

Kelsey Hauser¹ PhD, Daniel Negron, PhD¹; Bradley Abramson, PhD¹; Michael Digney¹; Rhea Premanand¹; Nicholas Tolli²; David Ashford, DVM, MPH, DSc²; Gabrielle Melton¹; Bryan Necciai^{3,4}, Katharine Jennings, PhD¹; Shanmuga Sozhamannan^{3,4}

¹Noblis, Inc., 2002 Edmund Halley Drive, Reston, VA; ²Noblis ESI, 14425 Penrose Pl, Chantilly, VA; ³Joint Program Executive Office for Chemical, Biological, Radiological and Nuclear Defense, Joint Project Lead for CBRND Enabling Biotechnologies; ⁴Joint Research and Development, Inc., Stafford, VA

INTRODUCTION

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

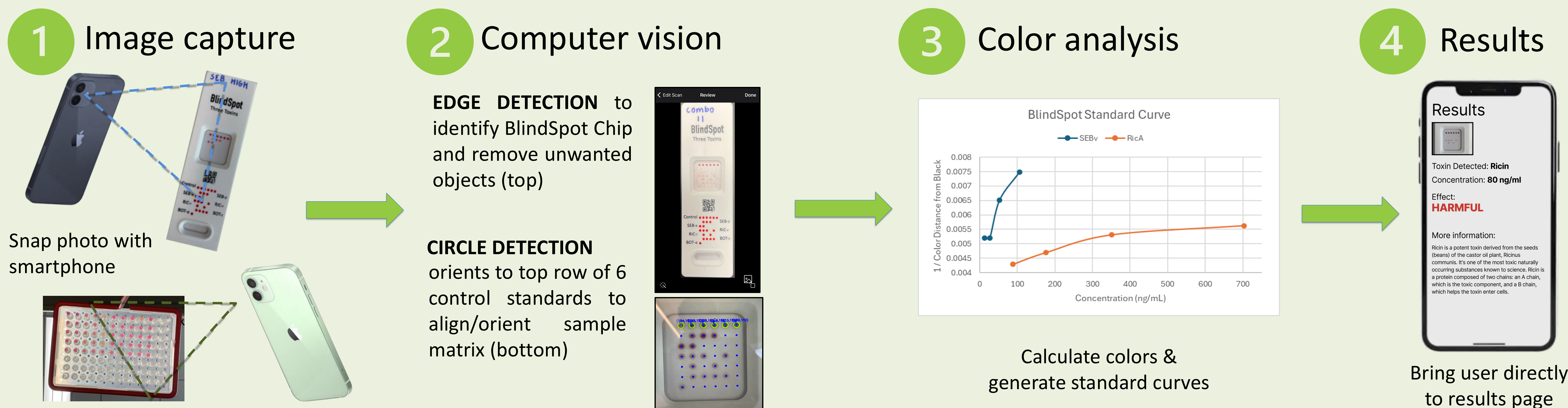
Lateral flow immunoassays (LFI) allow for rapid detection of diverse targets by leveraging capillary flow on portable, low-cost, and simple devices.¹ **BlindSpot Chips (MaximBio)** are multiplexed LFIs to detect toxins – up to 6 biological threats simultaneously.¹ Multiplexing improves sample quality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

