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## INTRODUCTION

Faster biothreat detection in resource-constrained environments enables faster countermeasure deployment, giving our warfighters unprecedented advantages.

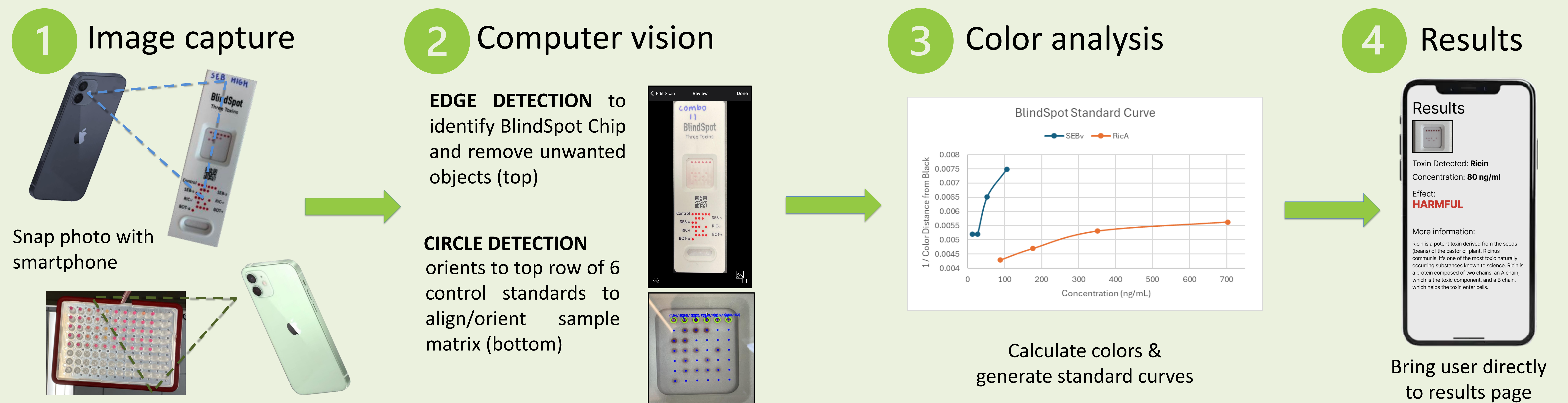
Lateral flow immunoassays (LFI) allow for rapid detection of diverse targets by leveraging capillary flow on portable, low-cost, and simple devices.<sup>1</sup> **BlindSpot Chips (MaximBio)** are multiplexed LFIs to detect toxins – up to 6 biological threats simultaneously.<sup>1</sup> Multiplexing improves sample quality and reduces time to results by cutting down number of tests.<sup>1</sup>

**Loop-Mediated Isothermal Amplification (LAMP)** assays are a cheaper, faster, and easier nucleic acid detection testing alternative to PCR.<sup>2</sup> LAMP assays enable high sensitivity and specificity visual target detection without the need for a thermocycler.<sup>2</sup>

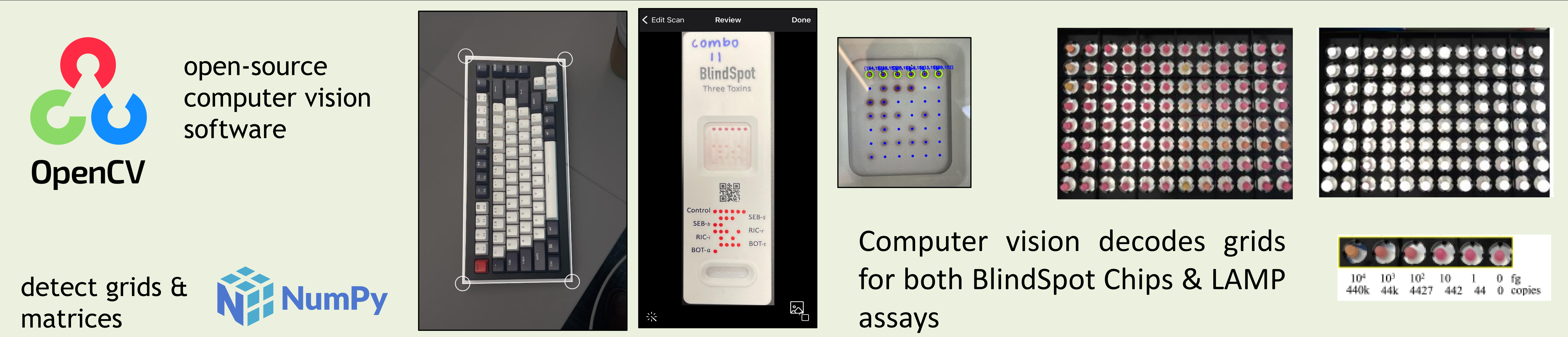
**CHALLENGES:** These tests are still prone to subjective interpretation due to issues like line bleeding and faint or weak positives. They also lack clear guidance for next steps and require costly, vendor-specific hardware.

