Liver Histology Predicts Liver Regeneration and Outcome in ALPPS

Novel Findings From A Multicenter Study

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Background and Aims: Alterations in liver histology influence the liver's capacity to regenerate, but the relevance of each of the different changes in rapid liver growth induction is unknown. This study aimed to analyze the influence of the degree of histological alterations during the first and second stages on the ability of the liver to regenerate.

Methods: This cohort study included data obtained from the International ALPPS Registry between November 2011 and October 2020. Only patients with colorectal liver metastases were included in the study. We developed a histological risk score based on histological changes (stages 1 and 2) and a tumor pathology score based on the histological factors associated with poor tumor prognosis.

Results: In total, 395 patients were included. The time to reach stage 2 was shorter in patients with a low histological risk stage 1 (13 vs 17 days, $P<0.01$), low histological risk stage 2 (13 vs 15 days, $P<0.01$), and low pathological tumor risk (13 vs 15 days, $P<0.01$). Regarding interval stage, there was a higher inverse correlation in high histological risk stage 1 group compared to low histological risk 1 group in relation with future liver remnant body weight ($r=-0.1$ and $r=-0.08$, respectively), and future liver remnant ($r=-0.15$ and $r=-0.06$, respectively).

Conclusions: ALPPS is associated with increased histological alterations in the liver parenchyma. It seems that the more histological alterations present and the higher the number of poor prognostic factors in the tumor histology, the longer the time to reach the second stage.

Keywords: ALPPS, colorectal liver metastases, histology, liver regeneration, liver resection, oncological outcomes

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Recently, a series of surgical techniques have been developed to induce efficient regeneration of future liver remnant (FLR) in staged liver resections.[1,2] Among them, associating liver partition

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and portal vein ligation for staged hepatectomy (ALPPS), induces accelerated liver hypertrophy compared with portal vein embolization (PVE) or classic 2-stage hepatectomy.[3–5] The major advantage of ALPPS is the reduction in drop-out rates (ie, progression to completion hepatectomy), particularly in colorectal liver metastases (CRLM), which is the most common indication.[6,7] The unique combination of portal vein deprivation and liver transection stimulates a complex pathophysiological regeneration pattern influenced by various factors.[8–16]

Histology plays a pivotal role in liver surgery. Patients with abnormal liver parenchyma who undergo hepatectomy have worse postoperative outcomes. However, discrepancies exist concerning the regenerative capacity of abnormal liver histology, particularly in the presence of fibrosis, cirrhosis, cholestasis, macrosteatosis, or chemotherapy-associated liver damage.[17–21] Alterations in liver histology influence the capacity of the liver to regenerate, but the relevance of each of the different changes in the scenario of rapid liver growth induction, as it occurs in ALPPS, is unknown.

ALPPS causes a series of histological changes in the FLR. In experimental models, histological analysis of the FLR revealed hepatic sinusoidal injury characterized by sinusoidal dilatation, microvesicular steatosis, ischemia, necrosis, hepatocellular atrophy, and centrilobular or perisinusoidal fibrosis.[22–25] This multicenter study aimed to analyze the influence of baseline FLR histological alterations and changes that occur between the first and second stages on the regenerative capacity of the liver, and perioperative outcomes in patients who underwent CRLM.

METHODS

Study Design

The ALPPS cohort study included from the International ALPPS Registry. The Registry has prospectively collected data on ALPPS cases since 2012, and is coordinated by the Department of Surgery at the University of Zurich, Switzerland. Approval to enter patients into the international ALPPS Registry was obtained from the Cantonal Ethics Committee of Zurich (KEK 2013-0326; Clinical Trials.gov. NCT01924741). The data extracted for analysis in February 2020. Only patients with CRLM were included in this study. To perform the analysis, all participating centers were contacted to complete a database with details on the histological data not captured in the current form of the registry. This study was conducted in accordance with the Declaration of Helsinki.

Endpoints

The primary aim of this study was to analyze the influence of the degree of histological alterations during the first and second stages and tumor pathology on the ability of the liver to regenerate. Secondary aims included intra- and postoperative outcomes, demographic variables (especially age, sex, weight, and comorbidities), and tumor profile characteristics. Histological samples were analyzed by experienced pathologists from centers specializing in complex hepatobiliary surgery.

Risk Score Definition Based on Histological Assessment

Ten types of histological changes were chosen for their relevance in defining the histological risk score for stages 1 and 2. Histological samples were analyzed and categorized according to the presence of fibrosis, macrosteatosis, microsteatosis, edema, sinusoidal dilatation, regeneration nodules, necrosis, hemorrhage, inflammation, and chemotherapy-associated steatohepatitis (CASH) (Fig. 1A). The degree of fibrosis was evaluated using Batts-Ludwig scale (0 = absent, 1 = portal, 2 = periportal, 3 = in bridges, and 4 = cirrhosis). The degree of macrosteatosis was classified according to the % of steatosis as 0 (0%), 1 (1%–33%), 2 (33%–66%), or 3 (> 66%). The degree of inflammation was classified as 0 (absence of inflammation), 1 (mild), 2 (moderate), or 3 (severe). The remaining alterations were classified according to their presence in the analyzed sample. Fibrosis was scored from 0 to 4 for the histological risk score calculations, and macrosteatosis and inflammation were scored from 0 to 3. The rest of each histological was assigned a value of 1 if it was present, or 0 otherwise. This score was applied to the intraoperative samples obtained during stage 1 (histological risk stage 1) and stage 2 FLR (histological risk 2). We defined histological risk as low when the score was < 2 and high when it was ≥ 3.

