Sex Disparities in Outcomes Following Major Liver Surgery

New Powers of Estrogen?

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Aim: To explore potential sex differences in outcomes and regenerative parameters post major hepatectomies.

Background: Although controversial, sex differences in liver regeneration have been reported for animals. Whether sex disparity exists in human liver regeneration is unknown.

Methods: Data from consecutive hepatectomy patients (55 females, 67 males) and from the international ALPPS (Associating-Liver-Partition-and-Portal-vein-ligation-for-Staged-hepatectomy, a two stage hepatectomy) registry (449 females, 729 males) were analyzed. Endpoints were severe morbidity ($\geq$3b Clavien-Dindo grades), Model for End-stage Liver Disease (MELD) scores, and ALPPS interstage intervals. For validation and mechanistic insight, female-male ALPSS mouse models were established. *t*, χ^2, or Mann-Whitney tests were used for comparisons. Univariate/multivariate analyses were performed with sensitivity inclusion.

Results: Following major hepatectomy (Hx), males had more severe complications ($P = 0.03$) and higher liver dysfunction (MELD) $P = 0.0001$) than females. Multivariate analysis established male sex as a predictor of complications after ALPPS stage 1 (odds ratio = 1.78; 95% confidence interval: 1.126–2.89; $P = 0.01$), and of enhanced liver dysfunction after stage 2 (odds ratio = 1.93; 95% confidence interval: 1.01–3.69; $P = 0.045$). Female patients displayed shorter interstage intervals (< 2 weeks, 64% females versus 56% males, $P = 0.01$), however, not in postmenopausal subgroups. In mice, females regenerated faster than males after ALPPS stage 1, an effect that was lost upon estrogen antagonism.

Conclusions: Poorer outcomes after major surgery in males and shorter ALPPS interstage intervals in females not necessarily suggest a superior regenerative capacity of female liver. The loss of interstage advantages in postmenopausal women and the mouse experiments point to estrogen as the driver behind these sex disparities. Estrogen's benefits call for an assessment in postmenopausal women, and perhaps men, undergoing major liver surgery.

Keywords: ALPPS, ALPPS registry, human and mice, liver regeneration, liver surgery, major hepatectomy, Omegaven, 2-stage hepatectomy, translational science

(*Ann Surg* 2022;276:875–881)

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Supported by project grant 310030_189262 and an MD/PhD fellowship (323530_199390/1) from the Swiss National Science Foundation.

This study is supported by the LGID (Liver and Gastrointestinal Disease) Foundation.

The authors report no conflicts of interest.

Supplemental Digital Content is available for this article. Direct URL citations appear in the printed text and are provided in the HTML and PDF versions of this article on the journal's website, www.annalsofsurgery.com.

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ISSN: 0003-4932/22/27605-0875

DOI: 10.1097/SLA.0000000000005635

The ability of the liver to regenerate has enabled the extensive surgical removal of hepatic tumors with curative intent. Successful regeneration is dependent on a sufficient functional volume of the liver remnant and is prerequisite for a swift recovery of patients. Indeed, an inverse relationship between the regenerative speed and the severity of postoperative outcomes exists: if regeneration is stalled, post-hepatectomy liver failure will develop, the most common cause of death due to liver surgery.[1–4]

Intriguingly, inferior outcomes after liver transplantation in female-to-male donations have been reported.[5] One explanation for these observations may be sex-dependent differences in recovery from liver injury. While sexual dimorphism is a common phenomenon observed for human diseases,[6–9] sex-related differences in the regenerative capacity of the human liver have not been explored.

Experimental evidence suggests regenerative sex disparities in animals. In rodents, however, evidence is controversial, with some reporting faster regeneration in males,[10–12] while others found females to have a superior regenerative capacity following Hx.[13–15] In contrast, a consistent observation

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across studies is the ability of estrogen to promote hepatocyte proliferation.[13,15–19]

Given the above role of estrogen, a plausible assumption is that the regenerative capacity of females is superior relative to males in humans. To pursue this hypothesis, we took advantage of patient data retrieved (i) from a trial on Hx patients, and (ii) from the international ALPPS registry. While Hx is an established inducer of regeneration, ALPPS surgery (Associating-Liver-Partition-and-Portal-vein-ligation-for-Staged-hepatectomy), a modified 2-stage Hx is intriguing, as it triggers markedly accelerated regeneration. Patient data on postoperative outcomes, liver function, survival, and and where available, volumetry was separately analyzed for males and females. A mouse model of female versus male ALPPS surgery was established to validate patient findings and obtain mechanistic insight.

