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Original Article

Prognostic significance of phosphoinositide 3-kinase p110α and p110β isoforms in non-small cell lung cancer

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Abstract: The proteins p110α and p110β are isoforms of the catalytic subunit of class I phosphoinositide 3-kinases (PI3Ks). Class I PI3Ks are involved in the regulation of cell survival, growth, proliferation, and migration, and their aberrant activation contributes to the oncogenesis of various human cancers. In this study, we assessed expression of p110α and p110β in non-small cell lung cancer (NSCLC) and their association with clinicopathological factors and patient survival. Seventy-six NSCLC cases were analyzed by immunohistochemical staining for p110α and p110β. Of the 76 tumors, 18 (23.7%) and 43 (56.6%) were classified in the high p110α and p110β expression groups, respectively. Expression of p110α was higher in smokers compared with non-smokers ($P = 0.042$). No other clinicopathological factors showed significant association with p110α or p110β expression. In univariate and multivariate survival analyses, high p110β expression was associated with worse overall survival (OS) in stage I NSCLCs ($P < 0.001$), whereas the high p110α expression group had shorter OS in stage II to IV NSCLCs ($P = 0.005$). Our results suggest that p110α and p110β play different roles depending on tumor stage, and that both p110α and p110β have potential as independent prognostic biomarkers of NSCLC.

Keywords: Non-small cell lung cancer, PI3K, p110α, p110β, prognosis, immunohistochemistry

Introduction

Phosphoinositide 3-kinases (PI3Ks) are a family of lipid kinases that phosphorylate the 3'-hydroxyl position of the inositol ring of phosphatidylinositol-4,5-bisphosphate in response to extracellular stimuli and generate phosphatidylinositol-3,4,5-triphosphate. PI3Ks are grouped into three classes on the basis of their structural features and substrate specificity but only class I PI3Ks have been linked to the oncogenesis in humans [1]. Class I PI3Ks are heterodimeric enzymes composed of a catalytic subunit and a regulatory subunit. The catalytic subunits of class I PI3Ks are p110α, p110β, p110γ, and p110δ [1]. These four isoforms have non-redundant functions and different expression patterns in different cell types. While p110α and p110β are ubiquitously expressed, p110γ and p110δ are expressed largely in cells of hematopoietic lineage [1, 2].

Class I PI3Ks together with their downstream molecules, including AKT and mammalian target of rapamycin (mTOR), comprise the central axis of a complicated and interconnected signaling network that integrates extracellular signals from growth factors, insulin, nutrients, and oxygen to play a pivotal role in controlling cell growth, proliferation, metabolism, survival, and motility [3-5]. It has been well-established that aberrant and constitutive activation of class I PI3Ks is involved in the oncogenesis of various types of cancers, including non-small cell lung cancer (NSCLC) [1, 5-7], and in resistance to receptor tyrosine kinase inhibitors, such as trastuzumab or imatinib, as well as to traditional chemo- and radiotherapy [8-11]. Thus, class I PI3Ks have promising potential as therapeutic targets in a diverse array of human cancers.

Many kinds of pan-PI3K and isoform-specific inhibitors have been evaluated in preclinical

studies or are under evaluation in clinical trials [6, 12]. However, their clinical benefits have not yet been clearly proven in solid cancers. This may be due to the existence of various PI3K isoforms and their differential functions in cancer biology and compensatory feedback activation of oncogenic pathways [13, 14].

With regard to NSCLC, most studies on aberrant activation of the PI3K/AKT pathway have focused on amplification or mutation of *PIK3CA,* a gene encoding p110α, or loss of function of phosphatase and tensin homolog (PTEN), a negative regulator of AKT [5, 15-17]. Only a limited number of studies have examined expression of the PI3K protein, especially isoform p110α, and its correlation with clinicopathological factors and associated molecular alterations in NSCLC [16-18]. Moreover, there are no available data for expression of other PI3K isoforms in NSCLC, although it has been suggested that overexpression of wild-type PI3K isoforms could contribute to the malignant transformation and disease progression of other types of cancers [19-23].

