REVIEWER RESPONSE

REVIEWER 1

This observational study of a large cohort of patients worked up for liver transplantation finds that the QT prolongation is associated with mortality independent of the MELD score. This is an interesting and important finding, as QT measurement is not formally considered in liver transplant clinics. The results of this study are interesting and relevant; however, I have several questions:

1. Patients transplanted for ANY indication were analyzed. What proportion of the cohort was cirrhotic? Was there difference between cirrhotic and non-cirrhotic individuals?

    a. All patients in our study were cirrhotics that were listed for transplant on the Organ Transplant Tracking Record (OTTR) database. Non-cirrhotic patients were not included in the study or analysis. This has been clarified in page 8 line 5.

2. When was the 'recent' ECG performed? It almost sounds as though biochemical parameters were tested during outpatient consultation, but the ECG was performed on the night of transplant, but in non-transplanted patients a 'recent' ECG was used? The ECG timing should be at the same time (outpatient consultation) as measurement of the other variables. Otherwise the relationship between biochemical variables and ECG will be inconsistent and thus uninterpretable.

    a. This is very important observation and the reviewer is correct. To reflect the relationship between the biochemical variables and ECG findings we used the most recent outpatient labs (typically within 90 days of baseline ECG) to reflect the mortality rate predicted by the MELD score. For patients who did not undergo a liver transplant, the most recent ECGs were used. For transplanted patients, the last ECG prior to transplant was used. This has been added and clarified in page 8 line 10 to 20.

3. Timing of biochemical variables in relation to ECG needs to be better clarified. Why are so many MELD variables missing in a cohort being evaluated for liver transplant? MELD score is a frequently measured test. Were these patients with missing MELD different from those patients with a valid MELD (ie were they the patients who never got listed for transplant?).

    a. Although the MELD score is frequently calculated for cirrhotics, acute biochemical changes are often observed during inpatient hospitalizations. In addition, since the MELD score reflects the predicted 90-day mortality in these patients, labs performed outside 90 days of the date of ECG may not accurately reflect mortality rate associated with the observed ECG changes.

MELD variables were unavailable from 102 out of the 406 patients (25%) due to lack of recent outpatient labs (within 90 days) in relation to their baseline ECG. We excluded patients that underwent inpatient lab testing to avoid other clinical factors (such as medication changes, acute electrolyte disturbances, or clinical illnesses) that may have acutely affected their MELD score as mentioned above. The authors would have preferred to analyze the MELD data on all 406 patients, but this was not feasible due to the reasons mentioned above. Despite unavailable MELD scores on the remaining 102 patients, our analyses re-confirmed what is already well established in current literature that higher MELD scores are associated with higher mortality in end stage liver disease patients.

b. Rather than its association to mortality, when the MELD components/score was evaluated with the presence of QT prolongation, our results did show that males with QT prolongation had higher creatinine, MELD, and MELD-NA scores than males without QT prolongation (Table 1). Although this may imply that males with worse MELD scores (ie sicker patients) had a higher prevalence of QT prolongation, the retrospective nature of this study does not establish a cause and effect relationship and the findings do not directly impact the main conclusion of our study where QT prolongation was an independent risk factor for mortality in ESLD. This has been noted as a limitation in the discussion section in page 15 line 2 to 18.

4. 323 patients evaluated for liver transplant were excluded from this study. Why? was it because they did not have a valid ECG or for other reasons??

a. The Organ Transplant Tracking Record (OTTR) database revealed that 729 patients were evaluated for a liver transplant at the University of Kentucky over a recent 12-year period. Of the 729 patients, 406 met the inclusion criteria for this study. The other 323 patients were excluded from the analysis due to the lack of a pre-transplant baseline ECG over the study period, conduction abnormalities (bundle branch block), atrial fibrillation, MI within 30 days of baseline ECG, or pacemaker use.

5. It would be important to note in the abstract that QT prolongation predicts mortality INDEPENDENTLY of the MELD score, as this is a very important finding.

a. We have revised the manuscript to reflect this important finding.

REVIEWER 2

1. This is retrospective study of 406 ESLD patients. With a great (25%) missing values for MELD components and MELD totals how the results can be reliable?

   a. Please see above response to Reviewer 1. Although the MELD score is frequently calculated for cirrhotics, acute biochemical changes are often observed during inpatient hospitalizations. In addition, since the MELD score reflects the predicted 90-day mortality in these patients, labs performed outside 90 days of the date of baseline ECG may not accurately reflect mortality rate associated with the observed ECG changes. MELD variables were missing from a 102 out of the 406 patients (25%) mainly due to lack of recent outpatient labs (within 90 days) in relation to their baseline ECG. We excluded patients that underwent inpatient lab testing to avoid other clinical factors (such as medication changes, acute electrolyte disturbances, or clinical illnesses) that may have acutely affected their MELD score as mentioned above. As stated by the reviewer, the authors would have preferred to analyze the MELD data on all 406 patients, but this was not feasible due to the reasons mentioned above. Despite missing MELD scores on the remaining 102 patients, we believe the effective sample size of 304 to be large enough to reflect reliable results for the overall cohort of patients analyzed in this study.

   b. Rather than its association to mortality, when the MELD components/score was evaluated with the presence of QT prolongation, our results did show that males with QT prolongation had higher creatinine, MELD, and MELD-NA scores than males without QT prolongation (Table 1). Although this may imply that males with worse MELD scores (ie sicker patients) had a higher prevalence of QT prolongation, the retrospective nature of this study does not establish a cause and effect relationship and the findings do not directly impact the main conclusion of our study where QT prolongation was an independent risk factor for mortality in ESLD. This has been noted as a limitation in the discussion section in page 15 line 2 to 18.