METHODS

Patients

Patient data was retrospectively retrieved from a subgroup of patients participating in the Omegaven trial (NCT01884948). Subgroup was defined by the complete presence of all data collected from patients that were subjected to minor or major Hx in Zurich during 2013 to 2018. Ethical approval was waived by the local Ethics Committee of Zurich (Nr. 2010-0038), notified by Swissmedic (2012DR3215), with the study protocol conforming to the 1975 Declaration of Helsinki.

From the international ALPPS registry, all adult cases from the 2012 to 2020 period period were included, safe for those with hepatoblastomas or unknown sex.

All patients provided written informed consent.

The primary endpoint was morbidity (Clavien-Dindo Classification)[20] and post-Hx liver failure risk reflected by MELD scores (bilirubin, international normalized ratio, creatinine) on day 5 postoperation/surgery [postoperative day (POD) 5]. Secondary analysis were performed for liver volume differences and mortality where available.

Animal Experiments

All animal experiments were performed in accordance with Swiss Federal Animal Regulations and approved by the veterinary office Zurich. Female and male C57BL/6 mice between between the age of 10 to 12 weeks were obtained from Harlan NL (Horst, The Netherlands) and used for all experiments. ALPPS stage 1 surgery was performed as previously described for male mice only.[21,22] Surgery on female mice required special attention due to the high variability in vascular structure. Tamoxifen (Sigma Aldrich, Buchs, Switzerland; T9262-1G, 4 mg/kg)[16] in methanol/saline (1:100) was intraperitoneally injected 24 hours before and at surgery. Liver weight gain was assessed by the future liver remnant-to-body-weight-ratio LW/BW. Proliferative activity was assessed by manually counting Ki-67-positive hepatocytes in 5 random high-power-fields at ×20 magnification in a blinded manner.

Statistical Analyses

The Shapiro-Wilk test was used to test for normality. Group comparison was performed with Student t test (mean ± SD), χ^2 (dichotomic endpoints), or Mann-Whitney/Wilcoxon test [non-normal distribution, median ± interquartile range (IQR)]. Logistic regression models were used for multivariable analysis with major complications being the dependent variable and a linear regression model for MELD points. The percentage of missing values across the variables was between 5% and 36.2% in random patterns. To handle missing data, 5 multiply imputed datasets with 10 iterations each using predictive mean matching were created by chained equation (mice package, v. 3.9.0). A multivariable logistic regression model was applied to the imputed datasets and final estimates were obtained by averaging the 5 estimates via Rubin's rules.[23] A sensitivity analysis including complete cases (40.1%) was done. The level for statistical significance was set at <0.05. Statistical analysis was performed with R Core team[24] and Prism 8.0. (GraphPad).

RESULTS

Female Patients Recover Better From Major Hepatectomy Than Males

The cohort consisted of 122 patients including 55 female and 67 male patients with a median age of 57 (IQR: 54–71.5) and 63 (IQR: 57–71), respectively. Postoperative complications, liver function, and survival were available for analyses and compared between the sexes (baseline comparisons in Table S1, Supplemental Digital Content 1, http://links.lww.com/SLA/E112).

In the overall cohort, male sex was associated with a higher postoperative morbidity compared with females as reflected by the frequency of major (= 3b) complications after both minor and major Hx (15.4% males vs 9.1% females, $P = 0.037$). If considering major Hx only, the sex difference in complications was further exaggerated (51.6% males vs 25.9% females, $P = 0.030$) but vanished for minor Hx only ($P = 0.96$).