Risk Score Definition Based on Tumor Pathology Assessment

Nine types of prognostic tumor factors were chosen for their relevance in defining the tumor pathology risk score. The tumor samples were analyzed and categorized according to the presence of: degree of tumor differentiation, tumor necrosis, peritumoral inflammation, lymphovascular invasion, perineural invasion, biliary invasion, sinusoidal invasion, resection margin, and chemotherapy response (Fig. 1B). The degree of tumor differentiation was classified as 0 (well differentiated), 1 (moderately differentiated), 2 (poorly differentiated), or 3 (undifferentiated). The degree of chemotherapy response was classified as 0 (absence of viable cells in all sections studied), 1 (1%–10% isolated tumor cells or small groups of tumor cells), 2 (11%–50% significant decrease in tumor cells), and 3 (50% minimal response). Resection margins were classified as 0 (R0), 1 (R1), and 2 (R2). The remaining alterations were classified according

A

Liver histological alterations	Score
Fibrosis	
Absent	0
Portal	1
Periportal	2
In bridges	3
Cirrhosis	4
Macrosteatosis	
0%	0
1-33%	1
34-66%	2
> 66%	3
Microsteatosis	1
Edema	1
Sinusoidal dilatation	1
Regeneration nodules	1
Necrosis	1
Hemorraghe	1
Inflammation	
Absence	0
Mild	1
Moderate	2
Severe	3
CASH	1

Histological risk	Score
Low	< 2
High	> 3

B

Tumor pathology alterations	Score
Degree of differentiation	
Well	0
Moderately	1
Poorly	2
Undifferentiated	3
Chemotherapy response	
100%	0
99-90%	1
89-50%	2
< 50%	3
Tumor necrosis	1
Peritumoral inflammation	1
Lymphovascular invasion	1
Perineural invasion	1
Biliary invasion	1
Sinusoidal invasion	1
Resection margin	
R0	0
R1	1
R2	2

Tumor pathological risk	Score
Low	< 4
High	> 5

FIGURE 1. Histological risk score value definition (A). Tumor pathology risk score value definition (B).

to their presence in the analyzed histological samples. For the tumor pathology risk score calculation, tumor differentiation, and chemotherapy response were scored from 0 to 3, and resection margins were scored from 0 to 3 and 0 to 2, respectively, according to their respective classifications. The rest of each histological change was assigned a value of 1 if it was present, or 0 otherwise. This score was applied to the tumor histology of liver metastases. We defined pathological risk as low when the score was <4 and high when it was ≥ 4.

Variables Studied

Patient biometric data (age, sex, body mass index, comorbidities, Charlson index), pre-stage-2 risk score, RAS and RAF mutational status, TNM, neoadjuvant chemotherapy, adjuvant chemotherapy, operative time, Pringle maneuver, blood loss and need for transfusions, surgical technique, complications, and length of hospital stay in both stages were extracted. The Clavien–Dindo classification was used to assess 90-day morbidity. Postoperative hepatic insufficiency (PHLF) was analyzed according to the International Study Group of Liver Surgery definitions. Volumetric data regarding FLR/body weight (BW) in both stages, sFLR in both stages, sFLR increase (%), and the time between stages 1 and 2 were also collected.

Statistical Analysis

All data included in the database were analyzed using a professional statistical package (R Project, ver. 3.6.1, GLP). A descriptive statistical analysis for the continuous variables was carried out using median [interquartile range (IQR)] and the mean ($\pm$ standard deviation), depending on their distribution. Frequencies and percentages were used as the qualitative variables. Intergroup differences in continuous variables were assessed using the Kruskal-Wallis H-test and categorical data were assessed using Pearson's chi-squared test. To study the relationship between the variables, the chi-square test was applied between 2 qualitative variables and the Pearson correlation was applied when the variable was quantitative. Statistical significance was considered with a P value of <0.05.

RESULTS

Definition of the Study Population and Perioperative Outcomes

In total, 395 patients with stage 1 and/or stage 2 biopsies were included in the study (Table 1). The mean age of the patients was 60 years ($\pm$ 15.47), 62.9% were female and the mean body mass index was 24.98 kg/m^2 ($\pm$ 4.91). In 73.2% of patients, there was bilateral tumor involvement with a mean total lesion size of 6 ($\pm$ 5). Ninety-one percent of the patients received chemotherapy before stage 1, with a mean of 7 cycles ($\pm$ 5). The mean operative time of the second stage was shorter than the first stage,156 versus 260 minutes, respectively, whereas the percentage of Clavien–Dindo complications $\geq$IIIb was higher in the second stage compared to the first stage, 21% versus 6.8%, respectively.

