Supplementary Information (SI) for Analytical Methods.

Supplementary Information:

Highly selective solution and film based sensor for colorimetric sensing of arginine in aqueous and biological samples.

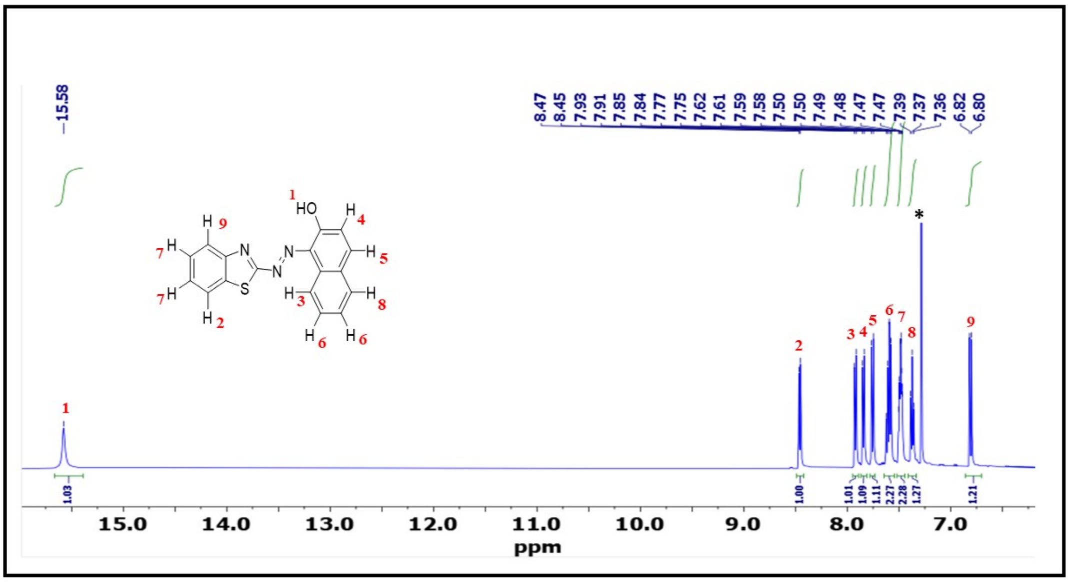


Figure S1: ^{1}H-NMR of BTAN

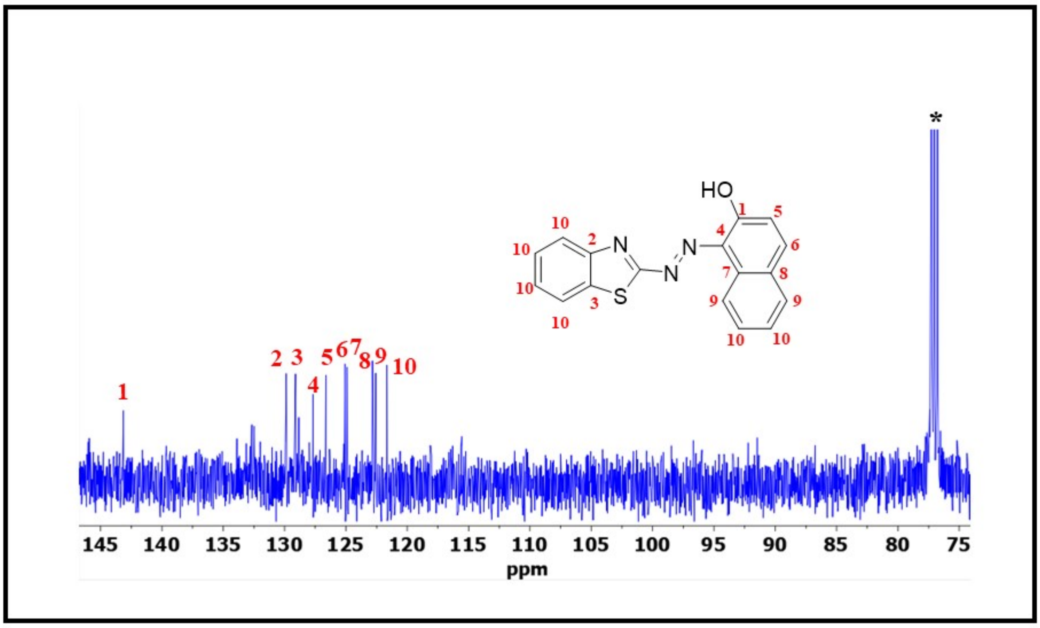


Figure S2: ^{13}C-NMR of BTAN

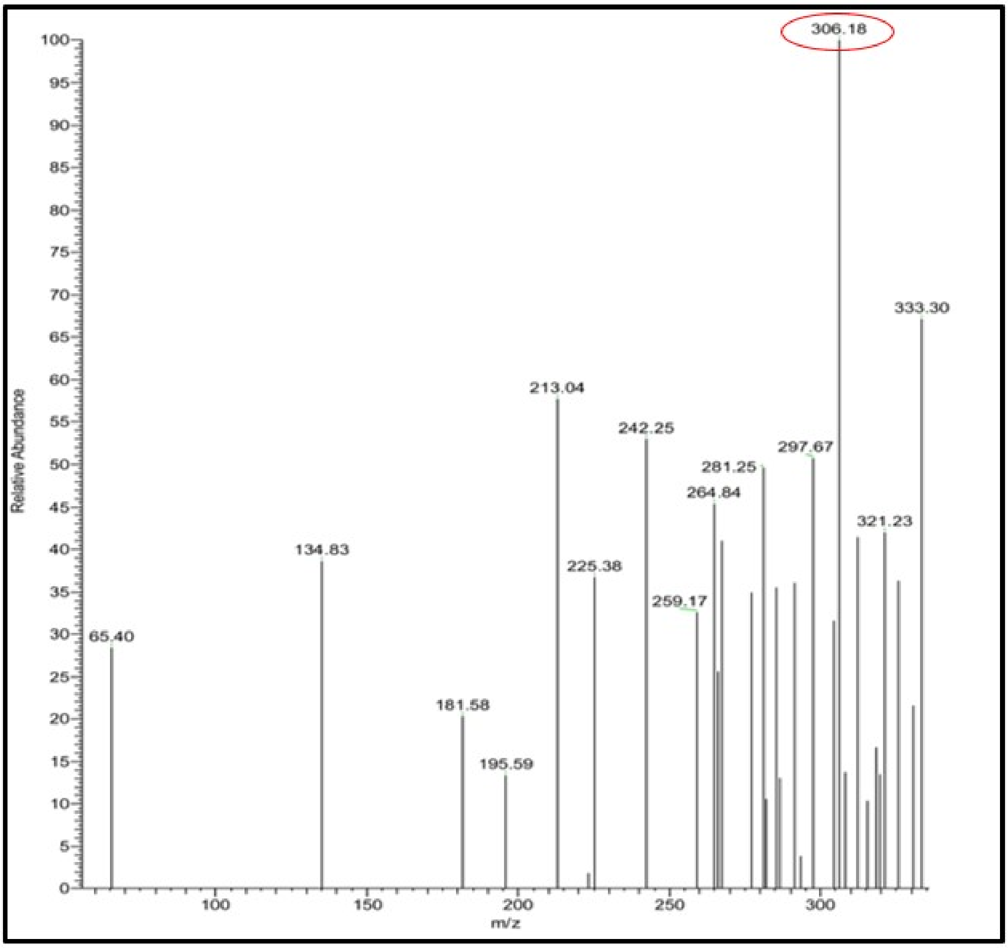


Figure S3: LC-MS spectra of BTAN showing [M+H$^+$] peak

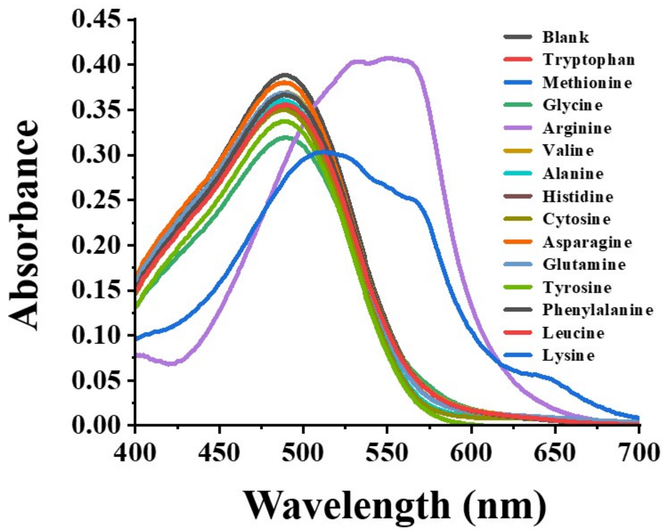


