Pathology & Oncology Research (2019) 25:1059–1066
https://doi.org/10.1007/s12253-018-0464-y

ORIGINAL ARTICLE

Ki67 and P53 in Relation to Disease Progression in Metastatic Pancreatic Cancer: a Single Institution Analysis

Sally Temraz[1] · Ali Shamseddine[1] · Deborah Mukherji[1] · Maya Charafeddine[2] · Arafat Tfayli[1] · Hazem Assi[1] · Miza Salim Hammoud[1] · Iman Makki[1] · Samer Nassif[3]

Received: 19 December 2017 / Accepted: 23 August 2018 / Published online: 5 September 2018

Abstract

We investigated the expression patterns of Ki67 and p53 in metastatic pancreatic adenocarcinomas and analyzed their relationship with disease progression-free survival (PFS) and overall survival (OS) in the overall study population and in patients treated with a gemcitabine-containing chemotherapy versus FOLFIRINOX chemotherapy. Patients with histologically confirmed stage IV adenocarcinoma of the pancreas treated at AUBMC were included after obtaining institutional review board approval (IRB ID: IM.ST.05). The ROC was plotted to identify the threshold Ki-67, p53 and CA19–9 value for disease progression, the identified value was further used in Kaplan Meier curves to compare PFS for both groups (gemcitabine versus FOLFIRINOX). A value of $p < 0.05$ was considered significant in all analyses. On univariate analysis, patients who had a Ki-67 > 12.5% or a p53 > 15% had significantly shorter PFS ($p = 0.034$ and $p = 0.016$, respectively). This effect was restricted to Gemcitabine or gemcitabine-combination treated patients. A decrease in CA19–9 levels 6–8 weeks after chemotherapy of >58% had significantly longer PFS ($p = 0.027$). On multivariate analysis after controlling for grade, age and P53, Ki-67 remained significant, for every one unit increase in Ki-67 the progression risk increases by 1.017 times. Our study highlights the negative impact of high P53 expression and Ki67 proliferation index on PFS in patients with metastatic pancreatic cancer.

Keywords Pancreatic cancer · Ki67 · P53 · CA19–9 · Prognosis

Introduction

Pancreatic cancer is the fourth leading cause of death from malignant disease and has an extremely poor prognosis with a 5-year survival rate less than 5% [1]. Chemotherapy for metastatic disease can suppress tumor growth however a significant proportion of cancers do not respond and all eventually develop resistance. Given the rapid lethality of this disease, identification of either molecular or immunohistochemical markers involved in response to chemotherapy is crucial in this patient population to identify subsets of patients who are more likely to respond to specific therapies and tailor therapeutic strategies accordingly.

Pancreatic cancers involve many genomic alterations, the majority of which include alterations in cell cycle regulators such as tumor suppressor gene p53 (*TP53*) [2]. The *TP53* gene is critical to normal cellular function and results in cell cycle arrest and apoptosis following DNA damage [3]. Mutated *TP53* leads to a p53 protein with increased stability. Thus high expression of the p53 protein is indicative of mutated *TP53*. While several retrospective studies failed to identify p53 overexpression a prognostic marker, a recent prospective trial, the CONKO-001 revealed that a high expression of p53 was significantly associated with shorter disease-free survival (DFS) and overall survival (OS) in the setting of resected pancreatic cancer [4]. The prognostic impact of p53 expression in the metastatic setting has yet to be determined.

Sally Temraz and Samer Nassif contributed equally to the work at hand

✉ Sally Temraz
st29@aub.edu.lb

[1] Department of internal medicine, division of oncology/hematology, American University of Beirut Medical Center, Riad El Solh, Beirut 110 72020, Lebanon

[2] Data Management and Clinical Research Unit, American University of Beirut Medical Center, Riad El Solh, Beirut 110 72020, Lebanon

[3] Department of Pathology & Laboratory Medicine, American University of Beirut Medical Center, Riad El Solh, Beirut 110 72020, Lebanon

Ki-67 protein is a cellular marker for proliferation which is present during all active phases of the cell cycle but is absent from resting cells [5]. In tumor cells, the expression of Ki-67 is indicative of tumor proliferation rates and is associated with initiation, progression, metastasis and prognosis [6–8]. In pancreatic cancer, Ki-67 evaluated by immunohistochemistry (IHC) has shown conflicting results with regard to its prognostic impact [9].

Protein expressions of p53 and Ki67 can be easily tested by IHC on archival formalin fixed paraffin embedded tumor specimens. Despite extensive research on these biomarkers, data on the prognostic impact of Ki67 and p53 in patients with metastatic pancreatic cancer remains limited. We therefore investigated the expression patterns of Ki67 and p53 in metastatic pancreatic adenocarcinomas and analyzed their relationship with disease progression-free survival (PFS) on first-line chemotherapy for metastatic disease and overall survival (OS). We further correlated tumor expression with PFS and OS in patients treated with a gemcitabine-containing chemotherapy versus FOLFIRINOX chemotherapy (5fluurouracil, irinotecan and oxaliplatin) in a retrospective analysis.

