PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Outpatient percutaneous native renal biopsy: safety profile in a large monocentric cohort
AUTHORS	Roccatello, Dario; Sciascia, Savino; Rossi, Daniela; Naretto, Carla; Bazzan, Mario; Solfietti, Laura; Baldovino, Simone; Menegatti, Elisa

VERSION 1 - REVIEW

REVIEWER	Simon Jiang Department of Renal Medicine The Canberra Hospital, Australia. John Curtin School of Medical Research, Australian National University, Australia.
REVIEW RETURNED	01-Dec-2016

GENERAL COMMENTS	The authors present an interesting study on the safety of PKB in a single centre, by a single operator, over a decade when transitioning from routine admission to ambulatory PKB. The paper has a large cohort and controls for operator variance by having single operator. They appropriately determine and analyse the variables influencing outcome. The authors also benefit from prospectively collecting data on biopsy outcomes. I have several questions regarding this work. Methods: 1. Please state whether the relevant ethics board or institutional review approval was obtained 2. For the retrospectivelly compiled cohort, how was this data obtained? Was there missing data and how was missing data handled? Were patients excluded? 3. Regarding outcomes, the authors report subcapsular haematoma <5cm spontaneously resolving was classified as a minor adverse event. How were haematomas >5cm not requiring classification reported, i.e do they automatically require intervention and are therefore major? 4. Please clarify what happens when greater than 3 passes with insufficient material from PKB - is the procedure abandoned? This is relevant in the context of the number of passes not being significant as risk climbs above 3-4 passes. 5. Did authors include kidney size in the prospectively collected data? This has been reported as a significant risk and would be important in the study 6. For multivariate analysis, could the authors please indicate the variables which they included in the model as this is essential to interpretation of results. Were haematological parameters included? 7. Did the authors test for variable inflation between creatinine and

	hypertension in their multivariate model, as renal function is known to be associated with development of hypertension and therefore these may be interacting and spuriously significant? 8. How was any missing data from the retrospective (or prospective) cohort handled in multivariate analysis? 9. It would probably be simpler to interpret data by combining table 1 and 2. 10. It would be helpful to have a table summarising the HR with 95CI for the variables included in the model. 11. Whilst generally well written, there a several typographical and grammatical errors that require review.

REVIEWER	Pornpimol Rianthavorn Faculty of Medicine Chulalongkorn University
REVIEW RETURNED	06-Dec-2016

GENERAL COMMENTS	Thank you so much for giving me the opportunity to review the article entitled “Point-of-Care Ultrasonography for Kidney Biopsy in Children”. The article is well written. However, several points require clarification. Major points: The study attempts to compare the safety or complication rates of PKB performed in inpatient setting vs. outpatient setting. Although this is an interesting and important issue, the study lacks novelty. Several studies have shown that outpatient PKB is safe when patients are well prepared. Material and Methods section 1. Safety of PKB varies between different reports depending on several factors including definitions of complications, biopsy techniques and characteristics of patients. For example, the incidence of post PKB hematoma depends on post PKB ultrasound protocol. When post PKB US was routinely performed, the incidence of post PKB hematoma would be high. Thus, the sentence “Postbiopsy imaging was done following the protocol or when clinically indicated at the discretion of the attending physician.” needs clarification. 2. Regarding the pre-ODS-PKB protocol, the algorithm for patient with hypertension needs clarification. Was the PKB cancelled or the hypertension was treated and PKB was performed when BP was under control? Statistical analysis 3. Please clarify factors including in the univariate analysis. 4. In multivariate analysis, please explain why the authors used the cutoff of serum creatinine of 3 mg/dL. Result section 5. In table 1 and 2, please defined uncontrolled hypertension and severe hypertension. 6. Could patients in the first era that required inpatient PKB included some patients who required hospitalization due to their disease and thus were sicker than patients in the second period as the rate of hypertension in the inpatient group was significantly higher (Table 2)? As the authors demonstrated that serum creatinine of >3 mg/dL and hypertension were independent risk factors for complications. The number of patients with serum creatinine > 3 mg/dL in each group should be compared as well. In the result section 7. For better understanding, the odds ration with 95% CI of factors

	including in the univariate and multivariate analysis should be demonstrated. Minor points 1. Please confirm the description of the automated biopsy gun used. Should the length of the biopsy gun be 15 cm not 15 mm? 2. In the abstract, Line 19 and Material and Methods Section, Line 11, the sentence should read “inpatient procedure”. 3. In the Material and Methods Section, Line 2, the sentence should read “Patients are placed in a prone position on the bed for at least 2 hours. Patients received i.v. hydration and are observed for symptoms of urine retention. 4. In the discussion section, Line 31, the sentence should read “Oviasu and Ugdodaga [5] from Nigeria reported in no complications in a series of 20 patients.