**OBJECTIVE:** Develop application to analyze Blindspot Chips and LAMP assays using computer vision for in-field interpretation without need for costly equipment and advanced training.

## WORKFLOW OVERVIEW



## COMPUTER VISION IMAGE ANALYSIS DISCRETIZES POSITIONS & DECODES GRID

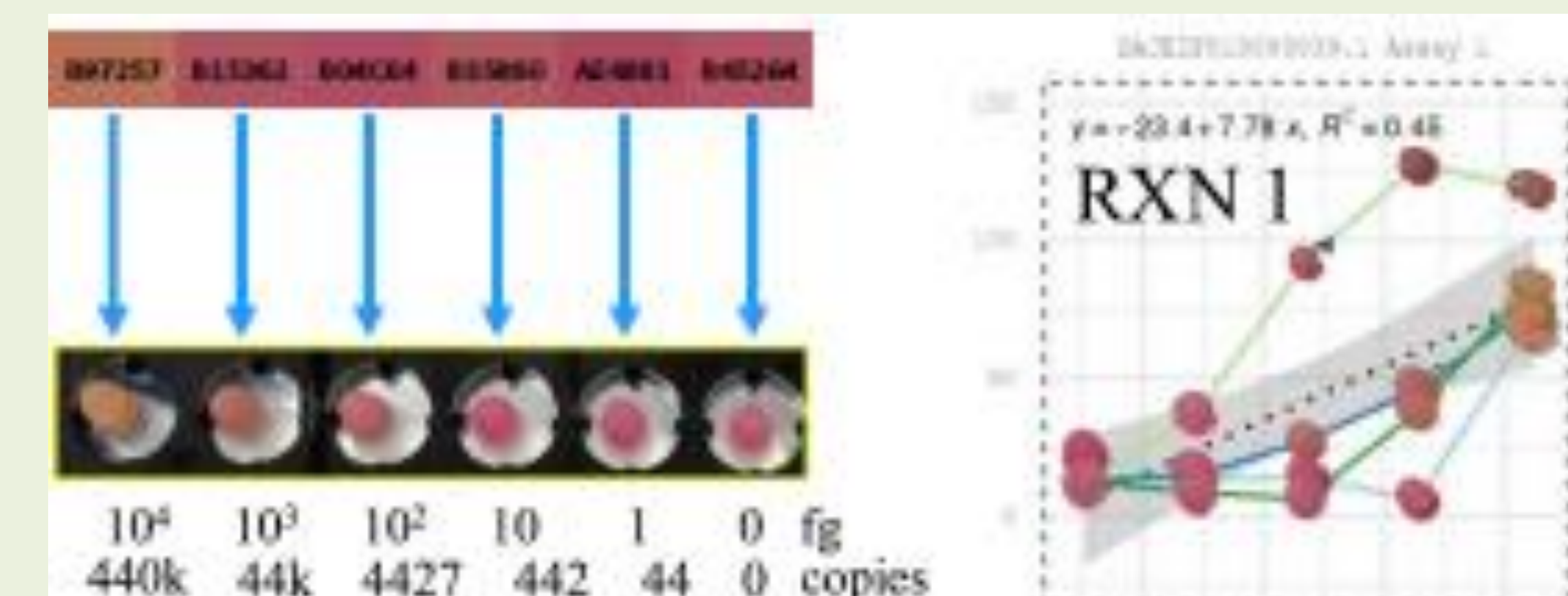
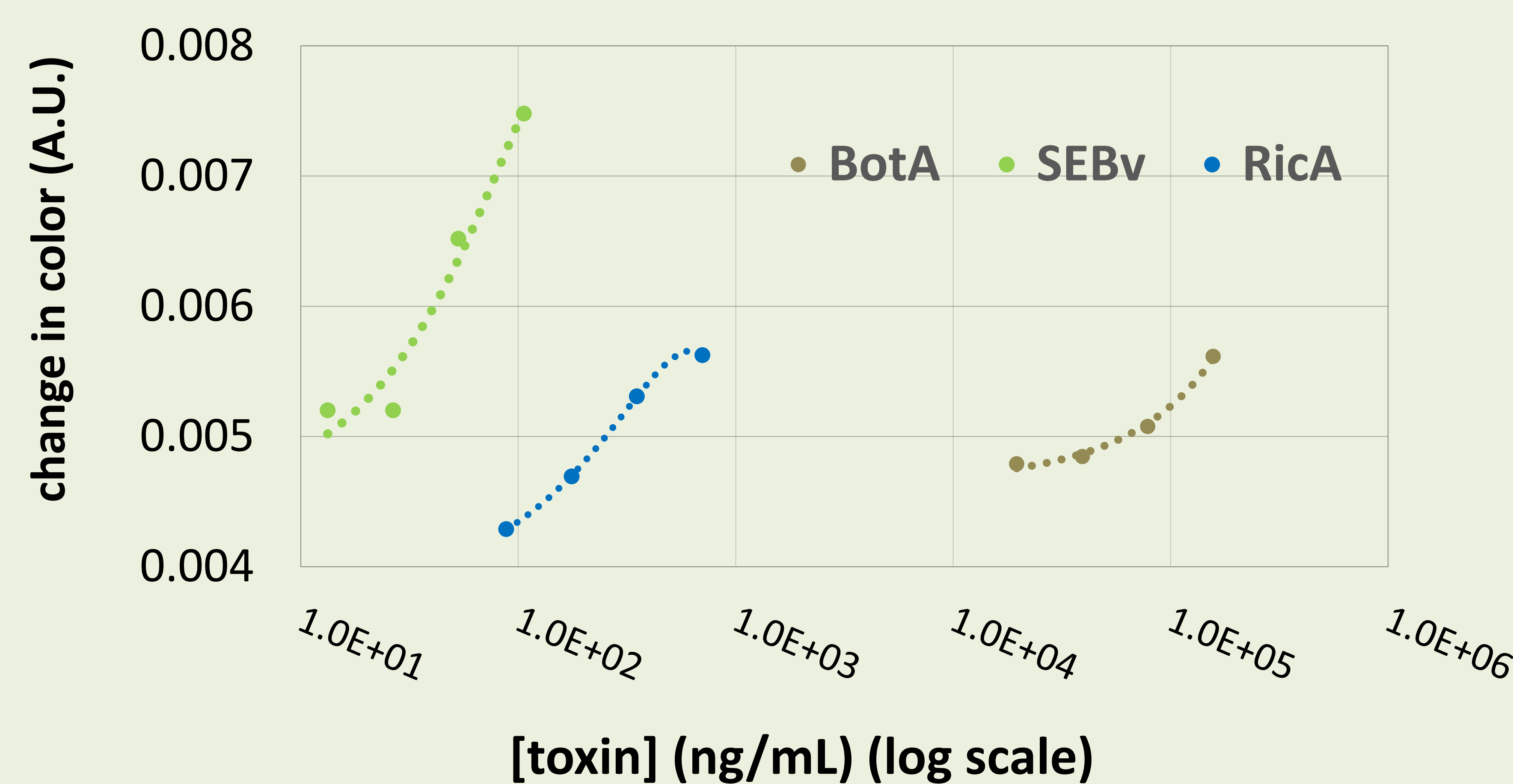


## COLOR ANALYSIS ALGORITHM DEFINES THRESHOLD FOR POSITIVE VALUES

First row (orange): Control standards

RICT 4					
#62343d	#613240	#5d2f3a	#592f37	#5d2f37	#5a3237
#92969f	#90949d	#91959e	#92969f	#90949d	#8d8e93
#9298a1	#9599a2	#91959e	#8f939c	#93939d	#8e8f94
#858891	#838790	#848790	#8b8f98	#82828c	#8e8f94
#92969f	#7f7f89	#8d919c	#828387	#8d8d97	#8b8c92
#8f939c	#8d919a	#8b8f98	#8a8e97	#8c8c96	#8b8b95
#8d919a	#8e929b	#8c9099	#898d96	#878b94	#8b8c91