In this study, we examined the differential expression of p110α and p110β in NSCLC and the correlation between their expression and clinicopathological factors, including patient survival. This study aimed to assess the potential of PI3K expression as a predictive biomarker for PI3K inhibitor therapy and as a prognostic biomarker of NSCLC.

Materials and methods

Patients and tissue samples

NSCLC tissue samples were obtained from 76 patients who underwent complete resection at the Samsung Changwon Hospital between January 2002 and December 2009. Demographic and clinicopathological data were collected from medical records and histopathological reports. The clinical stage was determined according to the 7th edition of the American Joint Committee on Cancer TNM staging system [24]. Follow-up data were included until December 2016 or until death or last follow-up with the patient. The study was approved by the institutional review board of our medical institution.

Tissue microarray and immunohistochemistry

Representative areas of the tumors were marked on hematoxylin- and eosin-stained slides and used for tissue microarray (TMA) construction. Tissue cores with diameters of 2 mm were taken from donor paraffin blocks and placed in blank recipient paraffin blocks. Two cores per tumor were arrayed. The TMA blocks were sectioned at 4 μm for immunohistochemical staining using a BenchMark XT automated staining platform (Roche-Ventana, Tucson, AZ, USA). All sections were deparaffinized and subjected to pretreatment with Cell Conditioning 1 solution (Roche-Ventana) for 30 min at 100°C. Sections were washed with reaction buffer followed by incubation with primary antibodies for 32 or 60 min at 37°C. Primary antibodies were against p110α (clone C73F8, 1:100, Cell Signaling Technology, Danvers, MA, USA) and p110β (clone EPR5515, 1:300, Epitomics, Burlingame, CA, USA). An UltraView Universal DAB kit (Roche-Ventana) was used according to the manufacturer's recommendations to detect the primary antibody, followed by counterstaining with hematoxylin (Roche-Ventana). Breast carcinoma was used as the positive control. The negative control was incubated with buffer instead of primary antibodies.

Immunostained slides were evaluated by an experienced pathologist (Lee, HW) blinded to the clinicopathological data. Cases were considered positive when 10% or more of the tumor cells expressed p110α or p110β. The staining intensity of the positive cases was scored as 1 (weak), 2 (moderate), or 3 (strong). For statistical analyses, the negative and weakly positive cases were clustered in the low expression group, while the moderately and strongly positive cases constituted the high expression group.

Statistical analysis

All statistical analyses were performed with SPSS Ver. 18 (SPSS Inc., Chicago, IL, USA). To evaluate possible relationships between immunohistochemical results and various clinicopathological parameters, we used Fisher's exact test for categorical variables and the Mann-Whitney test for ordinal variables. The impact of various parameters on overall survival (OS) was analyzed by the Kaplan-Meier method, and differences were compared using the log-rank test. Multivariate analysis for OS was performed with a Cox proportional hazards model. A P-value of < 0.05 was considered statistically significant.

Table 1. Correlation of p110α and p110β expression levels with clinicopathological factors in 76 patients with non-small cell lung cancer