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

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

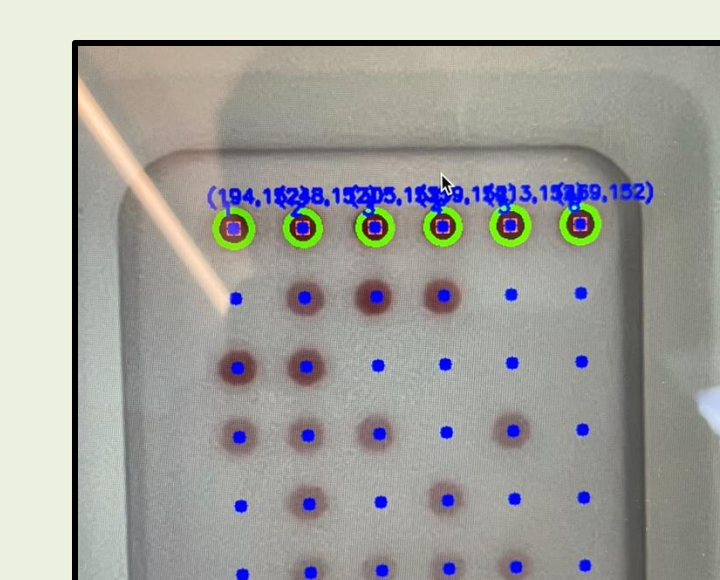
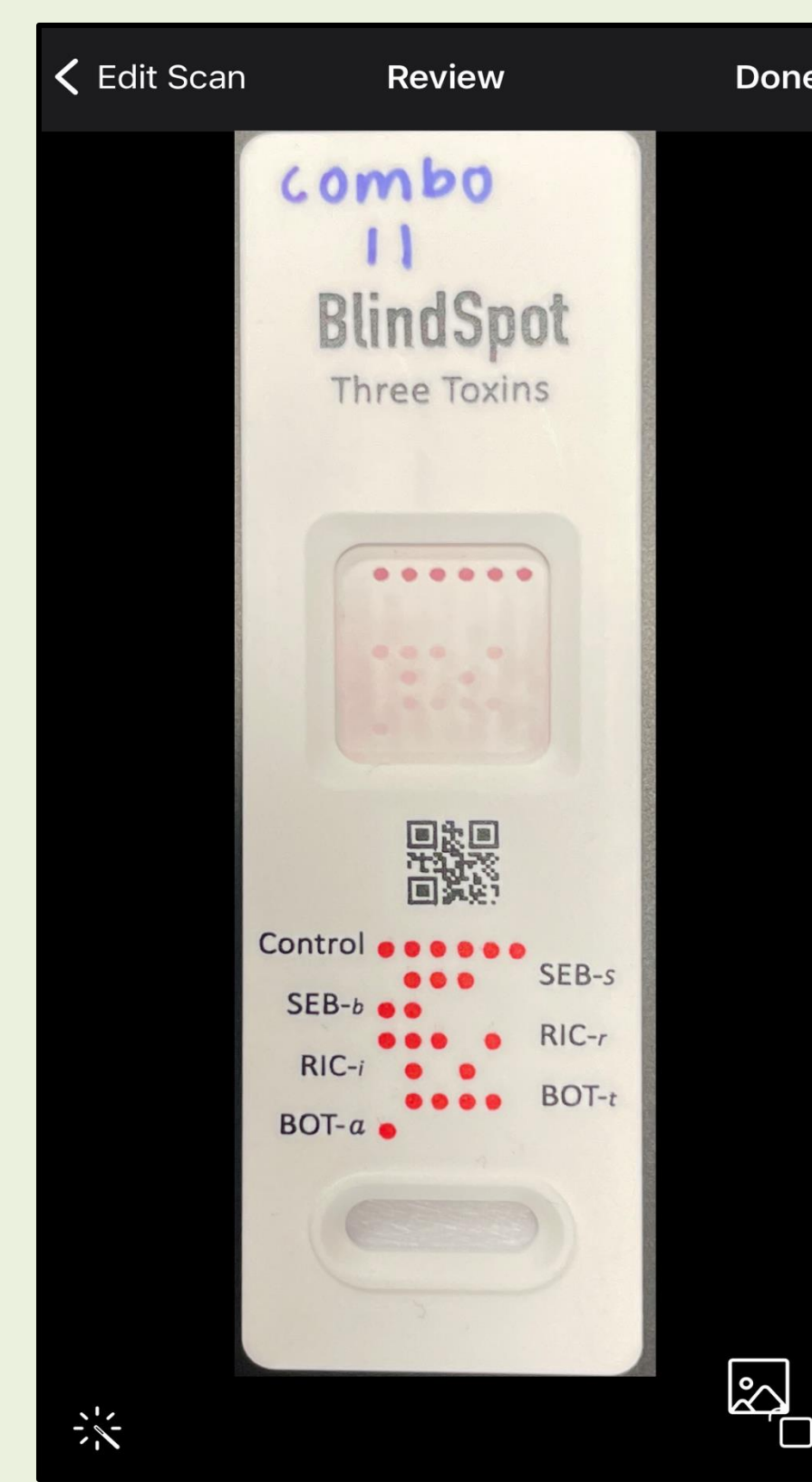
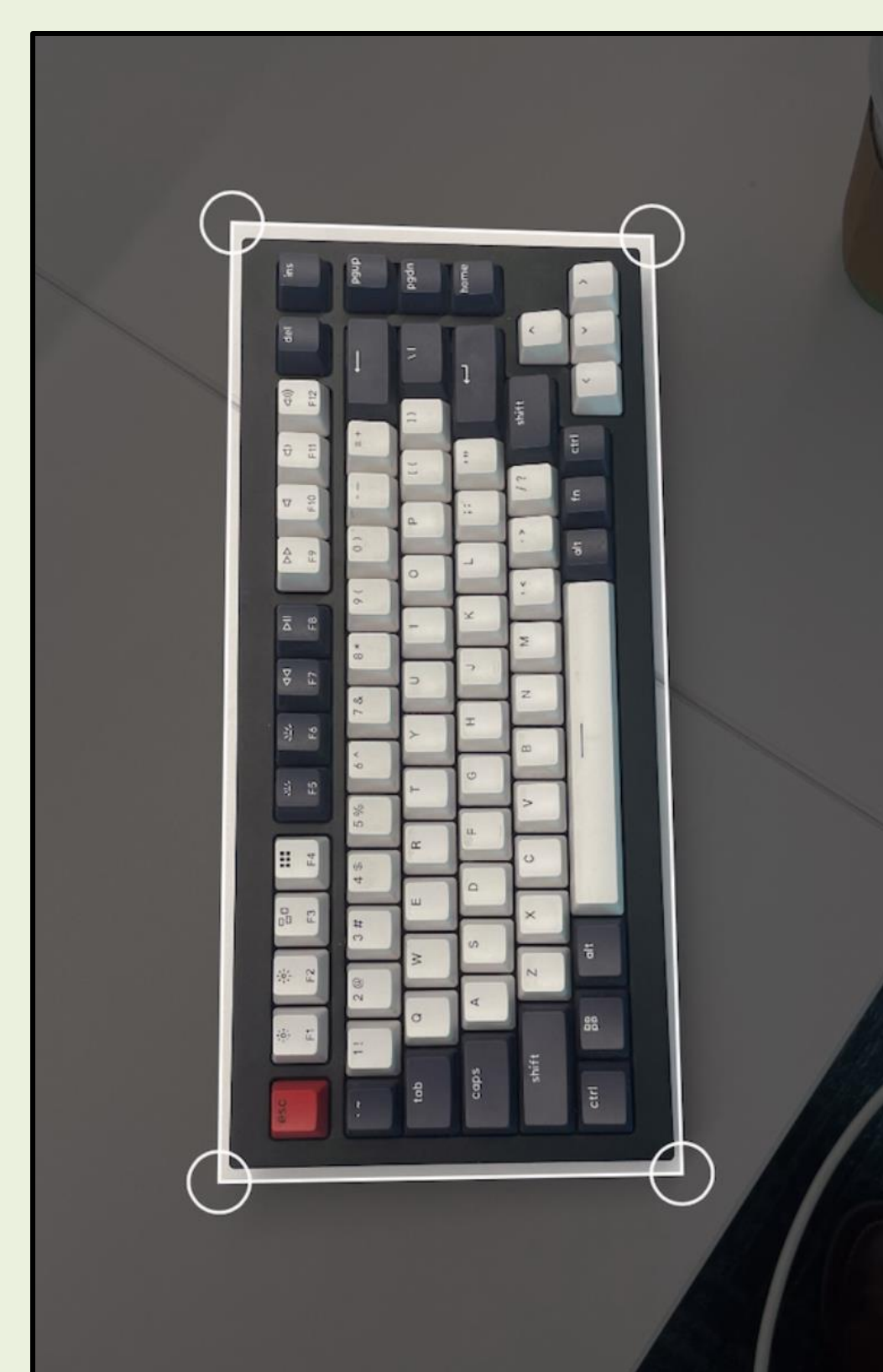


