00	0.99 - 1.01	0.95
GC shaping technique	0.53	0.02 - 11.75	0.69
The connection between the right and left gastroepiploic vessels	1.15	0.07 - 19.09	0.92
Poor blood supply of distal GC after the 1st injection	59.27	1.25 - 2802.03	0.04

When analyzing multivariable logistic regression, it was found that the more significant the proportion of GCs with poor perfusion, the higher the rate of anastomotic leakage (OR = 59.27; 95%CI = 1.25 - 2802.03; p = 0.04). Other factors such as age, protein, BMI, and the amount of blood loss are not related to anastomotic leak.

DISCUSSION

Based on real-time ICG flow signals in the GC to investigate the perfusion of the GC, it shows that the shorter the real-time, the better the perfusion of the GC. Normally, the ICG flow signal time will appear early in the pyloric region of the GC. This location of the GC is better perfused than other locations. Some authors have chosen short real-time ICG flow signals. This choice aims to ensure better perfusion of the GC and enough conditions to create an anastomosis. However, if we choose a short time for the ICG signal to appear, the GC's length is often not enough to reach the neck area. This means that the GC will be longer. Choosing locations where the ICG signal image will appear longer is necessary.

When injecting the first time, the GC was placed in front of the chest, the ICG appearance time of the entire GC (A-D) was 24 (21 - 59.3) seconds, segment (A-C) was 18.4 ± 5.1 seconds, and segment (B-C) was 12.17 ± 4.2 seconds. Evaluation of GCP revealed 17 patients with ICG signal time appearing in GC ≥ 60 seconds, a sign of poor blood supply in GC. Table 3 demonstrates that the central and peripheral intravenous routes are unrelated to the time the ICG flow signal takes in GC. When injecting the second time, the GC was placed in the posterior mediastinal tunnel, the time of total ICG appearance of the GC segment (A-D) was 25 (22 - 48) seconds, and segment (A-C) was 18, 9 ± 4.6 seconds (*Table 1*).

Table 5 shows that when comparing the ICG appearance time between different positions of the GC, we get the result that the ICG appearance time of the GC segment (B-C) in the anastomotic leak group is longer than the no-leak group. Anastomosis during the 1st injection, the difference between these 2 groups was statistically significant with p = 0.02. It is important to know that segment (B-C) is the location of GC that often has anemia and is also the area that needs to be investigated before GC shaping. The time to appear ICG signal of the entire GC (A-D) in the anastomotic leak group was longer than the group without anastomotic leak, 65 (43.5 - 73.5) seconds compared to 24 (21 - 41.5) seconds in the first injection and 65 (45.5 - 73.5) seconds in the no anastomotic leak group compared to 24 (21.5 - 39.5) seconds in the anastomotic leak group in the second injection.

Kumagai et al. proposed a 90-second rule to confirm a well-perfused GC position. The anastomosis was performed in the area of the GC with an enhanced IG flow signal less than 90 seconds from the appearance of the initial ICG signal at the origin of the right gastroepiploic artery. Based on this rule, poor perfusion GC had poor perfusion fecal resection in 50% (35/70), and the planned site of anastomosis was changed in 18 of those 35 cases. No patient underwent anastomosis creation at the site with ICG enhancement slower than 90 seconds. One case out of 70 cases (1.4%) showed anastomotic leakage when the anastomotic site where ICG appeared after 77 seconds [5].

Noma reported that if the GCP is enhanced by ICG within 20 seconds at that location of the GC that is considered well perfused, an anastomosis performed in this area will ensure good perfusion. The study showed that the anastomotic leak rate in patients using ICG was significantly lower than in patients who did not use ICG (8.8% vs. 22%, p = 0.03) [4].

In Rao-Jun Luo's study, with a total of 192 patients, 86 patients were in the ICG injection group, and 106 patients were in the non-ICG injection group. All patients in the ICG group underwent fluorescein angiography successfully. As a result, 32 patients out of 86 patients who received ICG injections found that GCs were judged to be well perfused within 60 seconds, meaning that these GCs did not have areas of poor perfusion. For the remaining 54 patients, there was an area of GC with poor perfusion with varying lengths of the GC following the 60-second rule. In the ICG group, the anastomosis was performed at the site where green fluorescent GC was observed for less than 60 seconds. ICG imaging confirmed blood flow, and perfusion was clearly visualized within 60 seconds. After 60 seconds, the fluorescence image of the GC was jagged and uneven, which was an inappropriate location for selecting the anastomotic area. Therefore, Rao-Jun Luo hypothesized that the risk of anastomotic leakage is minimized if the anastomosis is created in an area where the ICG is enhanced within 60 seconds. The poorly perfused GC area was excised before performing the anastomosis. The anastomotic leak rate in esophagectomy for EsC was 10.4% in the non-ICG group and was significantly higher than 1.2% in the ICG group. This suggests that the anastomotic leak rate can be reduced by approximately 9% by using ICG. This evidence strongly suggests that adequate perfusion of the GC is important for complete tissue healing at the anastomotic site [8].

Table 5 shows that the time to appear ICG signal in the segment (B-C) of the 1st injection of the anastomotic leak group was longer than the group without an anastomotic leak. This difference is statistically significant ($p = 0.02$).

EM de Groot demonstrated that in patients with anastomotic leakage, the median time to reach maximum ICG intensity at the tip of the GC was 56 seconds (30 - 83) compared with 34 seconds (12 - 66) in patients without anastomotic leakage. Based on the time of ICG appearance, the initial location chosen to perform the anastomosis had to be changed to another location in 14% of patients. The mean time to peak intensity was shorter at the base of the GC (25 seconds, ranging from 13 - 49) than at the distal tip (34 seconds, ranging from 12 - 83) [6].

Kazuya Yamaguchi uses the "90 to 60-second rule", meaning that the esophageal-gastric anastomosis will be performed where the ICG signal increases within 90 seconds (preferably within 60 seconds). The authors have demonstrated that using the 90 to

60-second ICG imaging rule reduces the anastomotic leakage rate. When performing the anastomosis in the enhanced ICG position within 90 seconds, anastomotic leakage occurred in 4 cases (3.1%) out of 129 cases, of which 3 cases (2.4%) out of 126 cases had junction sites where the ICG current signal enhanced within 60 seconds and 1 case (33.3%) out of 3 cases with time ICG enhancement exceeded 60 seconds (p = 0.09) [9].

