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Therapeutic drug monitoring of neoadjuvant mFOLFIRINOX in resected pancreatic ductal adenocarcinoma

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ABSTRACT

Background: Despite a potentially curative treatment, the prognosis after upfront surgery and adjuvant chemotherapy for patients with resectable pancreatic ductal adenocarcinoma (PDAC) is poor. Modified FOLFIRINOX (mFOLFIRINOX) is a cornerstone in the systemic treatment of PDAC, including the neoadjuvant setting. Pharmacokinetic-guided (PKG) dosing has demonstrated beneficial effects in other tumors, but scarce data is available in pancreatic cancer.

Methods: Forty-six patients with resected PDAC after mFOLFIRINOX neoadjuvant approach and included in an institutional protocol for anticancer drug monitoring were retrospectively analyzed. 5-Fluorouracil (5-FU) dosage was adjusted throughout neoadjuvant treatment according to pharmacokinetic parameters and Irinotecan (CPT-11) pharmacokinetic variables were retrospectively estimated.

Results: By exploratory univariate analyses, a significantly longer progression-free survival was observed for patients with either 5-FU area under the curve (AUC) above 28 mcg·h/mL or CPT-11 AUC values below 10 mcg·h/mL. In the multivariate analyses adjusted by age, gender, performance status and resectability after stratification according to both pharmacokinetic parameters, the risk of progression was significantly reduced in patients with 5-FU AUC $\geq$28 mcg·h/mL [HR = 0.251, 95% CI 0.096–0.656; p = 0.005] and CPT-11 AUC <10 mcg·h/mL [HR = 0.189, 95% CI 0.073–0.486, p = 0.001].

Conclusions: Pharmacokinetically-guided dose adjustment of standard chemotherapy treatments might improve survival outcomes in patients with pancreatic ductal adenocarcinoma.

1. Background

Pancreatic cancer is the fourth leading cause of cancer death and is expected to become the second by 2030 [1]. Approximately 50% of patients have metastatic disease at diagnosis and only 10–20% are considered resectable [2]. Even in the latter, the 5-year survival rate is poor and has not shown much improvement over the last decades [1].

Upfront resection with adjuvant chemotherapy has long been the standard of care for patients with resectable pancreatic ductal adenocarcinoma (PDAC) [3–7]. However, increasing data suggest a meaningful role for a neoadjuvant approach in this setting [8,9]. Potential advantages include an increased R0 resection rate, early exposure to systemic therapy in all patients, treatment of micrometastatic disease and improved patients' selection for resection. Several randomized trials have suggested an outcome benefit with the use of either preoperative chemotherapy or chemoradiotherapy (CRT) over upfront surgery [10–12]. Regarding the systemic treatment within the neoadjuvant approach, modified FOLFIRINOX

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(mFOLFIRINOX) might be of choice in fit patients due to its beneficial effect over Gemcitabine-based regimens in both the metastatic and adjuvant settings [13,14].

5-Fluorouracil (5-FU) dosage is usually based on body surface area (BSA) [15], but the BSA dosing method leads to both wide variability of individual systemic exposure (up to 30-fold in systemic clearance) and an inappropriate dosage [16]. 5-FU has been considered a perfect candidate for pharmacokinetic-guided (PKG) dosing as drug clearance and area under the curve (AUC) have been correlated with both toxicity and therapeutic efficacy [16].

Irinotecan (CPT-11) is a prodrug of the topoisomerase-1 inhibitor SN-38 that, in turn, can be conjugated by UDP-glucuronosyltransferase (UGT) enzyme encoded by the *UGT1A1* gene to form the inactive derivative SN-38 glucuronide (SN-38G) [17]. The concentration of SN-38 in individual patients is highly variable and is, in part, associated with a variant allele in the proximal promoter region of *UGT1A1* that causes a reduced enzyme expression. Moreover, as with 5-FU, CPT-11 dosing by BSA is not significantly related to SN-38 exposure due to individual pharmacokinetic variability [18,19].

PKG algorithms minimize this pharmacokinetic variability, ensure individual systemic exposure within the optimal range and contribute to a better therapeutic index [20]. The impact of these PKG algorithms in patients with resected PDAC after preoperative therapy is currently unknown.

The aim of this study was to assess whether PKG adjustment of mFOLFIRINOX treatment in the neoadjuvant setting of potentially resectable PDAC correlates with clinical outcomes.

2. Methods

2.1. Patient eligibility

All patients diagnosed of potentially resectable PDAC treated with neoadjuvant approach with mFOLFIRINOX-based induction polichemotherapy (IPCT), followed by CRT and surgery, and included in an institutional protocol of therapeutic anticancer drug monitoring were retrospectively analyzed. At patient diagnosis, initial work-up included: clinical examination, laboratory tests including serum CA-19.9 levels, endoscopic ultrasound (EUS) with guided fine needle aspiration (FNA) biopsy and a CT-scan to define the extension of the disease. Patients were considered for neoadjuvant therapy if they had a good performance status ($\leq$1 according to Eastern Cooperative Oncology Group (ECOG)), an adequate hematological, renal and liver function, and a histologically confirmed resectable/borderline resectable/locally advanced PDAC. The criteria applied to determine resectability were based on the National Comprehensive Cancer Network [21].