THRESHOLD VALS					
126.6057	126.5109	119.256	114.6952	117.8261	116.7262
262.8631	259.4012	261.1322	262.8631	259.4012	248.3022
265.2188	268.056	261.1322	257.6703	260.513	250.034
239.1861	236.9008	237.4553	250.7469	231.0844	250.034
262.8631	225.8916	255.4251	228.6613	250.126	245.4323
257.6703	254.2086	250.7469	249.0161	248.3948	246.6637
254.2086	255.9394	252.4777	247.2853	243.8237	244.8387



- Euclidean formula to determine threshold values that represent positive readings → Build a data set & standard curve
- Color vision algorithm output: table of HEX color values corresponding to each spot in sample matrix

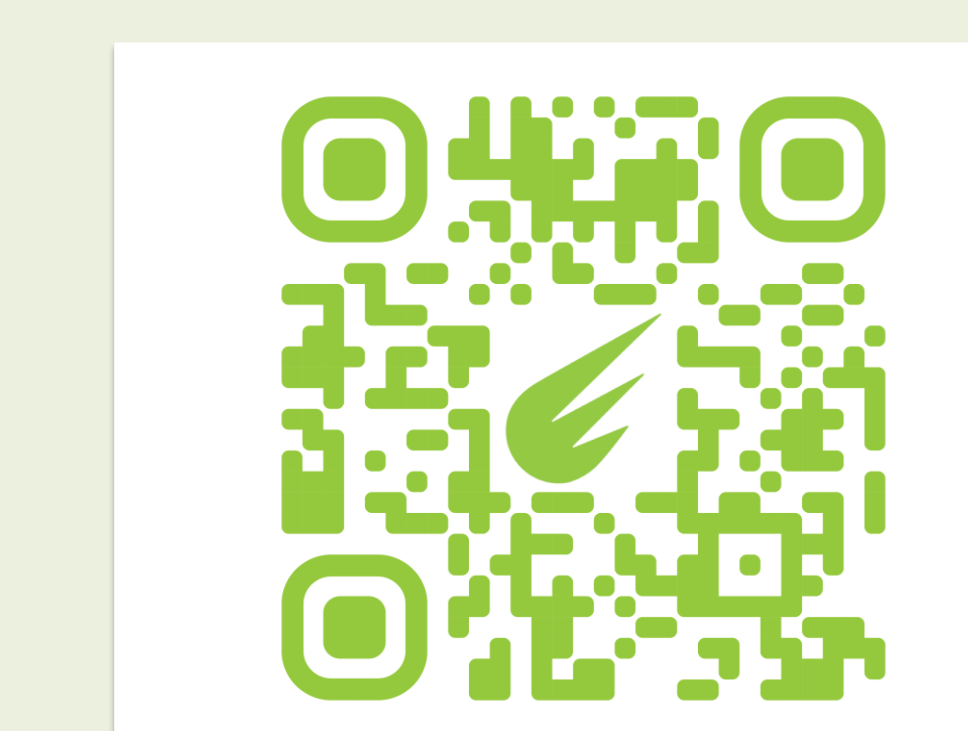
## CONCLUSIONS

Developed algorithm to decode BlindSpot and LAMP assay grids: This simplifies, automates, and quantifies interpretation with computer vision.

Workflow validated in virtual cell phone application that runs locally, without internet requirement.

**NEXT STEPS:** Develop application for iOS and Android operating systems to enable fast, easy, & accessible biothreat detection at the point-of-care.

## Learn More



## References

- [1] Hofmann, E. R., Davidson, C., Chen, H., Zacharko, M., Dorton, J. E., Kilper, G. K., & Sozhamannan, S. (2021). Blind spot: A braille patterned novel multiplex lateral flow immunoassay sensor array for the detection of biothreat agents. *ACS omega*, 6(35), 22700-22708.
- [2] Dao Thi, V. L., Herbst, K., Boerner, K., Meurer, M., Kremer, L. P., Kirrmaier, D., & Anders, S. (2020). A colorimetric RT-LAMP assay and LAMP-sequencing for detecting SARS-CoV-2 RNA in clinical samples. *Science translational medicine*, 12(556), eabc7075.

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