REVIEWER	Esther Lu Division of Public Health Sciences Section of Oncologic Biostatistics Department of Surgery Washington University School of Medicine USA
REVIEW RETURNED	17-Jan-2017

GENERAL COMMENTS	This paper is to evaluate the safety of performing percutaneous native kidney biopsy (PKB) as an outpatient procedure (implying an observation period of 6 hrs) compared to the traditional inpatient policy. One group includes the patients whose PKBs were performed in the outpatient department (2012-2016) and followed by 6 hours’ observation period and then by regular outpatient visits. The other group includes the retrospectively retrieved patients whose PKBs were performed and followed by at least 1-day hospital admission in the same Institution between January 2000 and November 2012 as in patient procedure. All biopsies were performed by a single nephrologist (DR) with the guidance of an expert radiologist. Several concerns about the statistical analysis. 1. Univariate analysis was performed to assess the association between complications and risk factors using the Pearson, $\chi 2$ and Fisher exact tests. Why wasn’t logistic regression model used to identify the risk factors in complication? 2. Instead multivariate Cox model was used to predict the complication. Manuscript says “The forward conditional techniques were used to find the final model.” 1) Is the variable of group forced to be in the model during forward selection? 2) Is univariate analysis in the Cox model considered? If the variable of group was forced to be in the model, then univariate analysis refers to the model in which includes group and the other factor. e.g. gender. P-value for gender need to be considered to keep gender in the next step or not. 3) The section of data analysis says “The results are expressed as ORs with 95% CI”. OR should be hazard ratio (HR)? 4) Add a K-M plot between two groups for probability of complication and p-value from log-rant test should be presented. 5) Add a table to present the results from Cox model including univariate and multivariate analysis, e.g p-value, HR, and 95% CI.

	3. Risk factors which were considered in the multivariate analysis should be reported in the section of data analysis. 4. Combine Tables 1 and 2? 5. Delete “p=” before numerical value in table 2. For example, p=0.01->0.01

VERSION 1 – AUTHOR RESPONSE

Reviewer 1

Reviewer 1. 1. Please state whether the relevant ethics board or institutional review approval was obtained
Reply 1.1 Percutaneous kidney biopsies were performed when needed as part of good clinical practise for patients refereed to our department. Data collection was performed according to the local legislation of the institutional review board. This was stated in the revised version.
Reviewer 1. 2. For the retrospectively compiled cohort, how was this data obtained? Was there missing data and how was missing data handled? Were patients excluded?
Reply 1.2. Data were collected from electronic clinical files system of the hospital and when missing, data were obtained through the original paper files or directly contacting the patients. Patients whose data set was not completed were excluded from our analysis. This applied to 3 cases and has been specified in the manuscript, to read: “Patients whose data set was not fully available were excluded from our analysis (3 cases)”.
Reviewer 1. 3. Regarding outcomes, the authors report subcapsular haematoma <5 cm spontaneously resolving was classified as a minor adverse event. How were haematomas >5cm not requiring classification reported, i.e do they automatically require intervention and are therefore major?
Reply 1.3. For the purpose of this study, we arbitrarily decided that subcapsular haematoma >5 cm and/or those requiring intervention (despite the size) were considered as major complications. This has been specified in the text, to read: “Subcapsular haematoma 5>cm and/or those requiring intervention (despite the size) were considered as major complications”.
Reviewer 1. 4. Please clarify what happens when greater than 3 passes with insufficient material from PKB - is the procedure abandoned? This is relevant in the context of the number of passes not being significant as risk climbs above 3-4 passes.
Reply 1. 4. When 3 passes for PKB retrieved insufficient material, the procedure was not abandoned. Table 1 reports mean±SD/median[range] values for passages. The upper limited of the range reads 5. Of note, it is worth mentioning that more than 3 passages were necessary only a minority of the patients. Besides, the number of passes was computed among the variables considered in the analysis.
Reviewer 1. 5. Did authors include kidney size in the prospectively collected data? This has been reported as a significant risk and would be important in the study.
Reply 1.5. Data on kidney size at sonographic investigations were available and computed in our analysis. We observed no statistical difference between patients with and without complications in terms of kidney size. The manuscript has been amended, to read “Conversely, we found no association of risk with the number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy nor the degree of proteinuria”.
Reviewer 1. 6. For multivariate analysis, could the authors please indicate the variables which they included in the model as this is essential to interpretation of results. Were haematological parameters included?
Reply 1.6 The text has been amended for clarity to read: “For univariate analysis, the following variables were included in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of

proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents. For the multivariate analysis included variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets agents (as described in table 1S)" .
Reviewer 1. 7. Did the authors test for variable inflation between creatinine and hypertension in their multivariate model, as renal function is known to be associated with development of hypertension and therefore these may be interacting and spuriously significant?
Reply 1.7 In our study we analysed the magnitude of multicollinearity between creatinine and hypertension in a set of patients with Kidney disease. We found a VIF of 1.83 and 1.76, respectively, compatible with a very low correlation.
Reviewer 1. 8. How was any missing data from the retrospective (or prospective) cohort handled in multivariate analysis?
Reply 1.8. As previously specified, patients whose data set was not fully available were excluded from our analysis (3 cases).
Reviewer 1.9. It would probably be simpler to interpret data by combining table 1 and 2.
Reply 1.9. Tables 1 and 2 have been now combined.

Reviewer 1. 10. It would be helpful to have a table summarising the HR with 95CI for the variables included in the model.
Reply 1.10: Table 1S has now been provided.

Reviewer 1. 11 Whilst generally well written, there a typographical and grammatical errors that require review.
Reply 1.11 The manuscript has been edited and amended when necessary by a native English speaker.

Reviewer: 2

Reviewer 2. 1. Safety of PKB varies between different reports depending on several factors including definitions of complications, biopsy techniques and characteristics of patients. For example, the incidence of post PKB hematoma depends on post PKB ultrasound protocol. When post PKB US was routinely performed, the incidence of post PKB hematoma would be high. Thus, the sentence "Postbiopsy imaging was done following the protocol or when clinically indicated at the discretion of the attending physician." needs clarification.
Reply 2.1: The sentence has been amended for clarity, to read: "Post-biopsy imaging was done in all the patients following the protocol. Additional imaging investigations, including additional sonography were performed when clinically indicated at the discretion of the attending physician"
Reviewer 2. 2. Regarding the pre-ODS-PKB protocol, the algorithm for patient with hypertension needs clarification. Was the PKB cancelled or the hypertension was treated and PKB was performed when BP was under control?
Reply 2.2. As stated in the protocol, we proceed in performing the PKB if blood pressure is 170/95 mm Hg or lower at the day of the procedure. If the patient presents with values above 170/95 mm Hg, we administer nifedipine oral drops (up to 10 mg) and/or captopril 25 mg and monitor blood pressure every 10 minutes. PKB is performed if blood pressure is stably controlled at 170/95 mm Hg or lower, otherwise PKD is postponed till blood pressure stabilization.
Those information has been added in the protocol accordingly.

Statistical analysis
Reviewer 2.3. Please clarify factors including in the univariate analysis.

Reply 2.3. A table summarising factors included in the analysis and the OR with 95CI for both univariate and multivariate analysis has been included and provided as supplementary material for this submission.

Reviewer 2.4 In multivariate analysis, please explain why the authors used the cutoff of serum creatinine of 3 mg/dL.
Reply 2.4. The cut off was arbitrary chosen. However, this is consistent with previous experimental data (e.g. J Am Soc Nephrol 15: 142–147, 2004).
Reviewer 2.5. In table 1 and 2, please defined uncontrolled hypertension and severe hypertension.
Reply 2.5. Tables and 1 and 2 has been merged and amended for clarity. Missing definitions have been provided.
Reviewer 2.6 Could patients in the first era that required inpatient PKB included some patients who required hospitalization due to their disease and thus were sicker than patients in the second period as the rate of hypertension in the inpatient group was significantly higher (Table 2)? As the authors demonstrated that serum creatinine of >3 mg/dL and hypertension were independent risk factors for complications. The number of patients with serum creatinine > 3 mg/dL in each group should be compared as well.
In the result section
Reply 2.6: While we cannot exclude that there could have been a bias of selection of patients between the two periods of admission, however for our Center hospital policy, all the patients undergoing PKB between 2000 and 2012 were hospitalized for at least 24h, potentially reducing a selection bias; in other words, the clinical setting at entrance was not a determinant impacting on the choice whether a patient needed hospitalization. In line with this comment, no statistical significant difference at baseline was observed between the two groups.
Besides, the number of patients serum creatinine > 3 mg/dL was added in table 1.