2.2. Neoadjuvant therapy

Neoadjuvant treatment consisted of 3–7 cycles of biweekly mFOLFIRINOX chemotherapy (oxaliplatin 85 mg/m^2 in 2-h infusion, leucovorin 400 mg/m^2 in 1h30′ infusion, irinotecan 165 mg/m^2 in 1h30' infusion and 5-FU 3200 mg/m^2 in 46-h infusion). After the first cycle, 5-FU dosage was adjusted according to individual pharmacokinetic parameters and treatment tolerance. The criteria used for dose adjustment were based on previously described clearance data and dose modifications guidelines in colorectal cancer [22].

Before each chemotherapy cycle, all patients underwent routine workup including physical examination, blood tests and treatment-induced adverse events assessment. mFOLFIRINOX chemotherapy was administered with the patient admitted in the hospital for the first two cycles to perform pharmacokinetic monitoring of 5-FU during the 46-h infusion. The pharmacokinetic monitoring in the second cycle was to evaluate the intercyclical variability and to confirm the results obtained.

In case of non-progressive disease after mFOLFIRINOX-based IPCT, three-dimensional conformal external beam radiotherapy (3D-RT) or an intensity-modulated technique (IMRT) was planned [23]. Treatment planning was performed as previously detailed [24]. Briefly, four fields with 15-MV photons were employed to deliver 50 Gy over 4–5 weeks with conventional daily fractions of 1.8–2 Gy, 5 days per week, with concurrent capecitabine and oxaliplatin.

2.3. Pharmacokinetic sampling

Before treatment initiation, for early identification of patients at risk of developing serious 5-FU toxicity, a baseline determination of endogenous pyrimidines (uracil (U) concentration, dihydrouracil (UH_2) concentration and its ratio) was performed as a phenotypic evaluation of individual dihydropyrimidine dehydrogenase (DPD) activity [25]. In patients with a DPD phenotype deficiency, an inferential dosage adjustmen was performed based on pyrimidines values identified as predictors of toxicity.

For the pharmacokinetic analyses of 5-FU, blood samples were taken at 15 and 30 hours after 5-FU infusion initiation. 5-FU plasma concentrations were analyzed by high performance liquid chromatography (HPLC), according to the technique developed by the Clinical Pharmacokinetics Unit from our Center [26]. The individual pharmacokinetic parameters of 5-FU were estimated by Bayesian methodology using as preliminary information a linear monocompartmental population model developed by the Clinical Pharmacokinetics Unit in 2002 and implemented in the USC* PACKv11.2 program package of the University of Southern California. The model was developed and validated in patients with colorectal and biliopancreatic cancer. Initially, both linear and non-linear pharmacokinetic models were explored, without finding significant differences in the selection criteria of the models. Therefore, similarly to other models described [27,28], a linear monocompartmental model was selected for the individual estimation of 5-FU pharmacokinetic parameters *(*Appendix A*)*.

CPT-11 pharmacokinetics were estimated based on a bicompartmental model developed and validated by the Clinical Pharmacokinetics Unit from our Centre in 2002. In that model, patient exposure to CPT-11 metabolites (SN-38 and SN-38G) were estimated by non-compartmental methods. Subsequently, in 2019, the previous model was optimized to a multicompartmental model including CPT-11, SN-38 and SN-38G data [18]. The original model has been the one used in clinical practice to date, but further studies for CPT-11 metabolites prediction are being currently developed. Multiple linear regression predictive equations for CPT-11 clearance (Cl) and CPT-11 AUC were developed to avoid the requirement of serum drug concentration data. As shown in Fig. S2, its predictive behavior is optimal.

2.4. Outcomes

The primary endpoint of the study was progression-free survival (PFS) with overall survival (OS) and pathologic tumor regression as secondary endpoints. Progression-free survival (PFS) was defined as the time from diagnosis to the date of radiologic progression (local and/or distant), death (all causes) or last contact until 5-year follow-up. Radiologic progression was defined according to RECIST 1.1 criteria [29]. Overall survival (OS) was defined as the time from diagnosis until death from all causes or last contact when still alive until 5-year follow-up. Pathologic tumor regression was evaluated according to the College of American Pathologists (CAP) grading

scheme [30]. R0 was reported when no tumor cells were evidenced within 1 mm of the circumferential resection margin (CRM) [31].

2.5. Statistical analysis

A descriptive analysis of the study variables was performed using absolute and relative frequencies for categorical variables, and central tendency and dispersion measures for quantitative variables. Correlation analyses were performed using Pearson's correlation coefficient.

