Author's response to reviews

Title: The Apparent Diffusion Coefficient (ADC) Ratio: Can it be Used as an Adjuvant Tool for Prostate Cancer Assessment of Tumor Aggressiveness?

Authors:

Andrei Lebovici (andrei1079@yahoo.com)
Silviu A Sfrangeu (ssfrangeu@yahoo.com)
Diana Feier (diana_feier@gmail.com)
Cosmin Caraiani (ccaraiani@yahoo.com)
Ciprian V Lucan (lucan_valerian@yahoo.com)
Mihai Suciu (suciu_umf@yahoo.com)
Florin Elec (florinelec@hotmail.com)
Gheorghita Iacob (iacob.gheorghita@gmail.com)
Mircea Buruian (mburuian@radiologietgm.ro)

Version: 2 Date: 26 November 2013

Author's response to reviews:

Adrian Aldcroft
The BioMed Central Editorial Team
Cluj-Napoca, 20.11.2013

Dear Editor,

Thank you very much for investing your valuable time in handling our manuscript MS: 9104375321015565 entitled “The Apparent Diffusion Coefficient (ADC) Ratio: Can it be Used as an Adjuvant Tool for Prostate Cancer Assessment of Tumor Aggressiveness?” and considering its acceptance in BMC Medical Imaging after proper revision. We also would like to thank the reviewers for the helpful suggestions and for giving us the opportunity to improve our manuscript accordingly and we appreciate all your efforts. We have specifically addressed all issues and concerns outlined by yourself and the reviewers in a detailed point-by-point revision and made the appropriate changes and supplementations in the marked manuscript. Please find the details below.

Response to Reviewer’s remarks

Reviewer 1

We thank you again for your helpful and constructive comments. We detailed our responses as listed point by point below.

“LT1 “These relatively new therapeutic methods come as an alternative option to existing treatment modalities such as active surveillance,” The authors refer to HIFU as an alternative to active surveillance which is untrue.”
We agree with your comment. We redrafted the paragraph accordingly in the revised version of the manuscript by deleting the words “active surveillance”.

*LT2 “Several functional imaging techniques are available, like MR spectroscopy (9) and dynamic contrast enhancement MR (10), but not widely accessible.” MRS is not widely accessible but DCE-MRI is a routine sequence on all scanners and one which is recommended in Guidelines (Use of multi-parametric MR imaging for prostate cancer) for many countries in Europe and in the USA. It takes 5 minutes to acquire and although contrast is administered the cost is not exorbitant.

We agree with your comment. In our daily practice we try as much as possible to follow all recommended guidelines but, due to several financial issues we are not always able to perform DCE-MR. We are aware that the financial situation in our country is not representative and it should not interest all readers. We apologize for the misleading information and we redrafted the paragraph accordingly in the revised version of the manuscript.

*LT3 “shortage of available tools for contrast enhancement analysis, possible variability in results dependent on post-processing and a significant increase in the number of images for analysis and stocking make these two techniques time consuming and expensive and difficult to integrate in the routine examination in many centers.” All MR systems come equipped with the basic tools for analysis of contrast uptake characteristics. This can be performed on the operator console if a workstation is not available. Admittedly there is intra-and inter-reporter variability but this will also be true of analysis of DWI data.

As already mentioned in the previous answer, we agree with your comment, we redrafted the paragraph accordingly in the revised version of the manuscript.

*LT4 “The requirement for institutional review board approval was waived.” The author should state why this was allowed.

We are sorry for the confusion. Regularly, our patients give written, informed consent prior to any MR examination, including answering questions regarding pacemakers, implants, and, in particular, renal impairment and allergy in case of contrast-enhanced MRI. Furthermore, patients, who undergo interventional procedures, such as biopsy etc., also give written, informed consent. These informed consents were reported; however, the ethics committee waived an additional informed consent for the retrospective analysis of these data. We have clarified this situation in the manuscript as follows:

“Written, informed consent had been obtained from patients prior to the MR examinations, as well as before all therapeutic interventions, such as prostatic biopsy. However, the necessity to obtain written, informed consent for the retrospective analysis of the data was waived.”

