PEER REVIEW HISTORY

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ARTICLE DETAILS

TITLE (PROVISIONAL)	Second-generation antipsychotics and the risk of chronic kidney disease: a population-based case-control study
AUTHORS	Højlund, Mikkel; Lund, Lars; Herping, Jonas; Haastrup, Maija; Damkier, Per; Henriksen, Daniel Pilsgaard

VERSION 1 – REVIEW

REVIEWER	Mu-Hong Chen Taipei Veterans General Hospital, Taiwan
REVIEW RETURNED	10-Mar-2020

GENERAL COMMENTS	Dear Editors and authors, Several suggestions are listed behind. 1. 557 for Second-generation antipsychotics, but 74 for schizophrenia, 120 for bipolar disorder, 122 for dementia. What else diagnoses for Second-generation antipsychotics? Delirium? Or others? Whether the association between Second-generation antipsychotics and CKD is confounded by the unmeasured diagnoses needs further clarification, especially 1-2 of prescriptions is related to CKD. Actually, I think it is impossible for 1-2 of prescriptions with CKD risk. It is very strange. 2. What is the exact definition of recent use of NSAIDs? How recent? 3. It is also very strange for the SGA with CKD risk only noted in No Prior AKI, No Hypertension, no Diabetes, and no NSAID groups. It may imply the increased risk of CKD with SGA may occur in such a healthy group. Is it really possible? 4. SGA with mild or high, but not moderate, risk of metabolic syndrome is related to CKD. It is also very strange and conflicting. 5. Finally, I think the very small OR of SCA with increased CKD may be a pseudo-positive finding owing to many unmeasured confounding bias. Thanks.

REVIEWER	Ryan M Seals Optum Epidemiology
REVIEW RETURNED	03-Apr-2020

GENERAL COMMENTS	The authors present a brief and well-written report on the association between SGA and CKD using a case-control study. The analysis is straightforward. I have minor concerns about the selection process, along with possible suggestions for how to address these or investigate their potential impact on the results. Specific comments:

	Page 6 “To ensure that controls had not developed CKD since their last creatinine measurement, all controls were required to have a creatinine recorded at least one year after the index date.” -Is this correct? I am concerned that this requirement may create selection bias because of who receives eGFR. It would be informative to see either a sensitivity analysis without this requirement, or to see the characteristics of those who were otherwise eligible controls but without the normal eGFR measurement in the 1y following index date. The risk of inducing selection bias via this mechanism seems to me to outweigh the benefit gained from decreasing measurement error in the measurement of CKD using future eGFR. I would moreover discuss this sampling framework in the discussion, either explaining the validity or possibly raising limitations. Table 1 -Please include prior AKI in table 1 (and consider a formal positive control analysis using AKI as described below) Table 2 -How were the cumulative dose categories determined? Consider a more flexible approach (e.g. splines) to investigate the fuller dose-response curve particularly as the highest groups appears suggestive. Figure 1 -I applaud the use of these types of figures. There appears to be some discrepancy between the figure and the text, however. In the text, cases are required to have all eGFRs in the first 3m be < 60, as well as the first eGFR after 3m. The figure indicates the need for a median eGFR<60 between 3m and 1y following the first eGFR<60. These do not seem equivalent to me. -The figure also seems to indicate prior AKI as an exclusion criterion, which is not described in the text, and because it was assessed in subgroup analyses, I do not believe this is possible. Methods -Please add a section describing all covariates in one place as well as how they were assessed (NDC, ICD9/10, etc…) Results -The authors state that hypertension and diabetes were more prevalent in the cases than controls. It would be nice to see this as a more formal positive control analysis, in which those exposures are assessed in the same conditional logistic model as the main SGA exposure to ensure that the case-control scheme is correctly picking up the association with those known risk factors. The crude percentages are less informative in a matched case-control analysis. -I would further consider looking at other known medication risk factors for CKD to ensure that the proper risk factors are being captured -The authors may also consider a negative control exposure to further assess the validity of the case-control sampling scheme, particularly one that captures an individual’s propensity to engage with the healthcare system. One example may be the receipt of a regular cancer screening in the prior year, though others may be

	more appropriate. This may inform whether or not the controls are being preferentially selected from among healthy individuals with heavier healthcare engagement.