Materials and Methods

Patients

After institutional review board (IRB) approval (IRB ID: IM.ST.05), the medical records of patients with pancreatic cancer treated at the American University of Beirut Medical center from May 2000–November 2013 were retrospectively reviewed. Eligibility criteria included all patients with histologically confirmed stage IV adenocarcinoma of the pancreas. Patient baseline characteristics collected included sex, smoking status, treatment modality, grade of tumor, Body Mass index (BMI), disease history, Karnofsky Performance status and response to treatment. Follow up on all patients was made though clinic charts and medical records for disease and survival status. All data were de-identified and transferred into a password protected IBM SPSS v. 23 spread sheet for statistical analysis.

For every pancreatectomy specimen, all slides were reviewed to identify the optimal blocks to be used for immunohistochemistry. Only one block was selected for each case, based on amount of tumor present and optimal tissue preservation. For needle core biopsy cases, only one block was available and was used for this study.

Immunohistochemistry

Immunostaining for Ki-67 (MIB-1) and P53 (DO-7) was performed on the Ventana immunostainer using protocols established by the manufacturer and validated at our laboratory. With each run of immunostaining, positive control tissues were used in order to confirm adequate staining performance, and internal controls were noted, when applicable. For Ki-67 and P53 staining, only nuclear staining was considered positive. Ki-67 proliferation index was determined using the following method: 1-the tumor area was screened and foci with the highest nuclear staining were selected; 2-these foci were further subdivided into quadrants and examined at 400× magnification; 3- tumor nuclei were counted in each quadrants, and the percentage of tumor nuclei with positive staining was determined. P53 staining results were noted as an overall estimate (percentage) of positive tumor nuclei, throughout the entire tumor area in the stained section. There were only three surgical specimens and the rest were biopsies.

Tumor Marker CA19–9 Measurement

Peripheral venous blood samples were retrospectively reviewed for each patient at the time of presentation, prior to any chemotherapy regimen and then again 6–8 weeks after chemotherapy administration. The analysis was performed using an automated, commercially available electrochemiluminescent immunoassay (GI-MA CA 19–9, Corporation Cobas e601 Hitachi/ Roche, Germany).

Statistical Analysis

Numerical variables were summarized by their median, mean and range. Categorical variables were described by counts and relative frequencies. Mean comparison was done using independent sample t-test for patients who had progression vs. patients who did not have progression. OS was defined as the time from initiation of first-line chemotherapy to death (due to any cause) or the end of follow-up (censored observations). PFS was calculated from the time of initiation of first line chemotherapy to date of documented relapse. PFS curve was plotted using the Kaplan Meier curve, and the log-rank was used to identify significant difference between the studied groups. Cox regression multivariate analysis that includes time-to-event and controls for all variables in the model was used to examine variables that are influencing OS and PFS. Using the backward elimination method, the hazard ratios (HRs) and 95% CIs were calculated for variables that remained significant in the model. The Receiver operating curve (ROC) was plotted to identify the threshold Ki-67 value for disease progression, the identified value was further used in Kaplan Meier curves to compare PFS for both groups. A value of $p < 0.05$ was considered significant in all analyses. All p values were 2-sided; all statistical analysis was performed using the SPSS v.23.0 statistical package.

Table 1 Patient baseline characteristics

	N (%)	Median
Gender		
Males	38 (62.3%)	
Females	23 (37.7%)	
Smokers	35 (57.4%)	
BMI ≤ 30 (overweight)	36 (59%)	
BMI > 30 (Obese)	25 (41%)	
HTN	29 (47.5%)	
DM II	27 (44.3%)	
Karnofsky Performance status		
≤80%	12 (19.7%)	
>80%	49 (80.3%)	
Prior Whipple procedure	3 (4.9%)	
Adjuvant Chemotherapy	2 (3.3%)	
Gemcitabine/Gemcitabine-based		
Gemcitabine single agent	10 (16.3%)	6 cycles
Gemzar-Oxaliplatin	11 (18.0%)	5 cycles
Gemzar- Cisplatin	12 (19.7%)	5 cycles
Gemzar- Abraxane	6 (9.8%)	6 cycles
Folfirinox	22 (36%)	5 cycles
Response to first-line chemotherapy		
Stable	13 (21.3%)	
Progression	45 (73.7%)	
Partial response	3 (5.0%)	
Second line therapy	29 (47.5%)	
Death	53 (86.9%)	

Results

A total of 61 patients were included in the analysis, out of these patients 58 had upfront metastatic disease while 3 patients had prior whipple surgery before the time of disease recurrence. Out of the 3 patients who had prior whipple, only two received post-operative adjuvant Gemcitabine, Median time from surgery to recurrence was 17 months. Table 1 shows patients' baseline characteristics. Median follow up on all patients was 15 months. There were 39 patients who took Gemcitabine-based chemotherapy with a median of 5 cycles, and 22 patients took Folfirinox with a median of 5 cycles. Progression was noted in 45 (73.7%) patients; whereas 13 (21.3%) patients had stable disease and only 3 patients (5%) had partial response.

Means for all three markers Ki-67, P53 and Ca19–9 were significantly higher in patients who had disease progression compared to patients who did not have disease progression; Ki67 (31.0 vs 8.2; $p < 0.001$), P53 (46.9 vs 16.2; $p < 0.001$) and CA19–9 (8885.3 vs 1714.4; $p = 0.016$). Means for all three markers for patients who were alive compared to those who died was not significant.