*LT5 “All patients underwent endorectal MRI composed of T2W-imaging and DWI at 4 to 6 weeks after PCa confirmation by biopsy” It is recognised that interpretation of MR prostate examinations within this window is difficult and that haemorrhage may result in low SI areas on T2W imaging. Inflammatory changes
can occur surrounding areas of haemorrhage – do the authors know to what extent they may have influenced ADC measurements as a cellular infiltrate would increase the cell density and hence the ADC value.

We appreciate your comment and we understand your concern. Our protocol included T2W and T1W images which were viewed for detecting post biopsy hemorrhage and allowed us to detect and exclude 5 patients due to post-biopsy hemorrhage. This information is already included in the Material and Methods section.

*LT6 “Results were reported as cancer with an assigned Gleason score or as benign tissue.” The biopsy specimens would have included a variable amount of normal or benign tissue with the cancerous tissue. This would change the ADC values depending on the % of cancer within the cores. So for example would a core containing 90% Gleason grade 6 tissue give the same value as a core containing 25% of Gleason Grade 9 tumour. I think without this information the data is difficult to interpret.

Thank you for your valuable observation. At your suggestion we compared the length of biopsy specimen and percentage of tumor in biopsy cores with no statistically significant difference between segments with Gleason score of 6 and 7 and sectors with Gleason score of 8 and 9.

We also inserted the data in Table 2 and in the results section, as follows:

“We found no significant difference between the two studied groups regarding the length of biopsy specimen (p=0.8) and the percentage of tumor material in biopsy cores.”

*LT7 Table 1. MRI parameters’ description.

<table>
<thead>
<tr>
<th>T2W-MRI</th>
<th>DW-MRI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sequence</td>
<td>Fast spin echo</td>
</tr>
<tr>
<td>TR (ms)</td>
<td>5500</td>
</tr>
<tr>
<td>TE (ms)</td>
<td>104</td>
</tr>
<tr>
<td>Flipangle</td>
<td>150°</td>
</tr>
<tr>
<td>FOV (mm²)</td>
<td>180 x 180</td>
</tr>
<tr>
<td>Matrix</td>
<td>256 x 256</td>
</tr>
<tr>
<td>Voxel size (mm³)</td>
<td>0.8 x 0.8 x 3 2.3 x 2.3 x 3</td>
</tr>
<tr>
<td>Slice thickness (mm)</td>
<td>3 4</td>
</tr>
<tr>
<td>Gap (mm)</td>
<td>0.3 0.3</td>
</tr>
<tr>
<td>Spectral suppression</td>
<td>No</td>
</tr>
<tr>
<td>b-values</td>
<td>0/400/800</td>
</tr>
<tr>
<td>NEX</td>
<td>1 2</td>
</tr>
<tr>
<td>TA (min:sec)</td>
<td>4:09 2:40</td>
</tr>
</tbody>
</table>

In the table above the voxel size for DWI does not correspond with the slice.
thickness.

Thank you for your observation. We corrected the information in the revised version of the manuscript, with a voxel size for DWI of 2.3 x 2.3 x 4.

*LT8 “regions of interest (ROIs) were placed according to biopsy results and the prostate reporting scheme. Lesions with low signal intensity compared to surrounding tissue on ADC were considered malignant. ROIs were drawn to occupy around 75% of the lesions to be sure that lesion margins would not be calculated. ROIs were drawn also in the tumor free sectors, with a standard ROI size of 0.8 cm2.”

From the quality of the DWI images contained in the manuscript it would only have been possible to detect relatively large areas of malignancy – hence potentially the high pick-up rate of high grade disease. The method used to delineate a ROI seems haphazard – either dependent on the pathology results and the empirical chart or using areas of low SI seen on DWI. These low SI areas can also be due to extensive fibrous stroma – how did the authors differentiate the 2 pathologies if DCE-MRI was not obtained? When the authors state that the ROIs were drawn to incorporate 75% of the lesion how could they reliable do this without better delineation of the tumour boundary? This would also be problematic on T2W imaging so soon after biopsy. The authors would have been better advised to perform MRI prior to biopsy and then perform saturation biopsies of areas of abnormality, by trans-perineal template approach, if a method of co-registering the MRI with TRUS guided biopsies was not available.