According to table 6, multivariable logistic regression analysis shows that the higher the rate of poorly perfused GC (ICG appearance time ≥ 60 seconds), the higher the rate of anastomotic leak (OR = 59.27; 95%CI = 1.25 - 2802.03; p = 0.04) at the 1st injection. Other factors, such as age, protein, BMI, and blood loss, were unrelated to anastomotic leak. This further demonstrates the importance of ICG imaging in detecting GC perfusion.

CONCLUSION

Assessment of GC perfusion based on ICG flow signal timing is feasible, safe, and objective. The ICG flow signal time is an objective index that helps choose the appropriate GC location to create the anastomosis, and the anastomotic leak rate is low (7.1%). By ICG, 17 patients with GC were detected with poor perfusion. The time to appear ICG signal in segments (B-C) and segments (A-D) of GC in the anastomotic leak group was longer than that in the group without anastomotic leak (p < 0.05). Multivariable logistic regression analysis found that the higher the rate of anemic GC, the higher the anastomotic leak rate (OR = 59.27; 95%CI= 1.25 - 2802.03; p = 0.04).

Acknowledgment: We would like to express our gratitude to all officers and staff of the Gastrointestinal Surgery Department, Digestive Surgery Center, Military Hospital 103 and the Department of Gastrointestinal Tract Surgery, 108 Military Central Hospital, who made a great contribution to the success of the study.

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A CASE REPORT: METHODS TO ALLEVIATE REPERFUSION SYNDROME IN AMPUTATED ARM REPLANTATION

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Tran Hoang Dat[1], Le Quang Dao[1]

Abstract

Limb replantation has been performed for decades and is being clinically applied more prevalently. Reperfusion syndrome crucially affects the outcome of the replantation and sometimes poses a life-threatening risk to patients, especially in cases with major surgeries like arm amputations. The following report presents methods to mitigate the syndrome in amputated arm replantation.

INTRODUCTION

The anaerobic metabolism occurs in a limb as it gets amputated. Subsequently, some metabolites such as cytokine, IL-6, and lactic acid will flow back into the main circulation briefly after the replantation is performed, which causes reperfusion syndrome leading to many complications, metabolic acidosis, acute renal failure, ventricular fibrillation, etc. The complications potentially impair the success of the surgery and are capable of causing some grave results. Minimizing the syndrome will facilitate the success of the surgery, especially in cases involving arm or leg replantations. The report aims to: *Present a case of an amputated arm that was replanted and the methods applied to minimize the risk of the syndrome.*

CASE REPORT

A 44-year-old male patient with a left middle third arm amputated by a cable tow, the amputated limb was treated and stored medically properly, and the patient was admitted to the Military Hospital 103 at the 2nd hour post-injury. Along with conventional techniques performed to replant the amputated limb, some adjuncts were added to alleviate the syndrome, followed by the passage of the amputated limb being restored.

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- The limb was immediately sent to an operating room and treated by a surgical team. Wound debridement and dissection were being done while administrative formalities were being completed.

- Continuously rinsing amputated limb: Blood vessels of the limb were rinsed with normal saline diluted with heparin at a ratio (500mL NS/5000UI Heparin) following the completion of the debridement, dissection, and the exposure of all blood vessels and nerves. The rinsing was performed until the fluid oozing out of the subcutaneous veins was relatively transparent and was continued until the replantation was commenced.

- Draining residual blood out of the amputated limb via veins after arteries were unclogged: After the brachial artery and satellite veins were anastomosed, the blood flowed through the artery to the amputated limb, but the satellite veins were still clamped, keeping the blood from the main circulation. 30 seconds later, if the hemodynamics were maintained, the veins were unclamped following some subcutaneous veins being completely reattached.

Bio-chemistry tests, arterial blood gas, and complete blood count were administered. The results were in the normal range, especially since there was no hyperkalemia. All vital signs were good, the amputated limb is now viable. The patient is in physical rehabilitation 4-day post-operative.

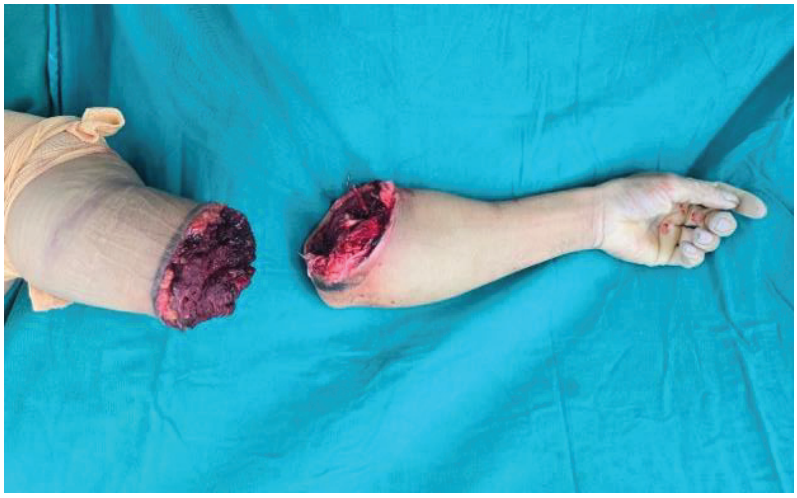
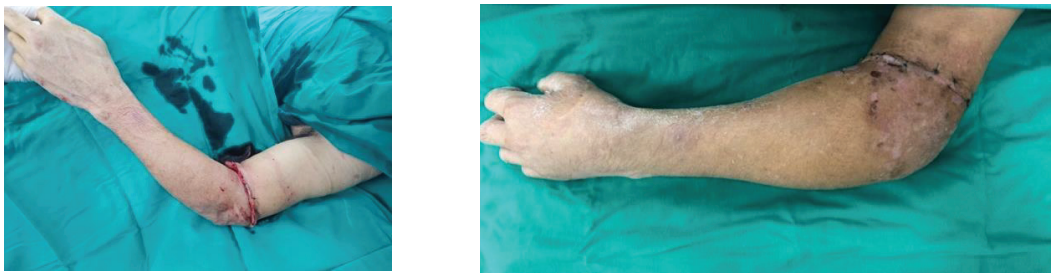


Figure 1. Preoperative amputated arm.



(A) (B)

Figure 2. Post-operative arm after the replantation.
(A): The arm right after the surgery; (B): The arm 14-day post-operative.

