Electronic Supplementary Information for:

Dengue diagnosis on laser printed microzones using smartphone-based detection and multivariate image analysis

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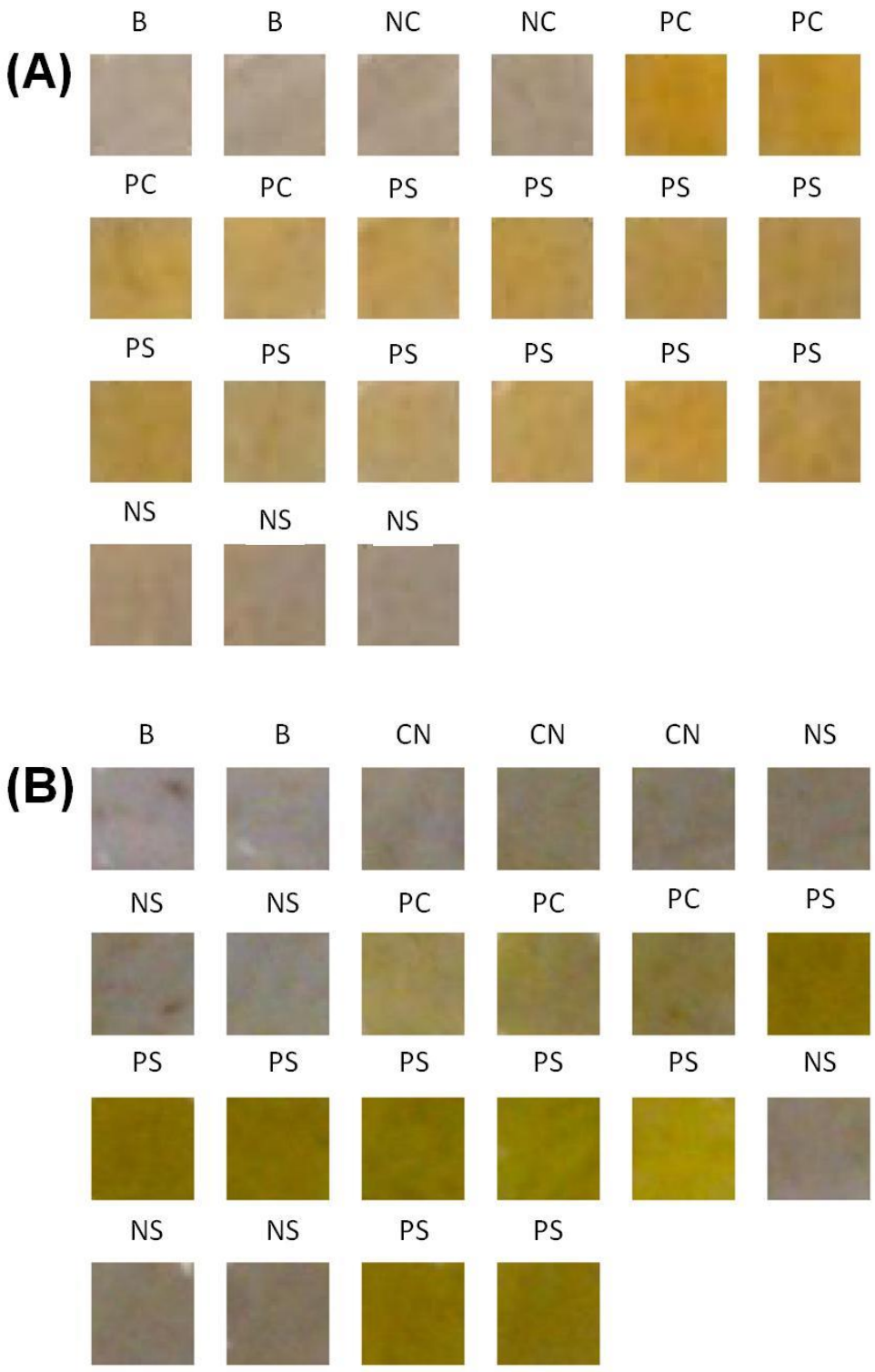

Fig. S1 Hue average color channel histograms for digital images captured after ELISA experiments dedicated to (A) IgM and (B) IgG detection. Abbreviations means: (B) blank, (PC) positive and (NC) negative controls and (PS) positive and (NS) negative samples.