High Proliferation Is Associated with Decreased Time to Disease Progression

To identify the cutoff for Ki-67 proliferation index after which the likelihood of progression increases, we plotted the ROC curve. The estimated cutoff of 12.5% was used in a Kaplan Meier curve for PFS and OS. The curve showed that in the overall study population, patients who had a Ki-67 greater than the cutoff 12.5% had significantly shorter PFS ($p = 0.034$) (Fig. 1). Most interesting, this effect was restricted to Gemcitabine or gemcibaine-combination treated patients ($p = 0.048$) while in patients treated with FOLFIRINOX, low proliferation was not associated with improved PFS ($p = 0.697$) (Fig. 2). Low proliferation was not associated with improved OS in the overall population ($p = 0.549$) (Fig. 1). This effect on OS remained insignificant even after comparing the Gemcitabine-treated and FOLFIRINOX-treated groups.

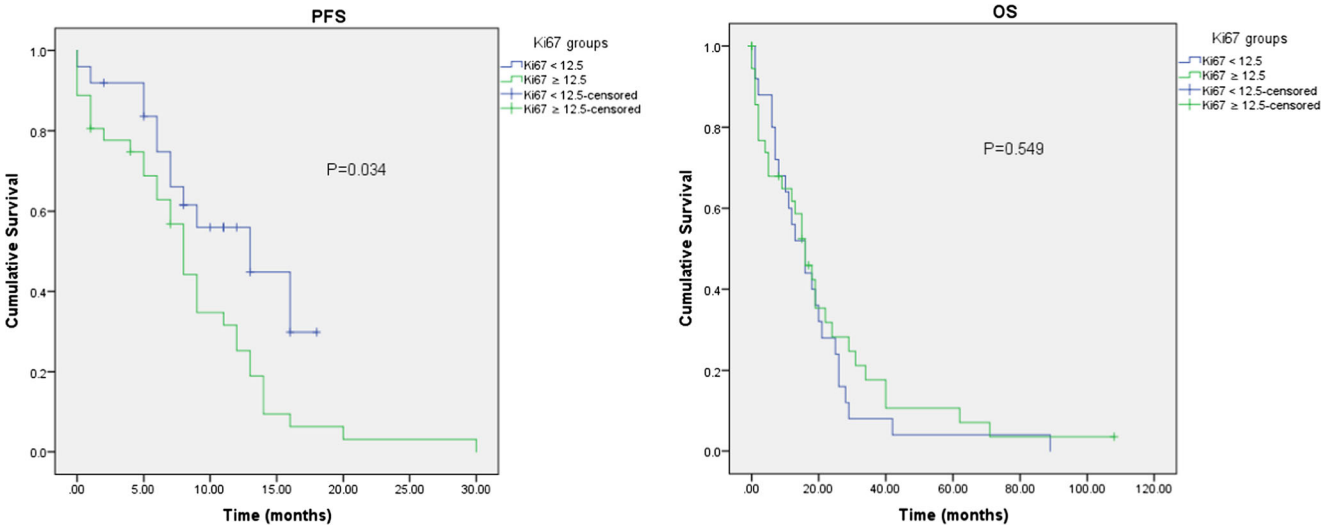

Fig. 1 progression free survival (PFS) and overall survival (OS) for the overall study group in dependence on high and low ki67 index

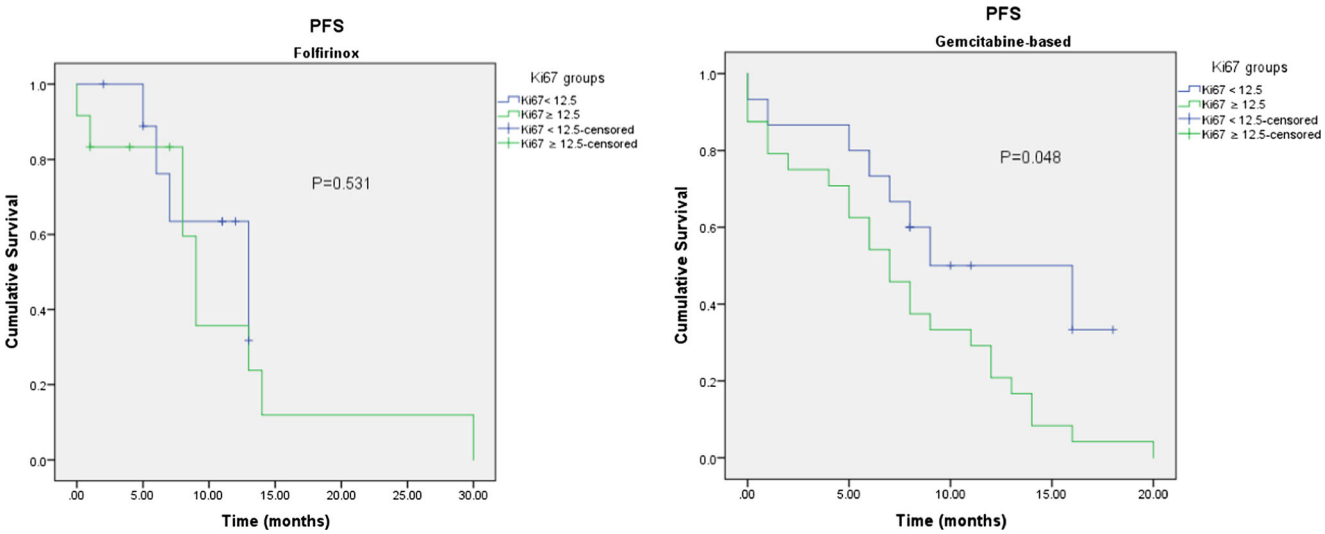

Fig. 2 progression free survival (PFS) in gemcitabine group and FOLFIRINOX group in dependence on high and low ki67 index