Variables	p110α			p110β		
	Low (%)	High (%)	*P*	Low (%)	High (%)	*P*
Age (years)						
< 65	29 (74)	10 (26)	0.790	16 (41)	23 (59)	0.817
≥ 65	29 (78)	8 (22)		17 (46)	20 (54)	
Sex						
Male	45 (73)	17 (27)	0.166	27 (44)	35 (56)	1.000
Female	13 (93)	1 (7)		6 (43)	8 (57)	
Smoking						
Nonsmokers	22 (92)	2 (8)	0.042	11 (46)	13 (54)	0.807
Smokers	36 (69)	16 (31)		22 (42)	30 (58)	
Histological type						
AC	22 (76)	7 (24)	1.000	13 (45)	16 (55)	0.844
SCC	30 (77)	9 (23)		16 (41)	23 (59)	
Others	6 (75)	2 (25)		4 (50)	4 (50)	
Differentiation						
Well	22 (82)	5 (18)	0.522	10 (37)	17 (63)	0.660
Moderately	25 (74)	9 (26)		17 (50)	17 (50)	
Poorly	11 (73)	4 (27)		6 (40)	9 (60)	
Tumor size (cm)						
≤ 3	20 (71)	8 (29)	0.577	12 (43)	16 (57)	1.000
> 3	38 (79)	10 (21)		21 (44)	27 (56)	
Pleural invasion						
Negative	47 (78)	13 (22)	0.510	24 (40)	36 (60)	0.270
Positive	11 (69)	5 (31)		9 (56)	7 (44)	
Lymphovascular invasion						
Negative	44 (80)	11 (20)	0.240	21 (38)	34 (62)	0.196
Positive	14 (67)	7 (33)		12 (57)	9 (43)	
Lymph node metastasis						
Negative	31 (74)	11 (26)	0.600	18 (43)	24 (57)	1.000
Positive	27 (79)	7 (21)		15 (44)	19 (56)	
Distant metastasis						
Negative	52 (77)	16 (23)	1.000	28 (41)	40 (59)	0.283
Positive	6 (75)	2 (25)		5 (63)	3 (37)	
TNM stage						
I	19 (76)	6 (24)	0.975	11 (44)	14 (56)	0.549
II	10 (83)	2 (17)		4 (33)	8 (67)	
III	19 (70)	8 (30)		11 (41)	16 (59)	
VI	10 (83)	2 (17)		7 (58)	5 (42)	
p110α						
Low				24 (41)	34 (50)	0.591
High				9 (59)	9 (50)	
p110β						
Low	24 (73)	9 (27)	0.591			
High	34 (79)	9 (21)				
Total	58 (76)	18 (24)		33 (43)	43 (57)	

AC, adenocarcinoma; SCC, squamous cell carcinoma.

Results

Clinicopathological characteristics

Of the 76 patients with NSCLC, 62 were male and 14 female. At the time of diagnosis, the median age of these patients was 64 years (range 26-77 years). Fifty-two patients (68.4%) were current or former smokers, while 24 (31.6%) were non-smokers. Histologically, the tumors consisted of 29 adenocarcinomas (AC) (38.2%), 39 squamous cell carcinomas (SCC) (51.3%), 4 sarcomatoid carcinomas (5.3%), 2 adenosquamous cell carcinomas (2.6%), a mucoepidermoid carcinoma (2.6%), and an unclassified NSCLC. Twenty-seven tumors (35.5%) were well differentiated, 34 (44.7%) moderately differentiated, and 15 (19.7%) poorly differentiated. Median tumor size was 3.5 cm (range 1.3-10.5 cm). Twenty-four tumors (31.6%) were stage I, 12 (15.8%) stage II, 27 (35.5%) stage III, and 12 (15.8%) stage IV. Pleural invasion, lymphovascular invasion, and nodal metastasis were detected in 16 (21.1%), 21 (27.6%), and 34 cases (44.7%), respectively. Eight patients (10.5%) had distant metastasis at the time of diagnosis. These clinicopathological characteristics are summarized in **Table 1**.

Correlation of p110α and p110β expression with clinicopathological factors

Isoform p110α was expressed in the cytoplasm

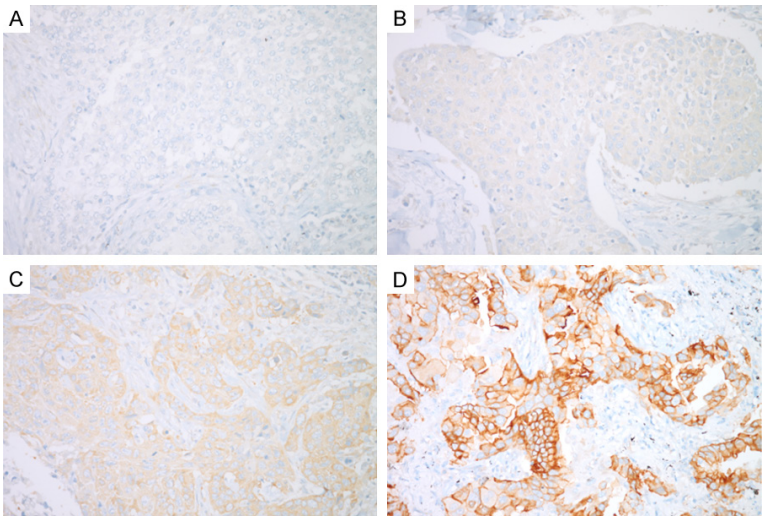

Figure 1. Immunohistochemical staining for p110α in non-small cell lung cancer: negative expression of p110α (A), weakly positive expression (B), moderately positive expression (C), and strongly positive expression (D).

