creative commons
COMMONS DEED

저작자표시-비영리-변경금지 2.0 대한민국

이용자는 아래의 조건을 따르는 경우에 한하여 자유롭게

- 이 저작물을 복제, 배포, 전송, 전시, 공연 및 방송할 수 있습니다.

다음과 같은 조건을 따라야 합니다:

저작자표시. 귀하는 원저작자를 표시하여야 합니다.

비영리. 귀하는 이 저작물을 영리 목적으로 이용할 수 없습니다.

변경금지. 귀하는 이 저작물을 개작, 변형 또는 가공할 수 없습니다.

- 귀하는, 이 저작물의 재이용이나 배포의 경우, 이 저작물에 적용된 이용허락조건을 명확하게 나타내어야 합니다.
- 저작권자로부터 별도의 허가를 받으면 이러한 조건들은 적용되지 않습니다.

저작권법에 따른 이용자의 권리는 위의 내용에 의하여 영향을 받지 않습니다.

이것은 이용허락규약(Legal Code)을 이해하기 쉽게 요약한 것입니다.

Disclaimer

dCollection

연세대학교
YONSEI UNIVERSITY

The effect of glycolytic enzyme expression and thyroiditis on the aggressiveness of papillary thyroid carcinoma

Kim, Won Woong

Department of Medicine

Graduate School

Yonsei University

연세대학교
YONSEI UNIVERSITY

The effect of glycolytic enzyme expression and thyroiditis on the aggressiveness of papillary thyroid carcinoma

Advisor Nam, Kee-Hyun

A Dissertation Submitted
to the Department of Medicine
and the Committee on Graduate School
of Yonsei University in Partial Fulfillment of the
Requirements for the Degree of
Doctor of Philosophy in Medical Science

Kim, Won Woong

June 2025

연세대학교
YONSEI UNIVERSITY

The effect of glycolytic enzyme expression and thyroiditis on the aggressiveness of papillary thyroid carcinoma

This Certifies that the Dissertation of Kim, Won Woong is Approved

Committee Chair	**Koo, Ja Seung**
Committee Member	**Nam, Kee-Hyun**
Committee Member	**Kim, Daham**
Committee Member	**Kim, Jae-Woo**
Committee Member	**Chung, Ki-Wook**

Department of Medicine
Graduate School
Yonsei University
June 2025

연세대학교
YONSEI UNIVERSITY

ACKNOWLEDGEMENTS

This thesis would not have been possible without the support, guidance, and encouragement of many individuals, to whom I owe my deepest gratitude.

First and foremost, I would like to express my sincere appreciation to my advisor, Prof. Kee-Hyun Nam, whose mentorship, expertise, and unwavering support have been the cornerstone of my research journey. I would also like to extend my heartfelt thanks to my thesis committee members — Professors Da-Ham Kim, Ja-Seung Koo, Jae-Woo Kim, and Ki-Wook Chung — for their invaluable feedback, thought-provoking discussions, and constructive suggestions that helped refine and elevate this research.

On a personal note, I would like to express my deepest gratitude to my family and loved ones. I'm deeply grateful to my parents and parents-in-law for their unwavering support and encouragement. To my spouse, Sejin Park, for your unending patience, understanding, and encouragement. Your support has been my foundation throughout this endeavor.

Finally, I dedicate this thesis to endocrine surgery team, whose presence, encouragement, and belief in my abilities have been a source of inspiration throughout this academic journey.

Thank you all for being a part of this journey and for helping me reach this milestone.

July, 2025

Won Woong Kim

연세대학교
YONSEI UNIVERSITY

TABLE OF CONTENTS

연세대학교
YONSEI UNIVERSITY

연세대학교
YONSEI UNIVERSITY

LIST OF FIGURES

연세대학교
YONSEI UNIVERSITY

LIST OF TABLES

연세대학교
YONSEI UNIVERSITY

ABSTRACT

The effect of glycolytic enzyme expression and thyroiditis on the aggressiveness of papillary thyroid carcinoma

Background: Glycolytic enzymes have been extensively studied in various cancer types, revealing their aggressive characteristics and roles in tumor progression. However, it remains unclear whether glycolytic enzyme expression and peritumoral enzyme activity correlate with the behavior of papillary thyroid carcinoma (PTC). This study aimed to determine whether the expression of glycolytic enzymes is associated with aggressiveness—such as lymph node metastasis (LNM) and extrathyroidal extension (ETE)—in the presence or absence of chronic lymphocytic thyroiditis (CLT).

Methods: The expression of hexokinase 2 (HK2), lactate dehydrogenase A (LDHA), pyruvate kinase isoform M2 (PKM2), glucose transporter 1 (GLUT1), and monocarboxylate transporter 4 (MCT4) was examined in 233 PTC tissue specimens by immunohistochemistry. We evaluated whether the expression of these glycolytic enzymes correlates with LNM, ETE, and recurrence rate, both with and without CLT. Disease-free survival was compared after 1:3 propensity score matching based on by age, sex, tumor size, ETE, multifocality, and cervical LNM. In addition, we analyzed the correlation between glycolytic enzyme mRNA expression and clinicopathological characteristics in PTC using The Cancer Genome Atlas (TCGA).

Results: All glycolytic enzymes and transporter proteins were significantly overexpressed in PTC compared with normal tissue. There was a linear correlation among all glycolytic enzymes and transporter proteins, except for HK2. PKM2 expression was most highly correlated with the others. High PKM2 expression was significantly linked to increased recurrence risk in patients without CLT (HR 1.76, 95% confidence interval (CI) 1.01–3.06, p=0.046), but this association was not observed in those with CLT. Univariate and multivariate analyses showed that PKM2 mRNA expression and T staging were significantly correlated with LNM in TCGA data.

Conclusion: Overexpression of glycolytic enzymes such as LDHA, PKM2, and GLUT1 is associated with PTC. Interestingly, CLT is associated with greater local invasiveness (gross ETE) yet paradoxically lower recurrence. LDHA expression was lower in the presence of CLT, whereas PKM2 remained consistently associated with a higher recurrence rate in the absence of CLT. Based on TCGA data, PKM2 mRNA expression may serve as a promising biomarker for predicting LNM in PTC. Notably, patients with CLT exhibited better prognostic outcomes, even with elevated PKM2 expression, suggesting a potential protective role of CLT in modulating disease progression. Among the glycolytic enzymes examined, PKM2 may serve as a valuable biomarker for identifying patients at higher risk of recurrence.

연세대학교
YONSEI UNIVERSITY

Key words : glycolytic enzyme, Warburg effect, thyroid cancer, metabolic reprogramming, lymph node metastasis, chronic lymphocytic thyroiditis

연세대학교
YONSEI UNIVERSITY

1. Introduction

Papillary thyroid carcinoma (PTC) generally has a favorable prognosis; however, approximately 40–90% of cases demonstrate metastatic potential, particularly involving the lymph nodes[1]. One major factor contributing to a poorer prognosis in PTC is the presence of the $BRAF^{V600E}$ mutation, which has been associated with increased lymph node metastasis (LNM), recurrence, and cancer-related mortality[2-4]. Although testing for the $BRAF^{V600E}$ mutation aids in diagnosis, its value as an independent prognostic marker remains inconsistent, especially in populations with a high mutation prevalence, such as the Korean population[3,5-7]. The $BRAF^{V600E}$ mutation is frequently observed in papillary thyroid carcinoma (PTC), particularly in Western populations, with prevalence rates between 48% and 79%[8-10]. While its prognostic significance remains debated, studies consistently show an association with older age, advanced tumor stage, and higher recurrence risk when combined with other high-risk features. Notably, $BRAF^{V600E}$ mutation alone is not an independent predictor of recurrence, but its co-occurrence with TERT promoter mutations significantly worsens prognosis, correlating with immune suppression and poor outcomes[8]. Some cohorts also report lower 5-year survival rates in BRAF-positive cases (67.5% vs. 82.1%)[9]. Therefore, $BRAF^{V600E}$ mutation status should be interpreted in the context of additional clinicopathologic and molecular features, rather than used as a sole prognostic marker[10]. Additional markers are therefore needed to more reliably predict aggressiveness in PTC.

Recent research has highlighted the role of key glycolytic enzymes, including pyruvate kinase isoform M2 (PKM2) and lactate dehydrogenase A (LDHA), in tumor metabolism. These enzymes support cancer cell proliferation through enhanced glucose uptake and lactate production, even under aerobic conditions—a phenomenon known as the Warburg effect[3,11-16]. They are frequently overexpressed in aggressive malignancies, including PTC, and have been linked to increased invasiveness and unfavorable clinical outcomes[17,18]. In particular, PKM2 has been shown to be highly expressed in cancer-associated fibroblasts within the tumor microenvironment, thereby driving glucose uptake and lactic acid production. These metabolic changes promote cellular movement and invasiveness[19-23].

Lactate accumulation in the tumor microenvironment, primarily facilitated by monocarboxylate transporter 4 (MCT4), leads to extracellular acidification that supports tumor growth, invasion, and immune evasion[24,25]. High levels of LDHA, often associated with the $BRAF^{V600E}$ mutation, further enhance aggressiveness in PTC by promoting aerobic glycolysis. Preliminary studies have demonstrated that LDHA expression correlates with $BRAF^{V600E}$ mutational status, suggesting its potential as a prognostic biomarker [26,27].

In studies comparing recurrence rates and survival outcomes based on glycolytic enzyme expression across different thyroid cancer types, findings have indicated significant associations between enzyme overexpression and poor prognosis[28-30]. In medullary thyroid carcinoma (MTC), HK2 and MCT4 protein overexpression has been linked to lower survival rates. In poorly differentiated thyroid carcinoma (PDTC), MCT4 and glucose transporter 1 (GLUT1) protein overexpression have been associated with reduced survival rates. These findings suggest that the glycolytic shift may contribute to tumor aggressiveness and poor clinical outcomes beyond PTC[28].

연세대학교
YONSEI UNIVERSITY

In an anaplastic thyroid carcinoma (ATC) cell line study, MCT4 inhibition was reported to suppress ATC growth[29]. Another ATC cell line study found that PKM2 plays a role in enhancing metastasis and promoting aerobic glycolysis[30].

In one study, a significant increase in glycolytic enzymes such as GLUT1, HK2, PKM2, and LDHA was observed in thyroid inflammation compared to the normal control group, which was associated with an increased extracellular acidification rate and oxygen consumption rate. These findings suggest that thyroid inflammation may play a significant role in shaping the peritumoral microenvironment[31].

In a previous study, we examined whether glycolysis was increased in PTC by comparing glycolytic enzyme expression in cancerous versus matched normal thyroid tissues using immunohistochemistry (IHC). Patients with chronic lymphocytic thyroiditis (CLT) were excluded from that prior work. We subsequently compared glycolytic enzyme expression according to $BRAF^{V600E}$ mutational status. We found that LDHA levels were significantly higher in the $BRAF^{V600E}$ mutation group, whereas PKM2 and GLUT1 levels did not differ significantly between the $BRAF^{V600E}$ -mutated and BRAF wild-type groups. Additionally, we investigated whether the $BRAF^{V600E}$ mutation upregulates LDHA expression via activation of the mitogen-activated protein kinase signaling pathway in human thyroid cell lines, as well as whether inhibiting BRAF reduces LDHA expression in these cell lines. Furthermore, we indirectly confirmed that high LDHA mRNA expression is associated with PTC aggressiveness using data from The Cancer Genome Atlas (TCGA) thyroid cancer database [26].

The present study investigates the correlation between the expression of glycolytic enzymes, including LDHA, PKM2, hexokinase 2(HK2), GLUT1 and MCT4, and features of PTC aggressiveness such as lymph node metastasis and extrathyroidal extension according to the presence or absence of CLT. Our goal is to improve prognostic accuracy and inform clinical decision-making for patients with PTC.

연세대학교
YONSEI UNIVERSITY

2. Materials and methods

2.1. Patients

From January 2013 to December 2016, a total of 233 patients who underwent thyroid surgery and were preoperatively diagnosed with PTC by fine needle aspiration at Yonsei University Severance Hospital were enrolled. Inclusion and exclusion criteria were applied to ensure that only patients with available pathological results were included. All samples were obtained as formalin-fixed, paraffin-embedded tissue blocks.