High Expression of p53 Is Associated with Decreased Time to Disease Progression

To identify the cutoff for P53 percentage after which the likelihood of progression increases, we plotted the ROC curve. The estimated cutoff of 15.0% was used in a Kaplan Meier curve for disease progression and overall survival. The curve showed that in the overall study population, patients who had a P53 greater than the cutoff 15.0% had significantly shorter PFS ($p = 0.015$) (Fig. 3). Also, this effect was restricted to Gemcitabine treated patients ($p = 0.016$) while for patients treated with FOLFIRINOX, low expression was not associated with improved PFS. Low P53 expression was not associated with improved OS in the overall population ($p = 0.652$) (Fig. 4). This effect on OS remained insignificant even after comparing the Gemcitabine-treated and FOLFIRINOX-treated groups.

Decreasing Levels of CA19–9 of ≥58% Is Associated with Prolonged PFS

To identify the cutoff for CA19–9 after which the likelihood of progression increases, we plotted the ROC curve. The estimated cutoff of 58% was used in the Kaplan Meier curve for disease progression and overall survival. The curve showed that in the overall study population, patients who had a decrease in CA19–9 levels 6–8 weeks after chemotherapy of >58% had significantly longer PFS ($p = 0.027$) (Fig. 5). Also, this effect was restricted to Gemcitabine-based treated patients only (P-0.044) (Fig. 6). In the overall study population, a decrease in CA19–9 levels of >58% was not associated with prolonged OS (Fig. 5). The value remained non significant even when comparing the Gemcitabine-treated group and the FOLFIRINOX-treated group.

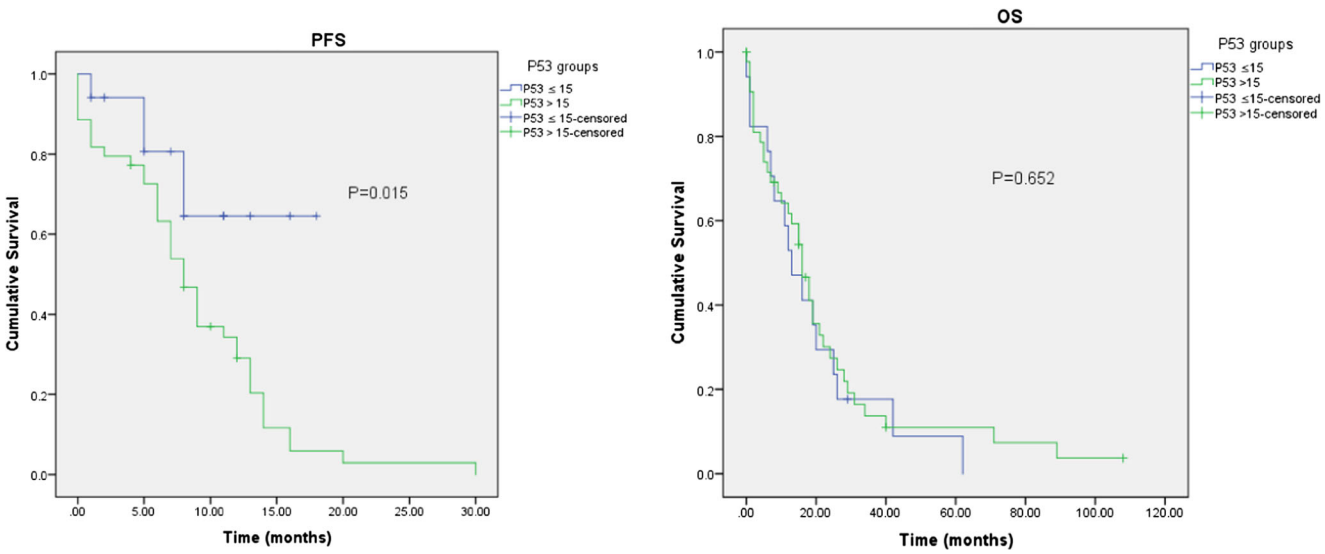

Fig. 3 progression free survival (PFS) and overall survival (OS) for the overall study group in dependence on high and low p53 expression