Figure S4: Spectral changes of BTAN upon addition of 1 equiv. of different amino acids.

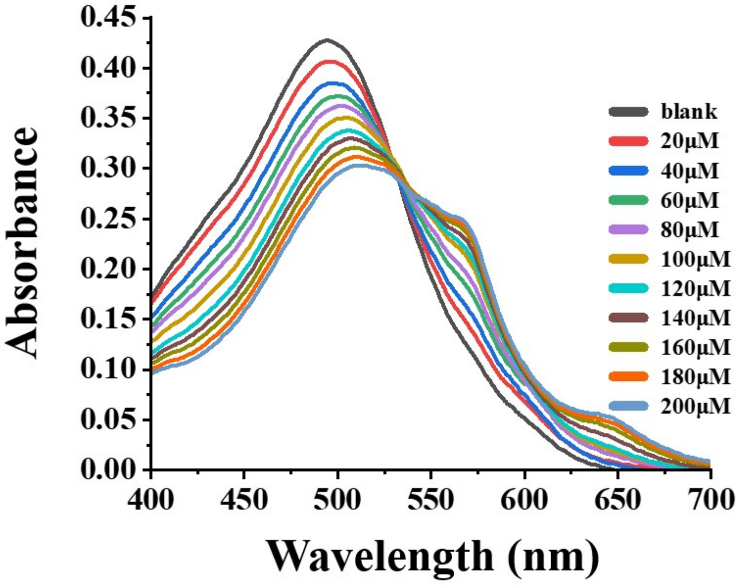


Figure S5: Overlaid absorption spectra of BTAN (10^{-5} M) on aliquot addition of lysine (10^{-4} M) in DMSO-H_2O, (1:1, v/v).

Determination of Binding Constant

The binding constant of BTAN for arginine (Arg) was determined using Benesi-Hildebrand equation. To achieve this, a graph was plotted with the inverse of arginine concentration on the x-axis