Inclusion criteria:
1) $20 \leq$ age < 70
2) Papillary thyroid carcinoma (tumor size > 1 cm)

Exclusion criteria:
1) Age < 20 or age ≥ 70
2) Papillary thyroid microcarcinoma (tumor size < 1 cm)
3) Follicular, medullary, or anaplastic carcinoma
4) Distant metastasis
5) Insufficient histopathological results
6) History of radiation exposure

2.2. Propensity score matching process

Propensity score matching was used to create comparable groups of patients with and without recurrence, matching them in a 1:3 ratio based on age, sex, tumor size, extrathyroidal extension (ETE), multifocality, and cervical LN metastasis (LNM). Patients with similar propensity scores—representing comparable probabilities of recurrence—were matched to ensure balanced distributions of these six baseline covariates between the groups. After matching, seven patients were excluded due to a lack of formalin-fixed, paraffin-embedded tissue blocks.

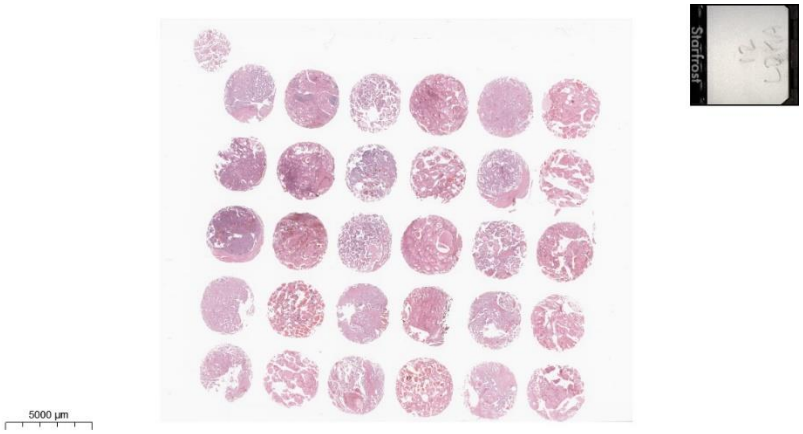
2.3. Preparation of tissue microarray

A 2-mm-diameter tissue microarray (TMA) apparatus (Tissue Microarray Set, Labro, Seoul, Korea) was used. Paraffin blocks containing vertically embedded thyroid tissue were carefully punched to obtain 2-mm-diameter, 5-mm-long paraffin cores. These cores, each containing representative tumor or normal thyroid tissue from a single patient, were embedded into a new paraffin block approximately 5–10 mm thick. A limitation of the Tissue Microarray (TMA) method is the potential heterogeneity in the distribution of thyroiditis, which may not be consistently captured during core sampling. To mitigate this issue, a pathologist carefully reviewed representative pathology slides to ensure that both tumor and peritumoral normal tissue were accurately represented in the selection process. The pre-manufactured plastic TMA cassette (2-mm lumen–sized TMA cassette, Labro, Seoul, Korea) contained thirty 2-mm-diameter sockets. Spacing

연세대학교
YONSEI UNIVERSITY

between transplanted tissues was maintained at 0.5 mm, allowing up to 30 tissue sections on a single slide.

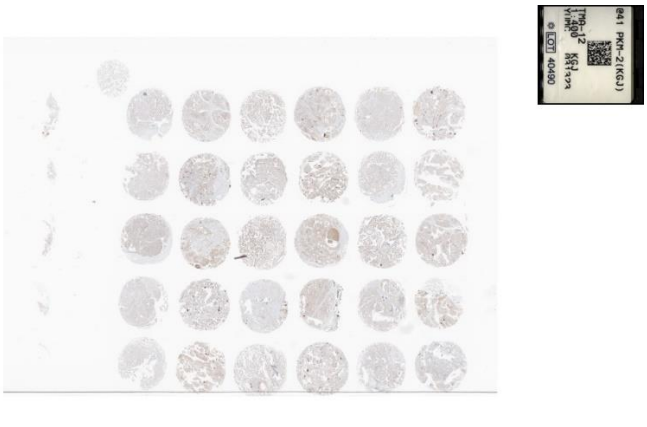
If a tissue core was not properly aligned in the paraffin block, the angle of the block was adjusted. After placing the paraffin cores into the cassette, it was immersed in molten paraffin (at approximately 65°C) within a metal mold designed for the TMA cassette. The mold was then placed on a hot plate for 5 minutes, allowing the paraffin tissue columns and the molten paraffin to fuse. Next, the mold was placed on a cold plate to solidify, creating a stable TMA block (Fig. 1).



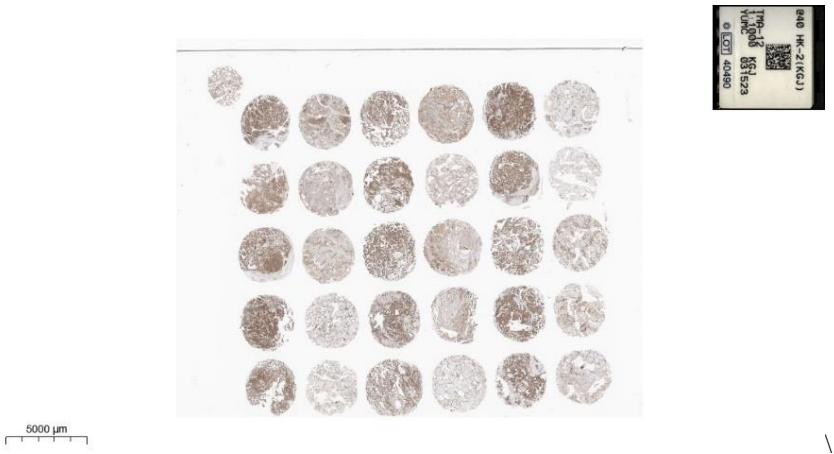
a) H&E stain

b) LDHA

연세대학교
YONSEI UNIVERSITY



c) PKM2



d) HK2

연세대학교
YONSEI UNIVERSITY

e) GLUT1

f) MCT4

Fig. 1. Immunohistochemistry (IHC) staining on Tissue microarray (TMA) including 15 paired cancer and normal tissue on a single slide. a)H&E stain, b)LDHA, c)PKM2, d)HK2, e)GLUT1, f)MCT4

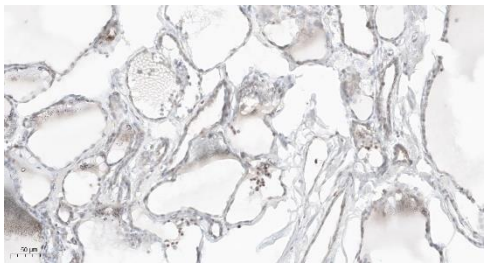
연세대학교
YONSEI UNIVERSITY

2.4. Immunohistochemistry(IHC) staining

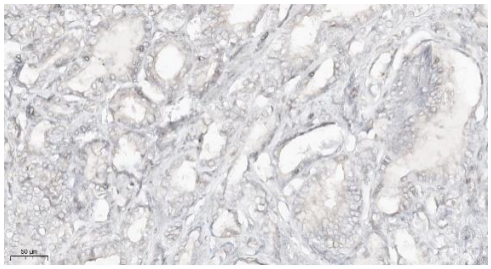
Paraffin-embedded tissue specimens were cut into 4-μm-thick sections. IHC staining was performed using a Discovery XT autoimmunostainer (750-701, Ventana, Tucson, AZ, USA) with monoclonal or polyclonal antibodies against GLUT1 (Dilution 1:100; Catalog No.: RM0063, Medaysis, USA), PKM2 (Dilution 1:100; Catalog No.: AF5234, Affinity, USA), LDHA (Dilution 1:400; Catalog No.: DF6280; Affinity, USA), hexokinase 2 (HK2; Dilution 1:1000; Catalog No.: BF0283, Affinity, USA), and MCT4 (SLC16A4; Dilution 1:100; Catalog No.: DF7145, Affinity, USA) according to the manufacturer's instructions.

IHC results were assessed by an expert using an immunoreactive score (IRS) derived by multiplying the intensity of staining (I) (0–3 points: absent, weak, moderate, and strong, respectively) by the percentage of positively stained tumor cells (P) (0–4 points: 0%, 1–10%, 11–49%, 50–80%, and 80–100%, respectively). The final IRS (H = P × I) was evaluated as low (0–4 points), moderate (6–8 points), or high (9–12 points). For risk factor analysis, the IRS was divided into low (0–4 points) and high (5–12 points) categories (Fig. 2).

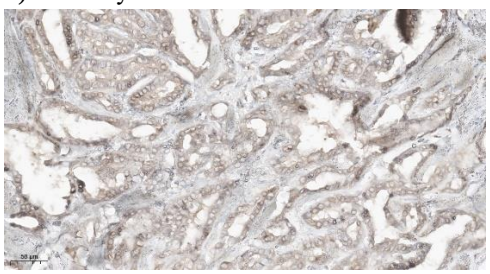
연세대학교
YONSEI UNIVERSITY



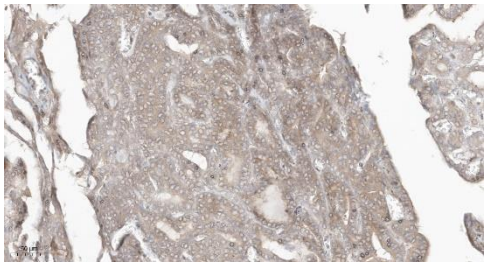
a) normal thyroid tissue



b) weakly



c) Intermediately



d) Strongly

Fig. 2. Immunohistochemial analysis of pyruvate kinase isoform M2 (PKM2) expression in normal and tumor tissue. a) normal thyroid tissue, b) weakly, c) intermediately, d) strongly stained papillary thyroid carcinoma (×400)

연세대학교
YONSEI UNIVERSITY

2.5. Data from TCGA thyroid cancer database

To investigate these associations in PTC, we used the TCGA dataset. Of the initial 507 thyroid cancer cases, we excluded patients with other malignancies or missing data on BRAF, LDHA, PKM2, and GLUT1 expression. Publicly available mRNA sequencing data, somatic mutation data, and clinical information from 465 patients with thyroid cancer were obtained from TCGA (version 2016_01_28; https://gdac.broadinstitute.org). Overall survival, disease-free survival, disease-specific survival, and progression-free survival data were downloaded from cBioPortal (TCGA, Firehose Legacy, and PanCancer Atlas). All data were fully anonymized prior to access. Thyroid cancer staging was based on the 7th edition of the American Joint Committee on Cancer (AJCC) staging system.

2.6. Primary outcomes assessment

This study examined whether glycolytic enzyme expression correlates with aggressiveness and oncological outcomes, such as recurrence, in intermediate-risk PTC larger than 1 cm. Furthermore, we evaluated whether glycolytic enzyme expression is associated with LNM, ETE, and recurrence in the presence or absence of CLT, a condition characterized by pathological lymphocytic infiltration of thyroid tissue and the presence of anti-thyroglobulin antibodies. In addition, this study aimed to evaluate whether glycolytic enzyme expression is correlated with aggressiveness, such as LNM and recurrence, using TCGA data.

2.7. Statistical analysis

Student's t-test was used to assess differences in continuous variables between groups. The χ^2 test or Fisher's exact test was used to compare categorical variables. Continuous variables are reported as the mean ± standard deviation with ranges, and categorical variables are expressed as percentages and absolute numbers. Univariate and multivariate analyses were performed to identify variables independently associated with recurrence, and odds ratios with 95% confidence intervals (CIs) were calculated. Differences with p-value < 0.05 were considered statistically significant.

Univariate and multivariate Cox proportional hazards modeling were conducted to assess the association between glycolytic enzyme expression and PTC aggressiveness. Hazard ratios (HRs) with 95% CIs were calculated. Statistical analyses were performed using SPSS Statistics for Windows, version 21.0 (IBM Corp., Armonk, NY, USA), with statistical significance defined as p-value < 0.05.

연세대학교
YONSEI UNIVERSITY

3. RESULTS

3.1. Expression of glycolytic enzymes in human thyroid cancer tissues

We assessed the expression of glycolytic enzymes and associated transporters in tissue microarrays, comparing normal thyroid tissue with cancerous tissue. HK2, PKM2, LDHA, and GLUT1 were all significantly elevated in tumor tissues. Although MCT4 levels also increased, this elevation was less pronounced than that observed for the other enzymes. These results indicate a prominent upregulation of glycolytic pathways in thyroid cancer (Table 1).

Given the observed differences in glycolytic enzyme expression, we subsequently explored the correlations among these enzymes to better understand their interactions within the glycolysis pathway.