DISCUSSION

The amputation cuts off the limb from the main blood source, leading to an ATP loss and the dysfunction of ion-dependent channels of cell membranes. The local ischemia causes acidosis, the accumulation of intra-cellular lactate, and the drainage of intra-cellular calcium. The success of the replantation heavily depends on the severity of the injury and the time of warm ischemia [1]. The longer the time of warm ischemia, the more severe the reperfusion would be. He (2022) reported that the time of cold ischemia within 6 hours for warm limbs or 12 hours for limbs stored in cold can increase the likelihood of surgical success as well as sustain the overall condition. In this case, the amputated limb was treated and stored properly before hospitalization, this early action helped decelerate the anaerobic metabolism in the amputated limb [2]. Nowadays, almost all amputated limbs will be stored properly thanks to communal medical education and media. The patient was admitted to the hospital very soon, 2 hours post-injury. Shortly after the hospitalization, the patient or his legal guardian was fully informed of his injury, potential complications, and how treacherous the replantation could be while the amputated limb and the patient were being prepared and closely monitored for ensuing surgery. This immediate action optimizes the overall state of the patient, enhancing the success rate of the surgery by minimizing the time of warm ischemia.

During the preparation, the limb was rinsed and cleaned with Heparin 5000UI dissolved into 500mL normal saline until the wound discharge through subcutaneous veins was nearly translucent. McCutcheon et al. (2002) reported a case of limb replantation in which the limb was rinsed with normal saline to clear off anaerobic metabolites and not to deteriorate blood vessels' injuries prior to blood vessel reattachment [3]. We assume that rinsing blood vessels with normal saline is a proper approach to eliminate anaerobic metabolites in the

amputated limb and mitigate the blood vessels' injuries, whereby stopping anaerobic metabolites from flowing back to the main circulation, decreasing the chance and the severity of the reperfusion.

Ischemic limb produces anaerobic metabolites, cytokine, IL-6, and lactic acid, and causes hyperkalemia and hypocalcemia [4, 5, 6]. The blood pH will surely be disrupted if these products travel back to the main circulation, impairing the cardiac function, and grave results would be inevitable. Disposing these products by rinsing the amputated limb will reduce the chance of reperfusion syndrome. Nevertheless, the rinsing cannot be performed during the replantation, so the metabolites will keep building up. Therefore, after the anastomosis was done, part of the residual blood in the amputated limb was got rid of by clamping the satellite veins and letting them drain via subcutaneous veins. This action was found efficient and proven by the following blood tests.

CONCLUSION

We concluded that immediate surgery, rinsing the amputated limb before replantation, and eliminating part of the residual blood in the limb before the passage of blood vessels is restored are applicable methods to lower the chance of reperfusion syndrome in cases with amputated limbs, especially major limbs.

Acknowledgment: The research was carried out according to the regulations of the host unit, Military Hospital 103. Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in the study.

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RESULTS OF BILIARY DILATION AND STONE REMOVAL VIA FLEXIBLE CHOLANGIOSCOPY FOR THE TREATMENT OF PRIMARY BILIARY STRICTURES AND STONES AT MILITARY HOSPITAL 103

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Abstract

Objectives: To evaluate the results of biliary dilation and stone removal via flexible cholangioscopy for the treatment of primary biliary strictures (BS) and stones. ***Methods:*** A prospective, descriptive, uncontrolled study was conducted on 62 patients with primary BS and stones treated by biliary dilation and stone removal via flexible cholangioscopy at Abdominal Surgery Centre, Military Hospital 103, from July 2021 to July 2024. ***Results:*** The mean age was 60.1 ± 14.1; the female/male ratio was 1.69/1. 75.8% of patients had a history of biliary stones. Most patients had multiple stones (79%), including choledocholithiasis and hepatolithiasis. BS were mostly in one location (90.3%), intrahepatic strictures (88.7%), and were all benign. The mean length and diameter of the strictures were 3.96 ± 2.9mm and 3.6 ± 0.7mm, respectively. Surgical methods were choledochotomy with intraoperative cholangioscopy (90.3%) and percutaneous cholangioscopy (9.7%). Stone removal was performed using baskets, electrohydraulic, and/or laser lithotripsy. BS was performed using balloon dilation; then, biliary-cutaneous stents were placed in 64.5% of cases at risk of recoil. Intraoperative complications accounted for 16.1%; postoperative complications accounted for 12.9%. The rate of stone clearance and successful stricture dilation after surgery was 83.9% and 87.1%. Rechecked at 1 month, 3 months, and 6 months after operation, the ratio of recurrent stones and BS was 0%, 0%, 5.8% and 1.9%, 7.4%, 11.1%, respectively. ***Conclusion:*** Stone removal and stricture dilation by flexible cholangioscopy is a safe and effective method for treating primary BS and stones.

Keywords: Flexible cholangioscopy; Biliary stricture; Primary bile duct stone; Biliary balloon dilator; Laser lithotripsy; Electrohydraulic lithotripsy.

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INTRODUCTION

Biliary stricture is a common disease in East Asia, Southeast Asia, and Vietnam. According to statistics, the rate of BS in patients with primary bile duct stones can reach 45.6 - 70% [1]. This disease greatly affects the treatment results of biliary stones and is the main cause of stone recurrence, requiring patients to undergo multiple surgeries.

In 2022, IHPBA (International Hepatobiliary-Pancreatic Association) defined: "BS is a localized reduction in the diameter of the bile duct compared to the adjacent biliary ducts, accompanied by the dilation above the stricture" [2]. In patients with both BS and stones, whether BS is the cause or consequence of stones has not been proven. They combine each other to create a pathological spiral. Although BS has always been considered a challenging problem, the diagnosis and treatment of BS were rarely mentioned in the past due to difficulties in directly investigated imaging. Until Shore first performed intraoperative flexible cholangioscopy in 1970, BS had gradually been mentioned. Since then, flexible cholangioscopy has become an effective method for diagnosing and treating BS by its ability to show clear images. Based on the cholangioscope, Lee SK proposed a classification of BS that has been widely applied nowadays [3]. In 2023, the ACG (American College of Gastroenterology) issued recommendations on the technique of cholangioscopy, BS dilation, and biliary stenting [4].