and the inverse of the change in the absorbance on the y-axis (1/ΔA vs. 1/[Arg]). The equation used for this calculation is given as follows:

$(1/\Delta A = 1/\{Ka(A_0-A_{max})\ [Arg]\} + 1/(A_0-A_{max})$

When plotting 1/ΔA vs. 1/[Arg], a linear graph is obtained. The binding constant, Ka, can be determined by Intercept/Slope obtained from the plot.

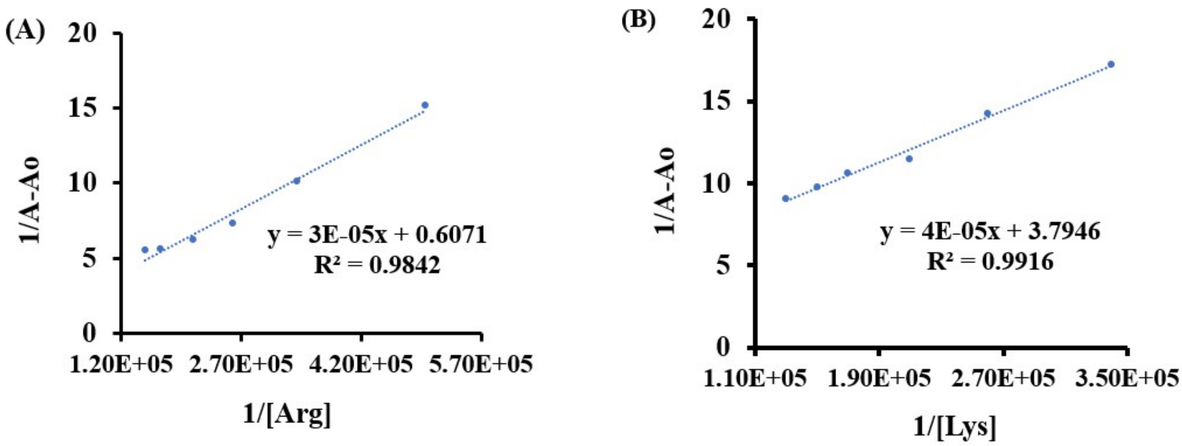


Figure S6: Binding constant plot of **BTAN** for (A) arginine at 530 nm (B) lysine at 567 nm

