Response to Reviewer #1:

We would like to thank you for your time and effort invested in reading our article, and above all, for the exhaustive and useful comments. Our responses are outlined below.

The topic is interesting and within the scope of the journal. It aims to determine the changes in the levels of ghrelin, CCK , some inflammatory markers with biliary obstruction and after drainage and how this change can play a role in the nutritional status of patients with biliary obstruction. Overall, it is a good topic and interested. However, it will be benefit from taking into consideration the following points:

1. Background: in addition to the effect of CCK on appetite, its effect on gallbladder evacuation is also important to be mentioned in this context.

RE: This is added to revised manuscript

2. Core tip: Furthermore, it is important to identify factors and mechanisms responsible for it's development : it's refers to what? Biliary obstruction or the malnutrition … needs to make it more clear.

RE: this section is changed in the revised version.

3. Plasma ghrelin and cholecystokinin levels were significantly higher in patients with obstructive jaundice which may be associated with the development of malnutrition during the inflammatory response. : Ghrelin stimulates appetite while CCK inhibits appetite … how to explain the increased levels of these two antagonists with malnutrition?

RE: Previous research indicates functional antagonism between ghrelin and CCK regarding food intake, but their interaction on secretion of both peptides is still not well understood. An animal study showed that intraduodenal ghrelin infusion increases CCK concentration, and human research indicates that CCK can suppress ghrelin secretion. Nonetheless, in our study group both high CCK and ghrelin concentrations persisted. A previous study showed that application of ghrelin after CCK infusion does not induce feeding and vice versa, CCK application after ghrelin infusion does not reduce food intake. This suggests that the efficacy of ghrelin and CCK signaling depends on their mutual balance. Disruption of that balance, such as it was seen in our patients with biliary obstruction, could lead to dysfunction in appetite regulation which, according to our results, can be restored, but through some other mechanisms. These findings were further expanded in the Discussion section.

5. English editing , as commas are missed in many sentences examples: Compared to patients with benign etiology, patients with malignant biliary obstruction were older,. Compared to control group patients with both malignant and benign etiology of biliary obstruction had greater body mass loss….. add , after group "Compared to control group, patients……

RE: the whole text underwent a spelling check once more.

6. Results: - Laboratory parameters Compared to the control group patients with biliary obstruction had statistically significant difference for most of the measured laboratory parameters…. Add comma after Compared to the control group, patients inflammation factors.. suggest to change into inflammatory markers –

RE: changed in the revised manuscript

7. -Multivariate analysis Predictive factors for having NRS 2002 score ≥3 were increased TNF-α concentrations… suggest to replace were by “included” to make it clear.

RE: this part was changed in the revised manuscript

8. -28 days were associated with malnourishment, namely increased CCK concentration reduced the chance of malnourishment 1.01 times… what does “malnourishment” refers exactly to here? How it is assessed?

RE: nutritional risk was assessed using validated nutritional risk screening tool NRS 2002- this part was changed to more precise term in the revised manuscript.

9.- It is stated that increased CCK concentration after 28 days, reduced the chance of malnourishment 1.01 times, and stated that 48 hours after drainage, higher CCK concentration and leukocyte count decreased the chance of improving appetite…. Are those results contradictory? Need to elaborated on under discussion.

RE: We have not establish any significant correlation between appetite and appetite regulating hormones or inflammatory markers tested during the biliary obstruction and 28 days after its resolution, suggesting the existence of additional factors of appetite regulation. The influence of CCK concentration 48 hours after biliary drainage on appetite was not confirmed in subsequent measurement, so its meaning is really questionable. Aside from appetite, the influnece of CCK on nutrional parameters and outcome was consistent across measurements with positive correlation. This was further expanded in the Discussion.