COMPUTER VISION IMAGE ANALYSIS DISCRETIZES POSITIONS & DECODES GRID



open-source computer vision software

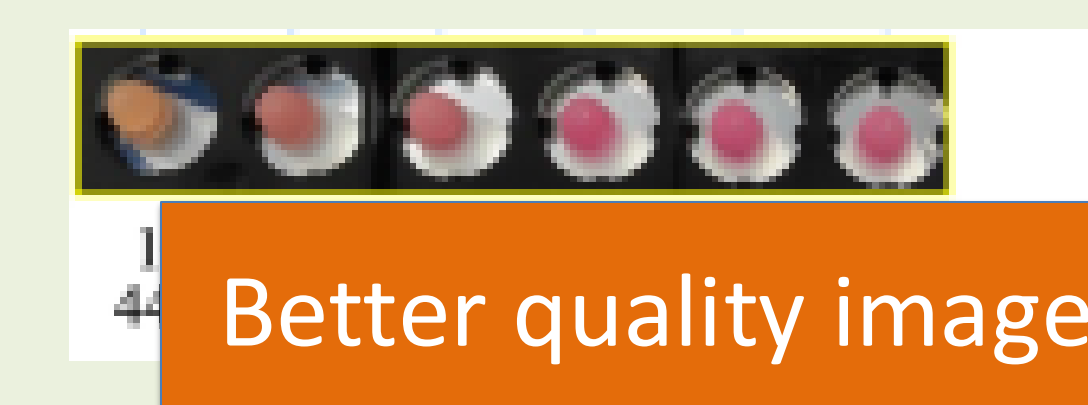
detect grids & matrices



Better quality image?