Reviewer 2.7. For better understanding, the odds ration with 95% CI of factors including in the univariate and multivariate analysis should be demonstrated.
Reply 2.7: A table summarising the ORs with 95CI has been included and provided as supplementary material for this submission.

Minor points
Reviewer 2. 1. 1. Please confirm the description of the automated biopsy gun used. Should the length of the biopsy gun be 15 cm not 15 mm?
Reply 2. 1. 1. Thank you for pointing this out. The information has been amended accordingly.
Reviewer 2. 1. 2. In the abstract, Line 19 and Material and Methods Section, Line 11, the sentence should read “inpatient procedure”.
Reply 2.1.2 Thank you for pointing this out. The sentence has been amended accordingly.
Reviewer 2. 1. 3. In the Material and Methods Section, Line 2, the sentence should read “Patients are placed in a prone position on the bed for at least 2 hours. Patients received i.v. hydration and are observed for symptoms of urine retention.
Reply 2.1.3 Thank you for pointing this out. The sentence has been amended accordingly.
Reviewer 2. 1. 4. In the discussion section, Line 31, the sentence should read “Oviasu and Ugdodaga [5] from Nigeria reported in no complications in a series of 20 patients.
Reply 2.1.4 Thank you for pointing this out. The sentence has been amended accordingly.

Reviewer 3

Reviewer 3.1. Univariate analysis was performed to assess the association between complications and risk factors using the Pearson, $\chi 2$ and Fisher exact tests. Why wasn’t logistic regression model used to identify the risk factors in complication? Instead multivariate Cox model was used to predict the complication.

Reply 3.1 Thank you for pointing out this aspect. Clinical complications for PKB are heterogeneous, and even among minor Vs. major complications, the spectrum of possible scenarios is extremely wide. Thus, the linear relationship between any continuous risk factors and the logit transformation of the complications could not be assumed.
With the above limitations in mind, and due to the ambispective nature of our study, we chose to limit our analysis to the comparison of the rate of the complication between the two groups.

Reviewer 3.2. Manuscript says “The forward conditional techniques were used to find the final model.” Is the variable of group forced to be in the model during forward selection? Is univariate analysis in the Cox model considered? If the variable of group was forced to be in the model, then univariate analysis refers to the model in which includes group and the other factor. e.g. gender. P-value for gender need to be considered to keep gender in the next step or not.
Reply 3.2. The variable of group was forced to be in the model and each variable found as statistically significant at the univariate analysis was considered for the next step of the analysis.

Reviewer 3.3. The section of data analysis says “The results are expressed as ORs with 95% CI”. OR should be hazard ratio (HR)?
Reply 3.2. As previously stated, due to the ambispective nature of our study, we chose to limit our analysis to the comparison of the rate of the complication between the two groups. For this reason and because only data on outpatient PKB outcomes were prospectively collected, we chose to express results as ORs with 95% CI.

Reviewer 3.4. Add a K-M plot between two groups for probability of complication and p-value from log-rang test should be presented.
Reply 3.4. A Kaplan-Meier analysis for the probability of complications has been performed. Results are shown herewith. P-value for log-rang test has been computed and no statistical significance was observed when comparing group 1 and 2.

[K-M figure provided as attachment]

Overall Comparisons
Chi-Square df Sig.
Log Rank (Mantel-Cox) ,025 1 ,875
Test of equality of survival distributions for the different levels of Group.

Reviewer 3.5. Add a table to present the results from Cox model including univariate and multivariate analysis, e.g p-value, HR, and 95% CI.
Reply 3.5. A table summarising the ORs with 95CI has been included and provided as supplementary material for this submission.

Reviewer 3.6. Risk factors which were considered in the multivariate analysis should be reported in the section of data analysis.

Reply 3.6. The text has been amended for clarity to read: “For univariate analysis, the following variables were included in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents. For the multivariate analysis included variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets agents. (as

described in table 1S)”

Reviewer 3.7. Combine Tables 1 and 2? Delete “p=” before numerical value in table 2. For example, p=0.01->0.01
Reply 3.7. Tables and 1 and 2 has been merged and amended for clarity.