Time to event endpoints were analyzed by Kaplan-Meier survival curves and Breslow tests (predominance of events at early follow-up) to assess the impact of the target cut-off values of 5-FU and CPT-11 AUCs. To quantify the association between statistically significant AUC values and survival outcomes, hazard ratios with two-sided 95% confidence interval were estimated with stratified Cox proportional hazards regression model by age, gender, resectability and ECOG. The assumption of proportional hazards was tested using log-minus-log survival plots. The co-secondary endpoint of pathologic tumor regression was compared between groups with the chi-squared test.

All statistical tests were conducted at a two-sided significance level of 0.05. Study results are expressed as absolute numbers and percentages, mean or median, interquartile range (IQR), standard deviation (SD) and 95% confidence intervals (CI).

Data processing and analysis were performed using the statistical packages for Windows IBM SPSS Statistics version 22.0.

3. Results

3.1. Patients characteristics

Forty-six patients diagnosed from January 2012 to February 2020 were retrospectively included, thirty men (65%) and 16 women (35%), with a mean age of 63 years old (SD 8.6). In twenty-eight patients (61%) the primary tumor location was at head or isthmus, and in 18 patients (39%) at pancreatic body or tail. Twenty-one patients (46%) presented resectable tumors, 17 patients (37%) borderline resectable and 8 patients (17%) non-resectable tumors according to NCCN guidelines. Table 1 describes patients' baseline characteristics.

After a median follow-up of 26.6 months, 26 (56.6%) recurrence events occurred, and 19 patients (41.3%) died. Median progression-free survival (mPFS) and overall survival (mOS) were 30.1 months (95% CI 14.2–45.9) and 45.7 months (95% CI 31.3–60.1), respectively. One-, 2- and 3-year PFS rates were 83% (95% CI 67.7–91.5), 44.3% (95% CI 28.5–59) and 38% (95% CI 22.7–53.1%), respectively.

3.2. Multimodal neoadjuvant therapy

Neoadjuvant treatment characteristics are described in Table 2. The median number of induction chemotherapy cycles administered were 4 (range 3–7). Three patients had high baseline uracil values suggesting a lower DPD mediated 5-FU metabolism by competitive mechanism. Consequently, 5-FU initial dosage was adjusted. Twenty-two patients had *UGT1A1* determination, with 1/28* heterozygosity in 11 patients and *28/*28 homozygosis in two patients. Mean 5-FU, CPT-11 and oxaliplatin dose intensity administered was 1360.3 mg/m^2/wk (SD 242.5), 69.1 mg/m^2/wk (SD 8.6) and 40.4 mg/m^2/wk (SD 3.6), respectively. Overall dosing did not correlate with clinical outcomes in terms of PFS in either of the three chemotherapy drugs.

After mFOLFIRINOX-based IPCT, thirty-seven patients (80%) received CRT, 32 patients with the IMRT technique and five with 3D-RT. In two patients who did not receive neoadjuvant radiotherapy, it was administered after surgery.

During IPCT, grade 3-4 adverse events were reported in 28 patients (61%). No grade 5 adverse events were described. Grade 3–4 toxicities included neutropenia (n = 21, 45.6%), diarrhea (n = 5, 10.9%), leukopenia (n = 2, 4.3%), anemia (n = 2, 4.3%), fatigue (n = 1, 2.2%), mucositis (n = 1, 2.2%) and rash (n = 1, 2.2%). A third of grade 3–4 neutropenia were febrile neutropenia. Granulocyte colony-stimulating factors were used in 26 patients (56.5%). Grade 3–4 CRT associated toxicity included thrombopenia (n = 4, 10.8%), lymphopenia (n =12, 32.4%) and mucositis (n = 1, 2.7%).

After the neoadjuvant approach, all patients underwent pancreatic surgery after confirming resectability by computed tomography (CT): thirty-one patients (67.4%) pylorus preserving pancreaticoduodenectomy (PPPC) and fifteen patients (32.6%) distal pancreatectomy. R0 resection was achieved in forty patients (87%). Tumor regression grade according to the CAP grading scheme was 0, 1, 2 and 3 in 3 (6.5%), 12 (26.1%), 23 (50%) and 8 (17.4)

Table 1
Baseline characteristics of patients with resected PDAC after neoadjuvant approach. GPT: glutamate-pyruvate transaminase.

Variable		Variable	
Age (years)		**Staging – n (%)**	
Mean (SD)	62.8 (8.6)	I	29 (63)
Gender – n (%)		II	8 (17.4)
Male	30 (65.2)	III	8 (17.4)
Female	16 (34.8)	**Resectability – n (%)**	
ECOG – n (%)		Resectable	21 (45.7)
0	21 (45.7)	Borderline resectable	17 (36.9)
1	23 (50)	Non-resectable	8 (17.4)
2	2 (4.3)	**Baseline CA19.9 (U/mL)**	
Tumor location – n (%)		Median (IQR)	106.2 (25.1–449.8)
Head-Isthmus	30 (65.2)	**Baseline creatinine (mg/dL)**	
Body-tail	16 (34.8)	Median (IQR)	0.8 (0.7–0.9)
Baseline T stage – n (%)		≤1.2 – n (%)	44 (95.7)
Tx	2 (4.3)	>1.2 – n (%)	2 (4.3)
T1	6 (13)	**Baseline bilirubin (mg/dL)**	
T2	26 (56.5)	Median (IQR)	0.69 (0.45–2.09)
T3	5 (10.9)	≤1.2 – n (%)	27 (58.7)
T4	7 (15.3)	>1.2 – n (%)	19 (41.3)
Baseline N stage – n (%)		**Baseline GPT (U/L)**	
Nx	1 (2.2)	Median (IQR)	25 (15–66)
N0	37 (80.4)	≤43.5 – n (%)	29 (63)
N+	8 (17.4)	>43.5 – n (%)	17 (37)

Table 2
Treatment characteristics of multimodal neoadjuvant treatment. G-CSF: Granulocyte colony-stimulating factor.