TABLE 1. Demographic Data, Tumor Characteristics, and Perioperative Outcomes of the Entire Cohort of 395 Patients

	n/Total	%	Missing (%)
Baseline			
Age, y, mean (SD)	60 (15.47)	—	
Gender, female	248	63	
BMI (kg/m^2), mean (SD)	24.98 (4.91)	—	1
Comorbidity	143	41	12
Charlson index, mean (SD)	7 (2)	—	18
Tumor diameter (T)			9
T1	6	1	
T2	38	10	
T3	233	60	
T4	81	20	
Node involvement (N)			9
N0	76	19	
N1	155	39	
N2	125	32	
Nx	23	1	
Bilobar tumor involvement	289	80	8
Largest_liver_lesion, mm, mean (SD)	47 (40)	—	9
Total number lesion, mean (SD)	6 (6)	—	9
Synchronicity	278	76	8
K-Ras mutation	140	35	9
Chemotherapy before stage 1	361	93	2
CEA before chemotherapy (μg/L), mean (SD)	35.3 (175.3)	—	35
Liver first approach	77	21	9
Total number of cycles, mean (SD)	7 (5)	—	14
Duration of chemotherapy, mo, mean (SD)	4 (4)	—	
Monoclonal antibodies	262	71	6
Operative details			
ALPPS procedure classic	238	62	2
Transfusion RBC	128	34	5
Operative time stage 1, min, mean (SD)	260.5 (217)	—	25
Operative time stage 2, min, mean (SD)	156 (90)	—	28
Any complication stage 1	189	50	4
Any complication stage 2	216	59	8
Clavien–Dindo $\geq$IIIb stage 1	27	7	
Clavien–Dindo $\geq$IIIb stage 2	83	22	4
Postoperative liver failure	26	8	13
Hospital stay stage 1, d, mean (SD)	9 (7)	—	7
Hospital stay stage 2, d, mean (SD)	10 (2)	—	10

ALPPS indicates association liver partition and portal vein ligation for two staged hepatectomy;BMI, body mass index; CEA, carcinoembryonic antigen; RBC, red blood cells.

Scoring of Histological Features in Stages 1 and 2

A total of 337 biopsies from stage 1 and 390 biopsies from stage 2 were analyzed (Fig. 2). The most frequent histological alteration that reached its highest score in stage 1 (histological risk 1) was macrosteatosis, followed by microsteatosis, inflammation, and fibrosis. At stage 2, the highest scores (histological risk stage 2) were obtained for inflammation, followed by macrosteatosis, necrosis, and microsteatosis. The histological phenomena that revealed the greatest percentage increase between the first and second stages were hemorrhage (355%), nodule regeneration (163%), necrosis (96%), and inflammation (96%). Phenomena related to hepatic steatosis showed a small increase (Fig. 2).

Histological Risk Scores and Their Impact on Volumetry and Interstage Intervals

By reviewing the total score of the mentioned histopathological features, high- and low-risk categories were defined for histological risk stages 1 and 2. Patients with low histological risk stage 1 required a median of 13 days (IQR, 8–21) to reach the second stage compared to 17 days (IQR, 11–32) for patients

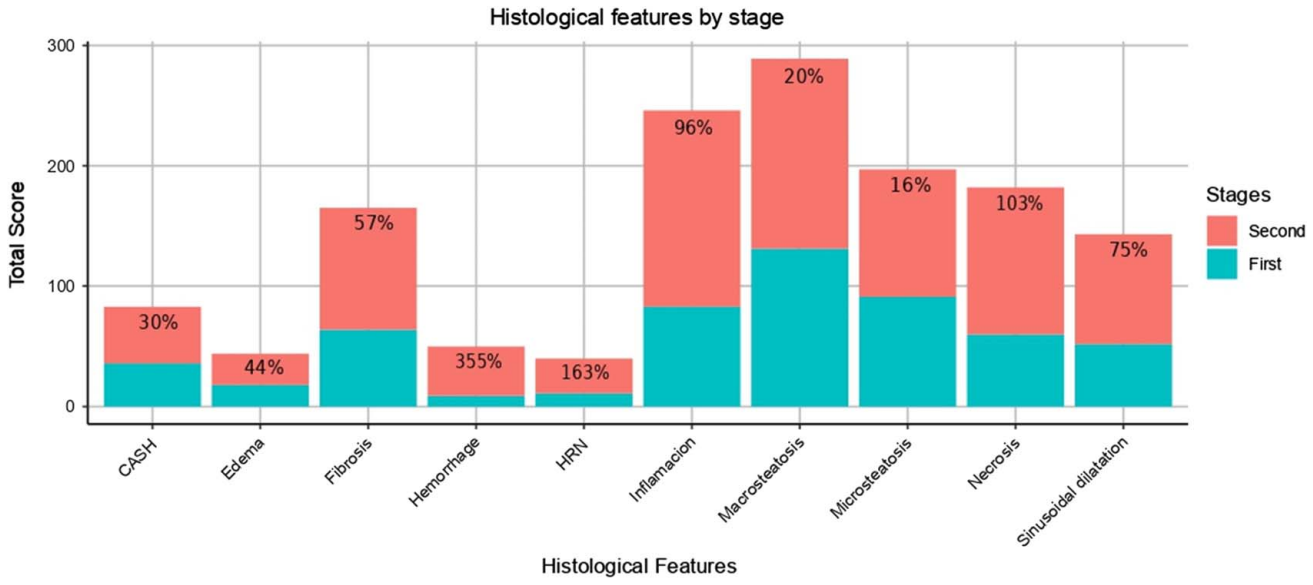

FIGURE 2. Histological features changes between stages 1 and 2 according to the established histological risk score.

with high histological stage risk 1 ($P < 0.01$) (Fig. 3A). Furthermore, patients with a low histological risk stage 2 needed less time than patients with a high histological risk 2 to reach the second stage, with a median of 12 days (IQR, 8–20) versus 15 days (IQR, 11–26), respectively ($P < 0.001$) (Fig. 3B).