VERSION 1 – AUTHOR RESPONSE

COMMENTS FROM REVIEWER #1:

Comment #1: 557 for Second-generation antipsychotics, but 74 for schizophrenia, 120 for bipolar disorder, 122 for dementia. What else diagnoses for Second-generation antipsychotics? Delirium? Or others? Whether the association between Second-generation antipsychotics and CKD is confounded by the unmeasured diagnoses needs further clarification, especially 1-2 of prescriptions is related to CKD. Actually, I think it is impossible for 1-2 of prescriptions with CKD risk. It is very strange.

Author response:
Thank you for this comment. We have added additional categories on mental illness to table 1 but were not able to accurately capture the remaining SGA users. We have not added "history of delirium" to table 1 as this diagnosis is inconsistently reported in Danish Registers. The excess number of SGA users is then most likely due to SGAs being used for sedative-hypnotic properties which might not result in hospital diagnoses. The relatively high proportion of SGA users with few prescriptions might also reflect this use, and we certainly agree that this category might contribute to residual confounding and have added further clarification of this to the discussion.

Changes to manuscript:
Added to the discussion: "Furthermore, the observed increase in risk associated with use of few prescriptions is suggestive of some degree of residual confounding." (page 10, bottom) and "Our finding of modest increases in risk of CKD with SGAs, does not suggest any clear association between these. Furthermore, the presence of an increased risk with few antipsychotic prescriptions is indicative of some degree of residual confounding. Therefore, we do not believe that SGAs by themselves increases the risk of CKD, but rather contribute to metabolic disturbances which in the end results in kidney damage. The increased risk among SGA users with diabetes adds to this interpretation." (page 11, mid)

Comment #2: What is the exact definition of recent use of NSAIDs? How recent?

Author response:
Thank you for noting this shortcoming in our manuscript. Recent use of NSAIDs was defined as use of NSAIDs in the past year. We have added this clarification to a new subsection on covariates.

Changes to manuscript: Added a subsection to the methods section named "Covariates":
"We included the following potential confounders in our analysis: i) Age, sex and calendar time (accounted for by sampling procedure), ii) use of other drugs known to affect renal function (lithium, NSAIDs), iii) history of hypertension and diabetes, and iv) highest achieved level of education as a proxy for socioeconomic status. Use of lithium was defined as any filling of prescriptions for lithium before the index date. Recent use of NSAIDs was defined as filling of prescriptions within one year before the index date. Relevant ICD-10 diagnoses and ATC-codes are listed in appendix 1." (page 6, mid)

Comment #3: It is also very strange for the SGA with CKD risk only noted in No Prior AKI, No Hypertension, no Diabetes, and no NSAID groups. It may imply the increased risk of CKD with SGA may occur in such a healthy group. Is it really possible?

Author response:
The results referred in table 3 show a slightly increased relative risk (odds ratio) for individuals without prior acute kidney injury (AKI) compared to individuals with a history of AKI (OR 1.27 and 0.96). The same pattern is observed when stratifying individuals on age (<65 or ≥65 years, OR 1.50 and 1.13). At first, this may seem strange, as one can assume that the risk of CKD is lower in the groups with the higher relative risk (no prior acute kidney injury, age below 65) than in their counterparts. We explored the absolute risk of CKD in the Funen Laboratory Cohort for these groups and found a lower absolute risk in individuals younger than 65 years compared to individuals above 65 years (Risk: 3.4% and 16.1%) and the same for prior acute kidney injury (Risk: 4.6% and 40.8%). In this case, a small increase in absolute risk related to exposure to SGA will result in a larger increase in relative risk in the group with a lower risk of CKD. We have added these results on absolute risk to the results section and further discussion of this phenomenon in the discussion.

Regarding the differences in relative risk when stratifying on a prior history of diabetes and hypertension, the relative risks for both groups are very close with overlapping 95% confidence intervals, which we interpret as some clinically relevant risk for history diabetes (OR 1.24 vs 1.52) given the difference in point estimates, while the association with history of hypertension is not clear.

Changes to manuscript:
Added the following sentences to the results section:
The absolute risk of CKD in this population was 3.4% for individuals <65 years, 16% for individuals ≥65 years. For individuals with prior AKI the absolute risk was 40.8% versus 4.6% for individuals without prior AKI. (page 8, bottom)

Added the following sentences to discussion: “Our finding of the highest risk among the younger age group (table 3) might be explained by the low absolute risk observed in this age group, resulting in greater increases in relative risk, when exposed to SGAs.” (page 10, mid) and “Furthermore, the observed increase in risk associated with use of few prescriptions is suggestive of some degree of residual confounding.” (page 10, mid)

Comment #4: SGA with mild or high, but not moderate, risk of metabolic syndrome is related to CKD. It is also very strange and conflicting.