Variable	
Induction mFOLFOXIRI (cycles)	
Median (Rank)	4 (3–7)
5-FU dose intensity (mg/m^2/wk)	
Mean (SD)	1360.3 (242.5)
5-FU AUC (mcg·h/mL)	
Cycle 1 – mean (SD)	31 (5.5)
Cycle 2 – mean (SD)	28.7 (5.7)
5-FU dose adjustment between C1 and C2 – n (mean % change)	
Increase	9 (14.5)
Decrease	20 (11)
No changes	17
5-FU dose adjustment between C2 and C3 – n (mean % change)	
Increase	17 (8.9)
Decrease	11 (10.9)
No changes	18
Irinotecan dose intensity (mg/m^2/wk)	
Mean (SD)	69.1 (8.6)
Irinotecan AUC (mcg·h/mL)	
Cycle 1 – mean (SD)	14.8 (11.5)
Cycle 2 – mean (SD)	9.1 (4.7)
***UGT1A1* – n (%)**	
1/1	9 (19.5)
1/28*	11 (24)
28*/28*	2 (4.3)
Unknown	24 (52.2)
Oxaliplatin dose intensity (mg/m^2/wk)	
Mean (SD)	40.4 (3.6)
Neoadjuvant radiotherapy technique – n (%)	
3D-RT	5 (11)
IMRT	32 (70)
Unknown	2 (4.3)
No neoadjuvant RT	7 (15.2)
Type of surgery – n (%)	
Pylorus preserving pancreaticoduodenectomy	31 (67.4)
Distal pancreatectomy	15 (32.6)
G-CSF during neoadjuvant treatment – n (%)	
Yes	26 (56.5)
No	20 (43.5)
Adjuvant treatment – n (%)	
Yes	18 (39)
No	28 (61)

Table 3
Pathological findings after neoadjuvant multimodal treatment.

Variable	n (%)
Surgical margins	
R0	40 (87)
R1	6 (13)
ypT	
ypTx	10 (21.7)
ypT0	2 (4.3)
ypT1-T2	22 (47.8)
ypT3-T4	12 (26.1)
Pathological lymph nodes	
ypNx	3 (6.5)
ypN0	31 (67.4)
ypN1	11 (23.9)
ypN2	1 (2.2)
Tumor regression (CAP score)	
0	3 (6.5)
1	12 (26.1)
2	23 (50)
3	8 (17.4)
Vascular invasion	
Present	4 (7)
Absent	42 (93)
Perineural invasion	
Present	19 (41.3)
Absent	27 (58.7)

patients, respectively. Vascular and perineural invasion was observed in 4 (8.7%) and 19 (41.3%) patients, respectively. The median number of resected lymph nodes was 10 (IQR 5–14), with twelve patients (26.1%) presenting nodal infiltration (ypN+). *[Table 3].*

3.3. Exploratory analysis of mFOLFIRINOX pharmacokinetic parameters

5-FU pharmacokinetic monitoring was performed in the first two cycles among all patients included. Fourteen patients (30.4%) required additional monitoring beyond cycle 2 due to a wide variability in pharmacokinetic parameters. In the first cycle, mean 5-FU AUC was 31 mcg*h/ml (SD 5.5). Seventeen patients (37%) achieved an AUC between 25 and 30 mcg·h/ml with the BSA dosage method. For the second cycle, twenty-nine patients (63%) required dose adjustment: 5-FU dosage was increased and reduced in 9 and 20 patients, respectively. Mean AUC in the second cycle was 28.7 mcg·h/ml (SD 5.7). Up to 19 patients (41.3%) achieved a 5-FU AUC within the target values. No correlation was observed between the AUC of the PKG second cycle and the dose administered (r = 0.082; p = 0.587).