Figure 4A shows scatter plots with high ($0.5 < |r| < 1$), moderate ($0.3 < |r| < 0.5$), and low ($0.1 < |r| < 0.3$) correlations between the histological risk scores and volumetric data. Statistically significant correlations were found between the histological risk scores analyzed in the stage 1 samples (Fig. 4B and C). Regarding the interval stage, there was a higher inverse correlation in the high histological risk stage 1 group than in the low histological risk stage 1 group in relation to FLR/BW ($r = -0.1$ and $r = -0.08$, respectively), and sFLR1 ($r = -0.15$ and $r = -0.06$, respectively). On the other hand, in relation with the sFLR1, there was a higher increase of percentage of FLR ($r = -0.44$ and $r = -0.32$, respectively) and sFLR2 ($r = -0.58$ and $r = -0.48$, respectively) in the low histological risk 1 group comparing to higher histological risk 1. In stage 2 samples, a higher positive correlation was found between sFLR1 and sFLR2 ($r = 0.66$ and $r = 0.49$) in the low histological risk stage 2 group than in the high histological risk stage 2 (Figure 4D and E).

Histological Risk Scores Correlations With Demographic, Tumor, and Perioperative Characteristics

Patients with a low histological risk stage 1 and 2 received a median of 6 cycles (IQR, 6–11.5) and 6 cycles (IQR, 6–12), respectively, compared to the 8 cycles (IQR, 6–12) received by both groups of patients with a high histological risk stages 1 and 2 ($P = 0.02$ and $P < 0.01$, respectively) (Fig. 5A and B). High histological risk stage 2 was related to a superior stage 2 median operative time (180 vs 137 minutes, $P = 0.03$), and more complications after stage 2 (15.97% vs 5.76%, $P = 0.03$) and in both stages (18.62% vs 6, 65%, $P = 0.01$) than patients with low histological risk stage 2 (Fig. 5C, E, F). Patients with high histological risk stage 1 required a greater need for transfusions than those with low histological risk stage 1 ($P < 0.01$) (Fig. 3D).

Tumor Pathology Risk Score

Factors associated with poor tumor prognosis were related to histological findings in both stages. The group of patients with a high tumor pathological risk score was associated with a higher

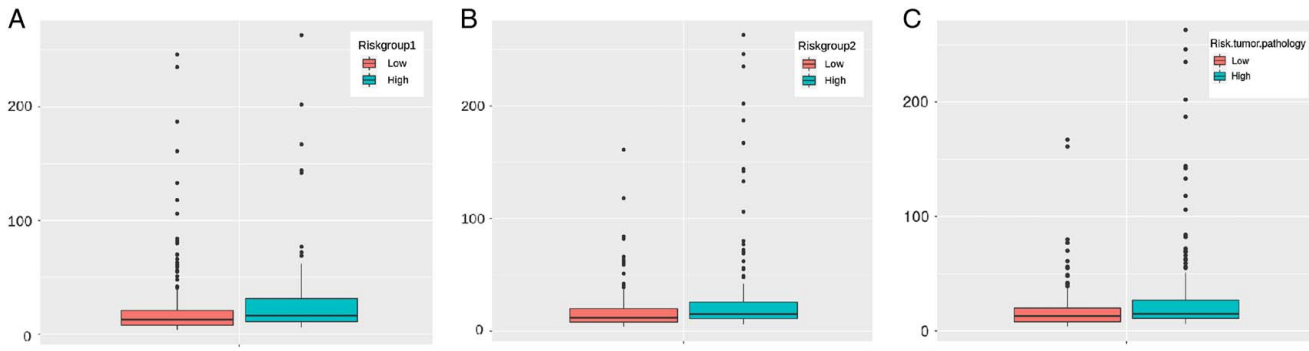

FIGURE 3. Relationship between the histological risk score in stage 1 and the interstage interval (A). Relationship between the histological risk score in the stage 2 and the interstage interval (B). Influence between the tumor pathology risk score and the interstage interval (C).