Author response:
The reviewer points out, that SGA with a mild (OR 1.21, 95% CI: 1.06-1.39) and high risk (OR 1.36, 95% CI: 1.11-1.68) of metabolic side effects are seemingly associated to CKD, while SGA with a moderate risk of metabolic side effects (OR 1.19, 95% CI: 0.96-1.48) are not.
This interpretation may be due to the lower bound of the 95% CI for mild and high risk SGA being greater than 1.00, while the 95% CI for moderate risk includes unity. This dichotomization of the confidence interval disregards the large overlap between the confidence intervals for mild and moderate risks SGAs. We rather think that these findings indicate no clinically relevant difference in relative risk between mild and moderate risk SGAs, but a tendency towards a slightly increased risk of CKD in SGAs with a high risk of metabolic side effects.

Changes to manuscript: None

Comment #5: Finally, I think the very small OR of SCA with increased CKD may be a pseudo-positive finding owing to many unmeasured confounding bias.

Author response:
Thank you for this comment. We agree with that the small increases in odds ratio is, most likely, due to residual confounding and have added further clarification of this precaution to the discussion as described above.

Changes to manuscript: please refer to “comment #3” for changes to manuscript.

COMMENTS FROM REVIEWER #2:

The authors present a brief and well-written report on the association between SGA and CKD using a case-control study. The analysis is straightforward. I have minor concerns about the selection process, along with possible suggestions for how to address these or investigate their potential impact on the results.

Authors response:
Thank you very much for the kind words about our manuscript and for your comments that certainly have improved it.

Page 6
“To ensure that controls had not developed CKD since their last creatinine measurement, all controls were required to have a creatinine recorded at least one year after the index date.”
-Is this correct? I am concerned that this requirement may create selection bias because of who receives eGFR. It would be informative to see either a sensitivity analysis without this requirement, or to see the characteristics of those who were otherwise eligible controls but without the normal eGFR measurement in the 1y following index date. The risk of inducing selection bias via this mechanism seems to me to outweigh the benefit gained from decreasing measurement error in the measurement of CKD using future eGFR. I would moreover discuss this sampling framework in the discussion, either explaining the validity or possibly raising limitations.

Author response:
Thank you, we agree that this choice may create a selection bias, as more frequent creatinine measurements may be due to higher disease severity or burden of disease. To explore the impact of this choice on our results, we have added a sensitivity analysis (as appendix 4) where eligible controls were not required to have a normal eGFR measured in the year after the assigned index date. Results were mainly unchanged, and we have added a short description of this additional analysis to the methods and results sections.

Changes to manuscript
Addition of appendix 4 - Sensitivity analysis with modified eligibility criteria for controls.

Addition of description to methods section: “We conducted a sensitivity analysis where eligible controls where not required to have normal eGFR measurement(s) in the year following the index date to assess the potential of selection bias with this criterion.” (page 6, bottom)

Addition to description to results section: “Sensitivity analyses: The risk of CKD in relation SGA exposure was largely unchanged, when including controls who were not required normal eGFR measurements in the year following their assigned index date (appendix 4).” (page 9, top)

Table 1
-Please include prior AKI in table 1 (and consider a formal positive control analysis using AKI as described below)

Author response:
Thank you for this comment. We have added “Prior AKI” among the characteristics in table 1.

Changes to manuscript: Addition of “Prior AKI” in table 1 (page 16).

Table 2
-How were the cumulative dose categories determined? Consider a more flexible approach (e.g. splines) to investigate the fuller dose-response curve particularly as the highest groups appears suggestive.

Author response:
Cumulative dose was stratified according to an assumed duration of use (1 Defined daily dose/10mg olanzapine per day, resulting in strata for 3 months, 3-6 months, 6-12 months and 1+ year of treatment).
To explore whether a more flexible modelling approach yielded a different dose response curve, we performed conditional logistic regression amongst all users of SGA, as suggested using restricted cubic splines. CKD case status was used as the dependent variable, while cumulative dose of SGA was used as the independent variable. Three knots were placed at the value for the 10th, 50th and 90th percentile.
As expected, this yielded a more uniform dose-response relationship (appendix 3), with risk of CKD increasing slightly with increasing cumulative dose of SGA until about 900-1000mg olanzapine equivalents.
Whether the restricted cubic spline or stratified analysis is closer to the true biological effect, cannot be determined. We prefer the stratified analysis, as fewer assumptions need to be satisfied using this approach.