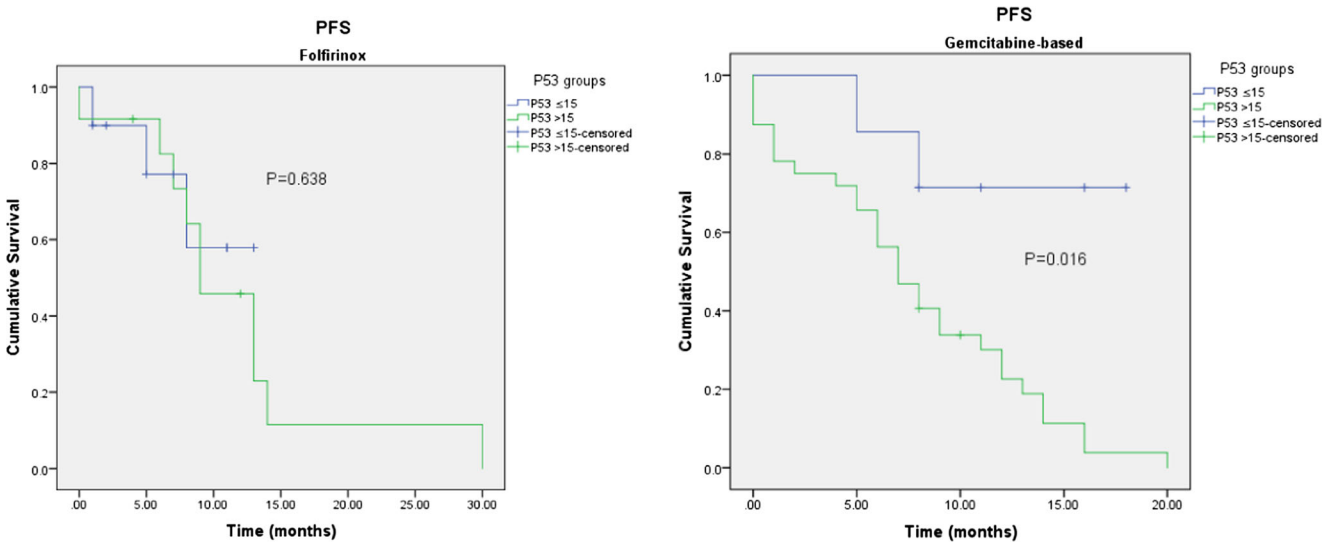

Fig. 4 progression free survival (PFS) in gemcitabine group and FOLFIRINOX group in dependence on high and low p53 expression

Multivariable Analysis

To test the variables influencing progression, a Cox regression was plotted for P53, Ki-67, grade, and age at diagnosis. Ki-67 and grade were shown to be significant, for every one unit increase in Ki-67 the progression risk will be increased by 1.017 times, controlling for other variables including grade, age, and P53 (Table 2). Grade was significantly associated with progression in the model; patients with grade 2 tumors were 7.163 times more likely to develop disease progression compared to patients with grade 1 tumors while patients with grade 3 tumors were 15.182 times more likely to develop disease progression compared to patients with grade 1 tumors (Table 2).

Multivariate Cox regression analysis for OS was adjusted for age, grade, P53, Ki-67 and Ca 19.9. Following the backward elimination method, ki-67 remained in the model although the *p* value was not significant at 0.249.

Discussion

Given the dismal prognosis of pancreatic adenocarcinomas and the very low 5-year survival rates, tumor biomarkers that could be used to predict which subset of patients are likely to benefit from chemotherapy would be useful to tailor therapy according to that individual patient's tumor characteristics.

P53 is a key mediator of DNA damage response by acting as a critical transducer of the DNA damage signal to p53 and other tumor suppressors [10]. The reported incidence of high P53 expression in pancreatic adenocarcinomas ranges

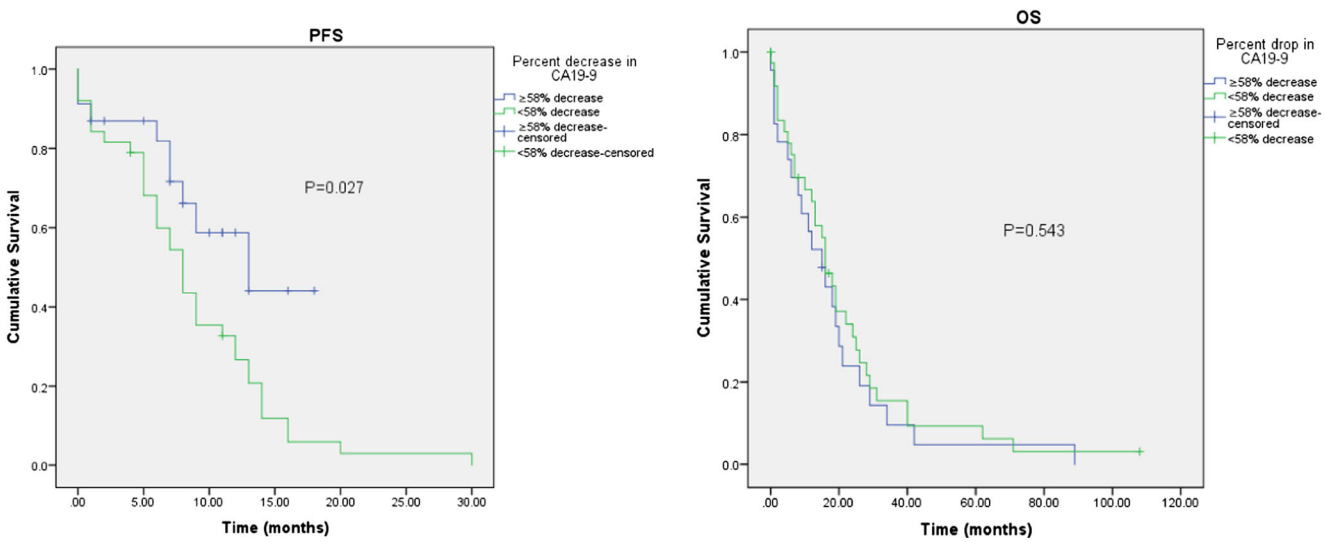

Fig. 5 progression free survival (PFS) and overall survival (OS) for the overall study group in dependence on >58% or < 58% decrease in CA19–9 after 6–8 cycles of chemotherapy