Up to now, very few Vietnamese authors have published researches that deeply evaluate the treatment results of BS. At Military Hospital 103, cholangioscopy has been performed since 2008 to treat stones. In recent years, we have developed a technique for dilating the BS with balloon dilation. The research problem was posed with the question: What is the effectiveness of the technique of stone removal combined with biliary dilation in the treatment of primary BS and stones? Because of that, we conducted this research with the aim to: *Evaluate the results of biliary dilation and stone removal via flexible cholangioscopy for the treatment of primary BS and stones.*

MATERIALS AND METHODS

1. Subjects

Including 62 patients with primary BS and stones treated by biliary dilation and stone removal via flexible

cholangioscopy at Abdominal Surgery Centre, Military Hospital 103 from July 2021 to July 2024.

* *Inclusion criteria:* Patients were diagnosed with primary BS and stones, treated by stone removal and stricture dilation via flexible cholangioscopy.

* *Exclusion criteria:* Patients had tumors or cancers in the bile duct, liver, pancreas's head or Vater's ampulla; patients had ASA score > 3 (classification of patient's health status before surgery according to the American Society of Anesthesiologists).

2. Methods

* *Study design:* A prospective, descriptive, uncontrolled study.

* *Research process:*

Patients were clinically examined and had para-clinical tests for preoperative diagnosis. If patients had primary BS and stones, met the selection criteria, and did not fall under the exclusion criteria, surgery was performed.

Surgical methods:

- If the patient had not previously undergone surgery, choledochotomy with intraoperative flexible cholangioscopy was performed to remove stones and dilate the stricture.

- If the patient had undergone percutaneous biliary drainage, percutaneous flexible cholangioscopy via the tunnel was performed to remove stones and dilate the stricture.

* *Stone removal and biliary dilation via flexible cholangioscopy technique:*

We used a cholangioscopic system with a Japanese Olympus CHF-V2 5mm diameter flexible cholangioscope, 2mm diameter instrument channel combined with irrigation way, 2-directions adjustable controller (up 160° - down 130°).

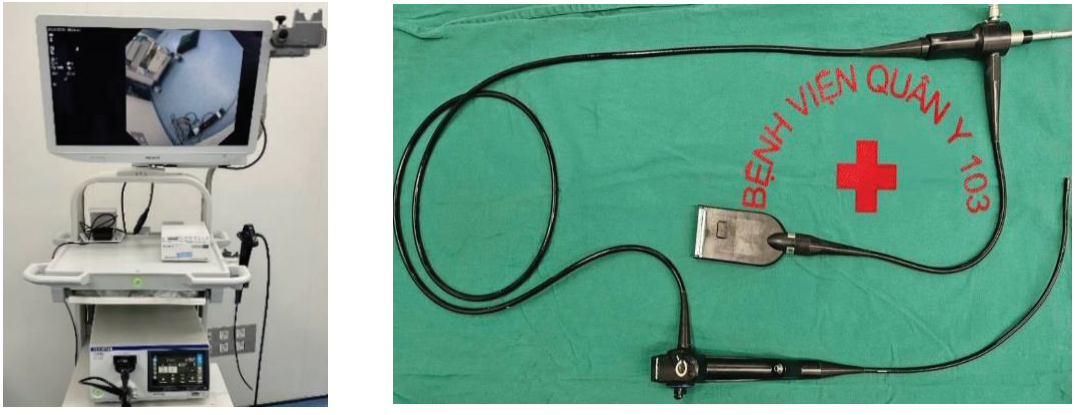

Figure 1. Cholangioscopic system and Olympus CHF-V2 flexible cholangioscope.

The surgeon took the cholangioscope with the right hand and held the controller with the left hand. Then, the cholangioscope was inserted into the bile duct. A continuously irrigated stream of NaCl 0.9% was used to dilate the bile duct and create a clean environment during cholangioscopy. The common bile duct was investigated first, then stones were removed to check the sphincter of Oddi and enterohepatic circulation. After that, the cholangioscope was controlled up to the common hepatic duct and the intrahepatic bile ducts sequentially. Within intrahepatic bile ducts, cholangioscopy was performed in order from the right hepatic duct, right anterior section, segments 5, 8, right posterior section, segments 6, 7, to the left hepatic duct, segments 2, 3, 4, and 1.

In this research, we performed stone removal using baskets, laser, and electrohydraulic lithotripsy and pumped small fragments outside or into the duodenum through the Oddi's sphincter. Stone removal was performed from near to far, to make a good enterohepatic circulation.

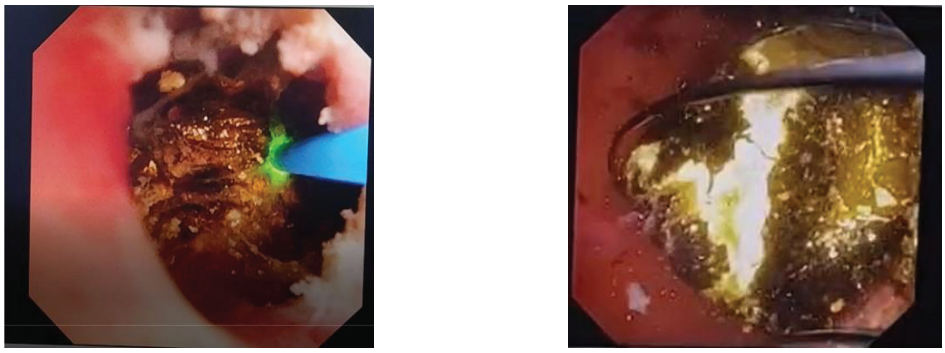

Figure 2. Stone removal with laser lithotripsy and basket.

After removing as many stones as possible, the bile ducts were examined for BS. When BS was found, its characteristics were evaluated, and the stricture was immediately biopsied with biopsy forceps. If the biopsy result was benign, the biliary dilation would be performed. In this research, we applied the balloon dilation method.

- Balloon dilation technique: Using a specialized 3-stage Biliary Balloon Dilation, 0.2 x 290cm, Model BD-410X Olympus, Japan. The dilation technique was applied according to the recommendations of Nunes T (2021) [5]. Based on the diameter of the BS, the appropriate type of dilation balloon was chosen, with a diameter 0.5 - 1mm

larger than the BS. The uninflated dilation balloon was inserted via the cholangioscope's instrument channel through the stricture position. Then, the balloon was slowly inflated with increasing pressure from 1 - 20atm, depending on the desired dilation size, and was held for about 2 minutes. This process was performed under the observation of the flexible cholangioscope to directly assess the result. The biliary dilation would be performed many times until the cholangioscope could go through the stricture. If the dilation was successful, we would continue to remove stones behind the stricture. After that, the indication for placing a biliary stent was considered. If the dilation was unsuccessful, consider other methods such as liver resection, biliary-enteric anastomosis, or conservative procedure.

