## **Electronic Supplementary Information**

## Accessory-free quantitative smartphone imaging of colorimetric paper-based assays

Tian Kong, Jae Bem You, Biao Zhang, Brian Nguyen, Farhang Tarlan, Keith Jarvi and David Sinton\*

## **Fabrication of Three-trial Colourimetric Device**

The device consists of 2 paper layers patterned with wax. The front layer is printed with both black reference spot boundaries and testing spot boundaries, while the back layer, a uniformly white reference scene, is printed with testing spot boundaries only. The back layer also prevents liquid penetration through the front paper layer. The device patterns were designed in AutoCAD, then printed on No. 1 Chromatography Paper from Sigma-Aldrich with a solid wax printer (ColourQube 8570N, Xerox Canada). Printed sheets of paper were placed in a preheated oven at  $120^{\circ}$ C for 3 minutes form a hydrophobic layer through the thickness of the paper. The diameter of the testing spots and background spot was 4 mm after heating.  $1\mu$ L of black ink was added and dried to the designated 1-mm-diameter dots to prepare for black reference spots. Double-sided tape was used to bond 2 layers such that the testing spots boundaries aligned with each other.



**Fig. S1** Comparison of two RGB colour quantification methods ( $\Delta$ RGB and intensity difference) before and after rescaling, without dominant light source. Images were obtained under various lighting conditions. The RGB values of 170 lux, 515 lux, 1000 lux were normalized to the value at 50 lux in their own category. (a) Green colour device imaged with iPhone 7 Plus. (b) Red colour device imaged with iPhone 7 Plus. (c) Green colour device imaged with Samsung Galaxy S7 edge. (b) Red colour device imaged with Samsung Galaxy S7 edge.



**Fig. S2** (a)  $\Delta$ RGB representation of the rescaled  $\Delta$ RGB values of the green devices imaged at different tilting angels. (b)  $\Delta$ RGB representation of the rescaled  $\Delta$ RGB values of the red devices imaged at different tilting angels. (a,b) Images were obtained from Huawei P8Lite ALE-L04, iPhone 7 Plus and Samsung Galaxy S7 edge.



**Fig. S3** The comparison between two imaging conditions, with LED light and without LED light. Images obtained under various lighting conditions for blue colour with (a) Huawei P8Lite ALE-L04, (b) Samsung Galaxy S7 edge, (c) iPhone 7 Plus



**Fig. S4** The comparison between two imaging conditions, with LED light and without LED light. ANOVA and Tukey HSD post-hoc test were used as statistical analysis method. P < 0.05 was considered as a statistically significant difference. \*\*p<0.01, \*\*\*p<0.001 (a-b) Images obtained with iPhone 7 Plus under various ambient lighting conditions (a) Green colour (b) Red colour (c-d) Images obtained with Samsung Galaxy S7 edge under various ambient lighting conditions (c) Green colour (d) Red colour device



**Fig. S5** Plot of the rescaled  $\Delta$ RGB values of red, blue and green colours together with (a) Samsung Galaxy S7 edge, (b) Huawei P8Lite ALE-L04, (c) iPhone 7 Plus



Plots of the calibration curves of yeast live cell concentration, its viability and  $\Delta RGB$  from iPhone and Samsung

**Fig. S6** Calibration curves for colour change on paper-based devices ( $\Delta RGB$ ) with respect to live cell concentration and viability measured by automated cell counter counting. (a-b) iPhone 7 Plus (c-d) Samsung Galaxy S7