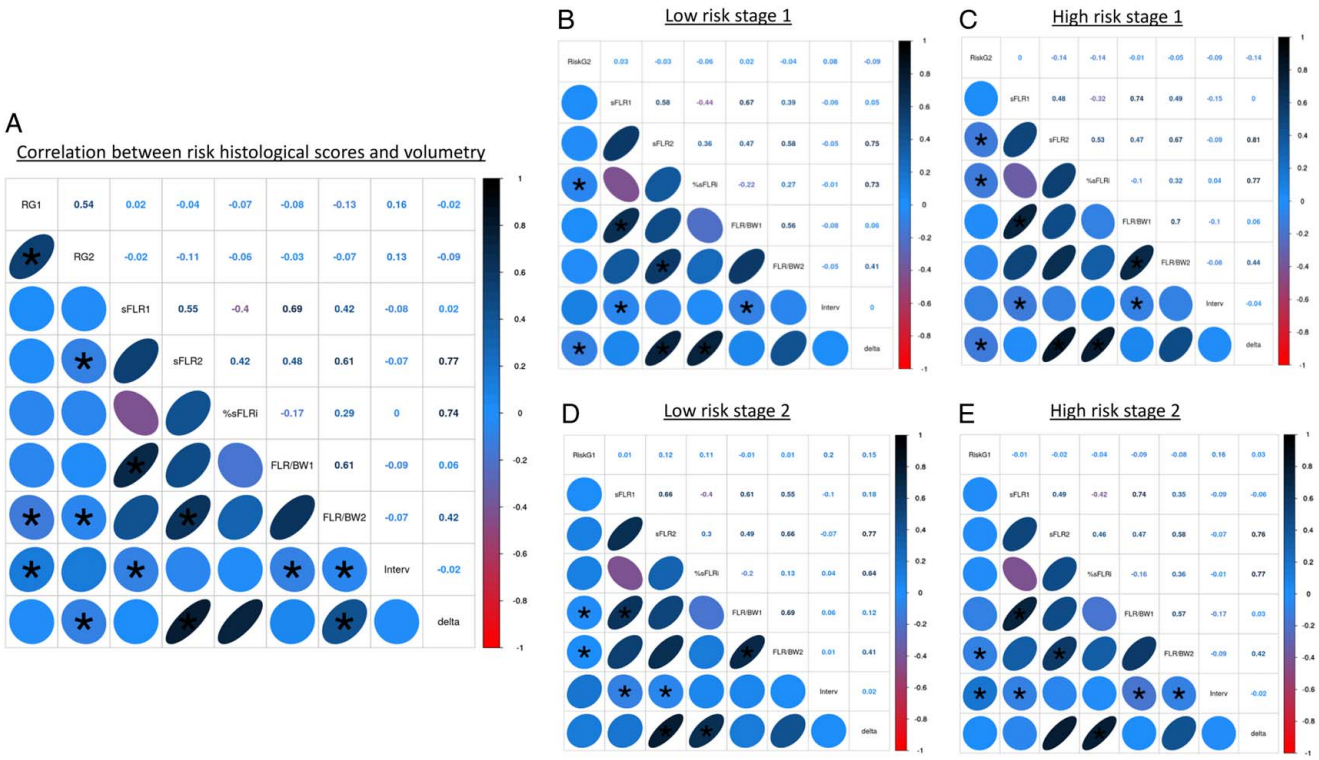

FIGURE 4. Scatter plots with correlations between the stage 1 and 2 risk histological scores and volumetric data (A). Separate analysis according to low and high histological groups in stage 1 (B and C). Separate analysis according to low and high histological groups in stage 2 (D and E).

proportion of patients with a high histological risk score of stage 1 than those with a low tumor pathological risk score (26.26% vs 11.36%, respectively) ($P < 0.01$). Similarly, the group of patients with a high tumor pathological risk score was associated with a higher proportion of patients with a high histological risk score stage 2 than those with a low tumor pathological risk score (39.9 vs 23.23%, respectively) ($P < 0.01$). In addition, similar to what happened with the histological risk score, patients with a low pathological tumor risk needed a median of 13 days (IQR, 8–20) to reach the second stage compared to 15 days (IQR, 11–27) for patients with a high pathological risk tumor ($P < 0.01$) (Fig. 3C).

DISCUSSION

This multicenter analysis of the histological alterations in ALPPS revealed that several changes in the liver parenchyma had a significant negative effect on the time required to reach a sufficient FLR. A single alteration does not affect regeneration or short-term results, but the presence of 2 or more alterations was associated with worse results, affecting the time interval necessary to safely reach the second stage. Furthermore, the interstage interval was affected by the presence of poor prognostic tumor factors in specimens of liver metastases.

The regenerating liver demonstrates a histology that is practically indistinguishable from the original features through lobular reorganization.[26,27] This process of liver regeneration can take a few days, several weeks, or even months and is associated with a series of changes in the architecture of the liver that depend on multiple factors and vary among patients. Although much remains to be learned about this phenomenon, it has been established that there are a series of growth patterns that depend on the characteristics of the patient (age and weight), growth stimuli (liver resection, ischemia-reperfusion, embolization or portal ligation, ALPPS), or liver characteristics (fibrosis, cirrhosis, steatosis, necrosis, or chemotherapy-related changes).

Liver growth-induction techniques used to achieve a sufficient liver volume before hepatectomy produce a series of changes in the liver parenchyma. Different authors have analyzed the histological changes that occur in the FLR and the deportalized lobe. In experimental models of ALPPS, the main histological features of the atrophic lobes are periportal congestion, sinusoid dilation, areas of ischemia, and necrotic or apoptotic hepatocytes.[22] In contrast, Shi et al,[24] found an FLR on postoperative day 7 in hepatocyte mitosis and hepatic sinusoidal injury, characterized by sinusoidal dilatation, microvesicular steatosis, hepatocellular atrophy, and centrilobular or perisinusoidal fibrosis. In humans, in the analyzed specimens obtained from 8 patients treated with ALPPS and from 14 patients treated with hepatectomy after PVE,[10] the areas of FLR hepatocyte brightness, sinusoidal narrowing and hepatocyte cell density were observed more frequently in the ALPPS group than in the PVE group, while the hepatocyte size was smaller in the ALPPS group. In the present study, we observed an increase in all histological risk factors analyzed, except for macrosteatosis. Furthermore, we found that rapid hypertrophy associated with ALPPS induced a greater increase in hemorrhagic phenomena, regenerative nodular hyperplasia, necrosis, and inflammation in the liver parenchyma. Histological alterations at stage 2 can only be interpreted in synopsis, with stage 1 histology serving as the baseline. Standardized regenerative changes, excluding pathological ones, are difficult to differentiate in this patient