Table 1. glycolytic enzymes and transporter proteins expression level in normal and papillary thyroid carcinoma tissues

variable	normal(n=233)	tumor(n=233)	p-value
HK2 expression			0.001*
Low	3	0	
Moderate	54	0	
High	176	233	
PKM2 expression			0.001*
Low	233	178	
Moderate	0	46	
High	0	9	
LDHA expression			0.001*
Low	230	124	
Moderate	3	58	
High	0	51	
GLUT1 expression			0.001*
Low	233	181	
Moderate	0	42	
High	0	10	
MCT4 expression			0.008*
Low	232	221	
Moderate	1	11	
High	0	1	

HK2, hexokinase 2 ; PKM2, pyruvate kinase isoform M2 ; LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

3.2. Correlation between enzymes in the glycolysis pathway

LDHA maintains the Warburg effect by converting pyruvate to lactate. PKM2 regulates pyruvate production, influences glycolytic flux, and activates the HIF-1α pathway, which in turn affects the expression of LDHA and MCT4. MCT4 enhances the external release of lactate produced by LDHA, thereby shaping a microenvironment conducive to cancer cell survival. Overall, increases in enzyme levels were observed concurrently, except in the case of HK2. Among these enzymes, PKM2 displayed the strongest correlations with the others, including a particularly robust correlation with MCT4 (r = 0.613). This suggests that pyruvate processing and lactate transport are closely linked, underscoring the central role of PKM2 in cancer metabolism and the Warburg effect (Fig. 3).

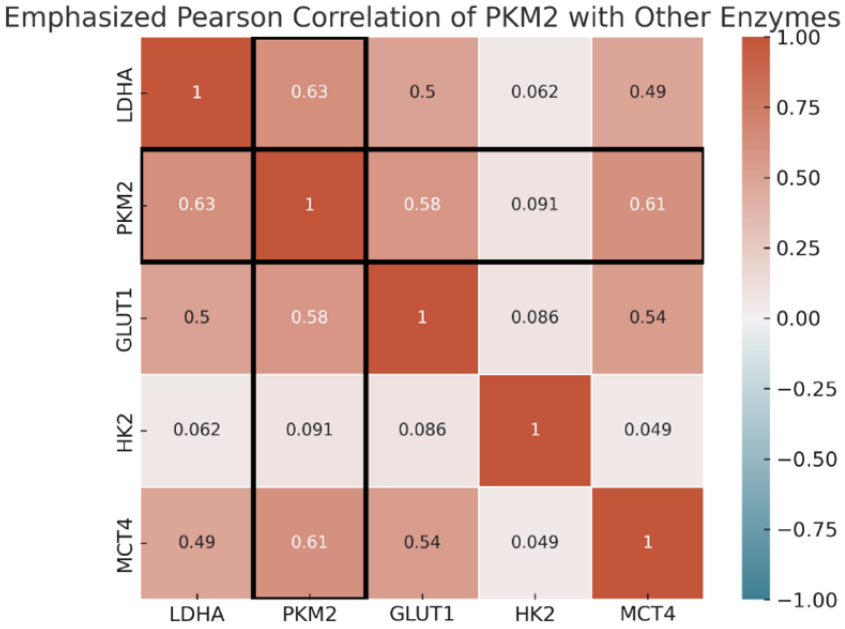


Fig. 3. Correlation among glycolytic enzyme expression levels in PTC

3.3. Clinical characteristics based on PKM2 expression levels

We compared the clinicopathological characteristics of PTC patients with low (n = 178) and high (n = 55) PKM2 expression. No significant differences were found between the two groups in terms of age, sex, tumor size, ETE, multifocality, lymphovascular invasion, $BRAF^{V600E}$ mutation or TNM staging. Expression levels of LDHA, GLUT1, and MCT4 were significantly elevated in the high PKM2 group (P =0.001), whereas HK2 levels showed no significant difference. However, patients with high PKM2 expression had a significantly higher recurrence rate (36.4%) than those with low

연세대학교
YONSEI UNIVERSITY

PKM2 expression (21.9%) (P = 0.031). Recurrences were predominantly regional lymph node events in both groups, and the sites of recurrence did not differ significantly (P = 0.579) (Table 2).

Thus, although PKM2 expression did not correlate with most clinicopathological factors, it was clearly associated with an increased risk of recurrence in PTC patients.

Having established that PKM2 correlates with recurrence, we then sought to identify additional factors contributing to direct invasion, a hallmark of aggressive thyroid cancer.

Table 2. Clinicopathological characteristics of patient with PTC according to PKM2 expression level

characteristic	low (n=178)	high (n=55)	p-value
Age	45.5 ± 13.5	43.4 ± 14.4	0.338
Sex			0.682
male	57 (32.0%)	16 (29.1%)	
female	121 (68%)	39 (70.9%)	
Tumor size, cm	1.75 ± 0.99	1.91 ± 1.84	0.398
Extrathyroidal extension			0.163
negative	33 (18.5%)	16 (29.1%)	
minimal	117 (65.7%)	34 (61.8%)	
gross	28 (15.7%)	5 (9.1%)	
Multifocality			0.36
negative	104 (58.4%)	38 (69.1%)	
unilateral	20 (11.2%)	5 (9.1%)	
bilateral	54 (30.3%)	12 (21.8%)	
Lymphovascular invasion			0.08
No	35 (19.7%)	17 (30.9%)	
Yes	143 (80.3%)	38 (69.1%)	
Thyroiditis			0.674
No	141 (79.2%)	45 (81.8%)	
Yes	37 (20.8%)	10 (18.2%)	
Clinical N stage			0.904
cN0	124 (69.7%)	38 (69.1%)	
cN1a	19 (10.7%)	5 (9.1%)	
cN1b	35 (19.7%)	12 (21.8%)	
T stage AJCC8th			0.107
T1	119 (66.9%)	37 (67.3%)	
T2	20 (11.2%)	11 (20%)	
T3	24 (13.5%)	2 (3.6%)	
T4	15 (8.4%)	5 (9.1%)	

연세대학교
YONSEI UNIVERSITY

N stage			0.769
N0	64 (36%)	20 (36.4%)	
N1a	76 (42.7%)	21 (38.2%)	
N1b	38 (21.3%)	14 (25.5%)	
M stage			0.947
M0	175 (98.3%)	54 (98.2%)	
M1	3 (1.7%)	1 (1.8%)	
$BRAF^{V600E}$ mutation			0.54
WT	10 (11.9%)	4 (16.7%)	
V600E	74 (88.1%)	20 (83.3%)	
Extent of surgery			0.269
Less than total	36 (20.2%)	15 (27.3%)	
Bilateral total thyroidectomy	142 (79.8%)	40 (72.7%)	
Recurrence			0.031*
No	139 (78.1%)	35 (63.6%)	
Yes	39 (21.9%)	20 (36.4%)	
Recurrence site			0.579
Regional lymph node	36 (90%)	20 (95.2%)	
Distant metastasis	4 (10%)	1 (4.8%)	
HK2 expression			0.999
Low	0	0	
High	178 (100%)	55 (100%)	
LDHA expression			0.001*
Low	115 (64.6%)	9 (16.4%)	
High	63 (35.4%)	46 (83.6%)	
GLUT1 expression			0.001*
Low	157 (88.2%)	24 (43.6%)	
High	21 (11.8%)	31 (56.4%)	
MCT4 expression			0.001*
Low	177 (99.4%)	44 (80.0%)	
High	1 (0.6%)	11 (20.0%)	

HK2, hexokinase 2 ; PKM2, pyruvate kinase isoform M2 ; LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

3.4. Expression of glycolytic enzymes in PTC with and without Thyroiditis

We assessed the expression of glycolytic enzymes and associated transporters in tissue microarrays, comparing cases with thyroiditis to those without thyroiditis. High LDHA expression was more prevalent in the thyroiditis-negative group (52.7%) than in the thyroiditis-positive group (23.4%). The expression levels of other glycolytic enzymes (HK2, PKM2, GLUT1, and MCT4) did not show significant differences based on thyroiditis status (Table 3).

Table 3. Glycolytic enzyme and transporter expression level according to thyroiditis

variable	Thyroiditis (-) (n=186)	Thyroiditis (+) (n=47)	p-value
HK2 expression			0.999
Low	0	0	
High	186 (100%)	47 (100%)	
PKM2 expression			0.674
Low	141 (75.8%)	37 (78.7%)	
High	45 (24.2%)	10 (21.3%)	
LDHA expression			0.001*
Low	88 (47.3%)	36 (76.6%)	
High	98 (52.7%)	11 (23.4%)	
GLUT1 expression			0.325
Low	147 (79.0%)	34 (72.3%)	
High	39 (21.0%)	13 (27.7%)	
MCT4 expression			0.132
Low	174 (93.5%)	47 (100%)	
High	12 (6.5%)	0	

HK2, hexokinase 2 ; PKM2, pyruvate kinase isoform M2 ; LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

3.5. Risk factors for gross ETE

Older age (>55), the presence of thyroiditis, and larger tumor size were significantly associated with an increased risk of gross ETE in both univariate and multivariate analyses. Age and thyroiditis were particularly influential, with ORs of 4.73 ($P < 0.001$) and 4.42 ($P < 0.001$), respectively. Other factors, including sex, $BRAF^{V600E}$ mutation and the expression levels of LDHA, GLUT1, and PKM2, were not significantly associated with gross ETE (Table 4).

Table 4. Risk factor for gross ETE invasion in PTC

Gross ETE	univariate			multivariate		
variable	OR	95% CI	p- value	OR	95% CI	p- value
Age(vs. <55)	3.34	1.52–7.33	0.003	4.73	1.96–1.41	0.001*
Male sex	1.24	0.56– 2.75	0.593			
Multifocality	1.57	1.04– 2.35	0.031	1.47	0.95–2.27	0.085
Thyroiditis	2.54	1.12–5.75	0.026	4.42	1.74–11.23	0.002*
Tumor size	1.65	1.20– 2.26	0.002	1.84	1.30–2.61	0.001*
$BRAF^{V600E}$ mutation	1.21	0.14 – 10.48	0.863			
LDHA high (vs. low)	1.25	0.59– 2.67	0.563			
GLUT1 high (vs. low)	0.81	0.32– 2.10	0.671			
PKM2 high (vs. low)	0.59	0.21–1.60	0.297			

LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; PKM2, pyruvate kinase isoform M2; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

3.6. Risk factors for clinical lymph node metastasis

In univariate analysis, male sex, multifocality, bilateral tumors, minimal and gross ETE, and larger tumor size were significantly associated with an increased risk of clinical LNM. In contrast, $BRAF^{V600E}$ mutation, multifocality, bilaterality and thyroiditis were not significant association with LNM. Following multivariate analysis, gross ETE emerged as the most robust independent predictor of LNM, while minimal ETE lost significance after adjusting for other factors. Elevated expression levels of LDHA, GLUT1, PKM2, and MCT4 were also not significantly associated with LNM in these analyses (Table 5).

Table 5. Risk factor for lymph node metastasis in PTC

LNM	Univariate			multivariate		
variable	OR	95% CI	p- value	OR	95% CI	p- value
Age (vs. >55)	1.94	1.04–3.60	0.036	2.33	1.19–4.56	0.013*
Male	2.37	1.27–4.44	0.007	2.53	1.30–4.92	0.006*
Multifocality	1.53	0.6–3.91	0.373			
Bilateral	1.04	0.57–1.91	0.894			
Thyroiditis	1.42	0.71–2.84	0.318			
Gross ETE (vs. minimal ETE and negative)	2.60	1.39– 4.89	0.003	1.129	0.39–3.27	0.824
Tumor size	1.09	0.86–1.38	0.460			
T stage						
T1	1			1		
T2	4.6	0.47–44.60	0.188	2.81	0.27–28.70	0.384
T3	2.07	1.05–4.07	0.036	1.93	0.96–3.88	0.064
T4	21.85	2.68–178.12	0.004	23.87	2.85–199.64	0.003*
$BRAF^{V600E}$ mutation	1.03	0.32 – 3.32	0.965			
LDHA high (vs. low)	0.65	0.38–1.12	0.120	0.72	0.41–1.27	0.259
GLUT1 high(vs. low)	0.88	0.46–1.65	0.681			
PKM2 high(vs. low)	0.98	0.52–1.84	0.956			
MCT4 high(vs. low)	1.14	0.33–3.89	0.841			

LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; PKM2, pyruvate kinase isoform M2; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

3.7. Risk factors for recurrence

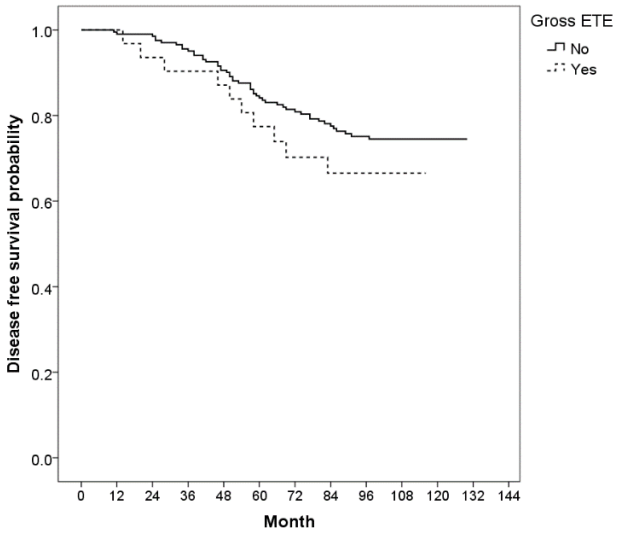
Both univariate and multivariate analyses identified male sex and high PKM2 expression as significant independent predictors of recurrence. Although larger tumor size was significant in univariate analysis, it did not maintain significance after adjustment. Other variables, including multifocality, ETE, $BRAF^{V600E}$ mutation and the expression of LDHA, GLUT1 and MCT4, were not significantly associated with recurrence. High PKM2 expression (OR 2.05, $P = 0.036$) and LNM (OR 3.90, $P = 0.001$) were linked to increased recurrence risk, whereas thyroiditis was associated with a reduced recurrence risk (OR 0.40, $P = 0.043$) (Table 6) (Fig. 4).