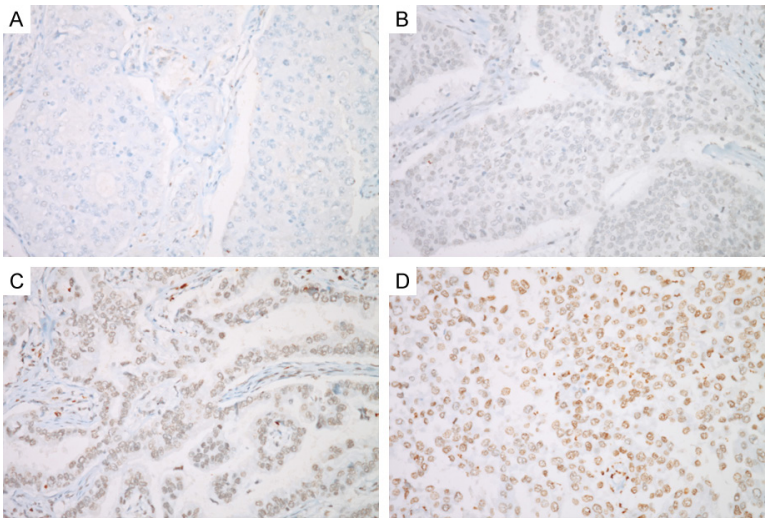

Figure 2. Immunohistochemical staining for p110β in non-small cell lung cancer: negative expression of p110β (A), weakly positive expression (B), moderately positive expression (C), and strongly positive expression (D).

(**Figure 1**), whereas p110β was expressed in the nucleus (**Figure 2**). Of the tumors, 36 (47.3%) and 65 (85.5%) were positive for p110α and p110β, respectively. Expression of p110α was weak in 18, moderate in 13, and strong in 5 tumors (**Figure 1**). Based on p110α expression, 58 tumors (76.3%) were classified in the low expression group, and 18 (23.7%) were placed in the high expression group. Expression of p110β was weak in 22, moderate in 25, and strong in 18 tumors (**Figure 2**). Among these, 33 tumors (43.4%) were classified in the low expression group and 43 (56.6%) in the high expression group.

Expression of p110α was significantly higher in smokers compared than in non-smokers (P = 0.042). However, no other clinicopathological factors showed significant associations with p110α or p110β expression (**Table 1**).

Correlation of p110α and p110β expression with patient survival

The median follow-up period was 27.5 months (range 1-176 months). During follow-up, 60 (78.9%) of the 76 patients died. In a univariate survival analysis for all patients, non-AC and non-SCC histological types (P < 0.001), higher grade (P = 0.021), lymphovascular invasion (P = 0.005), and higher TNM stage (P = 0.008) were significantly correlated with shorter OS. Expression of p110α and p110β had no statistically significant influence on OS, although there was a tendency towards decreased OS in the high p110α (**Figure 3A**; P = 0.173) and high p110β expression groups (**Figure 3B**; P = 0.179). However, when stratified by TNM stage, high p110β expression was significantly associated with worse OS in stage I NSCLCs (**Figure 3D**; P < 0.001), while p110α expression was not (**Figure 3C**; P = 0.494). Regarding stage II to IV NSCLCs, the high p110α expression group had significantly worse OS (**Figure 3E**; P = 0.005), whereas the high p110β expression group tended to have better OS, although it was not statistically significant (**Figure 3F**; P = 0.093).