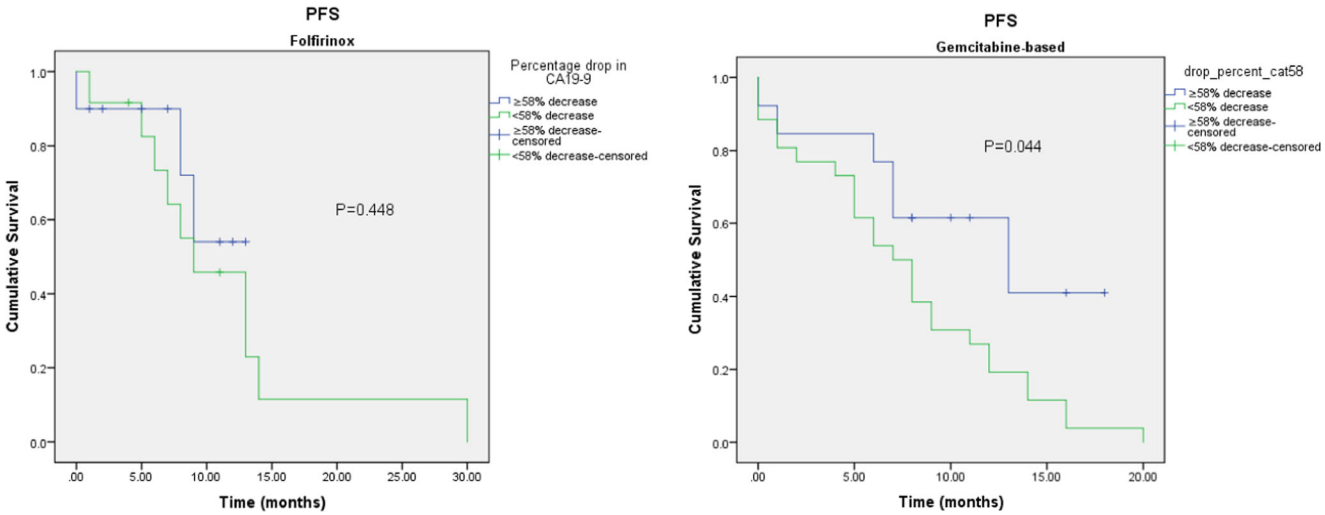

Fig. 6 Progression free survival PFS in gemcitabine group and FOLFIRINOX group in dependence on >58% or < 58% decrease in CA19–9

between 25 and 68% [11]. In previously published studies of resectable pancreatic adenocarcinomas, high P53 expression had no influence on OS [12–17]. Only few studies in resectable pancreatic adenocarcinomas receiving adjuvant chemotherapy reported that high P53 expression negatively influenced OS [4, 18]. Moreover, in a recently published study of metastatic pancreatic adenocarcinoma patients receiving FOLFIRINOX, patients with a strong tumor expression of P53 had a significantly lower OS compared to patients with no or weak expression of the protein [19]. In our cohort, high expression of P53 did not show any influence on OS whether treated with Gemcitabine-based or FOLFIRINOX treatments.

In our study, high P53 expression tends to negatively influence PFS in patients treated with Gemcitabine-based chemotherapy, but not in patients of the FOLFIRINOX group. In vitro studies revealed that mutant P53 confers resistance to gemcitabine treatment by stabilizing the mutant P53 protein in the nuclei of pancreatic adenocarcinomas [20, 21]. Thus, P53 expression might be used to predict which subset of pancreatic patients are more likely to respond to gemcitabine-based therapy. Despite the adverse activation of mutant P53 by gemcitabine, simultaneous treatment of pancreatic adenocarcinoma cells with gemcitabine and p53-reactivating molecules such as CP-31398 and RITA reduced growth rate and induced apoptosis [21].

Table 2 Multivariate Cox regression testing variables influencing disease progression

	p value	Hazard ratio	95.0% CI for Hazard ratio	
Ki-67 (%)	.037	1.017	1.001	1.034
Grade I (Ref)	.012			
Grade II	.069	7.163	.858	59.791
Grade III	.014	15.182	1.737	132.672

Ki-67 is a nuclear protein which is expressed in proliferating cells. Conflicting results pertaining to the prognostic significance of Ki-67 in resectable pancreatic adenocarcinomas have also been reported. To our knowledge, no study has reported on the prognostic significance of Ki-67 in advanced pancreatic cancers. Our results revealed that Ki-67 values had no influence on OS; however, in tumors with a high Ki-67proliferation index, the PFS was significantly poorer. Moreover, this negative effect was only seen in Gemcitabine-based treated group but not in the FOLFIRINOX group. In mouse models of pancreatic cancer, tumors showing low values of Ki-67 had increased sensitivity to gemcitabine [22]. This could partially explain the poorer PFS seen in high proliferating patients on gemcitabine therapy in our cohort. On multivariate analysis, Ki-67 remained significant with respect to disease progression and revealed that for every 1 % unit increase in Ki-67 the progression risk will be increased by 3%.

Several studies have addressed the efficacy of CA19–9 as a surrogate marker of response to chemotherapy in advanced pancreatic cancers. The majority of studies found that a decrease in CA19–9 levels of >20–50% correlated significantly with better OS [23–26]; however, in our cohort the cutoff of 58% did not affect OS but significantly improved PFS on univariate analysis.