Determination of limit of detection (LOD):

The detection limit for the arg detection was evaluated using the following formula:

Detection Limit = 3σ/K, where σ is the standard deviation of blank measurement, and K is the slope between the absorbance of host vs guest concentration.

σ = 0.000816

LOD for arginine = (3x0.000816/0.0317) 10^{-5}M = 0.7 x 10^{-6}M = 0.7 μM

LOD for lysine = (3x0.000816/0.0184) 10^{-5}M = 1.3 x 10^{-6}M = 1.3 μM

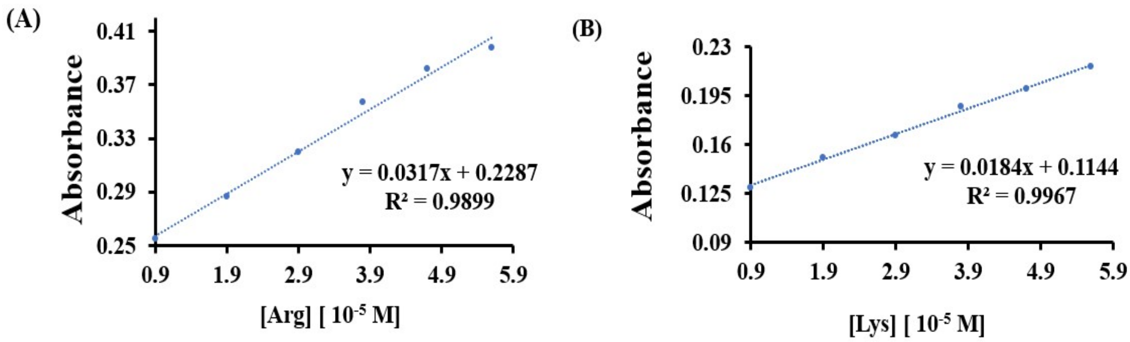


Figure S7: Detection Limit plot of BTAN for (A) arginine at 530 nm (B) lysine at 567 nm in DMSO-H_2O mix solvent

Determination of limit of quantification (LOQ):

The Limit of Quatification (LOQ) is the concentration level at which quantitative results can be obtained with a specified degree of confidence. Mathematically, the LOQ is defined as 10 times the standard deviation of the results from a series of replicates used to establish a justifiable detection limit. [1]

It can be calculated from the formula LOQ = 10 σ /K, where σ is the standard deviation of blank measurement, and K is the slope of calibration curve between the absorbance of host vs guest concentration.