In order to determine whether the 5-FU AUC in the second PKG cycle correlated with clinical outcomes, exploratory univariate analyses with different target AUC values were performed. In the multivariate analyses stratified by the significant AUC value and adjusted by age, gender, performance status and resectability, the risk of progression was significantly reduced in patients with 5-FU AUC ≥28 mcg·h/ml compared to patients with 5-FU AUC <28 mcg·h/ml, with a mPFS of 41.9 months (IQR 22.6 – not reached) and 19 months (IQR 12 – not reached), respectively [HR = 0.251, 95% CI 0.096–0.656; p = 0.005] *[Table 4, Fig. 1A]*. One-, 2- and 3-year PFS rates were 95%, 53.1% and 42.5% in patients with 5-FU AUC ≥28 mcg·h/ml and 71.6%, 36.2% and 36.2% in patients with 5-FU AUC <28 mcg·h/ml, respectively *[Table 4]*. No differences were seen in median overall survival, which was 44.7 months (IQR 31.4 – not reached) in patients with 5-FU AUC ≥28 mcg·h/ml and 51.2 months (IQR 18.2 – not reached) in patients below the 28 mcg·h/ml threshold (Breslow test = 0.242; p = 0.623) *[Table 4, Fig. 1B]*. No significant differences were also observed in tumor regression grade or pathological lymph node status (ypN) based on 5-FU AUC. CAP grade score was 0e1, 2 and 3 in 39.1%, 47.8% and 13% of patients with 5-FU AUC above 28 mcg$h\ml and in 26.1%, 52.2% and 21.7% of patients with 5-FU AUC below 28 mcg·h/ml, respectively *($\chi 2 = 1.143$, $df = 2$;* p = 0.565*) [Table 4]*.

CPT-11 pharmacokinetic variables were estimated for the first two cycles. Mean CPT-11 AUC in the first and second cycle was 14.8 mcg·h/mL (SD 11.5) and 9.1 mcg·h/mL (SD 4.7), respectively. As with 5-FU, no correlation was identified between CPT-11 AUC and the dose administered (r = 0.136; p = 0.366). Univariate analyses were also performed to assess whether a CPT-11 AUC threshold impacted in survival outcomes. Significant differences were observed at a cut-off value of 10 mcg·h/mL and multivariate analysis confirmed a significant benefit in terms of PFS. The mPFS of patients with CPT-11 AUC <10 mcg·h/mL in the second cycle was 38.3 months (IQR 19 – not reached) compared to 18.4 months (IQR 13.5–24.1) in patients with CPT-11 AUC ≥10 mcg·h/mL [HR = 0.189, 95% CI 0.073–0.486, p = 0.001] *[Table 4, Fig. 1C]*. One-, 2- and 3-year PFS rates were 82.4%, 56.3% and 46.9% in patients with CPT-11 AUC <10 mcg·h/mL and 84.6%, 17.3% and 17.3% in patients above the threshold, respectively *[Table 4]*. Similarly to 5-FU pharmacokinetics, no significant differences were observed in overall survival

Table 4
Survival outcomes and pathological variables according to 5-FU and CPT-11 AUCs thresholds.

	5-FU			CPT-11			5-FU & CPT-11			
Outcome variable	<28 mcg·h/mL	≥28 mcg·h/mL	*P*	<10 mcg·h/mL	≥10 mcg·h/mL	*P*	Group A	Group B	Group C	*P*
mPFS (m) – median (IQR)	19 (12 – NR)	41.9 (22.6 – NR)	0.047*	38.3 (10 – NR)	18.4 (13.5–24.1)	0.041*	NR (22.9 – NR)	24.1 (18 – NR)	13.5 (11.5–14.5)	<0.001*
1-y PFS rate - % (95% CI)	71.6 (47.4–86.1)	95 (69.5–99.3)	<0.001¯	82.4 (62.6–92.3)	84.6 (51.2–95.9)	0.568¯	91.7 (53.9–98.8)	83.5 (61.7–93.5)	60 (12.6–88.2)	<0.001¯
2-y PFS rate - % (95% CI)	36.2 (16.5–56.4)	53.1 (29.2–72.3)	0.016¯	56.3 (35.9–72.4)	17.3 (2.8–42.4)	<0.0001¯	66.7 (33.7–86))	41.5 (21–60.8)	–	<0.001¯
3-y PFS rate - % (95% CI)	36.2 (16.5–56.4)	42.5 (20.7–62.9)	0.384¯	46.9 (26.9–64.6)	17.3 (2.8–42.4)	<0.0001¯	50 (20.9–73.6)	41.5 (21–60.8)	–	0.256¯
mOS (m) – median (IQR)	51.2 (18.2 – NR)	44.7 (31.4 – NR)	0.623*	NR (28.8 – NR)	31.4 (18.2–43.5)	0.472*	NR (28.8 – NR)	45.7 (30.9 – NR)	18.2 (15 – NR)	0.216*
CAP 0/1 – n (%)	6 (26.1)	9 (39.1)	0.565¯	11 (33.3)	4 (30.8)	0.105¯	6 (40)	8 (30.8)	1 (20)	0.609¯
CAP 2 – n (%)	12 (52.2)	11 (47.8)		14 (42.4)	9 (69.2)		6 (40)	13 (50)	4 (80)	
CAP 3 – n (%)	5 (21.7)	3 (13)		8 (24.2)	0		3 (20)	5 (19.2)	0	
ypN0 – n (%)	16 (72.7)	16 (72.7)	1.000¯	24 (75)	7 (58.3)	0.281¯	11 (78.6)	18 (72)	3 (60)	0.720¯
ypN + - n (%)	6 (27.3)	6 (27.3)		8 (25)	5 (41.7)		3 (21.4)	7 (28)	2 (40)	