Table 6. Risk factor for recurrence in PTC

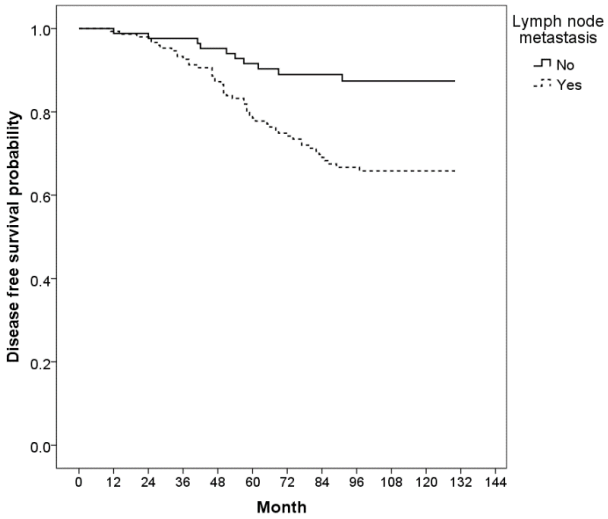
Recurrence	univariate			multivariate		
variable	OR	95% CI	p- value	OR	95% CI	p- value
Age (vs. <55)	0.80	0.39–1.65	0.551			
Male	1.93	1.04–3.57	0.036	1.64	0.86–3.13	0.133
Multifocality	0.90	0.64–1.26	0.524			
Thyroiditis	0.45	0.19–1.07	0.071	0.40	0.16–0.97	0.043*
Extrathyroidal extension(ETE)						
negative or minimal	1			1		
gross	1.49	0.66–3.37	0.342			
Tumor size	1.65	1.01–2.70	0.046	0.96	0.75–1.24	0.764
pN1	3.63	1.72–7.63	0.001	3.90	1.83–8.31	0.001*
$BRAF^{V600E}$ mutation	0.719	0.205-2.528	0.607			
LDHA high (vs. low)	1.36	0.75–2.46	0.306			
GLUT1 high (vs. low)	1.43	0.72–2.82	0.307			
PKM2 high (vs. low)	2.04	1.06–3.92	0.033	2.05	1.05–4.01	0.036*
MCT4 high (vs. low)	2.21	0.67–7.25	0.191			

LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; PKM2, pyruvate kinase isoform M2; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

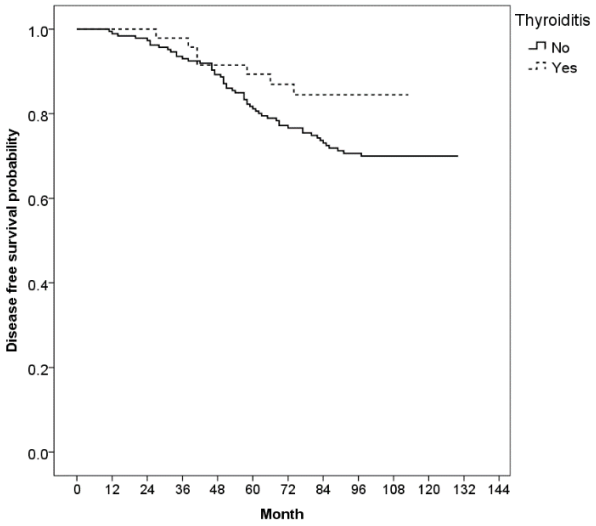


a) Gross ETE (*P*=0.253)



b) Lymph node metastasis (Hazard ratio(HR) 3.903, *P*=0.001)

연세대학교
YONSEI UNIVERSITY

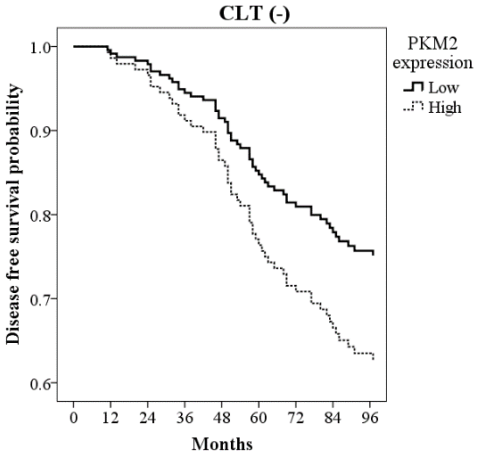


c) Thyroiditis (HR 0.415, *P* =0.034)

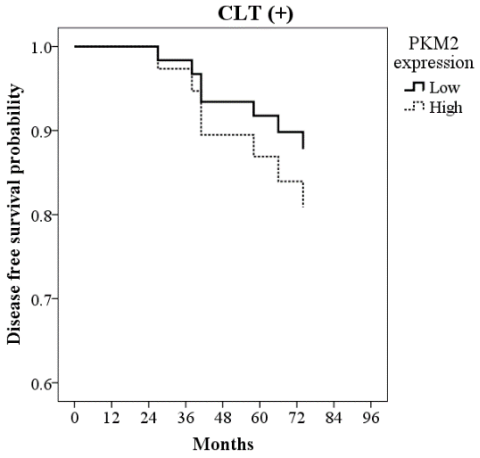
Fig. 4. Cox regression analysis evaluation the disease free survival (DFS) rate according to clinicopathological factors. a) gross ETE, b) lymph node metastasis, c) thyroiditis

Cox regression analysis of disease-free survival (DFS) based on glycolytic enzyme expression and the presence of chronic lymphocytic thyroiditis (CLT) revealed distinct prognostic implications. High PKM2 expression was significantly associated with an increased risk of recurrence in patients without CLT (HR 1.76, 95% confidence interval (CI) 1.01–3.06, *P*=0.046), whereas no such association was observed in those with CLT (HR 0.20, 95% CI 0.02–2.31, *P*=0.197). LDHA and GLUT1 expression had no significant impact on DFS, regardless of CLT status. These findings suggest that PKM2 expression may be a potential prognostic marker in the absence of CLT, while the presence of CLT may attenuate its influence on disease recurrence (Fig. 5).

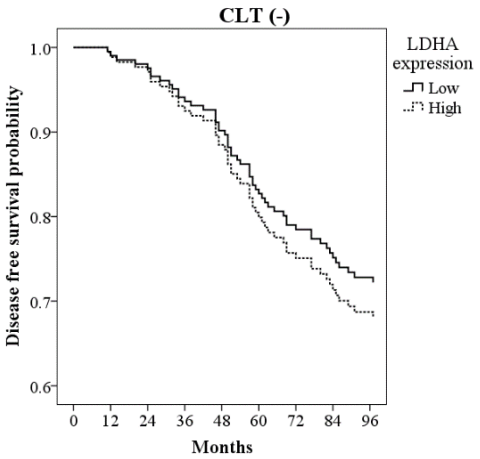
연세대학교
YONSEI UNIVERSITY



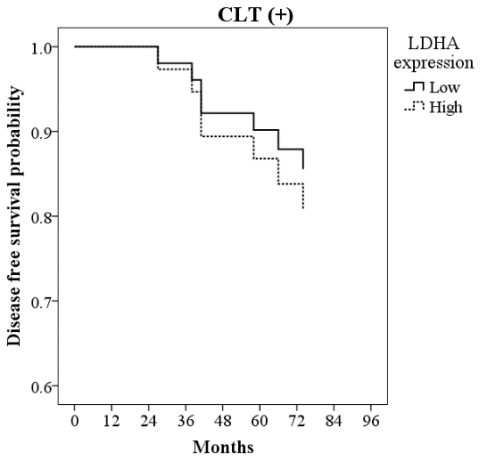
a) PKM2 expression without CLT
(HR 1.76 (95% CI 1.01-3.06), *P*=0.046)



b) PKM2 expression with CLT
(HR 0.20 (95% CI 0.02-2.31), *P* =0.197)

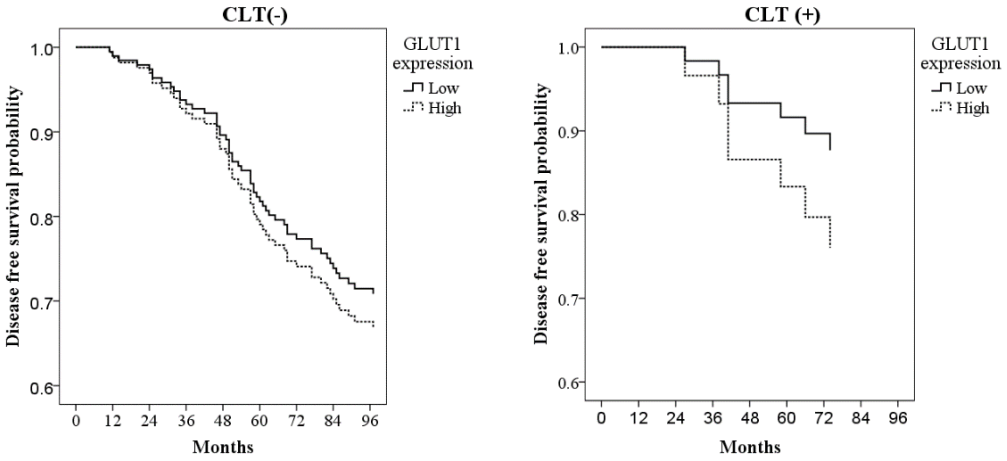


c) LDHA expression without CLT
(HR 1.18 (95% CI 0.69-2.03), *P* =0.544)



d) LDHA expression with CLT
(HR 1.37 (95% CI 0.27-7.07), *P* =0.708)

연세대학교
YONSEI UNIVERSITY



e) GLUT1 expression without CLT (HR 1.17 (95% CI 0.63-2.18), *P* =0.627)

f) GLUT1 expression with CLT (HR 2.08 (95% CI 0.47-9.31), *P* =0.337)

Fig. 5. Cox regression analysis of DFS rate based on glycolytic enzyme expression stratified by presence of chronic lymphocytic thyroiditis (CLT). a) PKM2 expression without CLT, b) PKM2 expression with CLT, c) LDHA expression without CLT, d) LDHA expression with CLT, e) GLUT1 expression without CLT, f) GLUT1 expression with CLT

연세대학교
YONSEI UNIVERSITY

3.8. Peritumoral microenvironment and cancer recurrence in relation to thyroiditis

Given the apparent association between thyroiditis and recurrence, we further investigated how the peritumoral microenvironment, particularly in the presence of CLT, influenced recurrence.
In cancer tissues from patients who experienced recurrence, the IRS values of LDHA, PKM2, and HK2 were similar regardless of CLT status. In cancer tissue from patients who did not experience recurrence, the IRS values of LDHA were significantly lower with CLT (P =0.049), suggesting that CLT may contribute to a metabolically reprogrammed and less aggressive tumor microenvironment. However, among patients with recurrence, the presence of CLT was linked to higher IRS of LDHA in peritumoral normal tissues (P =0.047) (Fig. 6).