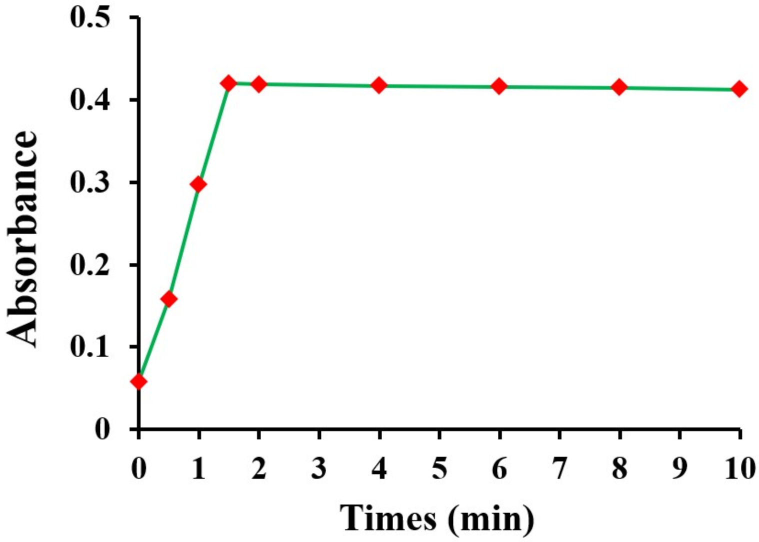
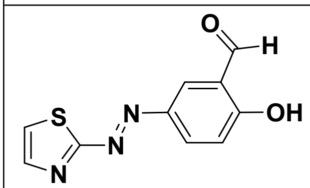
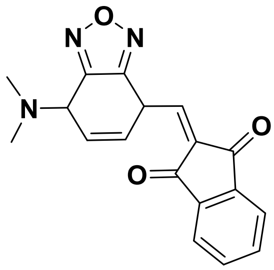
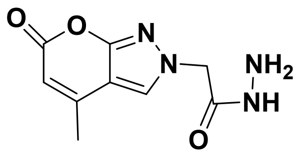
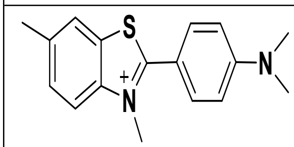
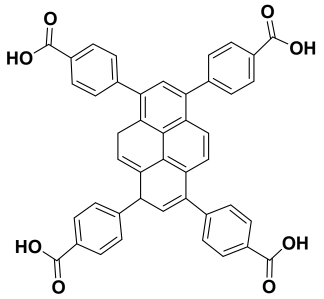
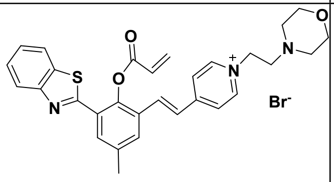
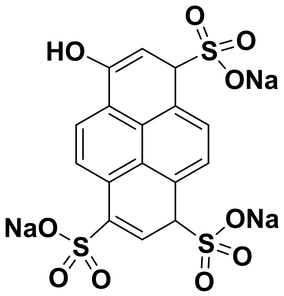
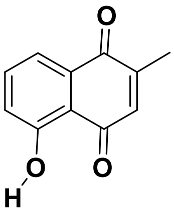
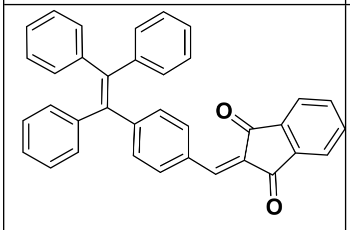
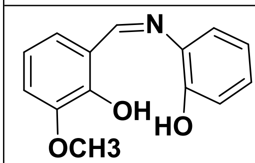
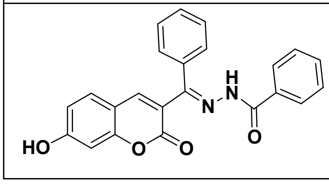
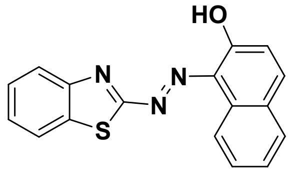


Figure S8: Response time of BTAN towards arginine in DMSO-H_2O, (1:1, v/v) at 564 nm.

Table S1: Comparison of different colorimetric and fluorescence probes utilized for the detection of arginine.

Sensing Probe	Medium	Method	LOD(μM)	Binding constant (M^{-1})	Ref
	ACN-H_2O (8:2 v/v)	Colorimetric & Fluorometric	1.62	1.32×10^5	**[2]**
	Na_2CO_3–Na HCO_3 buffer	Colorimetric & Fluorometric	1.39	NA	**[3]**
	DMSO	Colorimetric & Fluorometric	18	1.57×10^4	**[4]**
	H_2O	Fluorometric	50	NA	**[5]**

	H_2O	Fluorometric	2.3	6.8×10^5	**[6]**
	DMSO	Fluorometric	2.24	NA	**[7]**
	H_2O	Colorimetric & Fluorometric	1.94	NA	**[8]**
	H_2O	Colorimetric & Fluorometric	1	NA	**[9]**
	THF/H_2O	Colorimetric & Fluorometric	100	NA	**[10]**
	DMSO- H_2O (1:1 v/v)	Fluorometric	NA	NA	**[11]**
	CH_3CN- H_2O (9:1, v/v)	Colorimetric & Fluorometric	NA	NA	**[12]**

