TEXTURE AND MATHEMATICAL MORPHOLOGY FOR HOT-SPOT DETECTION IN WHOLE SLIDE IMAGES OF MENINGIOMAS AND OLIGODENDROGLIOMAS

Z. Swiderska*, T. Markiewicz*,†, B. Grala†, W.Kozlowski†

†Department of Pathomorphology, Military Institute of Medicine, Warsaw, Poland

*Warsaw University of Technology, Warsaw, Poland

Corresponding address: Warsaw University of Technology, Dept. of Electrical Eng, Pl. Politechniki 1, 00-661Warsaw, Poland, Tel.+48 22 234-72-17, e-mail: swidersz@ee.pw.edu.pl

Background

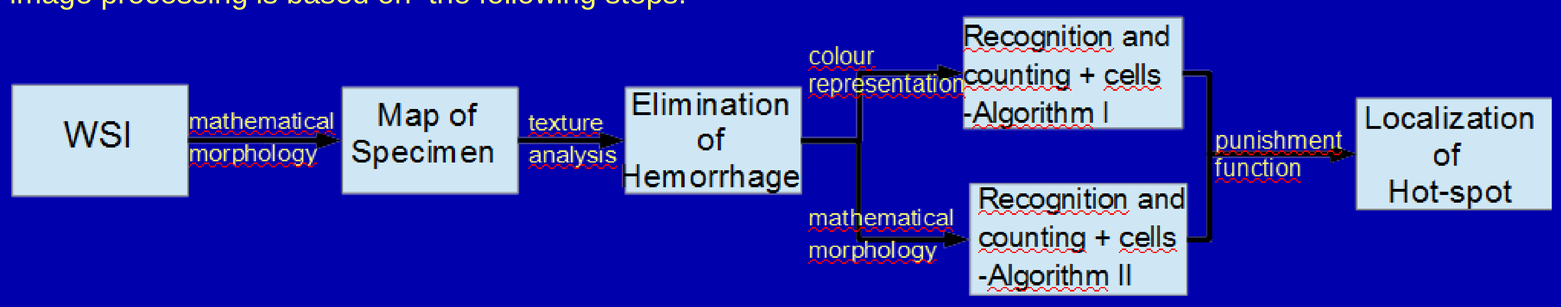
Histopathological examination of tissues using immunostaining tests is a basic method of identifying tumours. It often serves as a tool to support the choice of optimal therapy and defines the prognostic indicators. Tumour proliferation in central nervous system tumours can be characterised with the widely used Ki-67/MIB-1 marker. The immunopositive (proliferated) cell nuclei are marked with brown, whereas the cell nuclei are marked with blue, and their ratio gives the proliferation index. Thus, the automatic quantitative evaluation of the specimen can offer a very useful tool for the pathologists.

The areas with high immunopositive reactions are called hot-spots. For objective evaluation, some scattering of selected hot-spots is also recommended. One of the schemes is the choice of fields of view with the highest levels of Ki-67 and then choosing several adjacent regions with positive reaction in short distance from this field, followed by the search for other areas of specimen. The major problem in the tested formulations are hemorrhages, which are stained with brown creating false-positive results. Correct classification of tumour and hemorrhages areas allow for accurate identification of hot-spot areas and may help improve sample analysis.

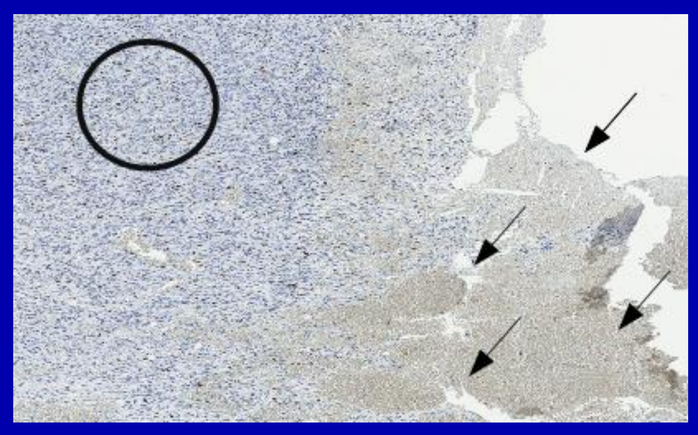
Methods

The fifteen cases of meningiomas and oligodendrogliomas subject to Ki-67/MIB-1 immunohistochemical staining were obtained from the archives of Department of Pathomorphology, Military Institute of Medicine in Warsaw, Poland. Acquisition of the whole slide images was performed on the Aperio ScanScope scanner. Due to the very large size of images, we have chosen eightfold reduction of the resolution to enable the evaluation performed both manually and by computer.

