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Multicenter pilot evaluation of a new AI-based metaphase finder
for application in karyotyping peripheral blood and bone marrow specimens

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Introduction

- Cytogenetics laboratories are experiencing a reduction in the workforce and challenges replacing highly experienced technologists. To address these drawbacks, there is a **growing demand for computer-aided karyotyping software** designed to enhance the efficiency of karyogram preparation, thereby optimizing resource allocation for higher-value tasks.
- **Finding and capturing metaphase images from G-banded slides at low magnification** is an important phase in the karyotyping workflow, both to expedite slide review and for determining which metaphases will be analyzed at higher magnification.
- **Traditional automatic metaphase finder classifiers have been used for several decades** but their specificity can be improved, especially for low mitotic index specimens where the need to identify any possible metaphase requires lowering detection thresholds, resulting in higher selection of debris.
- These default "out-of-the-box" classifiers require laboratory-specific fine-tuning to achieve optimal metaphase detection performance. It is therefore necessary to develop more generic and robust metaphase finder algorithms which are applicable to a variety of slide preparation methods, **reducing the need to perform custom adjustments**.
- Recent advancements in **artificial intelligence (AI)** have been integrated into digital karyotyping systems, particularly for the segmentation and classification of chromosomes at high magnification[1-3].
- However, the use of AI to reliably detect metaphases at 10x magnification is not yet well documented. The aim of this study is to assess the **accuracy of a new AI-based metaphase finder** algorithm compared to the conventional non-AI classifier for both peripheral blood (PB) and bone marrow (BM) specimens.

Methods

- G-banded slides of chromosomally **normal and abnormal PB and BM samples** were prepared by standard methods and scanned twice at 10x magnification using the HiBand system (Applied Spectral Imaging), once with the default metaphase finder, and a second time using a new AI-based computer-aided algorithm (Figure 1).
- A total of 13 slides from three different cytogenetics laboratories were included in this pilot evaluation. This set comprised 3 normal and 4 abnormal BM samples as well as 3 normal and 3 abnormal PB specimens.
- For the first 50 captured images, the total number of metaphases detected on each slide and the number of real metaphases counted, versus debris, was compared using both methods (Figure 2).
- The average, standard deviation, range and median were reported for each dataset. Statistical significance was assessed using either the Wilcoxon Signed-Rank two-tailed test for paired samples or the Mann-Whitney U test for independent samples. A p-value lower than 0.05 was considered significant.

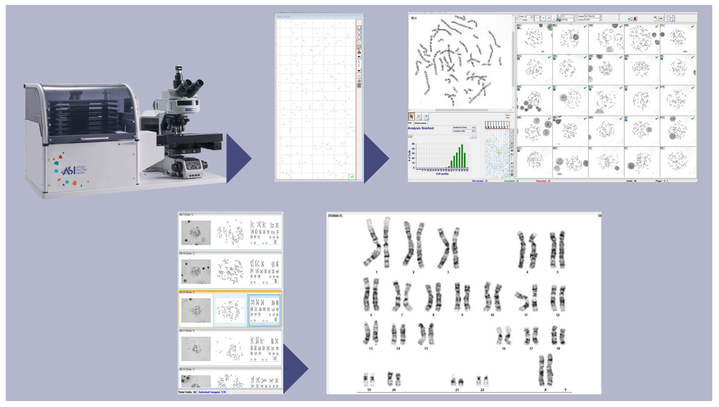

Figure 1
AI-based metaphase finder detects metaphase candidates following automatic slide scanning under 10x objective. Metaphase candidates, shown in the image gallery, can be sorted by index or by quality metric. Selected metaphases are then scanned under high magnification objective for review and analysis.