	DMSO- H_2O (1:1 v/v)	Colorimetric	0.7	0.12×10^6	**This work**

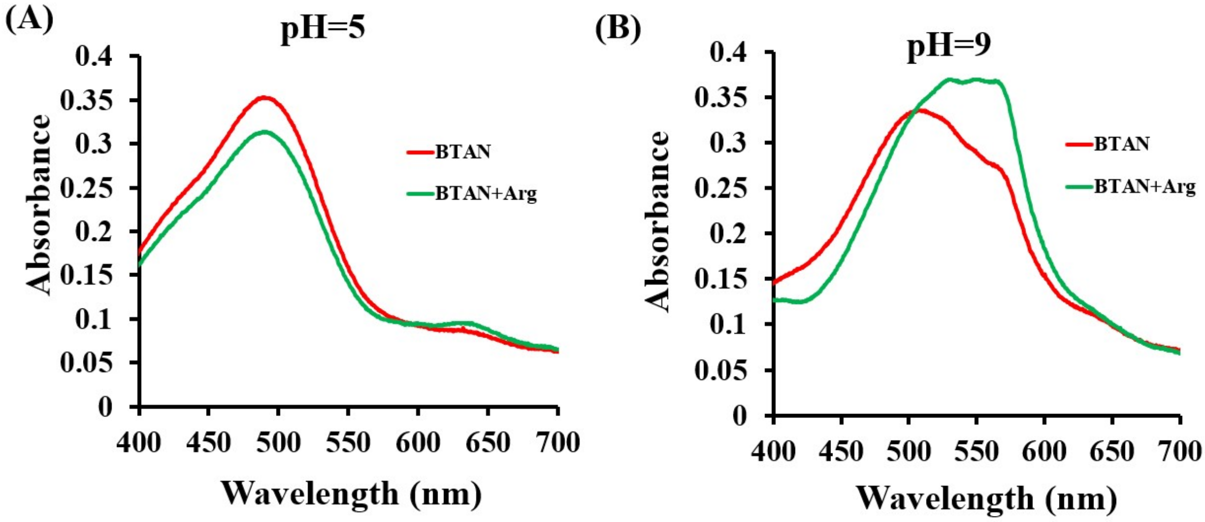


Figure S9: The effect of pH on the sensing performance of BTAN upon Arginine addition (A) pH=5 (B) pH=9

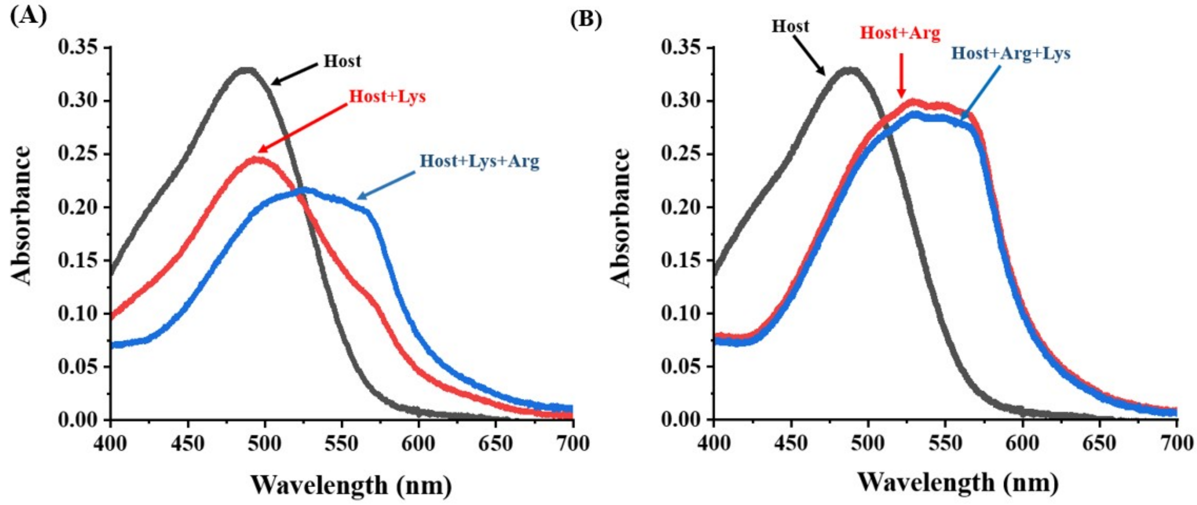


Figure S10: Competitive experiment of BTAN; (A) Arginine in the presence of Lysine (B) Lysine in the presence of Arginine