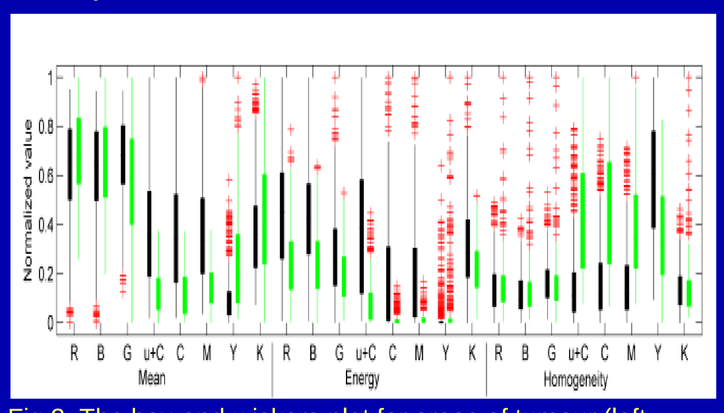
After digitalization of WSI (whole slide image), the selection of hot-spots was done manually by an expert and automatically with the proposed method of automatic hot-spot localization. The proposed scheme of the image processing is based on the following steps:



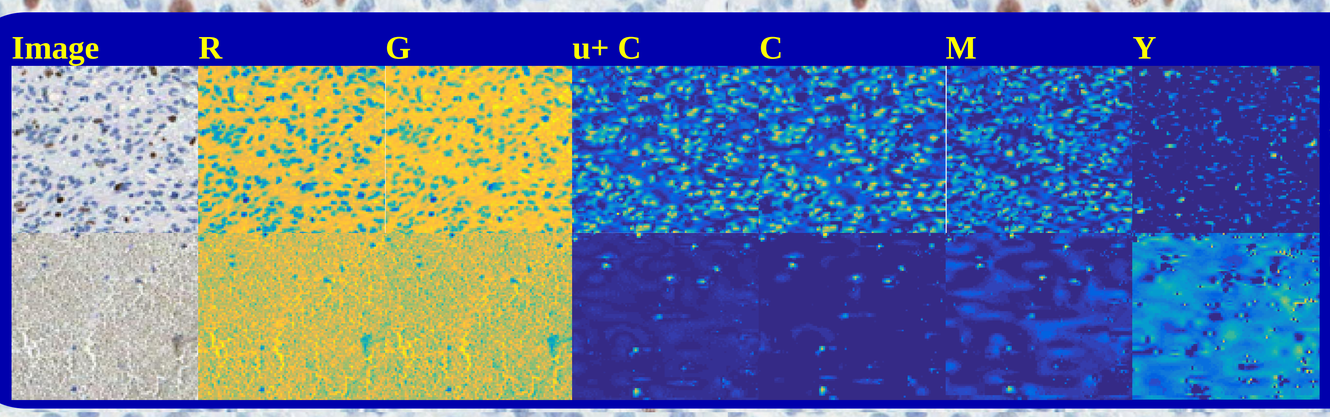
The final analysis of Ki-67 index is performed on the full resolution images.



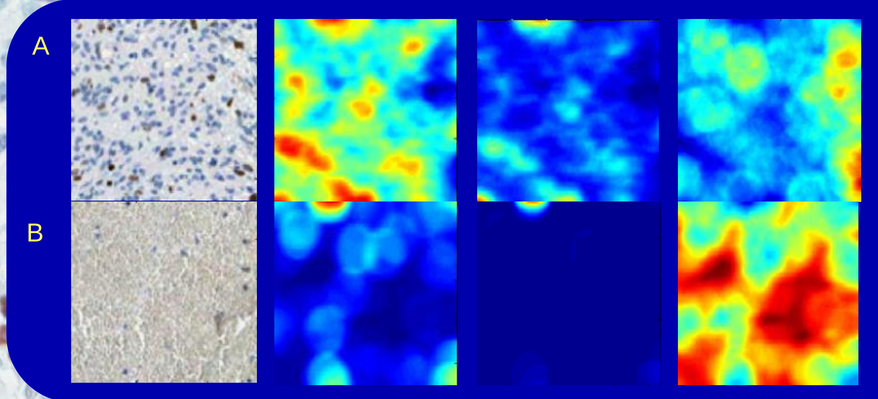
▲Fig.1. An example of a part of the whole slide image representing the brain tumour with the outlined hot-spot area. The hemorrhages are marked by the arrows.