MELD scores after major Hx were lower for females than males during all PODs (Fig. 1), including at times when liver function usually has recovered postsurgery [POD7: 7.4 (IQR: 6.4–7.4) for females vs 9.4 (IQR: 7.4–15.3) for males, $P = 0.0001$]. No significant changes between the sexes in MELD scores were observed after minor Hx (Fig. 2). Finally, females displayed a lower 90-day mortality, with all deaths occurring in males (0% female vs 7.5% male, $P = 0.1$).

Females Recover Better From ALPPS Surgery Than Males

The cohort of the ALPPS registry comprised 449 female and 729 male patients from 40 ALPPS centers over a 10-year period with a median age of 59 (IQR: 49.5–66) and 61 (IQR: 53–68), respectively (baseline comparisons in Table S2, Supplemental Digital Content 1, http://links.lww.com/SLA/E112). Available data included postoperative complications, liver function, and liver volume for stages 1 and 2.

Across all patients, males had more severe complications than females (9.5% vs 5.7%, $P = 0.027$) after stage 1. Following stage 2, the complications were similar between male and female patients (16.5% vs 15.7%, $P = 0.09$). Overall, liver function as estimated via MELD scores was superior for females after both stages 1 and 2 (Fig. 2). Survival post stage 2 was similar for both sexes, however, available data was limited [females: 40 months (IQR: 27.6–56.7) vs males: 36.8 months (IQR: 30.3–48.7), $P = 0.54$, with n = 591 missing observations, Fig. S2, Supplemental Digital Content 1, http://links.lww.com/SLA/E112].

After multiple imputation, the multivariable analysis demonstrated male sex to be a predictor of major complications [odds ratio (OR) = 1.66, 95% confidence interval (CI): 1.03–2.7, $P = 0.037$] and elevated POD5-MELD scores (OR = 2.10, 95% CI: 1.11–3.98, $P = 0.0225$) after stage 1. Complete case sensitivity analysis for stage 1 confirmed male sex as a predictor of severe

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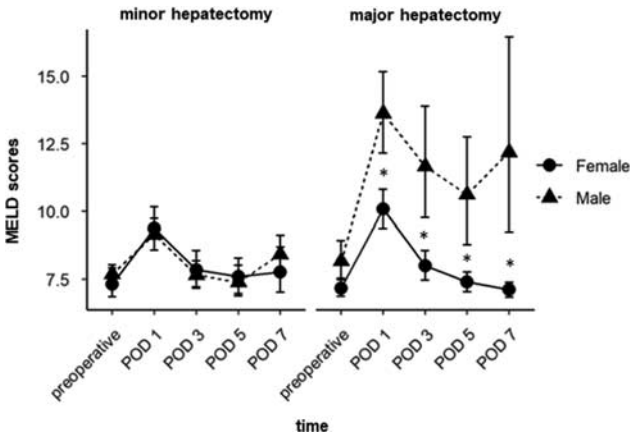

FIGURE 1. MELD scores from female and male Hx patients. MELD scores were similar for the sexes post minor Hx but lower for females compared with males after major Hx Median ± IQR is shown. MELD baseline is set at 6.4 corresponding to the corrected MELD score minimum. Mann-Whitney *U* test; $*P < 0.05$.

complications and of elevated POD5-MELD scores on both univariate (*complications*: OR = 1.78, 95% CI: 1.126–2.89, $P = 0.0163$; *MELD*: OR = 1.93, 95% CI: 1.01–3.69, $P = 0.045$) and multivariable analyses (*complications*: OR = 1.71, 95% CI: 1.04–2.92, $P = 0.0386$; *MELD*: OR = 2.06, 95% CI: 1.08–3.95, $P = 0.028$; adjusted for age, diabetes, body mass index, chemotherapy). Similar results were obtained for POD5-MELD after stage 2 in the univariate (OR = 2.79, 95% CI: 1.3–5.9, $P = 0.00827$) and multivariable analyses (OR = 2.8, 95% CI: 1.30–6.1, $P = 0.008829$).