In the whole cohort, multivariate Cox regression analysis including p110α, p110β, and clinicopathological variables that were significantly correlated with OS in univariate analysis

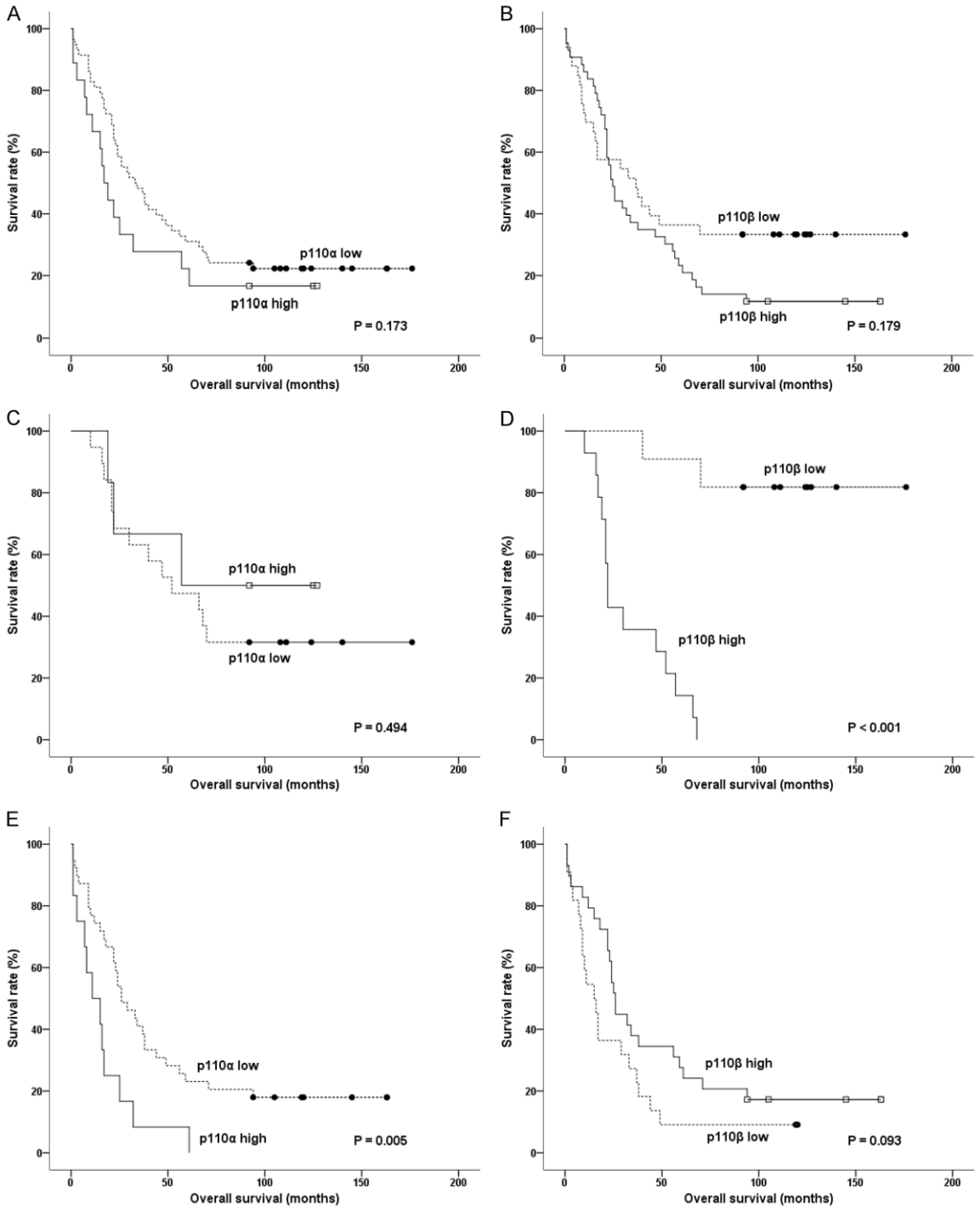

Figure 3. Survival curves using the Kaplan-Meier method: overall survival (OS) according to expression of p110α (A) and p110β (B) in all patients, OS according to expression of p110α (C) and p110β (D) in patients with stage I non-small cell lung cancers (NSCLCs), and OS according to expression of p110α (E) and p110β (F) in patients with stage II to IV NSCLCs.

showed that TNM stage and histological type were independent prognostic factors for OS, but p110α and p110β were not (**Table 2**). When additional multivariate analyses were performed after stratification according to TNM stage, p110α and histological type were independent prognostic factors for OS in patients with stage II to IV NSCLC, whereas p110β was the only independent prognostic factor for OS in those with stage I NSCLC (**Table 3**).