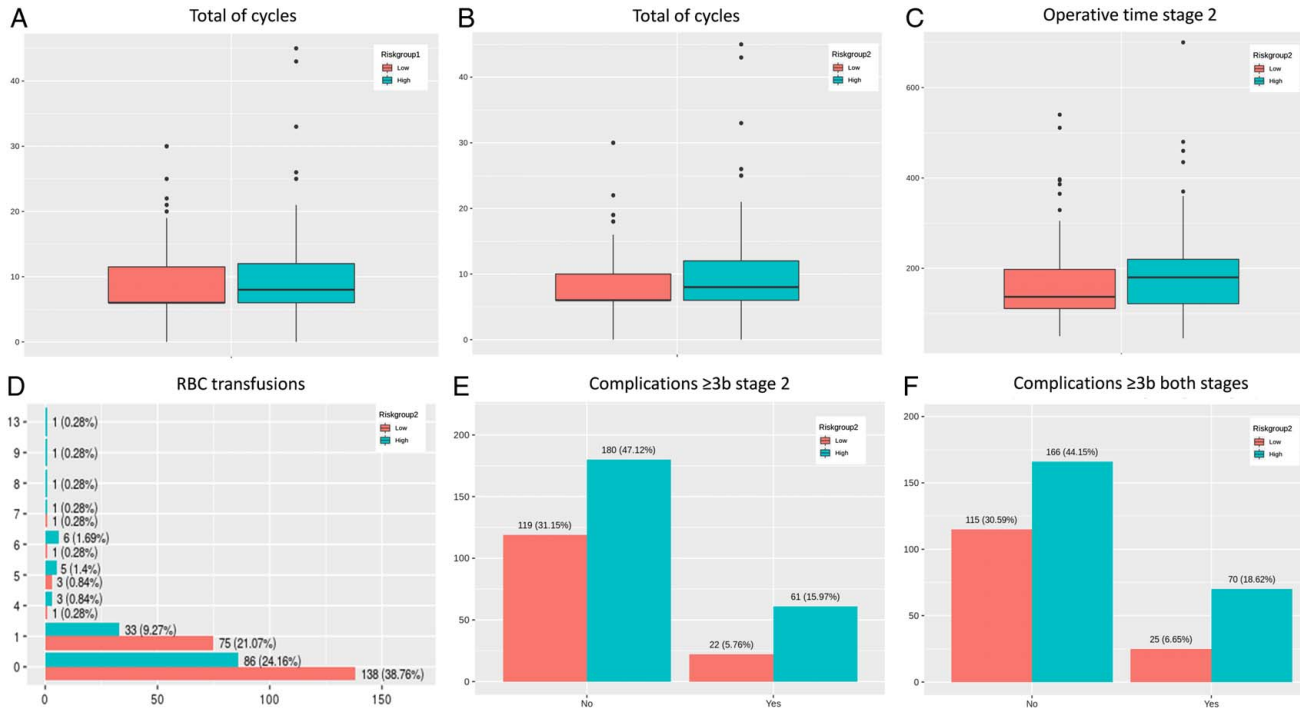

FIGURE 5. Relationship between the histological risk score in stage 1 and the number of chemotherapy cycles (A). Relationship between the histological risk score in stage 2 and the number of chemotherapy cycles (B). Relationship between the histological risk score in stage 2 and the operative time (C). Relationship between the histological risk score in the stage 2 and need of red blood transfusions (D). Relationship between the histological risk score in stage 2 and postoperative major complications after stage 2 (E). Relationship between the histological risk score in the stage 2 and postoperative major complications after both stages (D).

population. Therefore, we interpreted features such as increased hemorrhage and necrosis as a result of an impaired regeneration process due to pre-existing parenchymal damage.

One of the challenging questions that has been debated in relation to rapid hypertrophy is whether volumetric growth reflects the real proliferation of hepatocytes, or whether this increase in volume is related to venous congestion due to blood flow, interstitial edema, and associated intracellular steatosis. In an initial experience with 16 patients, there was efficient hepatocytes proliferation instead of an increase in edema and steatosis, and the authors speculated that the increase in water and steatosis could have an effect on mortality.[12] In our study, both edema and steatosis showed the least increase. Therefore, we conclude that hepatocyte proliferation occurs and that edema and steatosis do not depend on them.

The presence of steatosis is related to a poor worse capacity for liver graft regeneration or function.[18,28] In this study, the histological risk score was related to the time required to safely reach the second stage. Patients with a high-risk score were characterized by more severe alterations and, presumably, lower regenerative capacity. Both macrosteatosis and microsteatosis were the most frequent histological alterations found in the baseline biopsy, but no direct relationship was found with greater complications, liver failure, or longer regeneration time because steatosis higher than 66% in the samples analyzed was very rare. Transient regeneration-associated steatosis (TRAS) occurs in every regenerating liver and is recognized as an essential component of successful recovery following tissue loss. Generally, TRAS is a mandatory component for successful regeneration, with fat as the main regenerative fuel during periods of low liver capacity.[29] Thus, although steatosis plays a role in liver regeneration and proper functioning of the hypertrophied liver, it is not sufficient and depends on its interaction with other histological alterations to affect the regeneration and postoperative results in ALPPS.