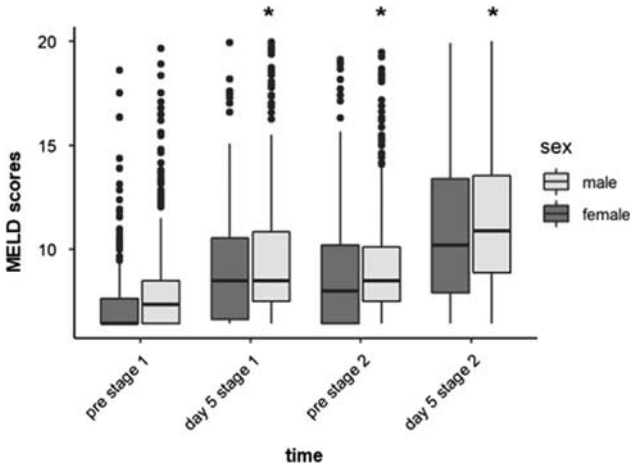

FIGURE 2. MELD scores from female and male ALPPS registry patients. MELD scores for females were significantly lower than male scores at pre-stage 1 ($P = 5.032e{-}06$), POD5 stage 1 ($P = 0.025$), pre-stage 2 ($P = 0.023$), POD5 stage 2 ($P = 0.002$). Median ± IQR is shown. MELD baseline is set at 6.4 corresponding to the corrected MELD score minimum. Mann-Whitney *U* test; $*P < 0.05$.

Similar Liver Volume Gain But Shorter Interstage Time for Females After ALPPS

No significant differences were observed for post stage 1 liver volume before stage 2 when corrected for baseline values (Fig. S1, Supplemental Digital Content 1, http://links.lww.com/SLA/E112), reflecting that all patients eligible for stage 2 have gained the predefined volume required to proceed with resection.

The interstage interval—the time interval after stage 1 until the required liver volume to enable stage 2 is gained – was shorter for females than for males, with a significant enrichment of females with times <2 weeks (Fig. 3, 64% females vs 56% males, $P = 0.01$). Intriguingly, this difference was gradually lost when comparing older (ie, postmenopausal) subgroups (above 60 years, $P = 0.048$; above 65 years, $P = 0.20$; above 70 years, $P = 0.34$).

Female Mice Regenerate Faster Than Males After ALPPS Stage 1, an Effect Related to Estrogen

To assess whether the observed human sex differences in the recovery from ALPPS surgery can be recapitulated in other species, we established ALPPS surgery for female mice and performed the operation separately in groups of females and males.

Our previous research has demonstrated that in mice liver weight gain is most pronounced at 24 hours after ALPPS stage 1 surgery.[21,22] Future liver remnant (FLR) weight gain (LW/BW) assessment and ALPPS surgery were performed akin to human surgery. At 24 hours post ALPPS stage 1, LW/BW was markedly elevated in female relative to male mice [median: females = 1.4 (95% CI: 1.39–1.77) vs males = 1.0 (95% CI: 0.78–1.14), n = 15/group, $P = 0.002$, Fig. 4A, Fig. S3, Supplemental Digital Content 1, http://links.lww.com/SLA/E112]. Consistent with the increased LW/BW, female liver displayed increased Ki-67 counts [marking proliferating hepatocytes; median: females = 11 (95% CI: 9.25–12.5) vs males = 2 (95% CI: 1.0–3.75), $P = 0.023$, Fig. S4, Supplemental Digital Content 1, http://links.lww.com/SLA/E112]. Therefore, female mice regenerate faster than males after ALPPS stage 1 and accordingly would need less time to reach the desired volume enabling stage 2 resection.

To assess whether the superior regenerative capacity of female mice may relate to estrogen abundance, we repeated ALPPS stage 1 surgery in the presence or absence of the estrogen receptor (ER) inhibitor tamoxifen. Treatment reduced median LW/BW in females from 1.39 (95% CI: 1.35–1.39) in vehicle controls to 0.67 (95% CI: 0.59–0.74) in the tamoxifen group (n = 5/group, $P = 0.032$, Fig. 4B). In male, tamoxifen had little effect on LW/BW [0.92 (95% CI: 0.82–1.04) in vehicle vs 0.95 (95% CI: 0.82–1.03) in tamoxifen groups, $P = 0.67$, Fig. 4C).