The FOLFIRINOX regimen has been shown to be superior to gemcitabine-based chemotherapy in metastatic pancreatic cancers [27]; however, our preliminary results reveal that the gemcitabine-based therapy could be effective in patients with low ki-67 and p53 values. The Ki-67 and p53 markers are easily obtained in the clinic and thus could serve as prognostic markers for metastatic cancer patients who are likely to respond to gemcitabine-based therapy instead of FOLFIRINOX. Thus, these two markers ought to be further

evaluated in prospective trials since prognosis in pancreatic cancer patients remains poor and treatment choices are still limited.

Limitations of this study are the retrospective nature of the study as well as the limited number of cases with no randomization between treatment groups. We also have a heterogeneous sample of patients including some patients who had prior pancreatic resection and adjuvant therapy. The use of second-line and subsequent therapy was not uniform and these factors may have influenced the overall survival results. These observations are preliminary and hypothesis-generating however prospective validation of these markers may help guide systemic therapy.

Conclusion

Our study highlights the negative impact of high P53 expression and Ki67 proliferation index on PFS in patients with metastatic pancreatic cancer. Additionally, Ki-67 remained an independent prognostic factor for PFS after controlling for other factors. This negative effect was especially seen in the Gemcitabine-based treated group. Further prospective studies are warranted to validate the prognostic and predictive value of Ki67 and p53 in advanced pancreatic adenocarcinoma with implications for systemic therapy treatment protocol selection.

Acknowledgements This work has been funded by the Seed grant from the American University of Beirut.

Funding This study was funded by the Seed grant from the American University of Beirut.

Compliance with Ethical Standards

Conflicts of Interest The authors report no direct conflicts of interest pertaining to the work. Other conflicts of interest not related to this work are as follows. ST reports receiving payment for development of educational presentations from Merck and travel support from MSD, Amgen and Roche. AS reports having received research grants, honorarium or educational support from Sanofi, Roche, Merck, Bristol Meyer, Amgen, Novartis, MSD and Piere Fabre. DM reports having received honoraria or travel support from Roche, Amgen, Pfizer, MSD and Novartis. MC reports having received research funding from GSK. AT reports having received research grant from Merck serono, travel support from MSK and Roche and consultancy from Pfizer, MSK, Lilly and Novartis. HA reports having received honoraria from Roche, Merck, MSD and AstraZeneca. For the remaining authors none were declared.

References

1. Siegel R, Ma J, Zou Z, Jemal A (2014) Cancer statistics, 2014. CA Cancer J Clin 64(1):9–29. https://doi.org/10.3322/caac.21208
2. Saiki Y, Horii A (2014) Molecular pathology of pancreatic cancer. Pathol Int 64(1):10–19. https://doi.org/10.1111/pin.12114
3. Shimamura A, Fisher DE (1996) p53 in life and death. Clin Cancer Res 2(3):435–440
4. Striefler JK, Sinn M, Pelzer U, Juhling A, Wislocka L, Bahra M, Sinn BV, Denkert C, Dorken B, Oettle H, Riess H, Blaker H, Lohneis P (2016) P53 overexpression and Ki67-index are associated with outcome in ductal pancreatic adenocarcinoma with adjuvant gemcitabine treatment. Pathol Res Pract 212(8): 726–734. https://doi.org/10.1016/j.prp.2016.06.001
5. Scholzen T, Gerdes J (2000) The Ki-67 protein: from the known and the unknown. J Cell Physiol 182(3):311–322. https://doi.org/10.1002/(SICI)1097-4652(200003)182:3<311::AID-JCP1>3.0.CO;2-9
6. Jalava P, Kuopio T, Juntti-Patinen L, Kotkansalo T, Kronqvist P, Collan Y (2006) Ki67 immunohistochemistry: a valuable marker in prognostication but with a risk of misclassification: proliferation subgroups formed based on Ki67 immunoreactivity and standardized mitotic index. Histopathology 48(6):674–682. https://doi.org/10.1111/j.1365-2559.2006.02402.x
7. Gentile V, Vicini P, Giacomelli L, Cardillo MR, Pierangeli A, Degener AM (2006) Detection of human papillomavirus DNA, p53 and ki67 expression in penile carcinomas. Int J Immunopathol Pharmacol 19(1):209–215
8. Lee CS (1996) Differences in cell proliferation and prognostic significance of proliferating cell nuclear antigen and Ki-67 antigen immunoreactivity in in situ and invasive carcinomas of the extrahepatic biliary tract. Cancer 78(9):1881–1887
9. Hruban RH, Iacobuzio-Donahue C, Wilentz RE, Goggins M, Kern SE (2001) Molecular pathology of pancreatic cancer. Cancer J 7(4): 251–258
10. Wang B, Matsuoka S, Carpenter PB, Elledge SJ (2002) 53BP1, a mediator of the DNA damage checkpoint. Science 298(5597):1435–1438. https://doi.org/10.1126/science.1076182
11. Smith RA, Tang J, Tudur-Smith C, Neoptolemos JP, Ghaneh P (2011) Meta-analysis of immunohistochemical prognostic markers in resected pancreatic cancer. Br J Cancer 104(9):1440–1451. https://doi.org/10.1038/bjc.2011.110
12. Bold RJ, Hess KR, Pearson AS, Grau AM, Sinicrope FA, Jennings M, McConkey DJ, Bucana CD, Cleary KR, Hallin PA, Chiao PJ, Abbruzzese JL, Evans DB (1999) Prognostic factors in resectable pancreatic cancer: p53 and bcl-2. J Gastrointest Surg 3(3):263–277
13. Dergham ST, Dugan MC, Joshi US, Chen YC, Du W, Smith DW, Arlauskas P, Crissman JD, Vaitkevicius VK, Sarkar FH (1997) The clinical significance of p21(WAF1/CIP-1) and p53 expression in pancreatic adenocarcinoma. Cancer 80(3):372–381
14. Nio Y, Dong M, Uegaki K, Hirahara N, Minari Y, Sasaki S, Takamura M, Iguchi C, Tamura K (1999) Comparative significance of p53 and WAF/1-p21 expression on the efficacy of adjuvant chemotherapy for resectable invasive ductal carcinoma of the pancreas. Pancreas 18(2):117–126
15. Nio Y, Dong M, Iguchi C, Yamasawa K, Toga T, Itakura M, Tamura K (2001) Expression of Bcl-2 and p53 protein in resectable invasive ductal carcinoma of the pancreas: effects on clinical outcome and efficacy of adjuvant chemotherapy. J Surg Oncol 76(3):188–196
16. Makinen K, Hakala T, Lipponen P, Alhava E, Eskelinen M (1998) Clinical contribution of bcl-2, p53 and Ki-67 proteins in pancreatic ductal adenocarcinoma. Anticancer Res 18(1B): 615–618
17. DiGiuseppe JA, Hruban RH, Goodman SN, Polak M, van den Berg FM, Allison DC, Cameron JL, Offerhaus GJ (1994) Overexpression of p53 protein in adenocarcinoma of the pancreas. Am J Clin Pathol 101(6):684–688