In this study, patients exposed to more cycles of chemotherapy presented with greater histological damage, which was significantly associated with the presence of a high-risk score in stages 1 and 2. In general, patients with a greater number of lesions receive neoadjuvant chemotherapy and undergo surgery based on a partial response to chemotherapy. Chemotherapy produces a series of changes in the liver parenchyma, which have been associated with a higher rate of complications following liver resection, without an associated increase in mortality. With chemotherapy, a high percentage of patients develop hepatic steatosis, indicating an altered lipid metabolism through the synthesis of lipoproteins in hepatocytes. It is also frequent, especially in relation to oxaliplatin, in a sinusoidal lesion that varies from sinusoidal dilation to sinusoidal hepatic obstruction syndrome and can progress to regenerative nodular hyperplasia. It has also been reported that patients treated with preoperative chemotherapy present with hepatocyte atrophy, hepatocyte necrosis, and an independent effect of fibrosis stage.[30] Furthermore, these alterations are more aggressively related to the number of cycles received, especially when more than 6 cycles are received, and can cause irreversible hepatocellular damage through the recruitment of inflammatory cells.

Initially, the morbidity in the published series and the ALPPS registry was very high, ranging from 53% to 100%, with a severe complication rate exceeding 30%.[31] High-volume blood loss increases the likelihood of postoperative complications, whereas blood transfusions exert immunosuppressive effects that can predispose patients to infectious complications and impair liver regeneration.[32] A decade later, after an improvement in the selection of patients and refinement of the technique, it was possible to significantly improve both intra- and postoperative results.[33,34] Histological alterations may have an underestimated impact on the continued improvement of the results of the ALPPS technique. In our series, the histological risk score had an impact on the perioperative results. Patients with high histological risk scores had a greater need for blood transfusions, a higher percentage of severe complications, and a longer surgical time. Therefore, prior histological examination of the liver, both before stages 1 and 2, could identify patients at a higher risk of complications or slower hypertrophy. Based on their experience with patients who presented worse results in their series, some groups recommended performing liver biopsy before proceeding to liver partition to avoid performing an ALPPS procedure in cases of fibrosis or cirrhosis.[35]

This study has some limitations. We did not evaluate the weight of each item because we do not know exactly which of them plays a more important role. The score allowed us to assess whether the sum of the number of histological or tumor alterations present and their severity had a representative impact on postoperative results and liver regeneration. In short, the score is not representative of the weight of each of the items studied but of the number of alterations and their severity.

In summary, the rapid hypertrophy caused by ALPPS was associated with an increased histological alteration in the liver parenchyma. Although steatosis is the most frequent histological alteration in the liver, other histological phenomena associated with rapid regeneration, such as inflammation, necrosis, hemorrhage, and regeneration of nodules, must be considered. Grouping patients with a greater number of tumor risk factors on histology also affects the interstage interval. It seems that the more histological alterations present and the higher the number of poor prognostic factors in the tumor histology, the longer the time to reach the second stage. Studying liver histology at different surgical times can provide a holistic picture of the structure and function of the liver to guide surgical resection and other therapeutic decisions.