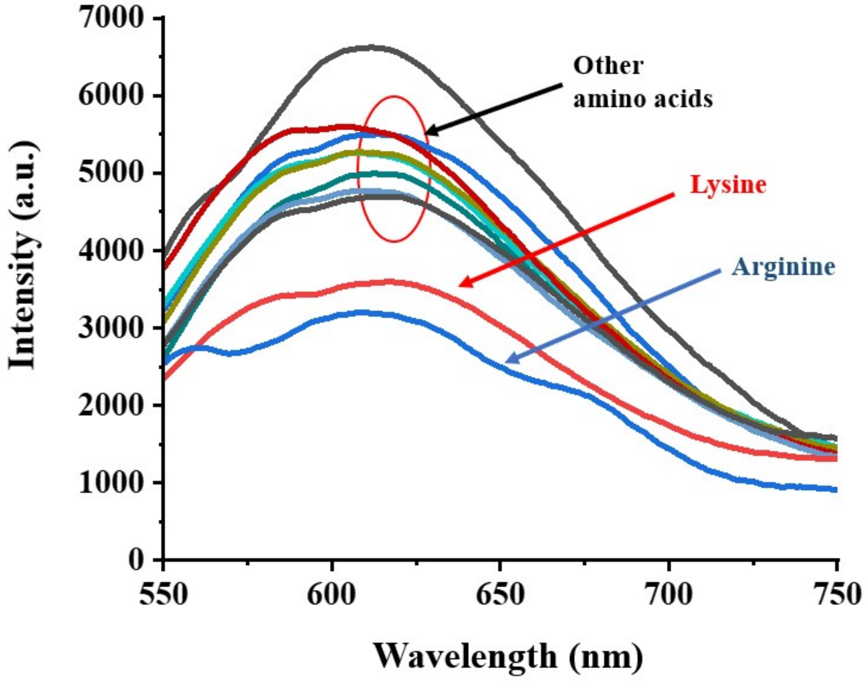


Figure S11: Fluorescence emission of BTAN upon addition of different amino acids, when excited at 472 nm.

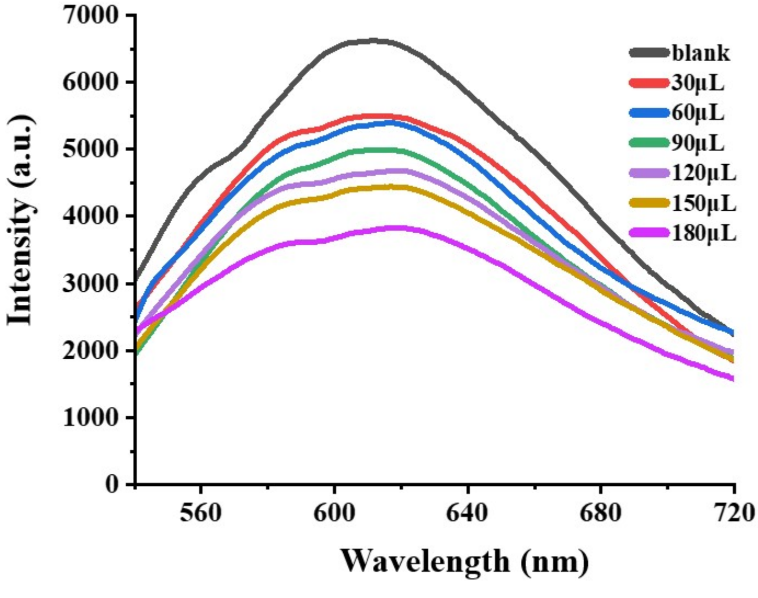


Figure S12: Overlaid emission spectra of BTAN (10^{-5} M) with lysine (10^{-4} M) in DMSO-H_2O, (1:1, v/v).