18. Ahrendt SA, Brown HM, Komorowski RA, Zhu YR, Wilson SD, Erickson BA, Ritch PS, Pitt HA, Demeure MJ (2000) p21WAF1 expression is associated with improved survival after adjuvant chemoradiation for pancreatic cancer. Surgery 128(4):520–530. https://doi.org/10.1067/msy.2000.108052
19. Vitellius C, Eymerit-Morin C, Luet D, Fizanne L, Foubert F, Bertrais S, Rousselet MC, Caroli-Bosc FX (2017) Relationship between the expression of O6-Methylguanine-DNA methyltransferase (MGMT) and p53, and the clinical response in metastatic pancreatic adenocarcinoma treated with FOLFIRINOX. Clin Drug Investig 37:669–677. https://doi.org/10.1007/s40261-017-0522-3
20. Galmarini CM, Clarke ML, Falette N, Puisieux A, Mackey JR, Dumontet C (2002) Expression of a non-functional p53 affects the sensitivity of cancer cells to gemcitabine. Int J Cancer 97(4):439–445
21. Fiorini C, Cordani M, Padroni C, Blandino G, Di Agostino S, Donadelli M (2015) Mutant p53 stimulates chemoresistance of pancreatic adenocarcinoma cells to gemcitabine. Biochim Biophys Acta 1853(1):89–100. https://doi.org/10.1016/j.bbamcr.2014.10.003
22. Venkatasubbarao K, Peterson L, Zhao S, Hill P, Cao L, Zhou Q, Nawrocki ST, Freeman JW (2013) Inhibiting signal transducer and activator of transcription-3 increases response to gemcitabine and delays progression of pancreatic cancer. Mol Cancer 12(1):104. https://doi.org/10.1186/1476-4598-12-104
23. Ko AH, Hwang J, Venook AP, Abbruzzese JL, Bergsland EK, Tempero MA (2005) Serum CA19-9 response as a surrogate for clinical outcome in patients receiving fixed-dose rate gemcitabine for advanced pancreatic cancer. Br J Cancer 93(2):195–199. https://doi.org/10.1038/sj.bjc.6602687
24. Halm U, Schumann T, Schiefke I, Witzigmann H, Mossner J, Keim V (2000) Decrease of CA 19-9 during chemotherapy with gemcitabine predicts survival time in patients with advanced pancreatic cancer. Br J Cancer 82(5):1013–1016. https://doi.org/10.1054/bjoc.1999.1035
25. Ziske C, Schlie C, Gorschluter M, Glasmacher A, Mey U, Strehl J, Sauerbruch T, Schmidt-Wolf IG (2003) Prognostic value of CA 19-9 levels in patients with inoperable adenocarcinoma of the pancreas treated with gemcitabine. Br J Cancer 89(8):1413–1417. https://doi.org/10.1038/sj.bjc.6601263
26. Saad ED, Machado MC, Wajsbrot D, Abramoff R, Hoff PM, Tabacof J, Katz A, Simon SD, Gansl RC (2002) Pretreatment CA 19-9 level as a prognostic factor in patients with advanced pancreatic cancer treated with gemcitabine. Int J Gastrointest Cancer 32(1):35–41. https://doi.org/10.1385/IJGC:32:1:35
27. Conroy T, Desseigne F, Ychou M, Bouché O, Guimbaud R, Bécouarn Y, Adenis A, Raoul J-L, Gourgou-Bourgade S, de la Fouchardière C, Bennouna J, Bachet J-B, Khemissa-Akouz F, Péré-Vergé D, Delbaldo C, Assenat E, Chauffert B, Michel P, Montoto-Grillot C, Ducreux M (2011) FOLFIRINOX versus gemcitabine for metastatic pancreatic Cancer. N Engl J Med 364(19):1817–1825. https://doi.org/10.1056/NEJMoa1011923