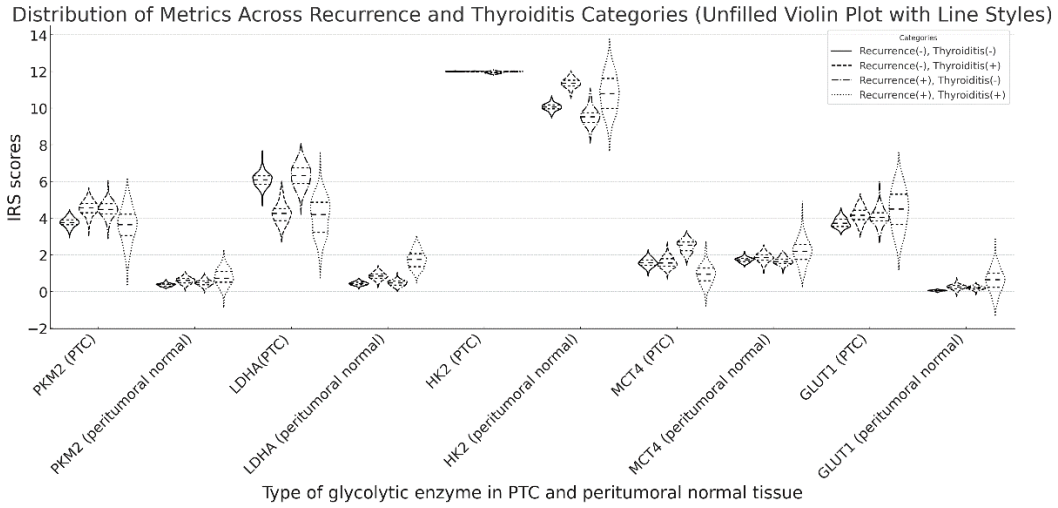
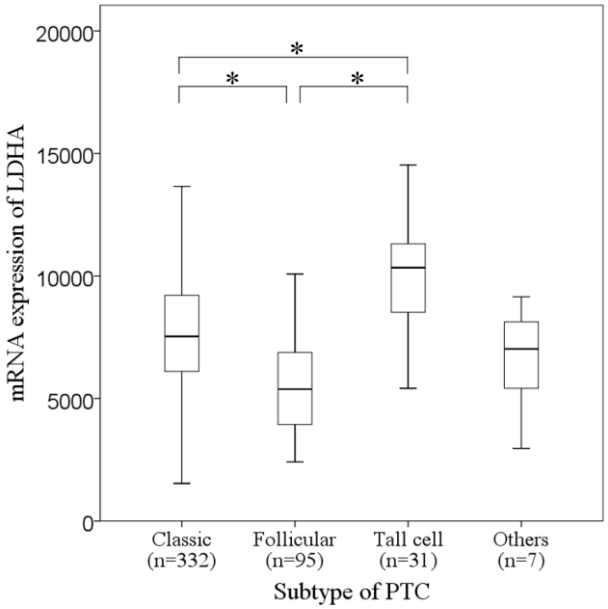


Fig. 6. Immunoreactive score (IRS) of glycolytic enzymes in peritumoral normal and cancer tissues accompanied by CLT in the recurrence group. PKM2, LDHA, HK2, MCT4, GLUT1

연세대학교
YONSEI UNIVERSITY

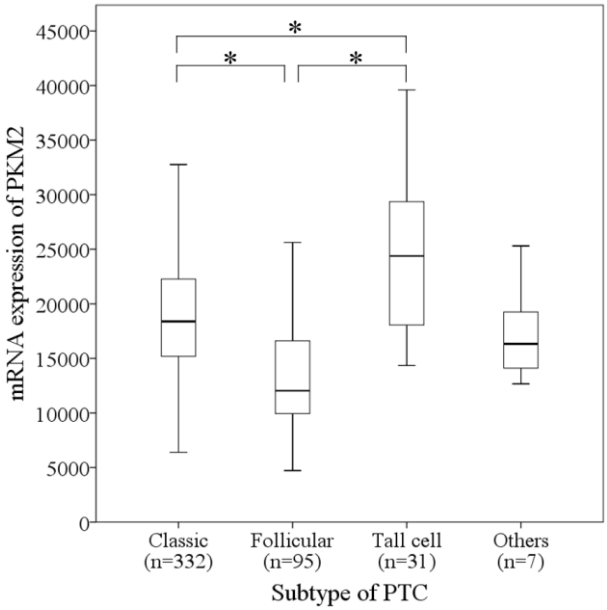
3.9. mRNA expression of glycolytic enzymes and transporter protein according to PTC subtype using TCGA thyroid cancer data

In PTC subtypes, significant differences in enzyme expression were observed. PKM2 and LDHA expression were highest in the tall cell subtype, followed by the classic, others, and follicular subtypes, with statistically significant differences among the classic, follicular, and tall cell subtypes ($P< 0.001$). Similarly, for GLUT1 expression, the tall cell and classic subtypes exhibited higher levels than the follicular subtype ($P < 0.001$). These findings suggest that glycolytic enzyme overexpression is associated with more aggressive PTC subtypes, particularly the tall cell subtype (Fig. 7).

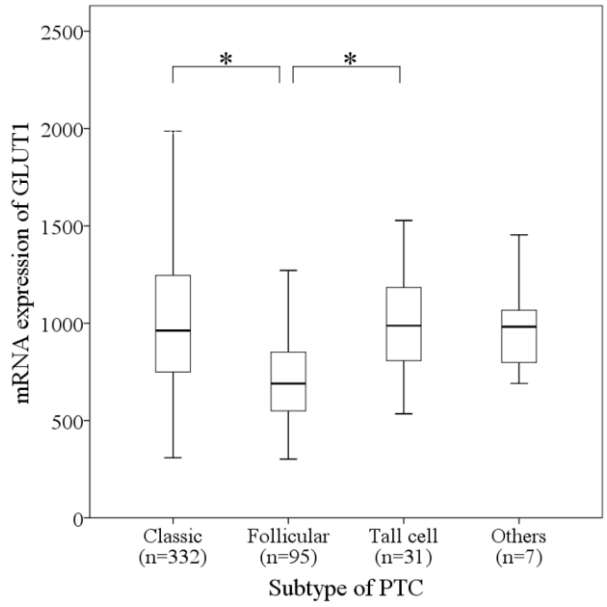


a) LDHA

연세대학교
YONSEI UNIVERSITY



b) PKM2



c) GLUT1

Fig. 7. mRNA expression of glycolytic enzymes and transporter proteins according to PTC subtype. glycolytic enzyme overexpression is linked to more aggressive PTC subtypes, particularly the tall cell subtype. a) LDHA, b) PKM2, c) GLUT1

* $p<0.05$

연세대학교
YONSEI UNIVERSITY

3.10. mRNA expression of glycolytic enzymes and transporter protein according to BRAF^{V600E} mutation using TCGA thyroid cancer data

mRNA expression levels of LDHA, PKM2, and GLUT1 were significantly elevated in PTC samples harboring the BRAF^{V600E} mutation compared to wild-type cases (P = 0.001 for all). High expression of LDHA, PKM2, and GLUT1 was markedly more frequent in the BRAF^{V600E} mutation group (34.1%, 34.6%, and 36.9%, respectively) than in the wild-type group (16.5%, 17.3%, and 14.5%). Conversely, low expression levels were predominant in wild-type tumors. These findings suggest a strong association between BRAF^{V600E} mutation and glycolytic pathway activation in PTC, underscoring the role of metabolic reprogramming in tumor progression (Table 7).

Table 7. mRNA Expression levels of glycolytic enzymes and transporter proteins according to the presence of BRAF^{V600E} mutation

Variable	BRAF wild type (n=248)	BRAF^{V600E} mutation (n=217)	*P*-value
LDHA mRNA expression			0.001*
Low	99 (39.9%)	19 (8.8%)	
Intermediate	108 (43.5%)	124 (57.1%)	
High	41 (16.5%)	74 (34.1%)	
PKM2 mRNA expression			0.001*
Low	102 (41.1%)	13 (6.0%)	
Intermediate	103 (41.5%)	129 (59.4%)	
High	43 (17.3%)	75 (34.6%)	
GLUT1 mRNA expression			0.001*
Low	101 (40.7%)	19 (8.8%)	
Intermediate	111 (44.8%)	118 (54.4%)	
High	36 (14.5%)	80 (36.9%)	

LDHA, lactate dehydrogenase A; PKM2, pyruvate kinase isoform M2; GLUT1, glucose transporter 1

연세대학교
YONSEI UNIVERSITY

3.11. Clinical characteristics of patients according to PKM2 enzyme expression levels using TCGA thyroid cancer data

The table compares clinicopathological characteristics among patients with PTC stratified by low, moderate, and high PKM2 mRNA expression. There were no significant differences in age, sex, tumor size, multifocality, or distant metastasis.

ETE was significantly associated with PKM2 expression, with gross extension occurring more frequently in the high-expression group (*P*=0.001). Additionally, significant differences in T stage and N stage were noted across PKM2 expression levels; the high-expression group demonstrated a greater proportion of advanced T stages (T3 and T4) and lymph node involvement (N1a and N1b) (*P*=0.016, *P*=0.001).

$BRAF^{V600E}$ mutation, as well as LDHA and GLUT1 mRNA expression, also correlated significantly with PKM2 expression. The high-PKM2-expression group exhibited a higher frequency of $BRAF^{V600E}$ mutations and elevated LDHA and GLUT1 mRNA levels (*P*=0.001). Subtype distribution varied with PKM2 expression levels, as the tall cell subtype was more common in the high-PKM2-expression groups, whereas the follicular subtype was more common in the low-expression group (*P*=0.001).

Overall, high PKM2 expression was associated with features indicative of aggressive tumor characteristics, including LNM, $BRAF^{V600E}$ mutation, and increased glycolytic enzyme and transporter protein expression, suggesting a potential role for PKM2 in the aggressiveness and poor prognosis of PTC (Table 8).

Table 8. Clinicopathological factors and glycolytic enzyme expression according to PKM2 mRNA expression using TCGA thyroid cancer data

characteristic	PKM2 expression			*P* value
	Low (n=115)	Moderate(n=232)	High (n=118)	
Age	47.2 ± 15.8	44.2 ±14.9	50.5 ±16.0	0.167
Sex				0.346
Male	24 (20.9 %)	62 (26.7%)	34 (28.8%)	
Female	91 (79.1%)	170 (73.3%)	84 (71.2%)	
Tumor size	3.02 ± 1.77	2.77 ± 1.54	3.03 ± 1.65	0.270
Extrathyroidal extension				0.001
Negative	96 (83.5%)	159 (68.5%)	59 (50.0%)	
Minimal	15 (13.0%)	60 (25.9%)	46 (39.0%)	
Gross	0	6 (2.6%)	10 (8.5%)	
Unknown	4 (3.5%)	7 (3.0%)	3 (2.5%)	

연세대학교
YONSEI UNIVERSITY

Multifocality				0.250
Negative	62 (53.9%)	116 (50.0%)	72 (61.0%)	
Positive	49 (42.6%)	111 (47.8%)	45 (38.1%)	
Unknown	4 (3.5%)	5 (2.2%)	1 (0.8%)	
T stage (AJCC 7th)				0.016
T1	37 (32.3%)	69 (29.7%)	26 (22.0%)	
T2	44 (38.3%)	79 (34.1%)	33 (28.0%)	
T3	31 (27.0%)	77 (33.2%)	49 (41.5%)	
T4	2 (1.7%)	7 (3.0%)	10 (8.5%)	
Tx	1 (0.9%)	0	0	
N stage				0.001
N0	74 (64.3%)	100 (43.1%)	38 (32.2%)	
N1a	19 (16.5%)	73 (31.5%)	46 (39.0%)	
N1b	7 (6.1%)	37 (15.9%)	26 (22.0%)	
Nx	15 (13.0%)	22 (9.5%)	8 (6.8%)	
Distant metastasis				0.087
M0	50 (43.9%)	137 (59.1%)	67 (56.8%)	
M1	3 (2.6%)	3 (1.3%)	1 (0.8%)	
Mx	61 (53.5%)	92 (39.7%)	50 (42.4%)	
$BRAF^{V600E}$ mutation	13 (11.3%)	129 (55.6%)	75 (63.6%)	0.001
LDHA mRNA expression				0.001
Low	78 (67.8%)	35 (15.1%)	5 (4.2%)	
Moderate	36 (31.3%)	149 (64.2%)	47 (39.8%)	
High	1 (0.9%)	48 (20.7%)	66 (55.9%)	
GLUT1 mRNA expression				0.001
Low	71 (61.7%)	38 (16.4%)	11 (9.3%)	
Moderate	41 (35.7%)	135 (58.2%)	53 (44.9%)	
High	3 (2.6%)	59 (25.4%)	54 (45.8%)	
Subtype of PTC				0.001
Classic	57 (49.6%)	180 (77.6%)	95 (80.5%)	
Follicular subtype	56 (48.7%)	34 (14.7%)	5 (4.2%)	
Tall cell subtype	0	14 (6.0%)	17 (14.4%)	
Others	2 (1.7%)	4 (1.7%)	1 (0.8%)	

LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; PKM2, pyruvate kinase isoform M2; MCT4, monocarboxylate transporter 4

연세대학교
YONSEI UNIVERSITY

3.12. Univariate and multivariate analysis of risk factors for lymph node metastasis using TCGA thyroid cancer data

Univariate analysis of TCGA data indicated that younger age (<55), male sex, multifocality, larger tumor size, ETE, advanced T stage, the $BRAF^{V600E}$ mutation, and elevated mRNA expression of LDHA, PKM2, and GLUT1 were significantly associated with LNM. In multivariate analysis, younger age, multifocality, advanced T stage (T3/T4), and higher PKM2 mRNA expression remained significant. Younger patients had more than twice the risk of LNM compared to older patients (OR 2.42, $P = 0.001$), and PKM2 emerged as the strongest predictor (OR 6.91, $P = 0.001$). Although LDHA and GLUT1 were significant in univariate analysis, they did not retain significance when adjusted for other factors, suggesting that their influence is not independent (Table 9).