Even if the printed image quality may look suboptimal, the image interpretation was performed on a PACS system with high quality imagines, good resolution and good lesion detection. The displayed image was initially provided in order only to demonstrate the way we calculated the ADC ratio. But we agree with your comment and realize that the information is misleading the reader therefore we replaced the image form Figure 1 with an other image correctly demonstrating placement of ROIs on approx. 75% of a lesion in a given segment. We also redrafted the information in the Material and Methods section, as follows:

“ROIs were drawn to occupy around 75% of the lesion in a given segment to be sure that lesion margins would not be calculated. “

In order to avoid measuring ADC values from fibrotic areas, we only considered for evaluation lesions from segments with confirmed prostate cancer according to biopsy results. The readers were aware to the results of biopsy and clinical data, as already mentioned in the manuscript.

We also accept the fact that the MRI examination should have been better performed prior to biopsy and we consider this as a limitation of our study. We were only able to perform the MRI examinations after biopsy procedures dew to the retrospective inclusion of patients with already confirmed prostate cancer.

In order not to confuse the reader, we also included this limitation in the discussion section of the manuscript, as follows:
“(4) Prostate biopsy was performed prior to MRI examinations, which might have influenced the ADC measurements.”

*LT9 “ADC-ratios were calculated by dividing tumoral ADCs and tumor-free ADCs form the peripheral and the central zone of the gland (Figure 2(A) and 2(B)). The highest ADCs of non-tumoral sectors were taken as reference.” Do the authors know exactly what type of tissue was present in the “tumour-free” ROIs? They might have included fibrosis which would lower that ADC value.

We understand your concern. The tumor-free zone was selected from areas with maximal signal intensities on ADC maps, therefore reducing the presence of fibrotic area. We also excluded segments were the pathologist reported fibrotic, atrophic or inflammatory changes.

In case of diffuse fibrotic changes involving the entire prostate, the decrease in signal intensity on ADC maps is excluded when measuring the ADC ratio, by being present both in tumor free zones as well as in malignant lesions. This is a good example of the practical use of the current ADC ratio, by eliminating background inter-patient variations of prostatic signal intensity.

*LT10 “On saturation biopsy, cancer was detected in 157 of the 440 cores (35.68%). After assigning the cancer positive cores to the 16 region standardized prostate reporting scheme, 128 out of 352 regions were cancer positive (36.07%). ... two patients had all 20 biopsy cores positive. “ Although 128 of the 352 regions biopsied were positive, 40 regions were derived from 2 patients i.e. 31.3% of the positive results were from 2 patients. Does this not significant bias the data? Would similar trends be apparent without these patients? The study numbers are small enough already.

We agree that the study population was relatively small, a fact that represents a limitation of our study.

The two patients with large tumors had different Gleason scores and different ADC values in different segments. We included these segments and considered them as individual tumors. These tumors are of great interest, showing high cellular variability with different Gleason scores. In this cases, we consider that it is important to lead the biopsy needle in the most aggressive areas with the lowest ADC values.

In order to answer your question regarding the bias of data, we tested the diagnostic accuracy of ADC measurements by performing AUROC analysis without including the segments of these two patients, obtaining similar results (new AUROC for tumor ADC 0.72, for ADC-CR=0.75 and for ADC-PR, p value=0.2, according to the pairwise comparison of ROC curves using the DeLong method)

*LT11 For a cut-off level of equal or lower than 0.82 x 10-3 ± 0.32 mm2/sec the tumor ADCs for Gleason 8 and 9 positive sectors showed a specificity and specificity of 72.7% and 70.6% while the AUROC was 0.75. Using a cut-off value of equal or lower then 0.57, the ADC-CR showed more promising results with
AUROC of 0.77 (Sensitivity=82.2% and Specificity=66.7%) (table 3). The best diagnostic performance was obtained for ADC-PR, with an AUROC of 0.84, when applying a cut-off value equal or less than 0.5. The authors make no mention of how these threshold values have been obtained and this should be corrected.

In our Material and Methods section we detailed the statistical analysis of our data. Optimal cut-off values were chosen to maximize the Youden index and sensitivity (%) and specificity (%) were computed from the same data, without further adjustments. Discriminative capability was examined per tumor unit and not per patient, due to the low number of available patients.

*LT12 Table 4. The cut-off values and diagnostic performance of ADC-Peripheral Ratio in predicting Gleason 8 and 9 stages, according to tumour location.