Table 2. Multivariate analysis of overall survival in all 76 patients with non-small cell lung cancer

Factors	HR	95% CI of HR	*P*
p110α			
High	1.68	0.89-1.35	0.107
Low	1.00		
p110β			
High	1.18	0.68-2.04	0.544
Low	1.00		
Histological type			
Others	5.92	2.42-14.45	< 0.001
AC and SCC	1.00		
Differentiation			
Poorly	1.82	0.92-3.59	0.084
Well to Moderately	1.00		
Lymphovascular invasion			
Positive	1.82	0.93-3.58	0.081
Negative	1.00		
TNM stage			
II-IV	2.07	1.01-4.25	0.047
I	1.00		

AC, adenocarcinoma; CI, confidence interval; HR, hazard ratio; SCC, squamous cell carcinoma.

Discussion

The PI3K/AKT pathway performs critical functions in the regulation of cell growth, proliferation, metabolism, survival and motility [3-5], and it is known that aberrant activation of this intracellular signaling pathway contributes to tumorigenesis of various types of human cancers [1, 5-7]. In NSCLC, oncogenic activation of the PI3K/AKT pathway has been also widely investigated, but the subjects of most previous studies have been restricted to mutation or amplification of *PIK3CA* and genetic or epigenetic inactivation of PTEN [5, 15-17]. There are only a limited number of published studies on the relationship between p110α protein expression and clinicopathological factors and associated genetic alterations [16-18], and the results were not notable. In addition, no information on p110β protein expression in NSCLC is available. In the present study, we found that expression of p110α and p110β isoforms had a significant association with clinicopathological factors and survival in patients with NSCLC regardless of *PIK3CA* mutation or amplification status.

In NSCLC, it has been suggested that the incidence of *PIK3CA* mutation or amplification is significantly higher in males, smokers, and SCC patients [15]. However, any association of p110α expression with gender, smoking history, or tumor histology has not yet been reported. In our study, p110α exhibited significantly higher expression in smokers than in non-smokers, although there was no correlation of p110α expression with gender or tumor histology. Our results are consistent with previous molecular studies on *PIK3CA* mutation or amplification in NSCLC. Taken together, *PIK3CA* genetic alteration or p110α overexpression seem to be more frequently involved in the carcinogenesis of smoking-associated NSCLC.

In our survival analysis, we found more unique and interesting effects of p110α and p110β expression on OS in patients with NSCLC. On the cohort as a whole, high p110α and p110β expression groups tended to have lower OS, but the differences were not statistically significant. When we assessed the prognostic value of p110α and p110β expression levels according to TNM stage, the levels had significant and strong associations with OS in different stage groups. In early disease (stage I), patients with high p110β expression had significantly shorter OS than those with low p110β expression, while p110α expression showed no association with OS. In advanced disease (stage II to IV), high p110α expression was significantly correlated with worse OS, whereas high p110β expression had a trend toward better OS. We further investigated the relationship between p110α and p110β expression levels and clinicopathological factors in the different stage groups. In stage I NSCLCs, tumors with high p110β expression had a larger size than those with low p110β expression (5.01 ± 2.62 cm vs. 2.62 ± 0.99 cm; $P = 0.006$), whereas in stage II to IV NSCLCs, high p110α expression was significantly associated with larger tumor size (4.71 ± 2.21 cm vs. 3.48 ± 1.61 cm; $P = 0.045$). Based on these results, the roles of p110α and p110β in the development and progression of NSCLC appear to change as disease stage progresses. Therefore, we can conclude that p110β might play a more predominant role in the growth and aggressiveness of early stage NSCLCs, but p110α mi-ght take on this role in advanced stages.