VERSION 2 – REVIEW

REVIEWER	Simon Jiang Department of Renal Medicine, Canberra Hospital, Australia Department of Immunology and Infectious Disease, John Curtin School of Medical Research, Australia
REVIEW RETURNED	04-Mar-2017

GENERAL COMMENTS	I thank the authors, my queries have been adequately addressed.

REVIEWER	Esther Lu WUSM
REVIEW RETURNED	01-Mar-2017

GENERAL COMMENTS	1. Regarding reply 3.1, “the linear relationship between any continuous risk factors and the logit transformation of the complications could not be assumed. With the above limitations in mind, and due to the ambispective nature of our study, we chose to limit our analysis to the comparison of the rate of the complication between the two groups.” However, the manuscript says :” Multivariate survival analysis was performed using the proportional hazards model (Cox model) to identify significant independent factors adjusted for the potential confounding risk factors able to predict a complication.” a. How do you solve the same issue using Cox model? b. Did you test cox proportional assumption holds for each independent factor? 2. Regarding reply 3.3 and 3.5, it is still confusing. If Cox model was used in the analysis, hazard ratio (HR) with 95% CI should be reported; If logistic regression model was used, ORs with 95% CI were reported. From the previous response, the logistic regression model was not used.However, ORs with 95% CI were reported throughout. Please clarify. 3. Regarding reply 3.6, it is more appropriate to say “ The following variables were considered in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents.”

VERSION 2 – AUTHOR RESPONSE

Reviewer: 3
Reviewer Name: Esther Lu
Institution and Country: WUSM
Please state any competing interests or state 'None declared': None declared

Point 1_Reviewer 3:
Regarding reply 3.1, "the linear relationship between any continuous risk factors and the logit transformation of the complications could not be assumed. With the above limitations in mind, and due to the ambispective nature of our study, we chose to limit our analysis to the comparison of the rate of the complication between the two groups." However, the manuscript says:" Multivariate survival analysis was performed using the proportional hazards model (Cox model) to identify significant independent factors adjusted for the potential confounding risk factors able to predict a complication."
a. How do you solve the same issue using Cox model?
b. Did you test cox proportional assumption holds for each independent factor?
Reply 1_Reviewer 3:
Thank you for your comment. The nature of our study is ambispective. Besides, clinical complications for PKB are heterogeneous. For these reasons, we chose to focus our analysis on the comparison of the rate of the complication between the two groups, as stated in the manuscript: "Univariate analysis was performed to assess the association between complications and risk factors using the Pearson, $\chi 2$ and Fisher exact tests". Multivariate logistic regression analysis was performed on the following variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets agents. Cox model was applied only for the prospective arm of the study; however, due to the small number of observed complications, the result section focuses on multivariate logistic regression analysis. For the prospective cohort, when analyzing with Cox model, proportional assumption holds were tested for each independent factor.
To add clarity, the manuscript has been amended , to read: "Univariate analysis was performed to assess the association between complications and risk factors using the Pearson, $\chi 2$ and Fisher exact tests. Multivariate logistic regression analysis was performed to identify significant independent factors adjusted for the potential confounding risk factors able to predict a complication, the results are expressed as Odds Ratios (OR) with 95% Confidence Interval (CI). Multivariate survival analysis using the proportional hazards model (Cox model) was performed in the prospective arm of the study."
Point 2_Reviewer 3:
Regarding reply 3.3 and 3.5, it is still confusing. If Cox model was used in the analysis, hazard ratio (HR) with 95% CI should be reported; If logistic regression model was used, ORs with 95% CI were reported. From the previous response, the logistic regression model was not used. However, ORs with 95% CI were reported throughout. Please clarify.
Reply 2_Reviewer 3:
As stated before, the results section focuses on the multivariate logistic regression analysis to compare the two cohorts of the study. For that reason, results from the multivariate regression analysis are expressed throughout the manuscript as Odds Ratio with 95% CI.
When applying a multivariate survival analysis using the proportional hazards model (Cox model) in the prospective arm of the study, we failed to observe any statistical significance. Consequently, results are not shown as HR in the manuscript.

Point 3_Reviewer 3:

Regarding reply 3.6, it is more appropriate to say " The following variables were considered in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations,

presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents."

Reply 3_Reviewer 3:
Thank you for your comment, the manuscript has been changed, to read: "For univariate analysis, the following variables were considered in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents."

