Registration accuracy between whole slide images and glass slides in eeDAP workflow

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Objective

Evaluate WSI-to-microscope registration accuracy

eeDAP: evaluation environment for Digital and Analog Pathology

- Register the whole slide image (WSI) and the microscope field-of-view (FOV).
- Collect pathologist evaluations at specific FOVs, cells, or features.

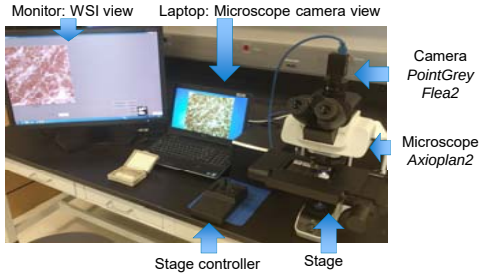


Figure 1 – eeDAP system
Registered to the same position on eyepiece and both displays

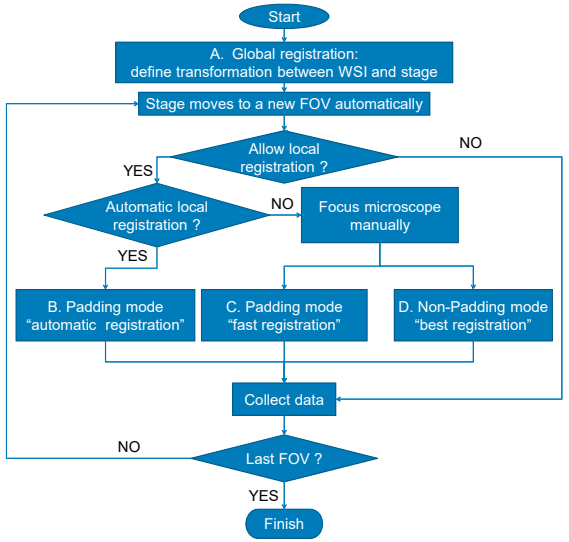


Figure 2 – eeDAP Workflow

Methods and Materials

Registration

- Use camera image and WSI FOV (after color-to-grayscale transform).
- Based on normalized 2D cross correlation.

Mode	Difference	Advantage
Padding	Padding the larger image with zeros	Find target on the boundary
Non-Padding	Do not padding zeros	Accurate registration for the center

Measurement accuracy for four registrations:
A. Global Registration
B. Local Registration with padding before focusing the microscope
C. Local Registration with padding after manually focusing the microscope
D. Local Registration without padding after manually focusing the microscope

Measurement & Slides

Two readers record distance-to-center on microscope, of a target (pre-identified and identifiable feature) on WSI for 2 study orders:
1. List order: Measure all FOVs on one slide and then go to the next slide.
2. Random order: Sequential FOVs potentially occurring on different slides.

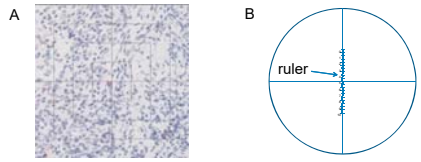
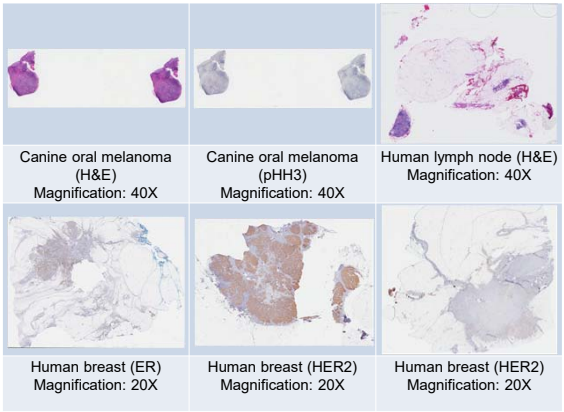


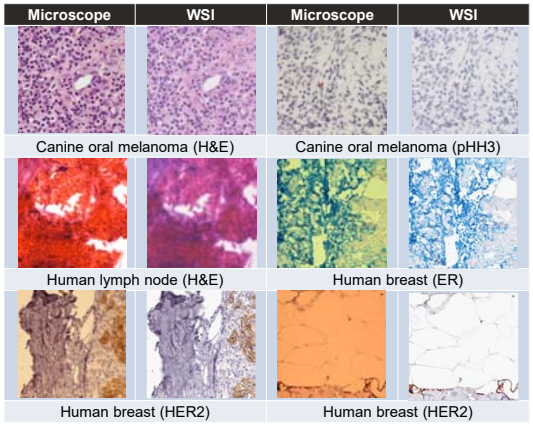
Figure 3 - A) Virtual reticle indicates the target in the eeDAP view of the WSI (top-right corner of a red cell). B) Microscope eyepiece ruler reticle for measurement (rotatable). The ruler total length: 10 mm with 100 divisions (250 µm at 40X, 500 µm at 20X)

Table 1 – Study slides, 10 FOVs per slide



Results

Figure 4 – Registration Results: Microscope (camera) vs WSI



Global Registration

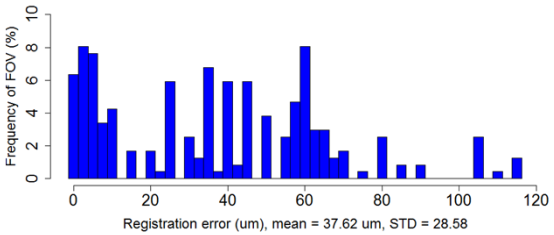


Figure 5 - Global Registration errors for 240 observations from 2 readers: measures Global Registration transformation and stage movement precision

Local Registrations

- Reduce most registration errors to smallest measurable. Histogram dominated by peaks for smallest bins 0-2.5 um and 2.5-5.0 um.
- Study order is important when registration is before focusing.
- Focusing improves registration, eliminates impact of study order.

Table 2 – Fraction of registration errors within 5.0 µm from two readers

	Padding mode (before focusing)	Padding mode (after focusing)	Without padding mode (after focusing)
List order	92/120 (76.7%)	113/120 (94.2%)	116/120 (96.7%)
Random order	62/120 (51.7%)	114/120 (95.0%)	119/120 (99.2%)

Discussion & Conclusion

- Global Registration: ok for large features (errors < 120um).
- Local Registration after focusing: most errors < 5um.
- No observed operator dependence.
- Registration accuracy does not appear to be affected by tissue types, stains, and scanning magnifications.
- The main factors affecting registration accuracy are the microscope focus quality, scan quality, and FOV content.
- Manual adjustment possible when registration error is large.
- *Future work: Investigate main factors and improve registration methods.*

Regulatory Impact

- eeDAP allows pathologists to evaluate the same FOVs, cells, or features in glass slides and WSIs. Such data allows technology comparisons (WSI vs. microscope) and AI algorithm training and testing.
- This study supports eeDAP's CDRH Medical Device Development Tool (MDDT) application, which will help medical device sponsors use eeDAP in the development and evaluation of medical devices.

References

- eeDAP is available now at **http://www.github.com/DIDSR/**

1. Gallas et al. *Evaluation Environment for Digital and Analog Pathology (eeDAP): a platform for validation studies*. JMI 1(3) 037501. 2014
2. eeDAP working group NCIP Hub. **https://nciphub.org/groups/eedapstudies/**
3. WSI working group NCIP Hub. **https://nciphub.org/groups/wsi_working_group/**

Acknowledgment

- All slides and images used in this study have been cleared by IRB and appropriate FDA channels.
- Mention of products herein cannot be construed as endorsement by FDA.
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