Table 3. Multivariate analysis of overall survival after stratification by TNM stage

Factors	Stage I NSCLC			Stage II-IV NSCLC		
	HR	95% CI of HR	*P*	HR	95% CI of HR	*P*
p110α						
High	1.01	0.25-4.01	0.991	2.26	1.06-4.83	0.035
Low	1.00			1.00		
p110β						
High	3.84	3.66-5.98	0.002	0.70	0.36-1.35	0.284
Low	1.00			1.00		
Histological type						
Others	1.66	0.40-6.87	0.481	6.52	1.51-28.24	0.012
AC and SCC	1.00			1.00		
Differentiation						
Poorly	1.31	0.31-5.59	0.712	1.28	0.50-3.32	0.606
Well to Moderately	1.00			1.00		
Lymphovascular invasion						
Positive				1.40	0.68-2.84	0.360
Negative				1.00		

AC, adenocarcinoma; CI, confidence interval; HR, hazard ratio; NSCLC, non-small cell lung cancer; SCC, squamous cell carcinoma.

Diverse pan-PI3K and isoform-specific inhibitors have been developed, and their efficacy and safety have been extensively tested in preclinical studies or clinical trials [6, 12]. However, the expected clinical benefits of such inhibitors have not yet been sufficiently proven. This unsatisfactory progress might be due to the existence of various PI3K isoforms and their differential functions in cancer biology [13]. This assumption is su-pported by our results that p110α and p110β are differentially expressed in each tumor and have different functions depending on disease stage. Therefore, to determine more effective PI3K inhibitors against individual NSCLCs, the differential expression of PI3K isoforms together with the disease stage should be considered. Thus, the influence of isoform selectivity under specific conditions, such as tumor stage, on the efficacy and safety of PI3K inhibitors in NSCLC needs to be examined in future studies.

To our knowledge, this is the first study to show significant associations between p110α and p110β expression levels and patient survival in NSCLC. Interestingly, high p110α and p110β expression levels correlated with worse OS in advanced and early stage NSCLCs, respectively; both isoforms functioned differently depending on the tumor stage. This result indicates that isoform selectivity should be seriously considered when PI3K inhibitors are investigated and adopted for NSCLC treatment. In addition, our study supports the potential of p110α and p110β as independent prognostic biomarkers of NSCLC. However, the study design was retrospective, included a relatively small number of cases, and lacked molecular validation. Large-scale, prospective studies with molecular validation are required to verify these results.

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Disclosure of conflict of interest

None.

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References

[1] Zhao L, Vogt PK. Class I PI3K in oncogenic cellular transformation. Oncogene 2008; 27: 5486-5496.

[2] Herman SE, Johnson AJ. Molecular pathways: targeting phosphoinositide 3-kinase p110-delta in chronic lymphocytic leukemia. Clin Cancer Res 2012; 18: 4013-4018.

[3] Engelman JA, Luo J, Cantley LC. The evolution of phosphatidylinositol 3-kinases as regulators of growth and metabolism. Nat Rev Genet 2006; 7: 606-619.