DISCUSSION

In our study on patients undergoing major liver surgery, outcomes in terms of complications and postoperative liver function were superior for females than for males after both Hx and ALPPS surgery. Importantly, ALPPS interstage intervals were shorter for female than male patients. As stage 2 is only performed when a predefined liver volume and function is gained, interstage intervals reflect the functional recovery speed after stage 1. When comparing older subgroups of females/males, the sex differences in interstage intervals were lost, suggesting hormonal contribution. In mice, females regenerated faster than males after ALPPS stage 1, an effect dependent on estrogen. Therefore, a superior regenerative capacity in females is driven by estrogen and plausibly explains the shorter interstage

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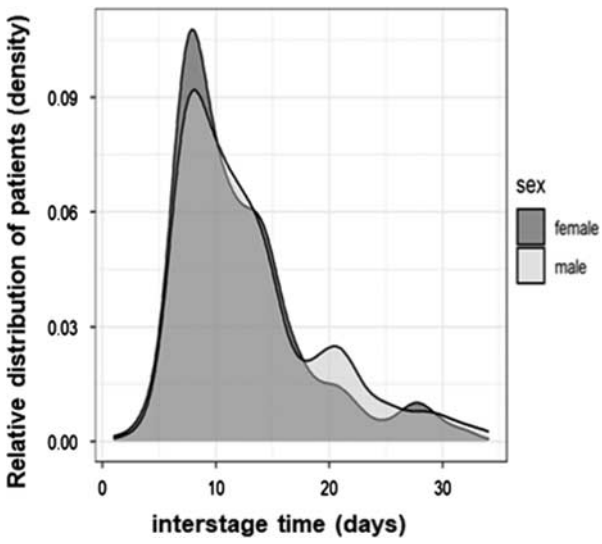

FIGURE 3. Sex-specific and age-specific distribution of interstage interval times ALPPS patients. Sex-specific distribution of interstage times below 70 years: females are enriched among patients with shorter (64% females vs 56% males, P=0.01). interstage intervals (the time period after stage 1 until a predefined volume is reached to enable stage 2 resection).

intervals as well as better outcomes in females post major resection.

This is the first study dedicated to sex-specific outcomes and regenerative parameters in settings of major liver resection. The data from major Hx uniformly indicated less complications and better liver function (MELD) for females, fully supported by their absent 90-day mortality. Unlike major Hx, minor Hx triggers only a limited regeneration response that seems to involve little hepatocyte proliferation.[11] The lack of sex differences after minor Hx, therefore, supports the view that better outcomes in females after major resection may relate to a superior proliferative capacity.

ALPPS stage 1 surgery elicits regeneration with unprecedented speed.[25,26] Again, females presented with less complications after stage 1 and better liver function (MELD) than males after either stage, as confirmed in the multivariate analysis and strengthened by the sensitivity analysis. The reason for the discrepancy between complications and MELD scores post stage 2 is unclear, however, MELD scores do not necessarily correlate with complications or perioperative morbidity after resection/surgery.[27] Moreover, only patients that have reached sufficient functional volume for a resection proceed with stage 2 (Fig. S1, Supplemental Digital Content 1, http://links.lww.com/SLA/E112); patients with severe complications post stage 1 (enriched with males) are thus less likely to undergo stage 2 surgery, explaining the lack of sex differences post stage 2 – and perhaps also the lack of survival differences post stage 2. Survival data additionally were weakened by the high percentage of missing data (Fig. S2, Supplemental Digital Content 1, http://links.lww.com/SLA/E112).