Changes to manuscript:
Addition to the methods section: “To explore a potential dose response relation, we performed a supplementary analysis, using conditional logistic regression amongst all users of SGA and restricted cubic splines with knots placed at the value for the 10th, 50th and 90th percentile for cumulative doses.” (page 6, bottom).

Addition to the results section: “Additional analysis of the association between cumulative dose of SGAs and the risk of CKD, yielded a somewhat uniform dose-response relationship, with risk of CKD increasing slightly with increasing cumulative dose of SGA until approximately 900-1000mg olanzapine equivalents (appendix 3).” (page 9, top).

Addition of appendix 3 – a figure showing the association between CKD and cumulated dose of SGA modeled using restricted cubic splines.

Figure 1
-I applaud the use of these types of figures. There appears to be some discrepancy between the figure and the text, however. In the text, cases are required to have all eGFRs in the first 3m be < 60, as well as the first eGFR after 3m. The figure indicates the need for a median eGFR<60 between 3m and 1y following the first eGFR<60. These do not seem equivalent to me.
-The figure also seems to indicate prior AKI as an exclusion criterion, which is not described in the text, and because it was assessed in subgroup analyses, I do not believe this is possible.

Author response:
Thank you for identifying this discrepancy between text and figure 1. We have corrected figure 1 to match the main text.

Changes to manuscript: Revision of figure 1 (removing prior AKI as exclusion criterion and the requirements for eGFR in the time following the index date).

Methods
-Please add a section describing all covariates in one place as well as how they were assessed (NDC, ICD9/10, etc…)

Author response:
We thank the reviewer for this suggestion to strengthen our reporting of the design. We have added a subsection entitled “Covariates” to the methods section (page 6).

Changes to manuscript: Addition of the subsection “Covariates”:
“We included the following potential confounders in our analysis: i) Age, sex and calendar time (accounted for by sampling procedure), ii) use of other drugs known to affect renal function (lithium, NSAIDs), iii) history of hypertension and diabetes, and iv) highest achieved level of education as a proxy for socioeconomic status. Use of lithium was defined as any filling of prescriptions for lithium before the index date. Recent use of NSAIDs was defined as filling of prescriptions within one year before the index date. Relevant ICD-10 diagnoses and ATC-codes are listed in appendix 1.” (page 6)

Results
-The authors state that hypertension and diabetes were more prevalent in the cases than controls. It would be nice to see this as a more formal positive control analysis, in which those exposures are assessed in the same conditional logistic model as the main SGA exposure to ensure that the case-control scheme is correctly picking up the association with those known risk factors. The crude percentages are less informative in a matched case-control analysis.

Author response:
Thank you for this suggestion. We have adapted this and included results for the association between risk factors and CKD as appendix 2.

Changes to manuscript:
Methods section:” Furthermore, we conducted control analyses to assess the association between CKD and risk factors (history of diabetes or hypertension, and use of lithium or NSAIDs), and between a negative control exposure and CKD (topical ocular antibiotics – not considered associated with CKD).” (page 7, top)

Results section: ”ORs for the association between risk factors and CKD presented in appendix 2.” (page 8, top)

Tables: Removal of crude percentages from tables 2 and 3 (page 17-18). Addition of appendix 2, a table showing OR association between risk factors, negative control exposure and CKD.

-I would further consider looking at other known medication risk factors for CKD to ensure that the proper risk factors are being captured

Author response:
We agree that it could strengthen the analysis to include use of other drugs known to cause kidney damage as risk factors. However, these are primarily used in hospitals (or dispensed from out-patient clinics) and thus not captured by our data sources (i.e., aminoglycosides, vancomycin, antiretrovirals, calcineurin-inhibitors, chemotherapy and x-ray contrast). Therefore, we are not able to include these in our analysis.

Changes to manuscript: Addition to the discussion (limitations):
“Secondly, we were not able to adjust for use other potentially nephrotoxic drugs26 (besides lithium and NSAIDs), as these are primarily used in hospitals (i.e., aminoglycosides, chemotherapy, or x-ray contrast) or dispensed from out-patient clinics (i.e., antiretrovirals, or calcineurin-inhibitors), and thus not captured in our data sources.” (page 11, top)

-The authors may also consider a negative control exposure to further assess the validity of the case-control sampling scheme, particularly one that captures an individual’s propensity to engage with the healthcare system. One example may be the receipt of a regular cancer screening in the prior year, though others may be more appropriate. This may inform whether or not the controls are being preferentially selected from among healthy individuals with heavier healthcare engagement.