10.- Considering that all of the patients with negative clinical outcome had malignant obstruction we conducted analysis only for patients in group with malignant etiology and higher initial levels of TNF-α (P = 0.018) and IL-6 (P = 0.003) were again observed in patients who died. This sentence is not clear

RE: this sentence is corrected in the revised manuscript

11.- Table1 Age What are these numbers ? is it meadian? 33-84 55-88 39-90

RE: Age is presented as median and 25th-75th percentile in the brackets.

12.- For weight loss%, how did you calculate? Over how long time?

RE: percentage of weight loss was calculated in the previous three months period.

13.- Is the significant difference between patients with malignant and benign etiology in many parameters is a confounding factor for the effect of drainage on the nutritional status, it is better to compare the results of these two groups separately.

RE: Although there were significant differences between patients with benign and malignant etiology the effect of the biliary drainage on these subgroups was not analyzed separately due to small sample size and lack of power. This explanation is added to study limitations in the revised manuscript.

14.- Write the full name of LPS.

RE: corrected

15.- Discussion - We hypothesise that in patients with advanced malignant disease and malnourishment prolonged inflammation secretion of IL-6 and TNF-α leads to suppression of CCK secretion, possibly through lower IL-1 activity… correct into “We hypothesise that in patients with

advanced malignant disease and malnourishment, prolonged inflammation, and secretion of IL-6 and TNF-α leads to suppression of CCK secretion, possibly through lower IL-1 activity"

RE: corrected

16.- but their interaction on secretion of both peptides is not well understood…need to reframe –

RE: : corrected

17.-Under conclusion, authors stated that" Patients with biliary obstruction, regardless of the etiology, have higher plasma concentrations of ghrelin as well as increased concentrations of TNF-α, IL-6 and CRP and worse nutritional status, however under discussion, authors stated that "When comparing the etiology of the obstruction, only CRP values were significantly elevated in patients with benign biliary obstruction (P = 0.001) while there was no difference in TNF-α and IL-6 concentrations. Those are contradictory.

RE: we admit the construction in the Conclusion is imprecise – when compared to control group patients with biliary obstruction had higher plasma concentrations of ghrelin as well as increased concentrations of TNF-α, IL-6 and CRP and worse nutritional status. But, when comparing both groups of patients, CRP values were significantly elevated in patients with benign biliary obstruction. The sentence in the Conclusion is corrected.

18.- Table 2: What is SE? is it sedimentation rate?

RE: Yes; this was explained in the text

19.- Table 4: suggest to change "During biliary obstruction" into "on admission" or " in the presence of biliary obstruction"

RE: changed

20. - Tables and figures are clear and easier to understand the findings than the the way the results have been written… need to improve and simplify the presentation of the results.

RE: the Result section have been significantly shortened and simplified and Discussion section have been modified as suggested by the second reviewer.

Response to Reviewer #2

We would like to thank you for your time and effort invested in reading our article, and above all, for the useful comments whic significantly improved our manuscript . Our responses are outlined below.

In this manuscript, the authors sought to investigate the influence of obstructive jaundice on the hormones controlling appetite and nutritive status. This paper has some significance for clinicians and researchers working. However, there are several issues that need attention.

1. How sample size was determined? It would be crucial for the authors to describe their sample size and Type I and II error calculations that resulted in the recruitment of 55 patients, with power.

RE: The power test analysis description is added to the Statistical analysis section as follows: "Analysis of test power for ANOVA (3 repeated measurements) with the following parameters: test power of 90%, α significance level of 0.05 and sample size effect of 0.2 with an assumed correlation between measurements of 0.5, estimated the total sample size to be 55. Additional analysis for the

independent t-test (difference between test and control groups) under the same conditions, found that the control group should consist of 40 healthy subjects"

2. Results---The results should be stated briefly and succinctly. Do not interpret the data here. Do not explain how you deduced the conclusion from the results obtained.

RE: the Result section has been significantly shortened and simplified.

3. Discussion---the discussion section is not well-written. The discussion opens with a section that would be most appropriate for the introduction.

RE: the Discussion section has been modified according to suggestions.

4.The authors did not note the limitations in their study.

RE: this part is added to the revised manuscript