Females after stage 1 reached the liver volume desired for stage 2 faster than males. As the pre-stage 2 volume requirements are same for sex (Fig. S1, Supplemental Digital Content 1, http://links.lww.com/SLA/E112), interstage intervals give a direct estimation of the regenerative speed after

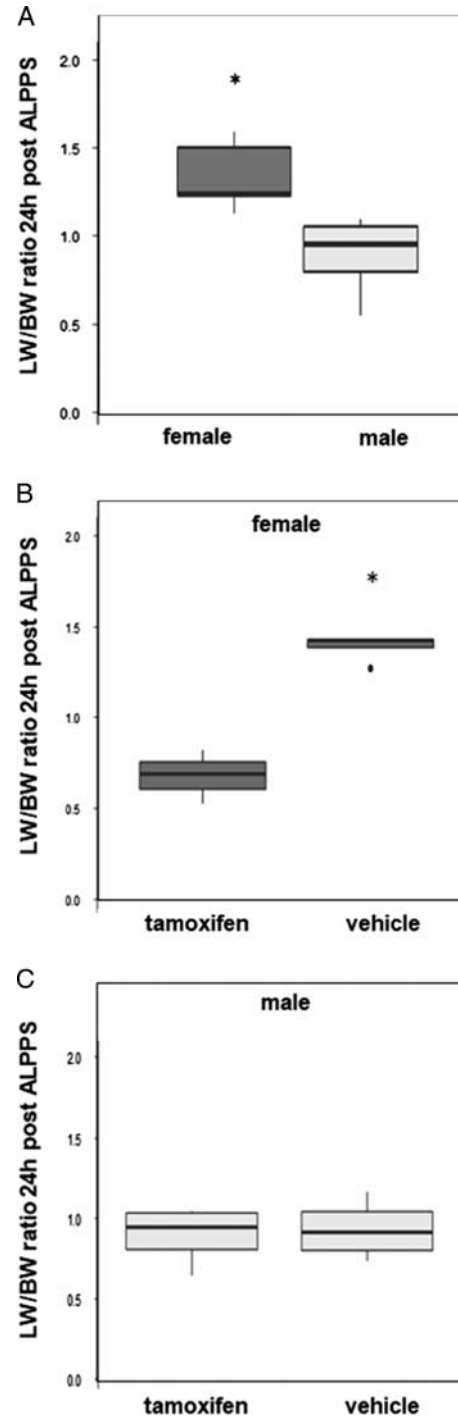

FIGURE 4. Liver weight gain 24 hours after ALPPS surgery in mice. (A) LW/BW in female and male mice. N = 6/group, $P = 0.0021$. (B) LW/BW in female mice treated or not with tamoxifen. N = 5/group, $P = 0.032$. (C) LW/BW in male mice treated or not with tamoxifen. N = 5/group, $P = 0.64$. For all, median LW/BW ± IQR is shown.

stage 1. Thus, the unique feature of ALPPS-induced regenerative acceleration reveals superior liver growth in female patients. Superiority, however, was limited to the age group of

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premenopausal women and gradually lost in female-male comparisons of older age. The importance of the female hormonal status was validated in our female-male ALPPS model, where the regenerative advantage of females was strongly dependent on estrogen signaling.

While our findings in mice are the first to document faster regeneration in females following ALPPS, some reports have found superior liver regeneration in rodent females post-Hx.[15,17] ERα (estrogen receptor alpha) on hepatocytes is thought to mediate the proliferative activity of estrogen.[15,18,28–30] In rats, ERα is induced on hepatocytes after Hx in male liver, however, much stronger in female liver – or male liver exposed to estrogen. Accordingly, estrogen can accelerate male liver regeneration to female levels following resection.[16,17] Estrogen signals also via ERβ, here to foster metabolic function of hepatocytes[31] – another reason why female patients may recover better from liver surgery. The mouse findings however are limited by the lack of data on estrogen effects in males. While according experiments were performed, technical issues precluded reliable findings.

As further limitations, no volumetric data was available for our Hx patients, and registry data as such were partially incomplete and not designed to pursue a specific hypothesis. ALPPS volumetry was not assessed by the same radiologists, and preoperative liver quality, impacting the outcome, was not always available. Therefore, a multicentric, prospective study on ALPPS/major Hx patients designed to assess true kinetic liver growth rates[32,33] in sex subgroups of different age classes considering sex hormone levels is needed. Nevertheless, our findings from patients undergoing 2 different kinds of Hx combined with our mouse data strongly suggest that females recover better from major surgery owing to a superior regenerative capacity.