Author response:
Thank you to the reviewer for this suggestion, which would indeed strengthen the analysis. However, we are not able to assess health care utilization as cancer screening or use of primary care, as we do not have these data available. Therefore, we have included an additional analysis among the appendices (appendix 2), where we use ocular topical antibiotics as exposure to test the validity of our sampling and analysis. This control analysis shows no association between use of ocular topical antibiotics and CKD (crude OR 0.95, 95%CI 0.90-1.00 and adjusted OR 0.93, 95%CI 0.88-0.98).

Changes to manuscript:
Addition of negative control analysis to the supplementary material in appendix 2 (page 3 of the supplement).

Other comments:
Comment #1: Please re-upload your Supplementary files in PDF format.

Changes to manuscript:
The supplementary material has been converted to pdf-format.

Comment #2: Figure 2 citation missing

Changes to manuscript:
Citation of figure 2 has been added to the paragraph on “Specific SGAs” in the results section (page 7) and to the discussion of these results (page 8).

VERSION 2 – REVIEW

REVIEWER	Ryan Seals Optum Epidemiology
REVIEW RETURNED	29-May-2020

GENERAL COMMENTS	The authors have successfully addressed many of the concerns raised in the initial review. However, some minor revisions should be made: In the dose-response analysis using splines, the range of cumulative use of olanzapine equivalents seem to reach a maximum at 1500. In Table 2 of the text, the two higher groups are both above this limit. Please explain why the ranges do not match, or extend the dose-response to the full range of data. I also

	suggest displaying a "rug plot" of exposure values to show visually the distribution of cumulative dose exposures. Please also provide the values for the 10th, 50th, and 90th percentiles of exposure, and explain if these were chosen among all (cases and controls) or only among cases. I recommend choosing the cut points among cases only. Please address and interpret the positive and negative control results. It appears that known risk factors are associated with CKD in your analysis, which lends support to the results. The near-null or slightly decreased is more difficult to interpret, but may suggest some degree of negative confounding.

VERSION 2 – AUTHOR RESPONSE

COMMENTS FROM THE REVIEWER:

Comment #1: The authors have successfully addressed many of the concerns raised in the initial review. However, some minor revisions should be made:

Author response: Thank you to the reviewer for accepting our revisions. This reviewer's comments and suggestions have certainly improved the manuscript.

Comment #2: In the dose-response analysis using splines, the range of cumulative use of olanzapine equivalents seem to reach a maximum at 1500. In Table 2 of the text, the two higher groups are both above this limit. Please explain why the ranges do not match, or extend the dose-response to the full range of data. I also suggest displaying a "rug plot" of exposure values to show visually the distribution of cumulative dose exposures.

Author response: Thank you to the reviewer for noting this discrepancy. We have extended the x-axis of the figure in appendix 3 to 15,000 mg olanzapine equivalents to cover the two higher dose categories. We agree that a rug plot would convey important information on the distribution of cumulative doses. Unfortunately, our data sources do not allow the display of individual-level data like this and we cannot include such visualization.

Changes to manuscript: Revised figure in appendix 3 with the extended x-axis.

Comment #3: Please also provide the values for the 10th, 50th, and 90th percentiles of exposure, and explain if these were chosen among all (cases and controls) or only among cases. I recommend choosing the cut points among cases only.

Author response: Thank you to the reviewer for this suggestion. We have included values for the percentiles in connection to the revised figure in appendix 3. Percentiles were chosen among cases and this information has been added to the methods description and the figure legend.

Changes to manuscript:

Methods section: "…among cases." (manuscript, page 6, bottom)

Appendix 3: Addition of percentile values to figure legend in appendix 3 along with clarification of cut points. "Percentiles were derived from cumulative doses among cases" (supplementary material, page 4, top)

Comment #4: Please address and interpret the positive and negative control results. It appears that known risk factors are associated with CKD in your analysis, which lends support to the results. The near-null or slightly decreased is more difficult to interpret, but may suggest some degree of negative confounding.

Author response: We agree with this interpretation of the positive control analysis – that the know/assumed risk factors are associated with CKD. We have added more text on this interpretation to the results section. Regarding the slightly reduced OR with the use of ocular topical antibiotics, we believe that it might arise from a population that is, on average, healthier than the other patients who have creatinine measurements. However, the magnitude of this OR is close to unity.

Changes to manuscript:

Results section: "Control analyses confirmed that each of the assumed risk factors included in the model was positively associated with increased risk of CKD and that a negative control exposure was not associated with increased risk of CKD (appendix 2)." (page 8, mid).