Table 9. Univariate and multivariate analysis of lymph node metastasis in patient with PTC in The Cancer Genome Atlas (TCGA) thyroid cancer data

LNM	Univariate			multivariate		
variable	OR	95% CI	P value	OR	95% CI	P value
Age<55 (vs. >55)	1.55	1.04–2.31	0.031	2.42	1.48– 3.94	0.001
Male sex	2.37	1.27– 4.44	0.007	1.42	0.87–2.33	0.163
Multifocality	1.46	1.01–2.12	0.046	1.62	1.09–2.42	0.017
Tumor size (cm)	1.20	1.06–1.35	0.003	1.07	0.91–1.26	0.442
Extrathyroidal extension						
Negative	1			1		
Minimal	2.60	1.69–3.99	0.001	1.51	0.74–3.08	0.256
Gross	11.95	2.67–53.51	0.001	0.94	0.27–3.29	0.921
T stage(AJCC 7^{th})						
T1	1			1		
T2	1.6	0.98–2.61	0.06	1.64	0.96–2.82	0.07
T3	2.93	1.80–4.77	0.001	3.16	1.83–5.46	0.001
T4	12.27	3.38–44.46	0.001	30.44	6.07–152.74	0.001
$BRAF^{V600E}$ mutation	1.47	1.02–2.12	0.041	0.89	0.55–1.44	0.647
LDHA mRNA expression						
Low	1			1		
Moderate	2.30	1.42–3.72	0.001	1.51	0.82–2.78	0.184
High	4.03	2.32–7.00	0.001	1.82	0.85–3.87	0.121

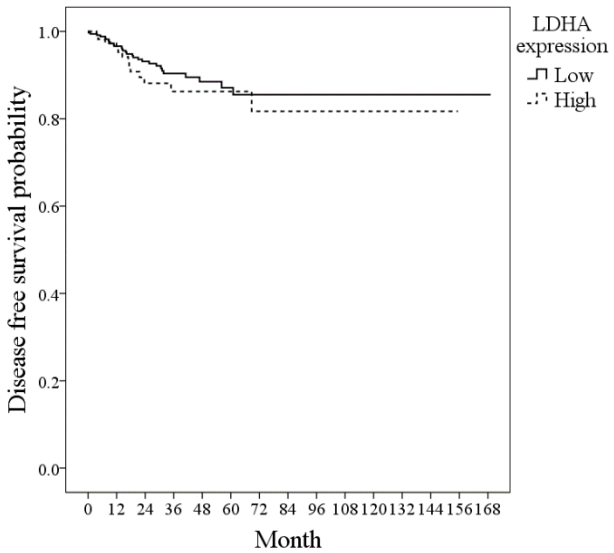
연세대학교
YONSEI UNIVERSITY

PKM2 mRNA expression						
Low	1			1		
Moderate	3.09	1.86–5.13	0.001	3.45	1.88–6.31	0.001
High	5.36	3.02–9.50	0.001	6.91	3.37–14.17	0.001
GLUT1 mRNA expression						
Low	1			1		
Moderate	2.22	1.40–3.54	0.001	1.24	0.70–2.19	0.466
High	2.17	1.27–3.69	0.004	0.69	0.35–1.37	0.292

LDHA, lactate dehydrogenase A; GLUT1, glucose transporter 1; PKM2, pyruvate kinase isoform M2; MCT4, monocarboxylate transporter 4; OR, odds ratio

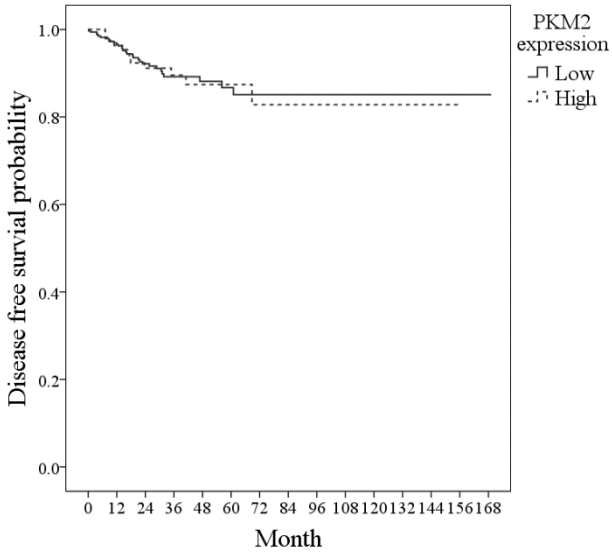
These data confirm that while LDHA and GLUT1 may have some impact, PKM2 remains the most critical independent predictor of LNM in PTC patients.

Figure 8 presents the disease-free survival curves of the two groups, demonstrating no significant differences in disease-free survival based on LDHA, PKM2, or GLUT1 expression. However, LNM (HR 1.86, P = 0.044) and ETE (HR 2.25, P = 0.008 for no vs. minimal ETE) were significantly associated with reduced disease-free survival probability in Kaplan–Meier analysis.

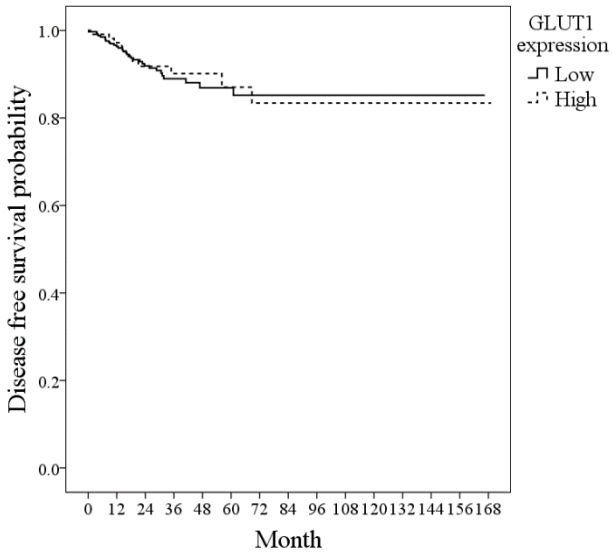


a) LDHA expression (P=0.377)

연세대학교
YONSEI UNIVERSITY

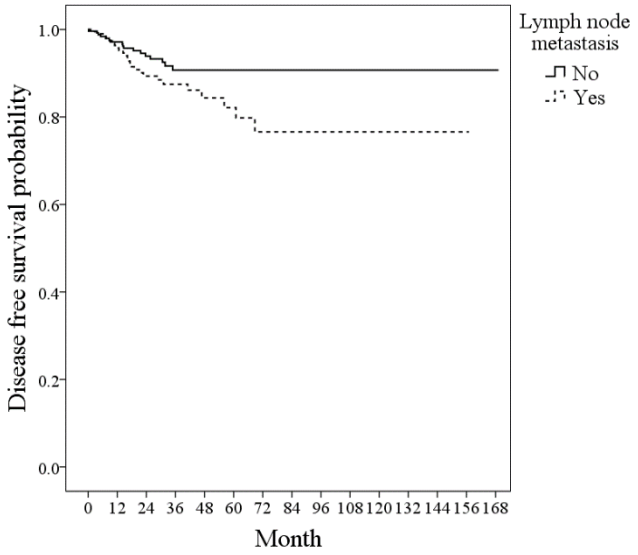


b) PKM2 expression (P=0.904)

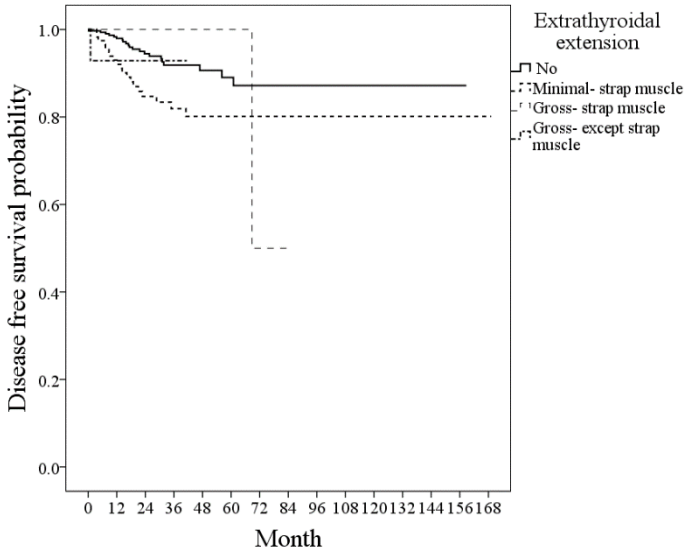


c) GLUT1 expression (P=0.872)

연세대학교
YONSEI UNIVERSITY



d) Lymph node metastasis (HR 1.863, P=0.044)



e) Extrathyroidal extension between No and minimal ETE (HR 2.25, P=0.008)

Fig. 8. Disease free survival rate from TCGA database. a) LDHA, b) PKM2, c) GLUT1, d) Lymph node metastasis, e) ETE

연세대학교
YONSEI UNIVERSITY

4. Discussion

Extensive research has demonstrated the roles of glycolytic enzymes in various cancers, linking their overexpression to more aggressive behavior and poor outcomes [18,25,27,32-36]. PKM2 and LDHA, for example, are frequently elevated in breast, lung, and colorectal cancers, and their inhibition is being investigated as a therapeutic strategy to curb tumor growth and metastasis [32,33,37,38]. In this study, we aimed to clarify whether the expression of glycolytic enzymes correlates with tumor aggressiveness—namely LNM and ETE—in the presence or absence of CLT.

Compared with normal cells, cancer cells exhibit increased glucose uptake and lactate production, even under aerobic conditions (the Warburg effect), thereby fueling proliferation and progression [11,25,39,40]. Here, we found marked overexpression of glycolytic enzymes and transporters in thyroid cancer tissues, reflecting a metabolic shift favoring enhanced glycolysis.

4.1. Metabolic reprogramming in thyroid cancer: increased glycolysis and glutaminolysis

In thyroid cancer, the heightened glycolytic flux in tumor cells is not matched by a corresponding increase in pyruvate oxidation. Instead, pyruvate is predominantly converted to lactate by LDHA. Lactate and pyruvate are then transported either into mitochondria or out of the cell via MCTs. The secreted lactate is often taken up by adjacent cancer cells, creating a feedforward loop that sustains tumor growth. Under hypoxic conditions, glycolysis enables cancer cells to survive in poorly vascularized areas [24,26,41-43]. This adaptation, however, leads to excessive lactate production and reduced extracellular pH, fostering an invasive microenvironment conducive to tumor cell migration.

Beyond glycolysis, many cancer cells rely heavily on glutamine metabolism to support the TCA cycle, thereby maintaining the biosynthesis of fatty acids and amino acids [24,40]. The interplay between glucose and glutamine metabolism allows tumor cells to thrive under metabolically challenging conditions. PKM2, a key enzyme in glycolysis, orchestrates this process by regulating the conversion of PEP to ATP and pyruvate. Numerous studies have linked PKM2 to increased tumor aggressiveness [18,36,44-47].