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REFERENCES

1. Clavien PA, Petrowsky H, DeOliveira ML, et al. Strategies for safer liver surgery and partial liver transplantation. *N Engl J Med*. 2007;356:1545–1559.
2. Del Basso C, Gaillard M, Lainas P, et al. Current strategies to induce liver remnant hypertrophy before major liver resection. *World J Hepatol*. 2021;13:1629–1641.
3. Lang H, de Santibañes E, Schlitt HJ, et al. 10th Anniversary of ALPPS—lessons learned and quo vadis. *Ann Surg*. 2019;269:114–119.
4. Schnitzbauer AA, Lang SA, Goessmann H, et al. Right portal vein ligation combined with in situ splitting induces rapid left lateral liver lobe hypertrophy enabling 2-staged extended right hepatic resection in small-for-size settings. *Ann Surg*. 2012;255:405–414.
5. Zhang L, Yang Z, Zhang S, et al. Conventional two-stage hepatectomy or associating liver partitioning and portal vein ligation for staged hepatectomy for colorectal liver metastases? A systematic review and meta-analysis. *Front Oncol*. 2020;10:1391.
6. Petrowsky H, Linecker M, Raptis DA, et al. First long-term oncologic results of the ALPPS procedure in a large cohort of patients with colorectal liver metastases. *Ann Surg*. 2020;272:793–800.
7. Robles-Campos R, Brusadin R, López-Conesa A, et al. Long-term outcome after conventional two-stage hepatectomy versus tourniquet-ALPPS in colorectal liver metastases: a propensity score matching analysis. *World J Surg*. 2019;43:2281–2289.
8. Hoffmann K, Nagel AJ, Tanabe K, et al. Markers of liver regeneration-the role of growth factors and cytokines: a systematic review. *BMC Surg*. 2020;20:31.
9. Robles-Campos R, Navarro-Barrios Á, Martínez-Caceres C, et al. The contribution of the deportalized lobe to liver regeneration in tourniquet-ALPPS. *Ann Surg*. 2020;271:e94–e96.
10. Matsuo K, Murakami T, Kawaguchi D, et al. Histologic features after surgery associating liver partition and portal vein ligation for staged hepatectomy versus those after hepatectomy with portal vein embolization. *Surgery*. 2016;159:1289–1298.
11. Matsuo K, Hiroshima Y, Yamazaki K, et al. Immaturity of bile canalicular-ductule networks in the future liver remnant while associating liver partition and portal vein occlusion for staged hepatectomy (ALPPS). *Ann Surg Oncol*. 2017;24:2456–2464.
12. Eshmuminov D, Tschuor C, Raptis DA, et al. Rapid liver volume increase induced by associating liver partition with portal vein ligation for staged hepatectomy (ALPPS): is it edema, steatosis, or true proliferation? *Surgery*. 2017;161:1549–1552.
13. Wakabayashi T, Tanaka K, Shiozawa T, et al. Liver regeneration after performing associating liver partition and portal vein occlusion for staged hepatectomy (ALPPS) is histologically similar to that occurring after liver transplantation using a small-for-size graft. *Surg Today*. 2021;51:374–383.
14. Schadde E, Tsatsaris C, Swiderska-Syn M, et al. Hypoxia of the growing liver accelerates regeneration. *Surgery*. 2017;161:666–679.
15. Moris D, Vernadakis S, Papalampros A, et al. Mechanistic insights of rapid liver regeneration after associating liver partition and portal vein ligation for stage hepatectomy. *World J Gastroenterol*. 2016;22:7613–7624.
16. Taki-Eldin A, Zhou L, Xie HY, et al. Liver regeneration after liver transplantation. *Eur Surg Res*. 2012;48:139–153.
17. Yang X, Yang C, Qiu Y, et al. A preliminary study of associating liver partition and portal vein ligation for staged hepatectomy in a rat model of liver cirrhosis. *Exp Ther Med*. 2019;18:1203–1211.
18. Selzner M, Clavien PA. Failure of regeneration of the steatotic rat liver: disruption at two different levels in the regeneration pathway. *Hepatology*. 2000;31:35–42.
19. Shindoh J, Tzeng CW, Aloia TA, et al. Optimal future liver remnant in patients treated with extensive preoperative chemotherapy for colorectal liver metastases. *Ann Surg Oncol*. 2013;20:2493–2500.
20. López-López V, Robles-Campos R, Brusadin R, et al. Tourniquet-ALPPS is a promising treatment for very large hepatocellular carcinoma and intrahepatic cholangiocarcinoma. *Oncotarget*. 2018;9:28267–28280.
21. Lopez-Lopez V, Robles-Campos R, Brusadin R, et al. ALPPS for hepatocarcinoma under cirrhosis: A feasible alternative to portal vein embolization. *Ann Transl Med*. 2019;7:691.
22. García-Pérez R, Revilla-Nuin B, Martínez CM, et al. Associated liver partition and portal vein ligation (ALPPS) vs selective portal vein ligation (PVL) for staged hepatectomy in a rat model. Similar regenerative response? *PLOS ONE*. 2015;10:e0144096.
23. Schlegel A, Lesurtel M, Melloul E, et al. ALPPS: From human to mice highlighting accelerated and novel mechanisms of liver regeneration. *Ann Surg*. 2014;260:839–846; discussion 46–47.
24. Shi JH, Hammarström C, Grzyb K, et al. Experimental evaluation of liver regeneration patterns and liver function following ALPPS. *BJS Open*. 2017;1:84–96.
25. Kawaguchi D, Hiroshima Y, Kikuchi Y, et al. Associating liver partition and portal vein occlusion, including venous congestion, induction in rats. *Anticancer Res*. 2017;37:2919–2925.
26. Michalopoulos GK. Hepatostat: Liver regeneration and normal liver tissue maintenance. *Hepatology*. 2017;65:1384–1392.
27. Michalopoulos GK, Bhushan B. Liver regeneration: Biological and pathological mechanisms and implications. *Nat Rev Gastroenterol Hepatol*. 2021;18:40–55.

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28. Marcos A, Fisher RA, Ham JM, et al. Liver regeneration and function in donor and recipient after right lobe adult to adult living donor liver transplantation. *Transplantation*. 2000;69:1375–1379.
29. Kachaylo E, Tschuor C, Calo N, et al. PTEN down-regulation promotes β-oxidation to fuel hypertrophic liver growth after hepatectomy in mice. *Hepatology*. 2017;66:908–921.
30. Kandutsch S, Klinger M, Hacker S, et al. Patterns of hepatotoxicity after chemotherapy for colorectal cancer liver metastases. *Eur J Surg Oncol*. 2008;34:1231–1236.
31. Schadde E, Raptis DA, Schnitzbauer AA, et al. Prediction of mortality after ALPPS Stage-1: an analysis of 320 patients from the international ALPPS registry. *Ann Surg*. 2015;262:780–785; discussion 5–6.
32. Bennett S, Baker LK, Martel G, et al. The impact of perioperative red blood cell transfusions in patients undergoing liver resection: a systematic review. *HPB (Oxford)*. 2017;19:321–330.
33. Linecker M, Björnsson B, Stavrou GA, et al. Risk adjustment in ALPPS is associated with a dramatic decrease in early mortality and morbidity. *Ann Surg*. 2017;266:779–786.
34. Robles-Campos R, Brusadín R, López-López V, et al. A new surgical technique variant of partial ALPPS (tourniquet partial-ALPPS). *Ann Surg*. 2021;273:e22–e24.
35. Vivarelli M, Vincenzi P, Montalti R, et al. ALPPS procedure for extended liver resections: a single centre experience and a systematic review. *PLoS One*. 2015;10:e0144019.