<table>
<thead>
<tr>
<th>Central zone tumors</th>
<th>Peripheral zone tumors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cut-off &lt;=0.42 &lt;=0.50</td>
<td>Cut-off &lt;=0.42 &lt;=0.50</td>
</tr>
<tr>
<td>Sensitivity 62.1 93.7</td>
<td>Sensitivity 62.1 93.7</td>
</tr>
<tr>
<td>Specificity 92.7 80</td>
<td>Specificity 92.7 80</td>
</tr>
<tr>
<td>AUROC 0.81 0.90</td>
<td>AUROC 0.81 0.90</td>
</tr>
</tbody>
</table>

p value <0.001 <0.001

Note: AUROC=Area under the receiver operating characteristic curve.

This table presumably refers to the results for both central gland and peripheral zone tumours.

Table 3 refers to DWi cut-off values and Performance Values of tumor ADC, ADC-CR and ADC-PR in predicting Gleason 8 and 9, independent of the location of the tumor. We updated the table legend for better understanding.

* Minor Essential Revisions

*LT13 There are large number of primarily grammatical errors and some spelling mistakes – too numerous to mention individually but which make the article frustrating to read. It does NOT flow! It seems to come to a rather abrupt ending with a list of potential limitations.

Thank you for your observation. We asked the help of a native English speaker in order to overcome our spelling mistakes.

Reviewer 2

*HM1 Normal central and peripheral zone ADC values in group with Gleason 6 and 7 were significantly lower than those with Gleason 8 and 9. This finding appears to be very interesting. Please explain the reason if possible, at least in part, speculation.

Thank you for your valuable comment. In our study we have shown that the ADC values in group with Gleason 6 and 7 were significantly lower than those with Gleason 8 and 9, which cannot be solely attributed to measurement variability.
As an additional observation, patients with Gleason 8 and 9 were significantly younger patients \( (p<0.0001) \). We hypothesize that the differences arise from natural variations in prostate physiology in younger patients with less significant fibrotic and atrophic changes of prostate glandular structure that would decrease the ADC measurements \( (1) \). We appreciate your observation and consider this aspect one worth to be mentioned, therefor we introduced the explanation in the Discussions section, as follows:

“When comparing the two studied groups, all ADC values in group with Gleason 6 and 7 were significantly lower than those with Gleason 8 and 9, which cannot be solely attributed to measurement variability. As an additional observation, patients with Gleason 8 and 9 were significantly younger patients \( (p<0.0001) \). We hypothesize that the ADC differences arise from natural variations in prostate physiology in younger patients, due to less significant fibrotic and atrophic changes of prostate glandular structure that would decrease the ADC measurements. \( ( ) \).”

*HM2 In this study, if you include T2W for detecting tumors, difference of AUC between two groups remained unchanged?

Thank you for your comment. All patients included in our study had conformed prostatic tumors. The purpose of our study was to assess the tumor aggressiveness and not their detection and therefore we did not test the diagnostic accuracy of T2w images. But in future, prospective studies we plan to perform such an analysis in order to compare it with the diagnostic accuracy of DWI. Adding morphological sequences like the T2w to functional ones, like DWI will surely increase the detection rate of prostatic tumors.

*HM3 Purpose of this study seems to use DW-MRI before biopsy. Please comments in the Discussion.

Thank you for your suggestion. We accept the fact that the MRI examination should have been better performed prior to biopsy and we consider this as a limitation of our study. We were only able to perform the MRI examinations after biopsy procedures due to the retrospective inclusion of patients with already confirmed prostate cancer.

In order not to confuse the reader, we also included this limitation in the discussion section of the manuscript, as follows:

“(4) Prostate biopsy was performed prior to MRI examinations, which might have influenced the ADC measurements.”

Again, we would like to thank the Editor and all the Reviewers for their contribution towards the improvement of the manuscript.

We would be grateful for consideration of the present manuscript to be published in your highly esteemed journal “BMC Medical Imaging”.

Diana Feier, MD
Radiology Department,
Emergency County Hospital,
Clinicilor, Nr. 3-5, 400006
Cluj-Napoca, Romania
diana.feier@gmail.com
Tel:+40-740-573-872