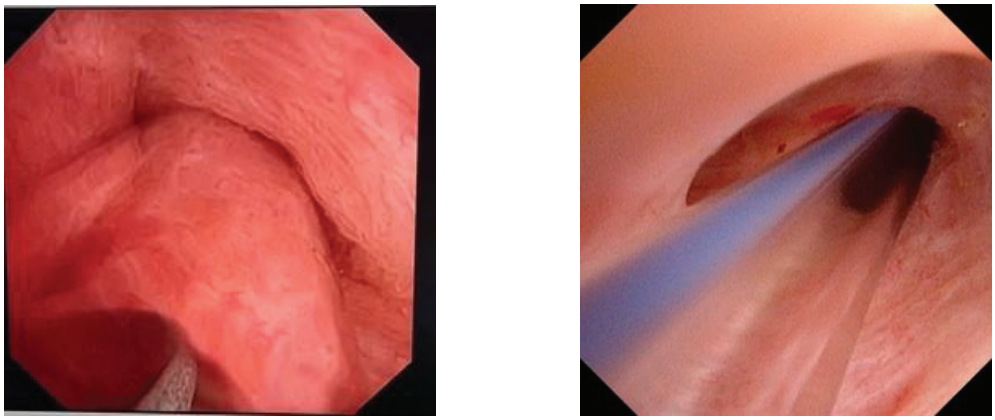

Figure 3. BS's balloon dilation and stent placement.

- Biliary stent placement: ACG clinical guideline 2023 recommended that if the BS was membranous, biliary stent placement would not be needed because it usually did not recoil after dilation. In other cases, the biliary stent should be placed [4]. In this study, we used a 16Fr biliary-cutaneous stent made of latex (T-tube drainage) or silicon.

- Patients who had completed stone clearance and BS dilation would be re-examined after 1, 3, and 6 months for re-evaluation. They would be clinically and para-clinically examined with ultrasound and magnetic resonance cholangio-pancreatography to check and compare with their surgery's results.

* *Data analysis:* The information was recorded and arranged in detail according to the research medical record form. The research data was processed using SPSS 20.0 software.

The percentages were calculated and presented in table form.

3. Ethics

This research has been approved by the Ethics Committee of Military Hospital 103 according to Decision No. 69/CNChT-HĐĐ, dated October 17th, 2022. The patients were clearly explained about the benefits and risks of the surgery, and agreed to participate in the study. Military Hospital 103 granted permission for the use and publication of the research data. The authors declare to have no conflicts of interest in this research.

RESULTS

From July 2021 to July 2024, we studied 62 patients.

* *Age and gender:* Patients' ages ranged from 20 - 86. The mean age was 60.1 ± 14.1. The female/male ratio was 1.69/1 (39/23). 75.8% of patients had a history of biliary stones, of which 54.8% had undergone biliary surgery.

* *Location of stones and BS:* Most patients had multiple stones (79%), including choledocholithiasis and hepatolithiasis. Stones were often located around the BS. The BS were mostly intrahepatic strictures. Left hepatic duct stricture accounted for the highest rate (30.6%). There were 69 BS among 62 patients.

Table 1. Characteristics of BS (n = 69).

Characteristics	Percentage (%)
Number of BS positions	
1	90.3
2	8.1
3	1.6
Classification of BS	
Mild	85.5
Moderate	8.7
Severe	5.8

All strictures were benign. The average length of BS was 3.96 ± 2.9mm. The average BS's diameter was 3.6 ± 0.7mm.

Table 2. Results of stone removal and BS dilation (n = 62).

Characteristics	Percentage (%)
Surgical methods	
Choledochotomy with intraoperative cholangioscopy	90.3
Percutaneous cholangioscopy	9.7
Stone removal methods	
Using basket only	11.3
Using baskets and electrohydraulic lithotripsy	21.0
Using basket and laser lithotripsy	37.1
Using basket, electrohydraulic, and laser lithotripsy	30.6
Balloon dilation of BS	
Success	87.1
Failure	12.9
Biliary-cutaneous stent placement	
T-tube 16Fr	30.6
Silicon tube 16Fr	33.9
No stent	35.5
Intraoperative complications	16.1
Bleed	14.5
Duodenal perforation	1.6
Postoperative complications	12.9
Bleed	3.2
Bile leakage	1.6
Surgical site infection	4.9
Acute pancreatitis	3.2
General results after surgery	
Completed stone clearance	83.9
Completed BS dilation	87.1

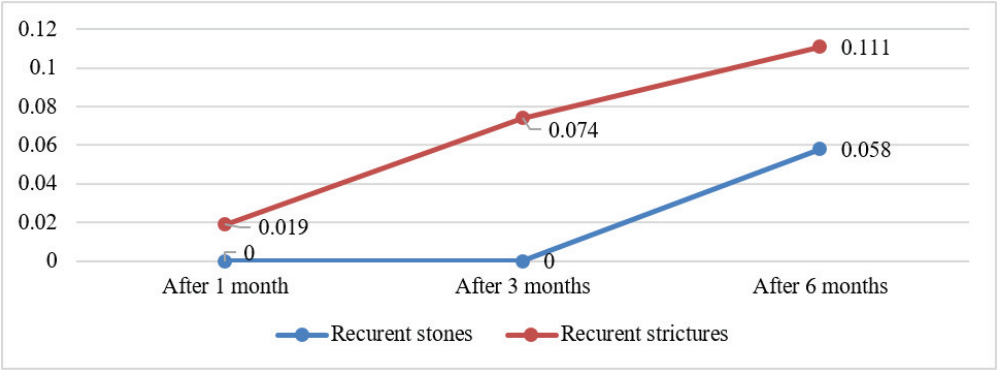

Chart 1. Recurrent biliary stones and strictures.

At 1, 3, and 6 months postoperative periodically, we scheduled follow-up examinations for 52 patients who had completed stone clearance and 54 patients who had completed BS dilation. The recurrence rates of stones and strictures were 0%, 0%, 5.8%, and 1.9%, 7.4%, 11.1%, respectively.

DISCUSSION

In this research, 75.8% of patients had a history of biliary stones, of which 54.8% had undergone biliary surgery. This showed that the prominent characteristic of BS patients was the high rate of residual and recurrence stones postoperative.