* Breslow test for Kaplan-Meier survival analysis.
¯The $\chi 2$ test for comparing categorical variables.

between patients below and above the CPT-11 AUC cut-off point of 10 mcg·h/mL *[Table 4, Fig. 1D]*. No differences were observed after stratification by *UGT1A1* genotype. Regarding pathological findings, tumor regression according to CAP score was 0-1, 2 and 3 in 33.3%, 42.4% and 24.2% of patients with CPT-11 AUC <10 mcg $h\mL, and in 30.8%, 69.2% and 0% of patients with with CPT-11 AUC ≥10 mcg·h/mL *[Table 4]*.

Subsequent analyses were performed considering both pharmacokinetic "*favorable*" parameters: 5-FU AUC ≥28 mcg·h/mL and CPT-11 AUC <10 mcg·h/mL in the second PKG cycle. All patients were stratified in three groups: Group A (n = 15): 5-FU AUC ≥28 mcg·h/mL and CPT-11 AUC <10 mcg·h/mL; group B (n = 26): 5-FU AUC ≥28 mcg·h/mL or CPT-11 AUC <10 mcg·h/mL; and group C (n = 5): 5-FU AUC <28 mcg·h/mL and CPT-11 AUC ≥10 mcg·h/mL. Table 5 shows baseline anthropometric and analytical variables of the 3 groups, as well as tumor and treatment characteristics. No significant differences were observed between groups except for a worse liver function tests in group C compared to group A *[Table 5]*.

Median PFS was not reached (IQR 22.9 – not reached) in patients of group A, 24.1 months (IQR 18 – not reached) in group B and 13.5 months (IQR 11.5–14.5) in group C (Breslow test = 16.497; p < 0.001) *[Fig. 1E]*. Median OS was not reached (IQR 28.8 – not reached), 45.7 months (IQR 30.9 - not reached) and 18.2 months (IQR 15 – not reached), respectively (Breslow test = 3.061; p = 0.216) *[Fig. 1F]*. No significant differences in prognostic pathological variables were observed between groups *[Table 4]*.

4. Discussion

The therapeutic window for most established cytotoxic anticancer agents is extremely narrow and their dosage remains largely empirical. Further adjustments in dose calculation according to individual parameters beyond BSA seem warranted. Our data point to a potential role of individual PKG adjustment of neoadjuvant 5-FU and CPT-11 to reach a specific threshold as a mean to improve outcomes in resected PDAC patients. Although the neoadjuvant induction regimen included oxaliplatin, metabolic alterations or polymorphisms that might significantly alter oxaliplatin exposure have not been described. Oxaliplatin pharmacokinetics includes a short initial distribution phase and a long terminal elimination phase, with a low interpatient variability [31]. In the present study oxaliplatin dose intensity did not correlate with PFS. For these reasons, a pharmacokinetic adjustment of this drug was not considered.

Considering several clinical and analytical prognostic factors in patients with potentially resectable PDAC, the combination of a "favorable" 5-FU and CPT-11 AUC seems to define a subgroup of patients with remarkable survival outcomes in terms of PFS. The absence of overall survival benefit in the present study might be influenced by other factors such as the use of adjuvant chemotherapy or treatments used at the disease recurrence. In the AGEO cohort, a retrospective multicenter study of neoadjuvant FOLFIRINOX, patients with CRT and surgery after induction therapy had benefit in disease-free survival but no significant benefit in OS [32]. Additionally, the low number of patients included might influence the non significant results of pharmacokinetic parameters in pathological findings.

5-FU is an optimal candidate for PKG dosing since therapeutic plasma levels are achieved in only 20–30% of patients with the BSA method [16,33]. In the present study, we empirically established a 5-FU AUC target range of 25–30 mcg·h/ml, mainly based on previous results in colorectal cancer suggesting a positive correlation between certain 5-FU AUC ranges and clinical benefit [22,33–35]. Scarce data is available about 5-FU pharmacokinetics in PDAC and their impact in terms of efficacy and toxicity. In the present study,