4.2. Role of CLT and the peritumoral microenvironment in PTC

Prior studies show that elevated glycolysis in CD4+ T cells in CLT is driven by enzymes such as GLUT1, HK2, PKM2, and LDHA. The immune response in CLT, particularly through thyroglobulin-specific CD8+ T cells, promotes cytokine release (e.g., IL-6, IL-1β, IL-8), attracting more lymphocytes to the thyroid gland [31,48-50]. Although the prognostic implications of CLT in PTC have been debated, it is often associated with both favorable and unfavorable outcomes [51-56].

A previous study found that the expression of glycolytic enzymes (GLUT1, HK2, PKM2, LDHA) was significantly elevated in thyroid inflammation compared to normal thyroid tissue. This increase correlated with higher extracellular acidification rates and oxygen consumption rates, suggesting that thyroid inflammation may contribute to shaping the peritumoral microenvironment [31].

연세대학교
YONSEI UNIVERSITY

In this study, PTC with CLT was significantly linked to higher rates of gross ETE, suggesting a more invasive local tumor behavior. However, CLT did not emerge as a risk factor for LNM, and patients with PTC and CLT surprisingly exhibited a lower recurrence rate [51,52,54,56]. Thus, while CLT may enhance local tumor invasion, it appears to limit metastatic potential. Inflammatory changes in the thyroid, driven by CLT, may alter cytokine profiles and metabolic pathways, promoting local invasion while simultaneously restraining distant dissemination.

Our data also indicate that glycolytic enzymes such as LDHA and PKM2 are more strongly expressed in the peritumoral normal thyroid tissue of patients with CLT than in those without thyroiditis. This finding suggests that the inflammatory milieu in CLT may drive metabolic reprogramming. While lactate-enriched environments facilitate local invasion, the chronic inflammatory state may also activate immune responses that constrain metastasis. This intricate interplay likely underlies the paradoxical prognostic effects of CLT, which promotes local invasiveness but reduces recurrence.

The fibrotic and inflammatory changes induced by CLT may also act as physical barriers, hindering the dissemination of tumor cells and thereby lowering recurrence rates [53,57]. Thus, the net effect of CLT on PTC outcomes may depend on a balance between metabolic facilitation of local invasion and immune-mediated constraints on distant spread.

4.3. PKM2 and other glycolytic enzymes in PTC prognosis

The $BRAF^{V600E}$ mutation is highly prevalent, but its utility as a prognostic marker for tumor aggressiveness—including gross ETE, LNM, and recurrence—remains limited. A significant upregulation of glycolytic enzyme mRNA expression was observed in tumors harboring $BRAF^{V600E}$ mutation. However, in multivariate analysis, only PKM2—not $BRAF^{V600E}$ mutation, LDHA, or GLUT1—emerged as an independent risk factor for LNM. These findings suggest that the overexpression of glycolytic enzymes, particularly PKM2, is more strongly associated with tumor aggressiveness than the presence of a $BRAF^{V600E}$ mutation. In line with this, we found that PKM2 overexpression was significantly associated with recurrence in PTC, a finding further supported by TCGA data. This dataset also demonstrated a specific correlation between PKM2 overexpression and LNM, reinforcing its role as a key driver of tumor progression.

Conversely, the overexpression of LDHA and GLUT1 was not significantly linked to LNM or recurrence. Regardless of the presence of thyroiditis, most glycolytic enzymes showed no significant difference in distribution. However, LDHA levels were notably lower in PTC with thyroiditis, and its IRS remained consistently low in recurrent cases when thyroiditis was present. PKM2 expression levels did not differ significantly based on CLT status; however, high PKM2 expression was a significant predictor of recurrence only in patients without CLT. These findings suggest that PKM2 may be a more robust indicator of tumor aggressiveness and recurrence than LDHA in PTC. Although LDHA is a critical component of glycolysis, PKM2 appears to have the strongest association with recurrence in PTC[26].

While thyroid cancer generally carries a favorable prognosis, PKM2 overexpression emerged as a key predictor of recurrence, underscoring the potential importance of incorporating glycolytic

연세대학교
YONSEI UNIVERSITY

enzyme profiles into long-term risk stratification.

Although several studies have investigated glycolytic enzyme expression and its link to thyroid cancer malignancy, few have evaluated its relationship with thyroid inflammation. In this study, we confirmed that glycolytic enzyme expression was elevated in malignant tissue, while LDHA expression was notably reduced in the presence of CLT. In contrast, PKM2 expression remained consistently associated with recurrence in patient without CLT. Interestingly, in cases with CLT, the prognosis tended to be better—even when PKM2 expression levels were comparable.

Unlike studies based on animal models or cell lines, this research utilized histological tissue samples from actual patients. To enhance the clinical relevance of the findings, protein-level analysis via immunohistochemistry was conducted, allowing for a more comprehensive evaluation of the influence of thyroiditis on tumor aggressiveness.

4.4. Limitations

This study had several limitations. First, although TCGA data reflect mRNA expression levels and our study evaluates protein expression via IHC, we did not directly correlate the two. As protein abundance can be influenced by post-transcriptional and post-translational modifications, it may not consistently mirror mRNA expression. This inherent difference between mRNA and protein expression should be acknowledged as a limitation.

Second, we did not establish standardized cutoff values for PKM2, LDHA, and GLUT1 expression, which may limit the interpretability and clinical applicability of our findings. Additionally, TCGA data did not provide information on CLT, precluding analysis of its influence on LNM and ETE in that dataset. Further investigation is warranted to elucidate the role of CLT in shaping the tumor microenvironment and influencing PTC outcomes. Despite these limitations, PKM2 emerged as a promising prognostic marker for PTC recurrence, supporting its potential utility in future risk stratification strategies.

연세대학교
YONSEI UNIVERSITY

5. Conclusion

Overexpression of glycolytic enzymes such as LDHA, PKM2, and GLUT1 is associated with PTC. Interestingly, CLT is associated with greater local invasiveness (gross ETE) yet paradoxically lower recurrence. LDHA expression was lower in the presence of CLT, whereas PKM2 remained consistently associated with a higher recurrence rate in the absence of CLT. Based on TCGA data, PKM2 mRNA expression may serve as a promising biomarker for predicting LNM in PTC. Notably, patients with CLT exhibited better prognostic outcomes, even with elevated PKM2 expression, suggesting a potential protective role of CLT in modulating disease progression. Among the glycolytic enzymes examined, PKM2 may serve as a valuable biomarker for identifying patients at higher risk of recurrence.

연세대학교
YONSEI UNIVERSITY

REFERENCES

1. Liu S, Liu C, Zhao L, Wang K, Li S, Tian Y, et al. A prediction model incorporating the BRAF(V600E) protein status for determining the risk of cervical lateral lymph node metastasis in papillary thyroid cancer patients with central lymph node metastasis. Eur J Surg Oncol 2021;47:2774-80.
2. Xing M, Alzahrani AS, Carson KA, Viola D, Elisei R, Bendlova B, et al. Association between BRAF V600E mutation and mortality in patients with papillary thyroid cancer. JAMA 2013;309:1493-501.
3. Chai YJ, Yi JW, Jee HG, Kim YA, Kim JH, Xing M, et al. Significance of the BRAF mRNA Expression Level in Papillary Thyroid Carcinoma: An Analysis of The Cancer Genome Atlas Data. PLoS One 2016;11:e0159235.
4. Gao Y, Yang F, Yang XA, Zhang L, Yu H, Cheng X, et al. Mitochondrial metabolism is inhibited by the HIF1alpha-MYC-PGC-1beta axis in BRAF V600E thyroid cancer. FEBS J 2019;286:1420-36.
5. Lee SE, Hwang TS, Choi YL, Han HS, Kim WS, Jang MH, et al. Prognostic Significance of TERT Promoter Mutations in Papillary Thyroid Carcinomas in a BRAF(V600E) Mutation-Prevalent Population. Thyroid 2016;26:901-10.
6. Yin DT, Yu K, Lu RQ, Li X, Xu J, Lei M, et al. Clinicopathological significance of TERT promoter mutation in papillary thyroid carcinomas: a systematic review and meta-analysis. Clin Endocrinol (Oxf) 2016;85:299-305.
7. Yang H, Park H, Ryu HJ, Heo J, Kim JS, Oh YL, et al. Frequency of TERT Promoter Mutations in Real-World Analysis of 2,092 Thyroid Carcinoma Patients. Endocrinol Metab (Seoul) 2022;37:652-63.
8. Sigarteu Chindris AM, Rivera M, Ma Y, Nair A, Liu Y, Wang X, et al. BRAFV600E/pTERT double mutated papillary thyroid cancers exhibit immune gene suppression. Front Endocrinol (Lausanne) 2024;15:1440722.
9. Hajjar R, Duta C, Faur IF, Prodan-Barbulescu C, Alhajjar H, Pantea S. The Correlation Between Biological Markers and Prognosis in Thyroid Cancer. Biomedicines 2024;12.
10. Brumfield A, Azar SA, Nordgren R, Cohen RN, Sarne D, Keutgen XM, et al. Prevalence and Clinical Impact of BRAF p.V600E Mutation in Papillary Thyroid Carcinoma. Endocr Pathol 2025;36:13.
11. Singer K, Kastenberger M, Gottfried E, Hammerschmied CG, Buttner M, Aigner M, et al. Warburg phenotype in renal cell carcinoma: high expression of glucose-transporter 1 (GLUT-1) correlates with low CD8(+) T-cell infiltration in the tumor. Int J Cancer 2011;128:2085-95.
12. Li Q, Wang Q, Zhang Q, Zhang J, Zhang J. Collagen prolyl 4-hydroxylase 2 predicts worse prognosis and promotes glycolysis in cervical cancer. Am J Transl Res 2019;11:6938-51.
13. Li R, Li P, Wang J, Liu J. STIP1 down-regulation inhibits glycolysis by suppressing PKM2 and LDHA and inactivating the Wnt/beta-catenin pathway in cervical carcinoma cells. Life Sci 2020;258:118190.
14. Guan H, Luo W, Liu Y, Li M. Novel circular RNA circSLIT2 facilitates the aerobic glycolysis of pancreatic ductal adenocarcinoma via miR-510-5p/c-Myc/LDHA axis. Cell Death Dis 2021;12:645.
15. Huo N, Cong R, Sun ZJ, Li WC, Zhu X, Xue CY, et al. STAT3/LINC00671 axis regulates papillary thyroid tumor growth and metastasis via LDHA-mediated glycolysis. Cell Death Dis 2021;12:799.