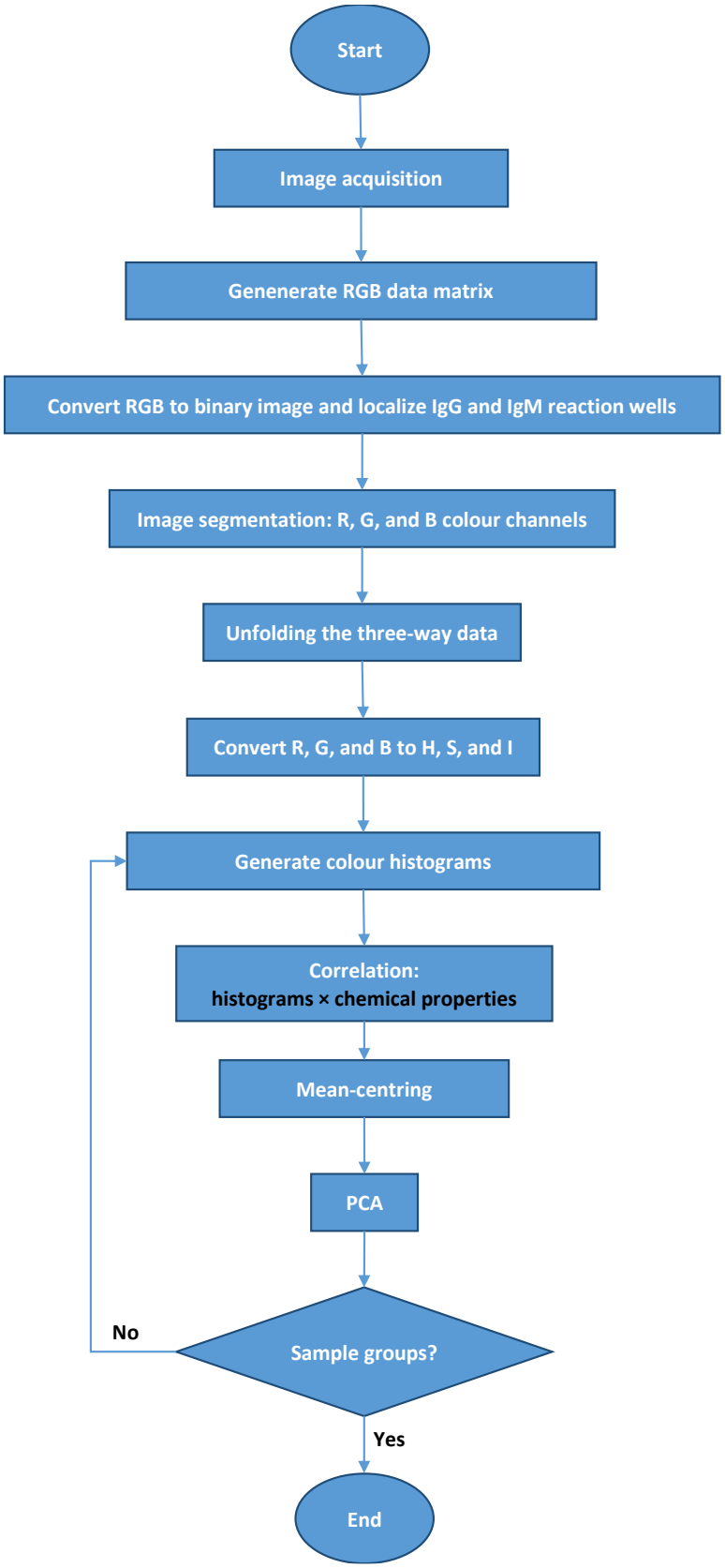

Fig. S2 Scheme of the in-house algorithms developed to analyze the data recorded with smartphone-based detection.