Notably, sex disparities are documented for hepatocellular carcinoma (HCC), viral hepatitis, cirrhosis, fibrosis and non-alcoholic fatty liver disease, with males displaying higher incidences and/or faster disease development relative to females.[6–9,34–37] Estrogen is thought to be one important determinant behind these differences,[6,36–38] and is likely central to our observations. Superior liver regeneration in females may add to above disparities, such as faster fibrosis/cirrhosis progression in males. Although counter-intuitive, faster liver regeneration might also relate to a lower HCC incidence in females, because unlike liver growth after tissue loss,[39,40] HCC originates from stem cells and indeed may counteract efficient liver regeneration.[41] Little evidence exists for a role of estrogen in liver surgery outcomes, besides the worse outcomes after female-to-male liver transplantation that have been associated with hormonal mismatch.[5,38,42] Intriguingly, however, male sex was reported as an independent risk factor for reduced volumetric liver growth after portal vein embolization,[43] fully consistent with a superior regenerative capacity of female liver.

In conclusion, this is the first clinical study to demonstrate sex differences in outcomes and liver regeneration parameters after major liver surgery. Our findings point to a regenerative pace that is faster in females than males, a phenomenon likely driven by estrogen. While prospective confirmation of our findings is needed, estrogen treatment is established and should be adapted to settings of major liver surgery. We therefore will test the regenerative benefits of estrogen on resected human livers maintained *ex vivo* in our long-term perfusion system.[44,45] Findings will also inform our randomized clinical trial planned to assess perioperative estrogen for postmenopausal women undergoing major resections. Given that estrogen can countervail the regenerative inferiority of male animals,[13] estrogen treatment will likewise be considered for men subjected to high-risk surgeries such as ALPPS.

ACKNOWLEDGMENTS

The authors thank Karin Pettersson for managing the Zurich ALPPS registry data and Susanne Gaal for administrative assistance.

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DISCUSSANTS

Guido Torzilli (Milano, Italy)

I congratulate the authors of this interesting paper, which investigates the potential role of estrogen in protecting against post-hepatectomy liver failure and major morbidity after major hepatectomies. In my view, the paper embraces an argument of interest, and the results are objectively of interest. However, methodologically patching the study into three parts is somehow confusing and conveys several potential biases, withdrawing rather than adding to the clarity of the message. The experimental study per se conveys information, which has already been reported, and does not add anything significant to the experimental part. Furthermore, the third part seems confusing, and rather supportive of the clinical results since an experimental background on this issue already exists in literature. On the other hand, the clinical results are biased in their retrospective setting, with significant data missing (i.e. volumetric data on the cohort with major hepatectomy).

In synthesis, wouldn't it have been better to wait more time, and focus on more complete data from a single arm, rather than patching data coming from different studies and clinical settings? Following your paper, do you think that we could have enough evidence to protect male and post-menopausal female patients with estrogen? Don't you think that this paper further maintains the idea of sustaining parenchyma sparing surgery, even for advanced disease?

Again, congratulations on your efforts to conduct such a complex study and for the privilege of the discussion.

Response from Dominique L. Birrer (Zurich, Switzerland)

Thank you very much, Professor Torzilli, for your comments and questions. You criticize the study design, which is based on two retrospective cohorts and novel experimental data. I agree that a single cohort study would be better than our mixed design, particularly in terms of power. Such a study would need to include suitable endpoints, such as longitudinal volumetry, and above all, the levels of sex hormones in patients. However, such data were not available to us and would require the design of a new study. Given that estrogen may have the power to improve surgical outcomes, we wanted to speed up this process, so we chose a pragmatic approach choose the data, which were available to us. Clearly, our approach comes with the disadvantages that you pinpointed, such as non-uniformity and missing data, but we think that we addressed the missing data by using multiple imputations, especially for the ALPPS registry. Our ALPPS data is solid because we also performed a sensitivity analysis, which confirmed the findings from our original data set. Another advantage of our study is that it adds some universality. The hepatectomy data, ALPPS data, and the mouse data all point to a superior recovery in females.