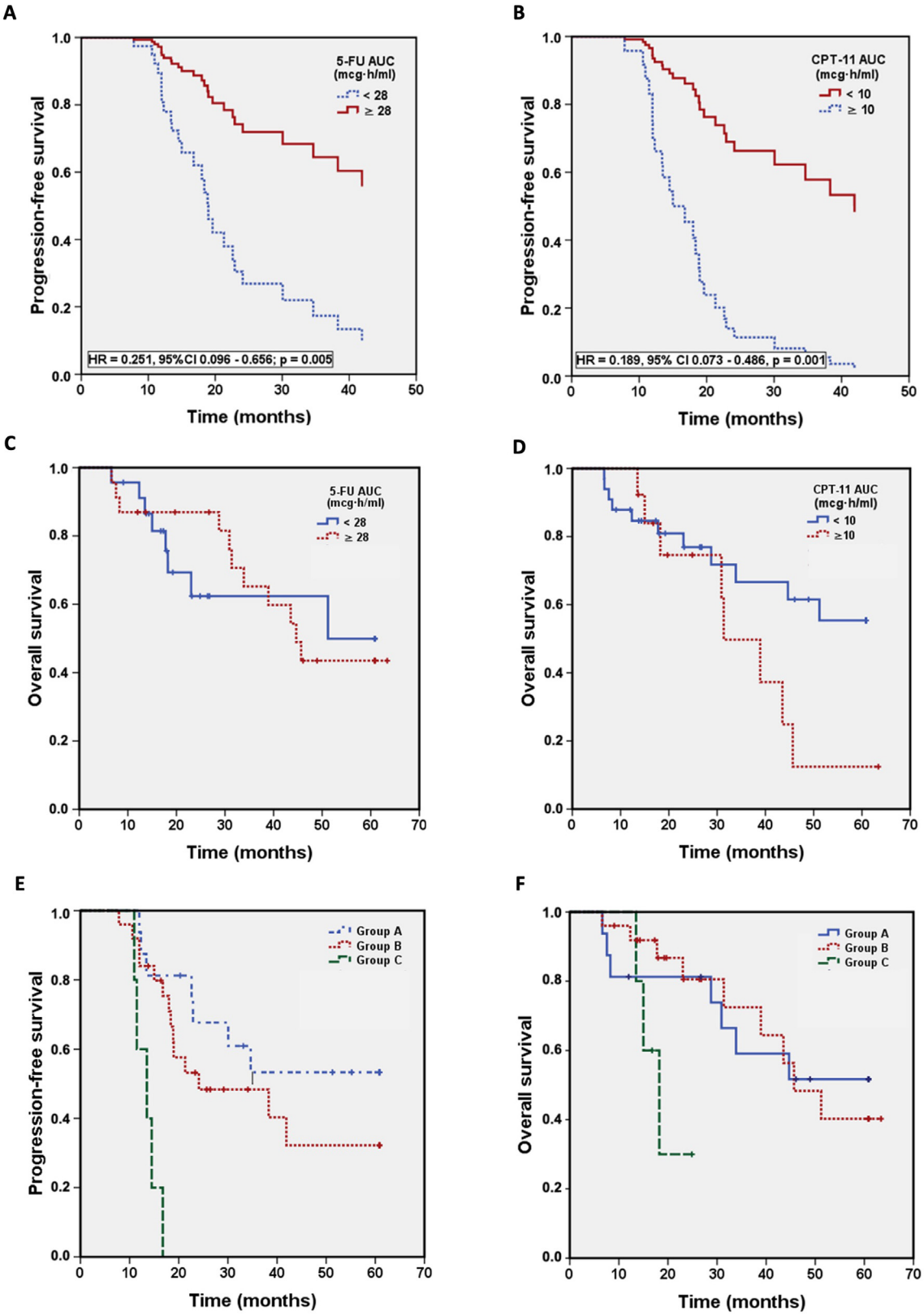

Fig. 1. Progression-free survival according to 5-FU **(A)** and CPT-11 **(B)** AUC. Kaplan-Meier survival curves for overall survival according to 5-FU and CPT-11 AUC **(C, D)**. Kaplan-Meier survival curves for progression-free survival **(E)** and overall survival **(F)** after stratification according to 5-FU and CPT-11 AUCs.
Group A (n = 15): 5-FU AUC ≥28 mcg·h/mL and CPT-11 AUC <10 mcg·h/mL; group B (n = 26): 5-FU AUC ≥28 mcg·h/mL or CPT-11 AUC <10 mcg·h/mL; and group C (n = 5): 5-FU AUC <28 mcg·h/mL and CPT-11 AUC ≥10 mcg·h/mL.

Table 5
Baseline and treatment characteristics of patients after stratification according to 5-FU and CPT-11 AUCs.

Variable	Group A (n = 15)	Group B (n = 26)	Group C (n = 5)
Age (years)			
Mean (SD)	60 (8.4)	64.3 (8.7)	61.6 (8.8)
Gender – n (%)			
Male	9 (60)	17 (65.4)	4 (80)
Female	6 (40)	9 (34.6)	1 (20)
ECOG – n (%)			
0	8 (53.3)	11 (42.3)	2 (40)
1	6 (40)	14 (53.8)	3 (60)
2	1 (6.7)	1 (3.8)	0
Tumor location – n (%)			
Head-isthmus	8 (53.3)	17 (65.4)	5 (100)
Body-tail	7 (46.7)	9 (34.6)	0
Staging – n (%)			
I	7 (46.7)	19 (73.1)	3 (60)
II	3 (20)	5 (19.2)	0
III	5 (33.3)	2 (7.7)	1 (20)
Resectability – n (%)			
Resectable	8 (53.3)	10 (38.5)	3 (60)
Borderline resectable	2 (13.3)	13 (50)	2 (40)
Non-resectable	5 (33.3)	3 (11.5)	0
Baseline creatinine (mg/dL)			
Median (IQR)	0.7 (0.6–0.9)	0.8 (0.7–0.9)	0.8 (0.8–0.9)
Baseline bilirrubine (mg/dL)			
Median (IQR)	0.5 (0.3–0.74)	0.89 (0.51–2.17)	2.17 (1.55–3.19)
Baseline GPT (UI/L)			
Median (IQR)	15 (10–33)	27.5 (19–81)	81 (55.5–134.5)
Neoadjuvant radiotherapy – n (%)			
Yes	11 (73.3)	22 (84.6)	5 (100)
No	4 (26.7)	4 (15.4)	0
Adjuvant treatment – n (%)			
Yes	6 (40)	9 (34.6)	3 (60)
No	9 (60)	17 (65.4)	2 (40)