Computer vision decodes grids for both BlindSpot Chips & LAMP assays



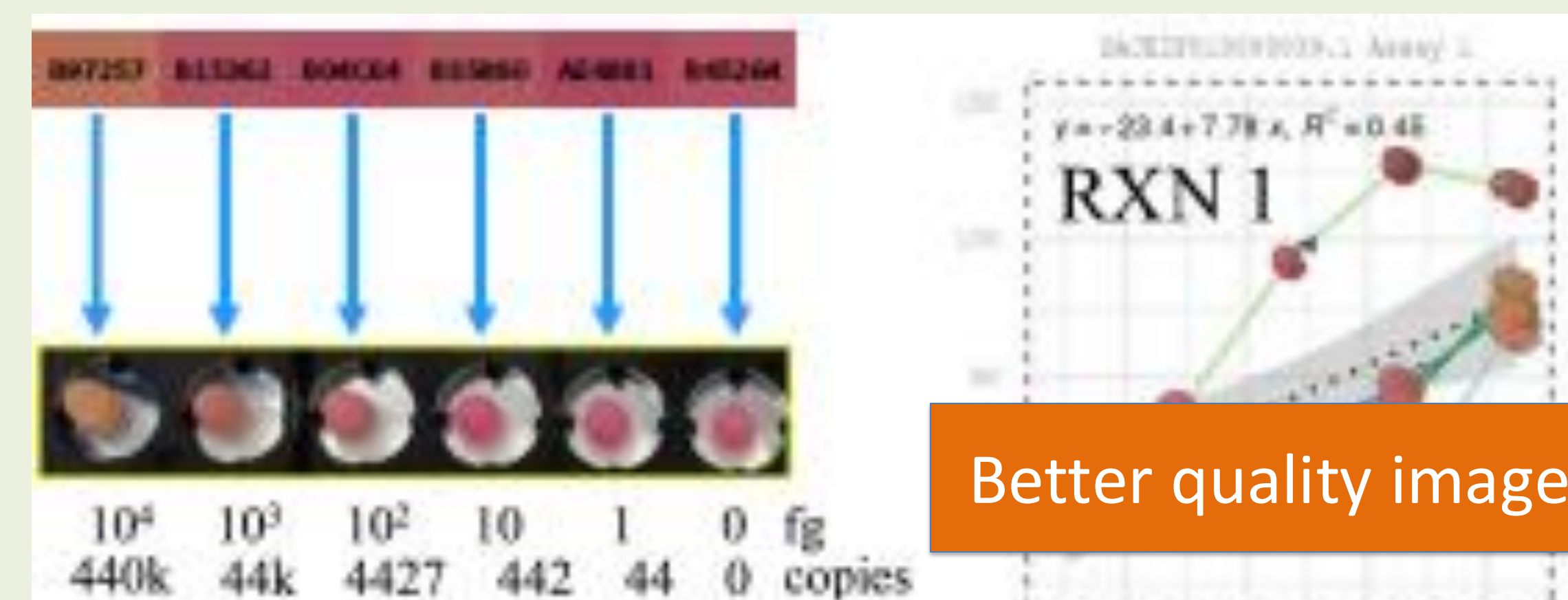
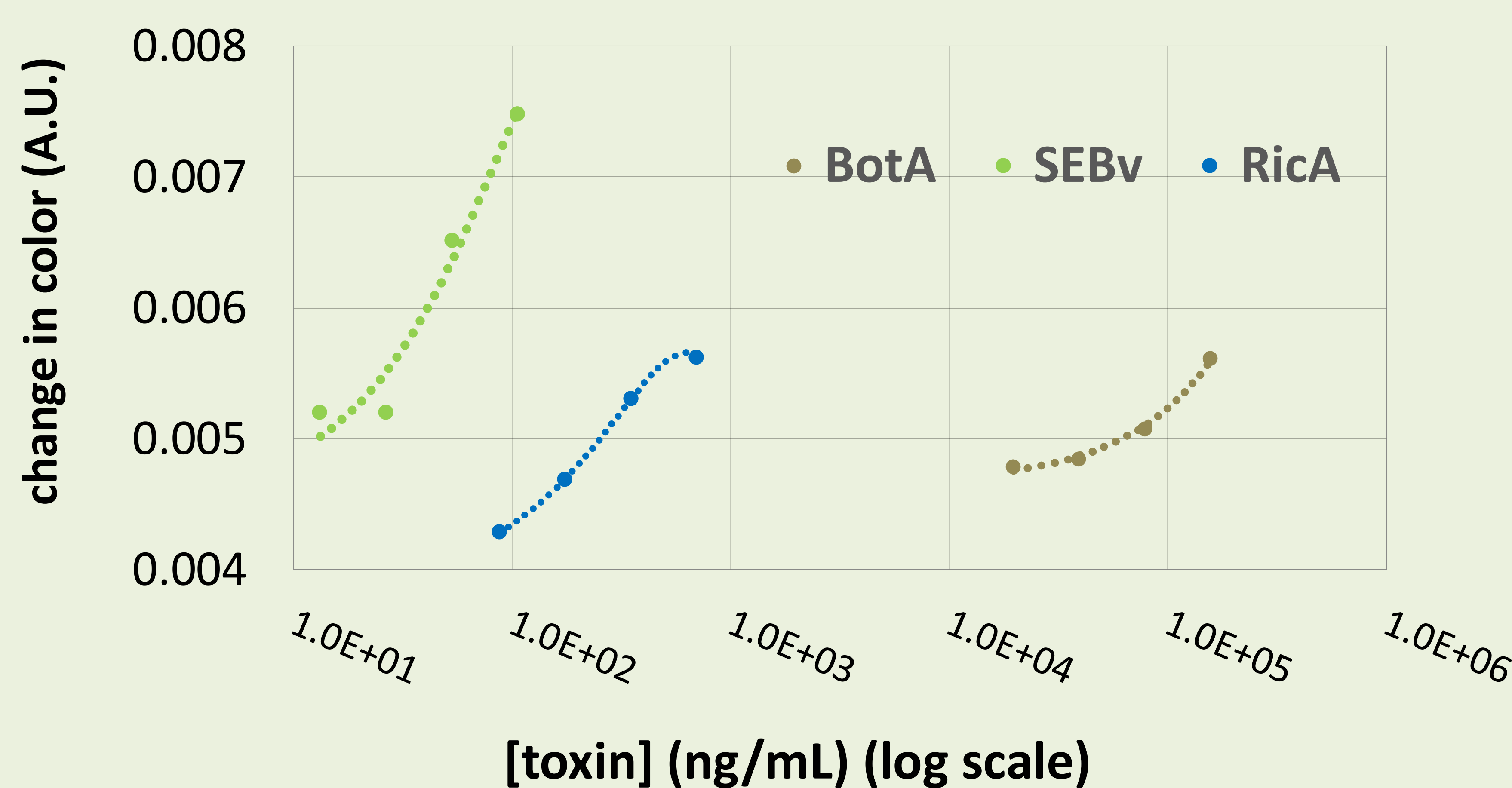
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COLOR ANALYSIS ALGORITHM DEFINES THRESHOLD FOR POSITIVE VALUES