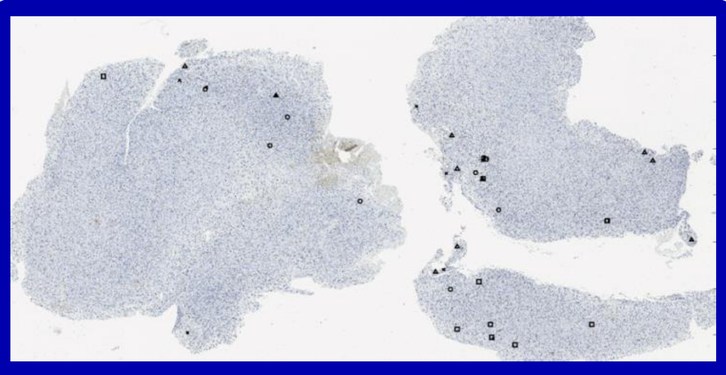
▲Fig.2. The box-and-wiskers plot for areas of tumour (left black boxes) and hemorrhages (right grey boxes), for colour componnents and three features: mean, energy, and homogeneity.



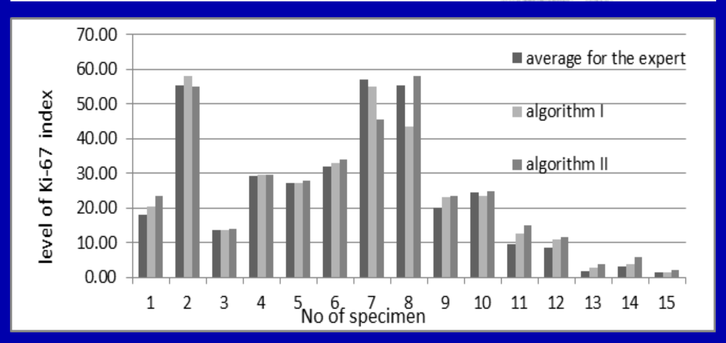
◄ Fig.3 The examples of maps with different colour components for tumour (A) and haemorrhage (B).



◄ Fig.4 The example colour maps with few different texture descriptions for tumour (A) and haemorrhage (B).



◄ Fig.5. Virtual slide with the hot-spot areas marked by an expert (Series I-□, Series II-○) and the hot-spots designated by the algorithm I (×) and algorithm II (Δ).



◄ Fig.6. The comparative results of analysis of 15 specimens for which areas of interest were determined by the medical expert and by the developed algorithms.

◄ Table 1. The quantitative analysis of Ki-67 index in the designated fields of view for the hot-spot areas selected by the expert (average for series I and II), and by the developed algorithms (algorithm I and algorithm II).

Case	Expert			Automatic	
	Series I,%	Series II,%	Average,%	Algorithm I,%	Algorithm II,%
1	18.39	17.69	18.04	20.45	23.64
2	56.11	54.54	55.32	57.96	56.03
3	13.61	13.43	13.52	13.58	14.07
4	28.47	30.30	29.39	29.56	29.67
5	27.28	26.91	27.10	27.23	27.99
6	29.41	34.31	31.86	33.13	34.02
7	54.71	59.35	57.03	55.03	45.39
8	60.12	50.48	55.30	42.60	58.18
9	21.11	19.40	20.26	23.21	23.64
10	24.03	24.06	24.45	24.51	24.71
11	9.82	9.43	9.62	12.64	14.88
12	8.28	9.05	8.66	10.96	11.73
13	1.72	1.75	1.74	2.73	3.83
14	2.95	3.06	3.01	3.68	5.83
15	1.61	1.39	1.50	1.63	2.19
Mean:			23.78	23.93	24.99

Results

Fifteen cases of meningiomas and oligodendrogliomas were subject to a quantitative analysis. The comparison of the average values of the Ki-67 index for the fields of view selected by an expert and by the developed algorithms I and II is presented in Fig. 6 and Tab 1.

The algorithm I appears to give results closer to expert's opinion, compared to algorithm II. However, the algorithm II can more accurately recognize the hot-spot areas with the higher Ki-67 index. The Wilcoxon matched pairs test confirms that no significant differences exist between the mean of experts results and Algorithm I, but there is a significant difference when compared with Algorithm II. Also, it should be noted that in the case of the analysis of low reaction specimens, fields selected by developed algorithms had similar or higher reaction than the reaction in fields selected by the expert. It is clear that in the low reaction specimen, finding hot-spot areas manually is much more difflcult compared to the high reaction cases.

Conclusion

We have presented the effective method for an automatic localization of the hot-spot areas in meningioma and oligodendrogliomas tumours. The algorithm II has shown the advantage over the algorithm I in accurate detection of hot-spot areas in the presence of staining artifacts. The use of maps of specimen and the elimination of hemorrhage areas have reduced the size of an image under analysis and also the computational time. The presented methods have good reproducibility (characterized by the repeatability of results) which gives them an advantage over the traditional, manual way of identification of hot-spot areas. Our results have confirmed the advantage of automatic evaluation over the manual assessment.

This work has been supported by the National Centre for Research and Development (PBS2/A9/21/2013 grant),