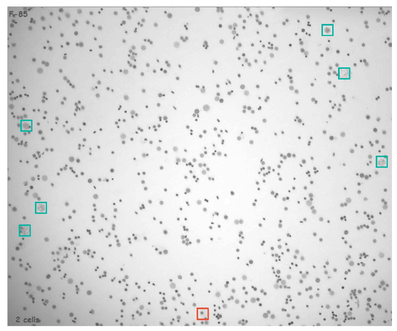

Figure 2
Example of detected metaphases of a bone marrow sample in a single 10x magnification frame. A total of 7 metaphase candidates were detected by the AI-based metaphase finder. Cyan squares represent "real" metaphases, while red squares represent non-genetic material (false-positive).

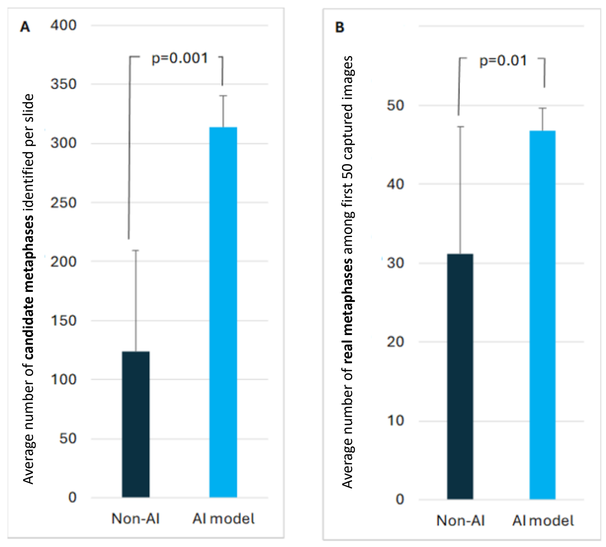

Figure 3
A: Graph comparing the average number of **candidate metaphases** (+/- standard deviation) identified per slide with the AI model (light blue) and the non-AI "out-of-the-box" classifier (dark blue). The p-value is indicated in the graph.
B: Graph comparing the average number of **real metaphases** (+/- standard deviation) among the first 50 captured images per slide with the AI model (light blue) and the non-AI "out-of-the-box" classifier (dark blue). The p-value is indicated in the graph.

Results

- For the 13 slides assessed, an average of **314±265 metaphase candidates per slide** (range 48-1,085, median 256) were detected with the AI model compared to 124±85 metaphases (range 9-269, median 131) with the non-AI "out-of-the-box" classifier. This **2.5-fold increase in number of metaphases detected** was significant (p=0.001) (Figure 3A).
- Among the first 50 captured images, an average of **47±3 analyzable metaphases** (range 40-50, median 48) were detected with the AI algorithm compared to 31±16 (range 7-50, median 29) with the default non-AI classifier (p=0.01) (Figure 3B).
- With the AI-based method, for the first 50 captured images, the average number of analyzable metaphases detected by the AI-based method was the **same for BM samples (46±4) and PB specimens (47±1)** (p>0.05).
- Furthermore, a **fourfold decrease in the number of captured images with non-metaphase material** was observed, from 23% (119 out of 524) with the non-AI classifier to 6% (40 out of 648) with the new AI algorithm.

Conclusions

- This multicenter pilot study demonstrates that for both bone marrow and peripheral blood specimens, the AI-based metaphase finder detects **2.5-fold more metaphases** than the conventional non-AI default classifier.
- **This increase in metaphase detection is critical, particularly in cases of low mitotic index**. Moreover, the AI algorithm reduces by fourfold the number of non-metaphase material images captured by the system, resulting in **more efficient and reliable metaphase image acquisition**.
- These findings suggest that integrating AI into metaphase detection workflows may improve abnormality detection and technologist productivity while **minimizing the need for time-consuming manual optimization across laboratories**.
- Further large-scale validation is warranted to confirm the robustness of this new method in routine cytogenetic analysis.

Disclosures

LSR, KP and RDB have no disclosure.
EZ, AT, OH, CS and YAG are employees of Applied Spectral Imaging.

References

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