First row (orange): Control standards

| RIC T4 | | | | | |
|---------|---------|---------|---------|---------|---------|
| #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 | #77f789 | #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 |



Better quality image?

- 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

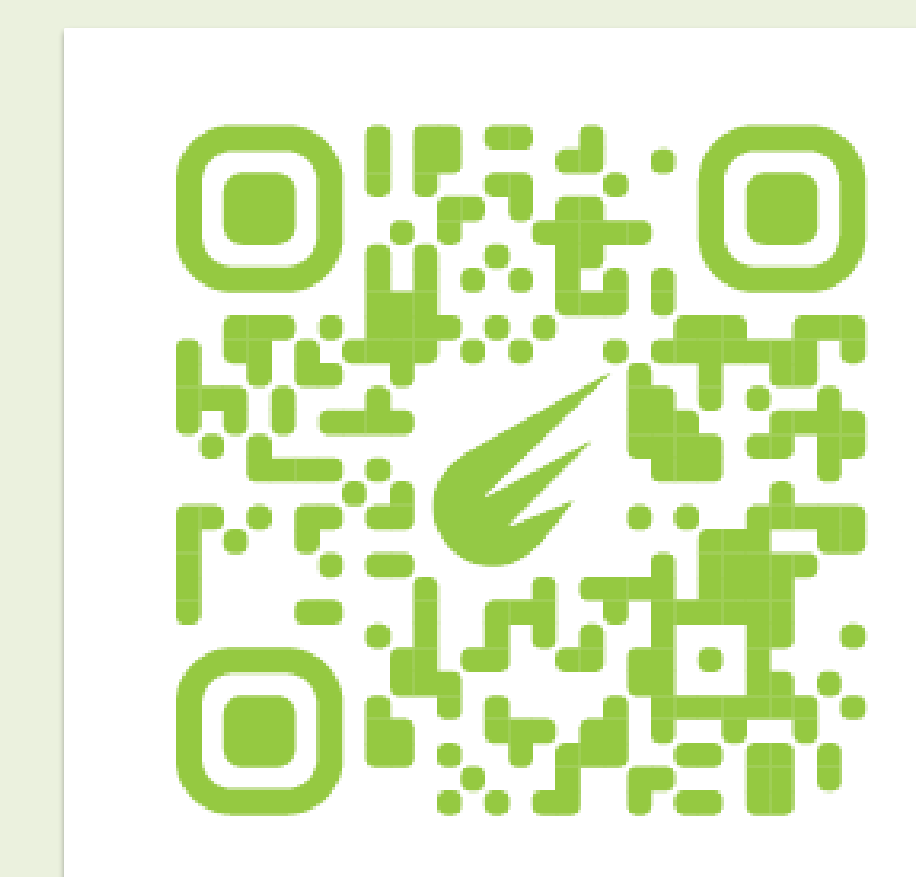
CONCLUSIONS

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 & Acknowledgements

- [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., Kirmmaier, 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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