연세대학교
YONSEI UNIVERSITY

16. Xing Z, Wang R, Wang X, Liu J, Zhang M, Feng K, et al. CircRNA circ-PDCD11 promotes triple-negative breast cancer progression via enhancing aerobic glycolysis. Cell Death Discov 2021;7:218.
17. Li X, Deng S, Liu M, Jin Y, Zhu S, Deng S, et al. The responsively decreased PKM2 facilitates the survival of pancreatic cancer cells in hypoglucose. Cell Death Dis 2018;9:133.
18. Feng J, Wu L, Ji J, Chen K, Yu Q, Zhang J, et al. PKM2 is the target of proanthocyanidin B2 during the inhibition of hepatocellular carcinoma. J Exp Clin Cancer Res 2019;38:204.
19. Kobierzycki C, Piotrowska A, Latkowski K, Zabel M, Nowak-Markwitz E, Spaczynski M, et al. Correlation of Pyruvate Kinase M2 Expression with Clinicopathological Data in Ovarian Cancer. Anticancer Res 2018;38:295-300.
20. Luo M, Luo Y, Mao N, Huang G, Teng C, Wang H, et al. Cancer-Associated Fibroblasts Accelerate Malignant Progression of Non-Small Cell Lung Cancer via Connexin 43-Formed Unidirectional Gap Junctional Intercellular Communication. Cell Physiol Biochem 2018;51:315-36.
21. Alquraishi M, Puckett DL, Alani DS, Humidat AS, Frankel VD, Donohoe DR, et al. Pyruvate kinase M2: A simple molecule with complex functions. Free Radic Biol Med 2019;143:176-92.
22. Dey P, Son JY, Kundu A, Kim KS, Lee Y, Yoon K, et al. Knockdown of Pyruvate Kinase M2 Inhibits Cell Proliferation, Metabolism, and Migration in Renal Cell Carcinoma. Int J Mol Sci 2019;20.
23. Gu J, Li X, Zhao L, Yang Y, Xue C, Gao Y, et al. The role of PKM2 nuclear translocation in the constant activation of the NF-kappaB signaling pathway in cancer-associated fibroblasts. Cell Death Dis 2021;12:291.
24. Coelho RG, Fortunato RS, Carvalho DP. Metabolic Reprogramming in Thyroid Carcinoma. Front Oncol 2018;8:82.
25. Pathria G, Scott DA, Feng Y, Sang Lee J, Fujita Y, Zhang G, et al. Targeting the Warburg effect via LDHA inhibition engages ATF4 signaling for cancer cell survival. EMBO J 2018;37.
26. Ban EJ, Kim D, Kim JK, Kang SW, Lee J, Jeong JJ, et al. Lactate Dehydrogenase A as a Potential New Biomarker for Thyroid Cancer. Endocrinol Metab (Seoul) 2021;36:96-105.
27. Shi L, Duan R, Sun Z, Jia Q, Wu W, Wang F, et al. LncRNA GLTC targets LDHA for succinylation and enzymatic activity to promote progression and radioiodine resistance in papillary thyroid cancer. Cell Death Differ 2023;30:1517-32.
28. Nahm JH, Kim HM, Koo JS. Glycolysis-related protein expression in thyroid cancer. Tumour Biol 2017;39:1010428317695922.
29. Zhao B, Aggarwal A, Im SY, Viswanathan K, Landa I, Nehs MA. Effect of Lactate Export Inhibition on Anaplastic Thyroid Cancer Growth and Metabolism. J Am Coll Surg 2022;234:1044-50.
30. Xu D, Yu J, Yang Y, Du Y, Lu H, Zhang S, et al. RBX1 regulates PKM alternative splicing to facilitate anaplastic thyroid carcinoma metastasis and aerobic glycolysis by destroying the SMAR1/HDAC6 complex. Cell Biosci 2023;13:36.
31. Zhao L, Wu Q, Wang X, Wang S, Shi X, Shan Z, et al. Reversal of Abnormal CD4+ T Cell Metabolism Alleviates Thyroiditis by Deactivating the mTOR/HIF1a/Glycolysis Pathway. Front Endocrinol (Lausanne) 2021;12:659738.
32. van Niekerk G, Engelbrecht AM. Role of PKM2 in directing the metabolic fate of glucose in cancer: a potential therapeutic target. Cell Oncol (Dordr) 2018;41:343-51.

연세대학교
YONSEI UNIVERSITY

33. Duan SL, Wu M, Zhang ZJ, Chang S. The potential role of reprogrammed glucose metabolism: an emerging actionable codependent target in thyroid cancer. J Transl Med 2023;21:735.
34. Ji X, Lv C, Huang J, Dong W, Sun W, Zhang H. ALKBH5-induced circular RNA NRIP1 promotes glycolysis in thyroid cancer cells by targeting PKM2. Cancer Sci 2023;114:2318-34.
35. Liang N, Mi L, Li J, Li T, Chen J, Dionigi G, et al. Pan-Cancer Analysis of the Oncogenic and Prognostic Role of PKM2: A Potential Target for Survival and Immunotherapy. Biomed Res Int 2023;2023:3375109.
36. Adem S, Rasul A, Riaz S, Sadiqa A, Ahmad M, Shahid Nazir M, et al. Pyruvate kinase modulators as a therapy target: an updated patent review 2018-2023. Expert Opin Ther Pat 2024;34:953-62.
37. Xintaropoulou C, Ward C, Wise A, Queckborner S, Turnbull A, Michie CO, et al. Expression of glycolytic enzymes in ovarian cancers and evaluation of the glycolytic pathway as a strategy for ovarian cancer treatment. BMC Cancer 2018;18:636.
38. Shi X, You L, Luo RY. Glycolytic reprogramming in cancer cells: PKM2 dimer predominance induced by pulsatile PFK-1 activity. Phys Biol 2019;16:066007.
39. Tahtouh R, Wardi L, Sarkis R, Hachem R, Raad I, El Zein N, et al. Glucose restriction reverses the Warburg effect and modulates PKM2 and mTOR expression in breast cancer cell lines. Cell Mol Biol (Noisy-le-grand) 2019;65:26-33.
40. Upadhyay S, Khan S, Hassan MI. Exploring the diverse role of pyruvate kinase M2 in cancer: Navigating beyond glycolysis and the Warburg effect. Biochim Biophys Acta Rev Cancer 2024;1879:189089.
41. Kachel P, Trojanowicz B, Sekulla C, Prenzel H, Dralle H, Hoang-Vu C. Phosphorylation of pyruvate kinase M2 and lactate dehydrogenase A by fibroblast growth factor receptor 1 in benign and malignant thyroid tissue. BMC Cancer 2015;15:140.
42. Hou X, Shi X, Zhang W, Li D, Hu L, Yang J, et al. LDHA induces EMT gene transcription and regulates autophagy to promote the metastasis and tumorigenesis of papillary thyroid carcinoma. Cell Death Dis 2021;12:347.
43. Tian T, Dai H, Zhang M, Su M, Chen X, Huang R. Lactate Dehydrogenase A is Associated with Elevated FDG Metabolism, Radioiodine Non-avidity, and Poor Prognosis in Differentiated Thyroid Cancer. Acad Radiol 2024 doi: 10.1016/j.acra.2024.04.033. PubMed PMID: 38866688.
44. Amin S, Yang P, Li Z. Pyruvate kinase M2: A multifarious enzyme in non-canonical localization to promote cancer progression. Biochim Biophys Acta Rev Cancer 2019;1871:331-41.
45. Gómez-Escudero J, Clemente C, García-Weber D, Acín-Pérez R, Millán J, Enríquez JA, et al. PKM2 regulates endothelial cell junction dynamics and angiogenesis via ATP production. Sci Rep 2019;9:15022.
46. Li TE, Wang S, Shen XT, Zhang Z, Chen M, Wang H, et al. PKM2 Drives Hepatocellular Carcinoma Progression by Inducing Immunosuppressive Microenvironment. Front Immunol 2020;11:589997.
47. Zhu S, Guo Y, Zhang X, Liu H, Yin M, Chen X, et al. Pyruvate kinase M2 (PKM2) in cancer and cancer therapeutics. Cancer Lett 2021;503:240-8.
48. Curry JM, Tassone P, Cotzia P, Sprandio J, Luginbuhl A, Cognetti DM, et al. Multicompartment metabolism in papillary thyroid cancer. Laryngoscope 2016;126:2410-8.

연세대학교
YONSEI UNIVERSITY

49. Gill KS, Tassone P, Hamilton J, Hjelm N, Luginbuhl A, Cognetti D, et al. Thyroid Cancer Metabolism: A Review. J Thyroid Disord Ther 2016;5.
50. Van Wilpe S, Koornstra R, Den Brok M, De Groot JW, Blank C, De Vries J, et al. Lactate dehydrogenase: a marker of diminished antitumor immunity. Oncoimmunology 2020;9:1731942.
51. Kim EY, Kim WG, Kim WB, Kim TY, Kim JM, Ryu JS, et al. Coexistence of chronic lymphocytic thyroiditis is associated with lower recurrence rates in patients with papillary thyroid carcinoma. Clin Endocrinol (Oxf) 2009;71:581-6.
52. Lee I, Kim HK, Soh EY, Lee J. The Association Between Chronic Lymphocytic Thyroiditis and the Progress of Papillary Thyroid Cancer. World J Surg 2020;44:1506-13.
53. Sakiz D, Sencar ME, Calapkulu M, Ozturk Unsal I, Aktas L, Ucan B, et al. The Effects of Chronic Lymphocytic Thyroiditis on Clinicopathologic Factors in Papillary Thyroid Cancer. Endocr Pract 2021;27:1199-204.
54. Issa PP, Omar M, Buti Y, Issa CP, Chabot B, Carnabatu CJ, et al. Hashimoto's Thyroiditis Minimizes Lymph Node Metastasis in BRAF Mutant Papillary Thyroid Carcinomas. Biomedicines 2022;10.
55. Santana VB, Kruger VM, Abrahao MCY, Cantu PLM, Brackmann RL, Pandolfi GM, et al. Chronic Lymphocytic Thyroiditis with Oncocytic Metaplasia Influences PD-L1 Expression in Papillary Thyroid Carcinoma. Head Neck Pathol 2024;18:14.
56. Yu J, Li X, He Z, He J, Rao S. Chronic Lymphocytic Thyroiditis is a Protective Factor for Lymph Node Metastasis in Papillary Thyroid Carcinoma: A Propensity Score Matching Analysis. Br J Hosp Med (Lond) 2024;85:1-14.
57. Song E, Jeon MJ, Park S, Kim M, Oh HS, Song DE, et al. Influence of coexistent Hashimoto's thyroiditis on the extent of cervical lymph node dissection and prognosis in papillary thyroid carcinoma. Clin Endocrinol (Oxf) 2018;88:123-8.

연세대학교
YONSEI UNIVERSITY

Abstract in Korean

Glycolytic enzyme의 발현과 갑상선염이 갑상선암의 악성도에 미치는 영향

배경: Glycolytic enzyme는 다양한 암 유형에서 광범위하게 연구되어 왔으며, 공격적인 특성과 종양 진행에서의 역할이 밝혀졌다. 그러나 glycolytic enzyme 및 peritumoral enzyme의 활성도 간의 연관성이 있는지는 아직 명확하지 않다. 본 연구는 만성 림프구성 갑상선염의 존재 여부에 따라 glycolytic enzyme의 발현이 림프절 전이 및 갑상선 외 침윤과 같은 공격성과 관련이 있는지를 규명하는 것을 목표로 하였다.

방법: 233개의 갑상선 유두암 조직 샘플에서 헥소키나제 2(HK2), 젖산 탈수소효소 A(LDHA), 피루브산키나제 M2 동형(PKM2), 포도당 수송체 1(GLUT1), 단일카복실산 수송체 4(MCT4)의 발현을 면역조직화학법 (Immunohistochemistry)을 통해 분석하였다. 이러한 glycolytic enzyme의 발현이 림프절 전이, 갑상선 외 침윤, 재발률과 연관이 있는지, 그리고 만성 림프구성 갑상선염의 유무에 따라 차이가 있는지를 평가하였다. 연령, 성별, 종양 크기, 갑상선 외 침윤, 다병소성, 경부 림프절 전이를 기준으로 1:3 성향 점수 매칭(Propensity score matching) 후 무병 생존율을 비교하였다. 또한, The Cancer Genome Atlas (TCGA)를 이용하여 glycolytic enzyme(HK2, LDHA, PKM2)발현과 갑상선 유두암의 임상병리학적 특성 간의 상관관계를 분석하였다.

결과: 정상 조직과 비교했을 때, 모든 glycolytic enzyme 및 transporter protein이 갑상선 유두암에서 비정상적으로 과발현되었다. HK2를 제외한 모든 glycolytic enzyme 및 transporter protein간에는 선형 상관관계가 있었으며, PKM2가 다른 효소들과 가장 높은 상관관계를 보였다. PKM2 과발현은 재발과 유의한 관련이 있었으나(OR 2.049, P = 0.036), 림프절 전이 또는 육안적 갑상선 외 침윤을 증가시키지는 않았다. The Cancer Genome Atlas(TCGA) 데이터를 활용한 단변량 및 다변량 분석에서 PKM2의 mRNA 발현과 T 병기(T staging)가 림프절 전이와 유의한 상관관계를 보였다. 그러나 glycolytic enzyme 발현과 무병 생존율 사이에는 유의한 차이가 없었다.

연세대학교
YONSEI UNIVERSITY

결론: LDHA, PKM2, GLUT1, MCT4의 과발현은 갑상선 유두암과 관련이 있다. 만성 림프구성 갑상선염은 높은 육안적 갑상선 외 침윤과 유의하게 관련이 있지만, 역설적으로 갑상선암의 낮은 재발률과 연관이 있다. 갑상선염이 있는 경우 LDHA 발현이 낮았으며, PKM2는 갑상선염증 상태와 관계없이 일관되게 재발 증가와 연관이 있었다. 그러나 갑상선염이 있는 갑상선암환자에서는 PKM2 발현이 높더라도 예후가 더 양호한 것으로 나타나, 갑상선염이 질병 경과에 잠재적인 보호 효과를 가질 수 있다. 본 연구에서 분석된 glycolytic enzyme중에 PKM2는 갑상선 유두암의 재발을 예측하는 바이오마커로 활용될 가능성이 있다.

핵심되는 말 : 해당과정 효소, 와버그 효과, 갑상선암, 대사 재프로그래밍, 림프절전이, 갑상선염