an AUC $\geq$ 28 mcg·h/ml correlated with a longer PFS. This higher AUC target compared to that described in CCR patients might be justified by the pancreatic cancer stroma, which hinders the access of chemotherapy drugs to cancer cells [36]. In addition to becoming a mechanical barrier to effective drug delivery, pancreatic stroma favors the formation of a poorly vascularized microenvironment and promotes the secretion of soluble factors and inflammatory cytokines related to the development of therapeutic resistance [37,38]. Therefore, higher drug concentrations might be required to exert an antitumoral effect. Indeed, positive relationships between dose intensity and survival outcomes have already been reported in pancreatic cancer, both in adjuvant setting and advanced disease [39,40]. In locally advanced and metastatic PDAC patients treated with FOLFIRINOX regimen, improved response rate (RR) and disease control rate (DCR) was observed above a relative dose intensity threshold [41]. Thus, PKG adjustment can ensure an individual systemic exposure to the antineoplastic agent with a potential survival benefit.

CPT-11 pharmacokinetic variability has also led to CPT-11 and SN-38 pharmacokinetic models' description [18,20,42]. The results of the present study suggest a longer progression-free survival in patients with CPT-11 AUC <10 mcg·h/mL. CPT-11 is promptly converted by carboxylesterase enzymes to its active metabolite SN-38 which has about a hundred times greater potency than CPT-11. The disposition of SN-38 has been shown to be dependent on the CPT-11 provisions in previous pharmacokinetic studies [43,44]. Thus, patients with CPT-11 AUC $\geq$ 10 might present lower time to progression due to less conversion and, therefore, lower exposure to the active metabolite SN-38. Additionally, a multicompartmental pharmacokinetic model of CPT-11 and its metabolites established a direct correlation between minor levels of CPT-11 AUC and analytical covariables such as platelets, leukocytes and neutrophils count, suggesting a higher exposure to the active metabolite SN-38 [18]. Further studies to predict CPT-11 metabolites exposure from the multicompartmental model are being currently performed to confirm the results described.

Pharmacokinetic and pharmacodynamic data exist for 5-FU and CPT-11 and both chemotherapy drugs have a narrow therapeutic window [20,34]. For this reason, it is particularly relevant to achieve an optimal drug exposure within the therapeutic range based on pharmacokinetic parameters. In the present work, dosage individualization of conventional chemotherapy by easily obtained pharmacokinetic parameters in the daily practice seems to improve time to progression in PDAC patients compared to previous studies with an equivalent neoadjuvant regimen [33]. In the subgroup analysis, no significant differences in baseline and treatment characteristics were observed except for a worse liver function in the subgroup with unfavorable pharmacokinetic parameters. Further analyses with increased sample size are required to confirm the results obtained between groups.

Considering tolerability, PKG induction mFOLFIRINOX treatment was associated with grade 3–4 toxicity in 61% of cases. No patient required neoadjuvant treatment discontinuation due to severe toxicity. The higher incidence of neutropenia registered in the present work compared to previously reported meta-analysis might be due to a lack of G-CSF use as primary prophylaxis [45].

The study presented has several limitations. First, it is a retrospective single-institution study with patient heterogeneity in terms of disease characteristics and treatment variables. Second, the low number of patients included has not permitted a subanalysis of treatment efficacy depending on pharmacokinetic parameters according to age and gender. Third, after patients' stratification based on pharmacokinetic parameters of 5-FU and CPT-11, one group contained a low number of patients which

prevented from further analysis. Finally, the CPT-11 AUC was estimated from a bicompartimental model based on previous studies. Further studies with prospective pharmacokinetic analysis of CPT-11 and its metabolites are required to elucidate its role in terms of treatment efficacy.

5. Conclusions

Personalized management is one of the cornerstones in patients with pancreatic ductal adenocarcinoma. While progressive advances are arising in new therapies development, pharmacokinetically-guided dose adjustment of standard chemotherapy treatments might have clinical benefits in this patient population.

Ethics approval and consent to participate

Informed consent for chemotherapy treatment was obtained from all subjects involved in the study. The clinical research ethics committee from Navarra, Spain, approved the present study (reference EO16/3). The study was performed in accordance with the Declaration of Helsinki.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.pan.2023.03.001.

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