Stones were both choledocholithiasis and hepatolithiasis. Stones were often located around the BS, which were mostly intrahepatic strictures. Left hepatic duct stricture was the highest rate (30.6%). Nguyen Quang Nam also found that the rate of intrahepatic stones and strictures was also higher than that of extrahepatic ones. The author assumed that left hepatic stones were more common than right hepatic stones (20.8% compared to 11.1%) [6].

In this study, when the majority of patients had multiple stones (79%), most of the cases were one-location BS (90.3%). We found that the average length and diameter of BS were 3.96 ± 2.9mm and 3.6 ± 0.7mm, respectively, and all BS were benign. Jeng KS published that the diameter of BS was the indication for choosing treatment methods, and the length of BS was the indication for biliary stent placement to prevent recoil [7]. We classify BS according to Lee SK [3]: 81.2% of BS were mild, and only 5.8% were severe.

Our most used surgical method was choledochotomy with intraoperative flexible cholangioscopy (90.3%). For patients who already had the percutaneous tunnel from previous interventions (8.7%), we performed flexible cholangioscopy via the existing channel without the

need for reoperation. Zhu J believed that the key to achieving a high stone clearance rate was using a variety of methods to remove the stones [8]. In this study, we used 3 methods to remove biliary stones via cholangioscopy such as baskets, electrohydraulic, and laser lithotripsy. The baskets were used alone only in 7 cases with few stones. The remaining cases used baskets combined with lithotripsy. Especially, we used both electrohydraulic and laser lithotripsy in 19 cases (30.6%) to take advantage of diverse stones (*Table 3*).

We used balloon dilation for BS. The results after dilation were evaluated using direct cholangioscopy. We successfully dilated 87.1% of cases and were able to continue removing stones in the bile duct behind the stricture. Only 8 cases (12.9%) failed to dilate because of severe (2 cases) and moderate (6 cases) strictures.

40/62 patients had biliary stents placed to prevent strictures recoiling. In patients with 2 or 3 BS, only 1 stent was placed through all strictures. We did not place stents on other cases with a low possibility of recoiling according to the ACG guideline [4] or BS dilation failure. We used 2 types of stents: T-tube 16Fr and Silicon tube 16Fr. In fact, we used the T-tube in cases of extrahepatic or right, left hepatic BS (33.9%). In cases of section or segment BS, we use a silicone tube (35.5%).

Intraoperative complications were 16.1%, with 9 cases of biliary bleeding after dilation and lithotripsy, all of which were treated with hemostatic drugs. The remaining case was duodenal perforation during dissection to find the choledoc on the patient who had previous surgeries. We performed a 2-layer suture duodenal repair. Then, the patient was in stable condition. The postoperative complication rate was 12.9%, with biliary bleeding (2 cases), bile leak (1 case), surgical site infection (3 cases), and acute pancreatitis (2 cases). These patients were all treated with medicine and were stable without the need for reoperation. According to Nunes T, in general, complications of cholangioscopy were rare because it was a minimal intervention [5].

Our completed stone clearance and BS dilation rates were 83.9% and 87.1%, respectively. 8 patients who failed dilation still had biliary strictures. Among the 10 patients with residual stones, 8 had remained strictures, and 2 had inaccessible angled bile ducts. Zhang W found that when the bile duct was highly angled, the cholangioscope could neither properly investigate nor remove stones [9].

At 1, 3, and 6 months postoperative periodically, we scheduled follow-up examinations for 52 patients who had completed stone clearance and 54 patients who had completed BS dilation. After

1 and 3 months, no cases of stone recurrence were found; however, there were 1 (1.9%) and 4 cases (7.4%) of BS recurrence, respectively. After 6 months, the rate of stones and BS recurrence increased, at 5.8% and 11.1%, respectively. Le Quan Anh Tuan also concluded that the longer the postoperative period, the higher the risk of stones and BS recurrence [10].

CONCLUSION

Biliary dilation and stone removal via flexible cholangioscopy were initially safe and effective methods for treating primary BS and stones. These methods had a high rate of completed stone clearance and stricture dilation and a low rate of complications and recurrence.

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OUTCOMES OF LONG-STEM CEMENTLESS HEMIARTHROPLASTY FOR UNSTABLE INTERTROCHANTERIC FEMORAL FRACTURES IN GERIATRIC PATIENTS: A TWO-YEAR FOLLOW-UP STUDY

Pham Ngoc Thang[1], Nguyen Quoc Cuong[1]
Nguyen Thanh Thao[2], Vu Anh Dung[1,2]*

Abstract

Objectives: To assess the clinical outcomes of primary long-stem cementless bipolar hip arthroplasty as a treatment option for unstable intertrochanteric femoral fractures in geriatric patients. ***Methods:*** A retrospective study was conducted on 67 elderly patients aged > 70 years old with unstable intertrochanteric femoral fractures. Clinical outcomes were assessed based on the duration of walking without support, length of hospital stay, mortality rate, Harris Hip scores (HHS), and postoperative complications. ***Results:*** The mean age of patients was 84.34 ± 6.67. Fracture types included 43 cases of A2.2, 19 cases of A2.3, and 5 cases of A3.1. The mean follow-up duration was 27.89 ± 10.24 months. The mean duration of hospitalization was 15.37 ± 5.09 days. Median HHS at the last follow-up was 81.61 ± 7.06. Postoperative complications were four cases of pulmonary infection and one case of postoperative dislocation. During follow-up, the postoperative mortality rate was 5.97%. ***Conclusions:*** Primary cementless bipolar hemiarthroplasty is a secure and effective choice for the treatment of intertrochanteric femoral fracture in the elderly.

Keywords: Hemiarthroplasty; Geriatric patient; Intertrochanteric fracture; Cementless bipolar; Long-stem.