With regards to the mice experiments, I agree that existing reports have found faster regeneration in female rodents. However, others have reported the contrary; hence, these findings remain controversial. On the other hand, estrogen as a promoter of hepatocyte proliferation is an established observation. I would like to further note that ALPPS surgery has never been performed in female mice to date. Therefore, I can say with confidence that faster, estrogen-dependent regeneration following ALPPS in female mice is a novelty.

You also asked whether we have sufficient evidence to give estrogen to females and males. In my view, we first need solid data on the actual sex hormone levels in male and female patients of various age groups and look at the association with liver function and surgical outcomes. Only then can we define the level of estrogen needed in humans for an optimal regenerative response.

Regarding your topic of parenchyma sparing surgery, indeed, it can be argued the more you spare, the less regeneration

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is induced, and the less promoting measures (such as estrogen supplementation) will be needed.

Martin K. Angele (Munich, Germany)

Thank you for this nice paper and interesting concept of the effect of sex hormones on liver regeneration. I have two questions. First, regarding the subgroup of females under estrogen replacement treatment, do you know anything about this subgroup in your collective? Perhaps, in this subgroup, regardless of size, you can see more regeneration, which would prove your principle in the patient cohort. Second, both male and female sex hormones really decrease after major surgery. At least, this was our observation when we measured them in the ICU; could you please comment on this? In the ALPPS procedure, liver regeneration is a process that takes 2-3 weeks; how do you explain that, even with decreasing sex hormone levels, you can still observe such differences?

Response from Dominique L Birrer (Zurich, Switzerland)

Thank you very much, Professor Angele, for your questions. First, we agree that the estrogen replacement therapy subgroup would have been a precious source of information. Such data were, however, not available. Your second question is very interesting, because I also read about hormonal levels in rodents subjected to hepatectomies. After a partial hepatectomy, testosterone levels are thought to decrease; however, estrogen levels seem to be more fluctuating and do not always decrease. Importantly, it is the estrogen receptors that are induced upon hepatectomy in both males and females (apparently more in the latter), and this is most likely the mechanism that makes the regenerating liver susceptible to estrogen. No data is available on ALPPS; however, I would assume that estrogen receptors would also be induced after this surgery, given that ALPPS and a hepatectomy share basic regenerative mechanisms.

Kjetil Søreide (Stavanger, Norway)

Thank you for a very nice presentation. I only have one general comment about the role of estrogen on outcomes in surgery. Some studies have shown that outcomes in surgery are better when performed by female surgeons. So, tongue in cheek, do you think we need more estrogen (=more women) in surgery?

Response Dominique L. Birrer (Zurich, Switzerland)

As a female surgical trainee, I am very much in agreement!

Georgios Sotiropoulos (Athens, Greece)

Thank you very much for this nice analysis. I would like to add a comment. In 2010, in *Liver International*, I published a clinical score on prognostic factors after liver resection in intrahepatic cholangiocarcinoma. In this multivariate analysis, we found three prognostic factors. One of the parameters that was a positive predictive factor for better outcomes was female gender. Your analysis informs or underlines this further.

Response Dominique L. Birrer (Zurich, Switzerland)

Thank you very much for this information, which, adds some validity to our findings.

Fabrizio Michelassi (New York, United States)

I noticed that liver regeneration was larger within the first two weeks in females than in males. I recall a paper by Kim Olthoff, in 2015, which demonstrated that liver regeneration ended up being the same in males and female at 3 months. Whatever the driver of regeneration might be, do you think that it is somehow speedier in females than males in the initial 2-3 weeks, but then the ultimate result is the same?

Response Dominique L. Birrer (Zurich, Switzerland)

This is a very interesting question, and indeed, liver volume post-surgery will ultimately return to its original size in both males and females, leading to both having a similar liver-to-body ratio. Different studies in liver regeneration support this. The ultimate volumes return to normal. The parameters that demonstrate superiority are speed of regeneration, which is the critical period for successful recovery. This is consistent with fewer complications in females, and less liver failure.

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