2. ''Most recent ECGs were used'': the time of ECGs should be defined clearly. Could it had been just before or during the terminal lethal complication of ESLD?

   a. Please see above response to Reviewer 1. For patients who did not undergo a liver transplant, the most recent ECGs were used. For transplanted patients, the last ECG prior to transplant and the average of the oldest and the most recent post-transplant ECGs were used to eliminate any outliers during the immediate post transplantation recovery period, vulnerable to medication changes or electrolyte imbalances.

3. How many of 187 patients who died had QTc prolongation (total, mild, moderate, or severe)? Some of them died during or after liver transplantation?

a. Out of the 187 patients who died 110 had QT prolongation. QT prolongation was a significant independent predictor of mortality (OR 1.69, 95% CI 1.03-2.77, p=0.039). Out of the 406 patients that met the inclusion criteria for this study, 98 patients underwent liver transplantation and the effective sample size for comparison of QT interval pre- and post-transplantation was 74. In total, 21 out of 98 transplanted patients (14 out of the 74 with interpretable ECGs for comparison) expired during the study period. None of the deaths occurred during the surgical procedure.

b. As mentioned by Reviewer 2, we also examined the association between the degree of QT_c prolongation and mortality. Patients with QT_c prolongation were subdivided into three categories for analytic purposes: mild (451 – 470 msec in males; 471 – 490 msec in females), moderate (471 – 490 msec in males; 491 – 510 msec in females), and severe (> 490 msec in males; > 510 msec in females).

c. Any level (mild, moderate, or severe) of QT_c prolongation significantly increased mortality. This is included in the manuscript in Table 2 which demonstrates the estimated odds ratio based on levels of QT_c prolongation. However, the risk of mortality did not exhibit a homogenously proportional increase with respect to the level of prolongation. Therefore, we did not include the details of the analysis, but made it available for reviewer below.

Predictor	p-values
mild vs none	0.0452
moderate vs none	0.0136
severe vs none	0.0437

Predictor	Estimated Odds Ratio	95% Confidence Interval	
mild vs none	1.670	1.011	2.759
moderate vs none	2.109	1.166	3.814
severe vs none	1.834	1.017	3.306

Male: Mild 451 - 470, Moderate 471 - 490, Severe > 490
Female: Mild 471 - 490, Moderate 491 - 510, Severe > 510

Table of Expired by qtccat					
Expired(Expired)	**qtccat**				
Frequency Percent Row Pct Col Pct	**0**	**1**	**2**	**3**	**Total**
N	122 30.05 55.71 61.31	45 11.08 20.55 49.45	25 6.16 11.42 43.10	27 6.65 12.33 46.55	219 53.94
Y	77 18.97 41.18 38.69	46 11.33 24.60 50.55	33 8.13 17.65 56.90	31 7.64 16.58 53.45	187 46.06
Total	199 49.01	91 22.41	58 14.29	58 14.29	406 100.00

Mortality by QTC prolongation status. 3 is severe (above 490 for males, above 510 for females), 2 is moderate (470 to 490 for males, 490 to 510 for females), 1 is mild (450 to 470 for males, 470 to 490 for females), and 0 is absent.

4. OR was greater in moderate than in severe QTc prolongation (2.11 times και 1.83 times respectively). A Kaplan–Meier survival plot would be useful including all 406 patients in 4 groups: 1st without QTc prolongation and 2nd, 3d and 4th with mild, moderate, or severe QTc prolongation. Similarly, a Log rank P for patients with and without QTc prolongation would be useful.

   a. See response Number 3 above illustrating the difference in mortality based on the severity of QT prolongation. We agree with the Reviewer that a Kaplan-Meier survival plot may have been useful; however, given the lack of direct association (proportional increase in mortality to severity of QT_c prolongation) we did not include these results in our study. If the reviewer strongly feels that the above results need to be included in the manuscript, we certainly can.

5. Survival should include the parameter of time (days, weeks, months, years). A Cox proportional hazards regression analysis would be useful for the mortality rate estimation in relation to the time.

   a. This is an important observation made by the reviewer and we agree with the potential usefulness of a Cox proportional hazards regression analysis. Unfortunately, the exact date of death for our patients was not readily available to us during the study. During the study, mortality (or survival) was determined by what was reported in the Organ Transplant Tracking Record (OTTR) database. Although, each patient's electronic medical chart was reviewed at the University of Kentucky Medical Center to account for any

inpatient deaths, we were unable to account for the exact date of expiration for many patients who had expired at outside hospitals.