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INTRODUCTION

Intertrochanteric femoral fractures are one of the most important causes of functional failure and death among senior citizens, often resulting from low-energy trauma in the setting of osteoporosis, with over 150,000 patients reported annually in the United States [1]. The mortality rate of the elderly was 11.7 times greater than that of a population with similar characteristics after the first-year follow-up [2] since a period of restricted functional results in chronic comorbidities, including cardiovascular and pulmonary diseases. At present, various treatment options are available for intertrochanteric fractures, such as hip hemiarthroplasty, bone fusion, and conservative management. Each approach comes with its unique set of benefits, drawbacks, and specific indications for use. The main purpose is to achieve preinjury ambulation status and functional recovery as early as possible. Consequently, bipolar hemiarthroplasty becomes popular in the elderly population with earlier weight-bearing training and the same functional outcome compared to internal fixation [3]. Nowadays, it is still debatable whether cementless or cemented is a better option. Cemented stem provides quick prosthesis stability, but there is a risk of cement-related fatal cardiovascular complications, especially in the elderly with chronic conditions. On the other hand, few reports are found investigating cementless long-stem arthroplasty in the treatment of intertrochanteric fracture in the elderly. While cementless long-stem was often used for revision, some orthopedic doctors thought that long-stem cementless prosthesis was an option for primary hemiarthroplasty with additional stability. Our study aims to: *Evaluate the clinical outcomes of bipolar hemiarthroplasty using cementless long-stem in the treatment of unstable intertrochanteric femoral fracture in geriatric patients.* By addressing the existing gaps in the literature, this study may contribute to optimizing treatment strategies and improving the quality of care.

MATERIALS AND METHODS

1. Subjects

Including 67 patients (> 70 years old) who underwent bipolar hemiarthroplasty using cementless long-stem for unstable intertrochanteric femoral fracture at Military Hospital 103 from September 2020 to August 2023.

Retrospective data collected before surgery includes age, gender, fracture site, AO fracture classification, time

from hospitalization to intervention, time from trauma to hospitalization, cause of fracture, follow-up period, type of anesthesia, and comorbidities.

Low-molecular-weight heparin (enoxaparin) is given to patients until 12 hours preoperatively and 5 days postoperatively. The patients were under the spinal cord or general anesthesia. Procedures were performed by a standard direct-lateral approach. For rehabilitation, passive range of motion exercises were started on the first postoperative day, and partial weight-bearing exercises were started on 4 postoperative days.

2. Methods

* *Study design:* A retrospective study.

* *Study variables:* For clinical assessment, HHS was investigated at the last follow-up examination; length of hospitalization, mortality rate, and time from surgery to walking without support (cane, sticks, or walker) were recorded. Complications related to surgery were sought for their presence.

* *Statistical analysis:* Our data were loaded into the Microsoft Excel program, analyzed, and calculated by the medical statistics software SPSS 26.0, and the results were expressed in median (minimum, maximum) and standard deviation.

3. Ethics

The study received approval from the Institutional Ethics Committee. The Department of Joint Surgery at Military Hospital 103 granted permission for the utilization and publication of the research data. The authors are responsible for all aspects of the work to ensure that issues regarding the accuracy of any part of this study. Patients' data were kept confidential throughout the study to protect the anonymity of their information, and all participants gave written informed consent at the time of participation. The authors received no financial support and declare no conflicts of interest regarding the research, authorship, and publication of this article.

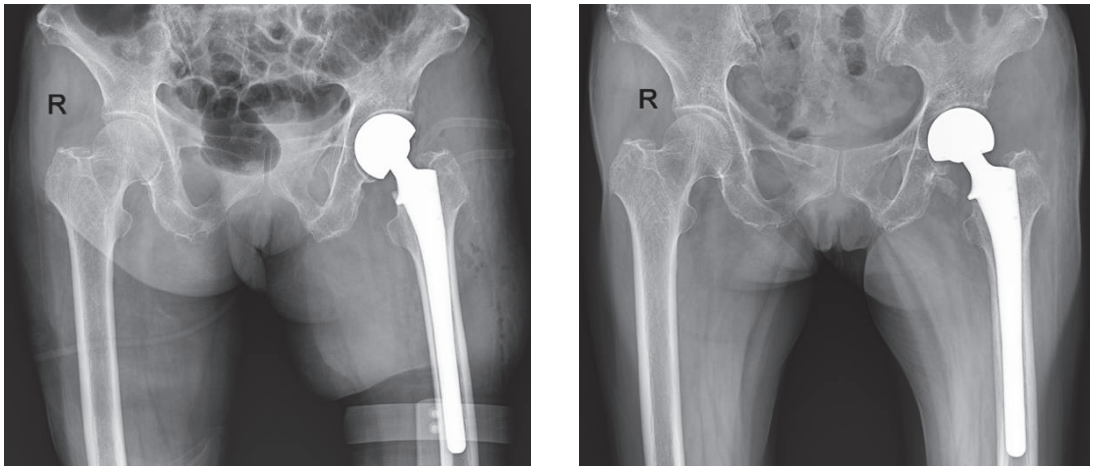
RESULTS

The preoperative data of 67 patients was shown in table 1. All patients underwent successful surgery, of which 31.34% were male and 68.66% were female. The average duration of hospitalization was from 8 - 38 days, and the mean follow-up period was from 12 - 44 months. Most patients with chronic preexisting diseases in which the group accounted for the highest rate of hypertension with 34%. According to the AO fracture classification, there were 64.17% type A2.2, 28.35% type A2.3, and 7.46% type A3.1.

Table 1. Demographic and clinical characteristics of patients (n = 67).

Characteristics	Values
Mean age (years)	84.34 ± 6.67 (71 - 98)
Male/female	21/46
Right/left side	23/44
Follow-up period (months)	27.89 ± 10.24
Time from hospitalization to surgery (days)	4.92 ± 2.96
Cause of fracture	
Road traffic accident	12 (17.9%)
Slip and fell	55 (82.1%)
Time from trauma to hospitalization	
< 24h	28 (41.8%)
24 - 48h	22 (32.8%)
> 48h	17 (25.4%)
AO/OTA fracture type	
A2.2	43 (64.2%)
A2.3	19 (28.3)
A3.1	5 (7.5%)
Metabolic disease (No. of patients)	
Cardiovascular disease	12 (17.9%)
Pulmonary disease	5 (7.4%)
Diabetes	12 (17.9%)
Hypertension	23 (34.3%)
Anesthesia mode (n)	
General	8 (11.9%)
Spinal	59 (88.1%)

The postoperative outcomes are shown in table 2. The average HHS was 81.61 ± 7.06 (range: 60 - 92) at the final follow-up, of which 16.41% were excellent and 58.2% were good. Before discharge from the hospital, all of them were treated. According to functional ambulation, 47.76% of patients walked without the help of aids at 3 months after the operation and 71.64% after 12 months. 19 patients delayed ambulation, 12 due to generalized weakness, and 7 due to associated concomitant fractures.



A B

Figure 1. Postoperative radiographs were taken immediately after surgery (A) and at the 2-year follow-up (B).