VERSION 3 – REVIEW

REVIEWER	Esther Lu WUSM
REVIEW RETURNED	07-May-2017

GENERAL COMMENTS	1. Do the following sentences in section of "data analysis" refer to logistic regression model or Cox model? "For univariate analysis, the following variables were considered in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents. For the multivariate analysis included variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents (as described in table 1S). The forward conditional techniques were used to find the final model." The current display looks they are for Cox model since they are put after the sentence of "Multivariate survival analysis using the proportional hazards model (Cox model) was performed in the prospective arm of the study". However, table 1S use the terms "OR" which should be obtained from logistic regression model. 2. "Multivariate survival analysis using the proportional hazards model (Cox model) was performed in the prospective arm of the study Did this Cox model analyze "time to complication? Need more details for this Cox model even if the results are not reported in the result section. 3. "For the multivariate analysis included variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents" should be "The final multivariate model includes variables: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets

	agents”?

VERSION 3 – AUTHOR RESPONSE

Reviewer: 3
Reviewer Name: Esther Lu
Institution and Country: WUSM
Please state any competing interests or state ‘None declared’: None declared

Point 1_Reviewer 3:
Do the following sentences in section of “data analysis” refer to logistic regression model or Cox model?
“For univariate analysis, the following variables were considered in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents. For the multivariate analysis included variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents (as described in table 1S). The forward conditional techniques were used to find the final model.”
The current display looks they are for Cox model since they are put after the sentence of “Multivariate survival analysis using the proportional hazards model (Cox model) was performed in the prospective arm of the study”. However, table 1S use the terms “OR” which should be obtained from logistic regression model.
Reply 1_Reviewer 3:
Thank you for your comment. The sentences refer to the logistic regression model; to add clarity, the manuscript text and order of paragraphs have been changed, to read:
“For univariate analysis, the following variables were considered in the model: number of biopsy passes, gender, age, diagnosis, kidney size at sonographic investigations, presence of haematuria before the kidney biopsy, the degree of proteinuria, haemoglobin level before the kidney biopsy, thrombocytopenia, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of anti-platelets, LMWH, anti-hypertensive agents.
Multivariate logistic regression analysis was performed to identify significant independent factors adjusted for the potential confounding risk factors able to predict a complication, the results are expressed as Odds Ratios (OR) with 95% Confidence Interval (CI). The final multivariate logistic regression model includes variables: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents (as described in table 1S). The forward conditional techniques were used to find the final model.
In order to analyze risk factors associated with time to complication, multivariate survival analysis was performed using the proportional hazards model (Cox model) in the prospective arm of the study. Risk factors included sex, diagnosis (categorised in primary glomerulopathy or systemic autoimmune condition), age ≥60, Kidney size < 8 cm at sonographic investigations, haematuria, nephrosic proteinuria, haemoglobin level (<10 g/dl), thrombocytopenia, severe arterial hypertension, serum creatinine >3 mg/dl, use of anti-platelets, LMWH, anti-hypertensive agents”
Point 2_Reviewer 3:
“Multivariate survival analysis using the proportional hazards model (Cox model) was performed in the prospective arm of the study Did this Cox model analyze “time to complication? Need more details for this Cox model even if the results are not reported in the result section.
Reply 2_Reviewer 3:
Thank you for your suggestion. The text has been changed, to read: “In order to analyze risk factors

associated with time to complication, multivariate survival analysis was performed using the proportional hazards model (Cox model) in the prospective arm of the study. Risk factors included sex, diagnosis (categorised in primary glomerulopathy or systemic autoimmune condition), age ≥60, Kidney size < 8 cm at sonographic investigations, haematuria, nephrosic proteinuria, haemoglobin level (<10 g/dl), thrombocytopenia, severe arterial hypertension, serum creatinine >3 mg/dl, use of anti-platelets, LMWH, anti-hypertensive agents".

Point 3_Reviewer 3:

"For the multivariate analysis included variables were included: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents" should be "The final multivariate model includes variables: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents"?

Reply 3_Reviewer 3:

Thank you for your suggestion. To add clarity to the text, the manuscript has been amended, to read: ". The final multivariate logistic regression model includes variables: gender, age, diagnosis, the degree of proteinuria, haemoglobin level before the kidney biopsy, severe arterial hypertension, serum creatinine level before the kidney biopsy, the use of antiplatelets agents."

VERSION 4 – REVIEW

REVIEWER	Esther Lu WUSM
REVIEW RETURNED	10-May-2017

GENERAL COMMENTS	No more concerns.