[4] Katso R, Okkenhaug K, Ahmadi K, White S, Timms J, Waterfield MD. Cellular function of phosphoinositide 3-kinases: implications for development, homeostasis, and cancer. Annu Rev Cell Dev Biol 2001; 17: 615-675.
[5] Solomon B, Pearson RB. Class IA phosphatidylinositol 3-kinase signaling in non-small cell lung cancer. J Thorac Oncol 2009; 4: 787-791.
[6] Marone R, Cmiljanovic V, Giese B, Wymann MP. Targeting phosphoinositide 3-kinase: moving towards therapy. Biochim Biophys Acta 2008; 1784: 159-185.
[7] Samuels Y, Ericson K. Oncogenic PI3K and its role in cancer. Curr Opin Oncol 2006; 18: 77-82.
[8] Wilks ST. Potential of overcoming resistance to HER2-targeted therapies through the PI3K/Akt/mTOR pathway. Breast 2015; 24: 548-555.
[9] Li J, Dang Y, Gao J, Li Y, Zou J, Shen L. PI3K/AKT/mTOR pathway is activated after imatinib secondary resistance in gastrointestinal stromal tumors (GISTs). Med Oncol 2015; 32: 111.
[10] Mayer IA, Arteaga CL. The PI3K/AKT pathway as a target for cancer treatment. Annu Rev Med 2016; 67: 11-28.
[11] Toulany M, Rodemann HP. Phosphatidylinositol 3-kinase/Akt signaling as a key mediator of tumor cell responsiveness to radiation. Semin Cancer Biol 2015; 35: 180-190.
[12] Jia S, Roberts TM, Zhao JJ. Should individual PI3 kinase isoforms be targeted in cancer? Curr Opin Cell Biol 2009; 21: 199-208.
[13] Liu N, Rowley BR, Bull CO, Schneider C, Haegebarth A, Schatz CA, Fracasso PR, Wilkie DP, Hentemann M, Wilhelm SM, Scott WJ, Mumberg D, Ziegelbauer K. BAY 80-6946 is a highly selective intravenous PI3K inhibitor with potent p110α and p110δ activities in tumor cell lines and xenograft models. Mol Cancer Ther 2013; 12: 2319-2330.
[14] Chandarlapaty S, Sawai A, Scaltriti M, Rodrik-Outmezguine V, Grbovic-Huezo O, Serra V, Majumder PK, Baselga J, Rosen N. AKT inhibition relieves feedback suppression of receptor tyrosine kinase expression and activity. Cancer Cell 2011; 19: 58-71.
[15] Martinez-Martí A, Felip E. PI3K Pathway in NSCLC. Front Oncol 2012; 1: 55.
[16] Scrima M, De Marco C, Fabiani F, Franco R, Pirozzi G, Rocco G, Ravo M, Weisz A, Zoppoli P, Ceccarelli M, Botti G, Malanga D, Viglietto G. Signaling networks associated with AKT activation in non-small cell lung cancer (NSCLC): new insights on the role of phosphatydil-inositol-3 kinase. PLoS One 2012; 7: e30427.
[17] Wang L, Hu H, Pan Y, Wang R, Li Y, Shen L, Yu Y, Li H, Cai D, Sun Y, Chen H. PIK3CA mutations frequently coexist with EGFR/KRAS mutations in non-small cell lung cancer and suggest poor prognosis in EGFR/KRAS wildtype subgroup. PLoS One 2014; 9: e88291.
[18] Usul Afsar C, Sahin B, Gunaldi M, Kılıc Bagir E, Gumurdulu D, Burgut R, Erkisi M, Kara IO, Paydas S, Karaca F, Ercolak V. Expression of fibroblast growth factor receptor 1, fibroblast growth factor 2, phosphatidyl inositol 3 phosphate kinase and their clinical and prognostic significance in early and advanced stage of squamous cell carcinoma of the lung. Int J Clin Exp Pathol 2015; 8: 9760-9771.
[19] Woenckhaus J, Steger K, Sturm K, Münstedt K, Franke FE, Fenic I. Prognostic value of PIK3CA and phosphorylated AKT expression in ovarian cancer. Virchows Arch 2007; 450: 387-395.
[20] Carvalho S, Milanezi F, Costa JL, Amendoeira I, Schmitt F. PIKing the right isoform: the emergent role of the p110beta subunit in breast cancer. Virchows Arch 2010; 456: 235-243.
[21] Edling CE, Selvaggi F, Buus R, Maffucci T, Di Sebastiano P, Friess H, Innocenti P, Kocher HM, Falasca M. Key role of phosphoinositide 3-kinase class IB in pancreatic cancer. Clin Cancer Res 2010; 16: 4928-4937.
[22] Cui W, Cai Y, Wang W, Liu Z, Wei P, Bi R, Chen W, Sun M, Zhou X. Frequent copy number variations of PI3K/AKT pathway and aberrant protein expressions of PI3K subunits are associated with inferior survival in diffuse large B cell lymphoma. J Transl Med 2014; 12: 10.
[23] Lee HW, Lee EH, Park MI, Kim MS, Jeong JS, Roh MS, Kim DC, Kim SJ, Pak MG, Rha SH. Differential expression of PI3K isoforms and correlation with clinicopathological factors in patients with gastric cancer. Int J Clin Exp Pathol 2016; 9: 1588-1597.
[24] Tsim S, O'Dowd CA, Milroy R, Davidson S. Staging of non-small cell lung cancer (NSCLC): a review. Respir Med 2010; 104: 1767-1774.