As a complication, there was 1 case of dislocation of the prosthesis head, which was manually reduced without any recurrence. 4 cases showed pulmonary infection, and 1 case showed urinary tract infection. There was not any case of surgical infection or deep vein thrombosis. In addition, 4 of them were lost by natural death (5 - 10 months post-surgery).

Table 2. Postoperative results of patients (n = 67).

Results	Values
Mean HHS	
Excellent (n)	11 (16.4%)
Good (n)	39 (58.2%)
Fair (n)	15 (22.4%)
Poor (n)	2 (3%)
Total hospitalisation (days)	15.37 ± 5.09
Postoperative hospital stay (days)	8.21 ± 4.43
Walking without support	
At 3 months (n)	32 (47.7%)
At 6 months (n)	40 (59.7%)
At 12 months (n)	48 (71.6%)

	Values
Mortality rate	
0 - 3 months (n)	0
3 - 12 months (n)	3 (4.5%)
12 - 24 months (n)	1 (1.5%)
Complications	
Dislocation (n)	1 (1.5%)
Pulmonary infection (n)	4 (5.9%)
Urinary tract infection (n)	1 (1.5%)

DISCUSSION

Unstable intertrochanteric fractures are among the most complex fractures in elderly patients. The primary goal of surgical treatment is to restore pre-fracture function and ambulatory status as quickly as possible. However, the optimal treatment for intertrochanteric fractures in the elderly remains a topic of debate. Osteosynthesis is often the preferred method; however, geriatric patients frequently have chronic comorbidities that increase the risk of osteosynthesis failure, necessitating revision to hemiarthroplasty. In our study, most patients had underlying conditions, including hypertension (34%), diabetes mellitus (17.9%), and cardiovascular accidents (17.9%). Primary hip replacement following intertrochanteric fracture is typically indicated for cases with degenerative hip disease, extensive comminution, or poor bone quality [4].

When compared with the results of proximal femoral nail anti-rotation, Chengkui Cai et al. assessed 70 patients aged > 70 years and reported that joint replacement allowed earlier mobilization post-surgery and better joint function within 12 months [5]. In our study, we utilized a cementless long femoral stem for elderly patients with intertrochanteric fractures, and 32/67 patients (47.76%) achieved independent ambulation within three months.

Shan Fan et al. investigated the clinical efficacy of locking compression plate fixation in 37 patients with femoral intertrochanteric fractures, reporting complications including deep vein thrombosis (3 patients), bedsores (1 patient), and delayed union (1 patient) [6]. Our findings indicated a general complication rate of 8.95%, with no cases of deep vein thrombosis.

Previous research suggests that cemented prostheses provide immediate

stability due to the cement-bone mechanical interlock. However, disadvantages such as prolonged surgical time and the risk of cement implantation syndrome, which can be life-threatening, have been noted [7]. Conversely, uncemented prostheses offer a faster and technically simpler procedure. Some authors have even argued that cementless implantation results in a lower risk of readmission, mortality, and postoperative complications compared to cemented procedures [8]. In our study, 4 patients (5.9%) were lost to follow-up due to natural death over a two-year period.

When compared to our findings, Qiang Mao et al. [9] observed a 6.9% mortality rate among 53 patients who underwent hemiarthroplasty using an uncemented long femoral stem for intertrochanteric fractures. They utilized the Peerless-160 long femoral stem for octogenarians, reporting an average HHS of 87.8 ± 6.1 at the final follow-up. Similarly, a retrospective study by Gema et al. [10] on 179 patients found a mean follow-up HHS of 85.28 ± 10.3, with a mean patient age of approximately 74.5 ± 8.1 years. In our study, the average HHS was 81.61 ± 7.06; however, our patients were older, with a mean age of 84.34 ± 6.67 years.

This study had several limitations, including a small sample size and a limited follow-up period. Additionally, the retrospective design makes the study susceptible to bias and confounding factors that may influence the results. Future research will explore these issues in greater detail.

CONCLUSION

The results of this study show that primary bipolar cementless long-stem hemiarthroplasty is the secure and effective option in the treatment of unstable intertrochanteric femoral fracture, yielding good functional outcomes. To enhance the reliability of the findings, a larger number of patient cohorts and a longer follow-up period should be required.

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SUBMISSION GUIDELINES FOR THE JOURNAL OF MILITARY PHARMACO-MEDICINE

The Journal of Military Pharmaco-medicine publishes 9 issues per year featuring research papers and review articles covering topics in military medicine, pharmacy, biomedicine, and socio-medicine, as well as domestic and international medical and pharmacological information, and research and training updates.

I. GENERAL GUIDELINES

1. The submitted scientific research paper should not have been previously published in any journal or accessible database.

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4. Manuscripts should be typed in English using Microsoft Word software, ensuring grammatical and spelling accuracy. The length of the manuscript, including tables, charts, figures, and references, should not exceed 10 A4 pages. Please use Times New Roman font, size 13 (Unicode font system), with line spacing set to 1.5, and margins according to the A4 standard. Manuscripts must include clear and centered page numbers.

5. Each article should contain no more than 5 tables, charts, and figures combined. Figures in formats such as .jpg or .pdf should be submitted as original files with captions. Table, chart, and figure captions should be integrated into the manuscript in their appropriate positions and numbered sequentially based on their order of appearance in the article.

6. References are arranged in the order they appear in the article. References are numbered and presented according to AMA regulations (refer to:https://libguides.usc.edu/ld.php?content_id=49818556). For Vietnamese authors, write their full names.

7. Terms used in the research paper should be unified according to the guidelines provided by the English Encyclopedia. Authors are expected to retain the original language when translating nouns from a foreign language. Abbreviations must be accompanied by captions that specify the full words represented by the abbreviations.

8. At the end of the article, the authors should include an acknowledgments section that clearly states the origin of the work, including where it was conducted, the timeframe of the research, and any support or sponsorship received from individuals or organizations.

II. SPECIFIC GUIDELINES FOR EACH TYPE OF SCIENTIFIC ARTICLES

1. Original research: According to IMRAD standards

- Title
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- Abstract
- Keywords
- Introduction
- Materials and methods
- Results
- Discussion (may combined with Results)
- Conclusion
- Acknowledgments
- References

2. Case report

- Title
- Authorship
- Abstract
- Keywords
- Introduction
- Case report
- Discussion
- Conclusion
- Acknowledgments
- References

3. Literature review

- Title
- Authorship
- Abstract
- Keywords
- Introduction
- Literature review
- Conclusion
- Acknowledgments
- References

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