Direct ELISA

Direct ELISA procedure was performed on printed microzones as recently described.[1] First, human IgG were immobilized on polyester surface using 10-µL aliquots from 0 to 100 µg/mL IgG solution prepared in carbonate buffer at pH 9.6. The sample was added on each zone and kept overnight at 4°C. Microzones were then washed and blocked the same procedure described before. After that, a 10-µL aliquot containing the peroxidase-conjugated secondary antibody (anti-human IgG) diluted (1:250) in blocking buffer was added to each zone and kept during 1 hour. The zones were washed twice with PBS-T solution. After washing step, 10 µL of the colorimetric substrate solution was added to the test zone and the colour-producing enzymatic reaction was allowed to proceed during 30 minutes under absence of light.

Digital images were acquired of direct ELISA and the region of interest (ROI) was decomposed and converted the same procedure described in the main text. The analytical parameters used to obtain analytical curve were performed through effective absorbance (eq. 1), where A_H, A_S, A_I are the effective absorbance for hue, saturation and intensity[2].

$$\begin{aligned} A_H &= -\log(H_c/H_b) \\ A_S &= -\log(S_c/S_b) \\ A_I &= -\log(I_c/I_b) \end{aligned} \qquad (1)$$

H_c, S_c, I_c, H_b, S_b and I_b are the average of the H, S and I channels. Afterwards, the total absorbance was found for each concentration. The calibration curve was performed using the total absorbance values (A_T), which represents the sum of A_H, A_S and A_I. The concentration values (predicted values) achieved with smartphone were compared to the values found with microplate reader. As it can be seen in Figure S2, the relationship

exhibited a linear behavior with R^2 value equal to 0.990 in a human IgG concentration range between 0 and 100 µg/mL.

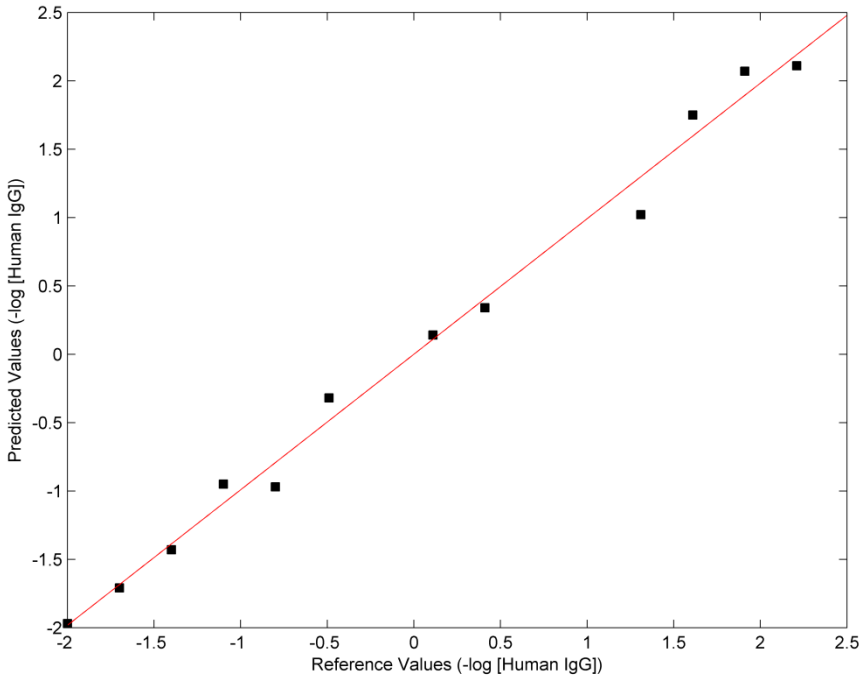

Fig. S3 Presentation of the predicted *versus* reference values for IgG detection using direct ELISA procedure. Predicted and reference values are associated with the concentration values achieved with smartphone and microplate readers, respectively.

References

1. K. A. Oliveira, C. R. de Oliveira, L. A. da Silveira and W. K. T. Coltro, Analyst, 2013, 138, 1114-1121.

2. L. F. Capitan-Vallvey, N. Lopez-Ruiz, A. Martinez-Olmos, M. M. Erenas and A. J. Palma, Anal Chim Acta, 2015, 899, 23-56