Table S2: RGB content of probe alone, on Arg addition and followed by TBAS addition.

Sample	Red	Green	Blue
BTAN	252	64	23
BTAN + Arg	181	49	113
BTAN + Arg + TBAS	224	60	39

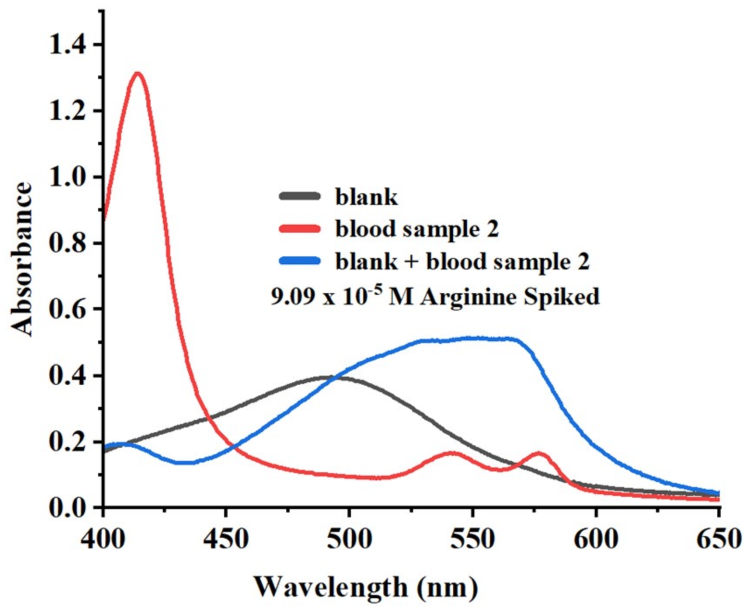


Figure S13: Absorption spectra of **BTAN** with 9.09 x 10^{-5} M arginine spiked blood sample

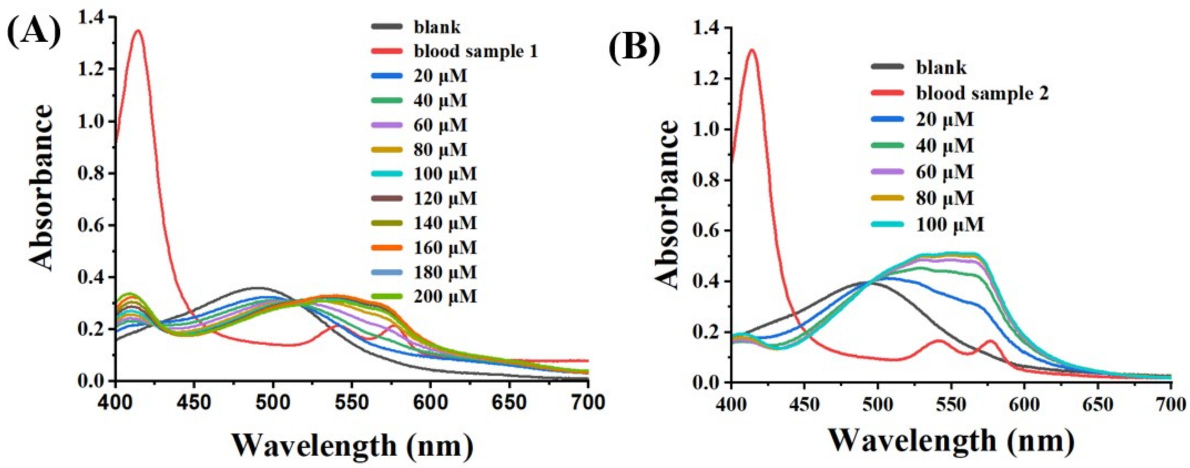


Figure S14: Absorption spectra of BTAN upon aliquot addition of spiked blood samples, (A) 2.7 x 10^{-5} M arginine spiked and (B) 9.09 x 10^{-5} M arginine blood sample.

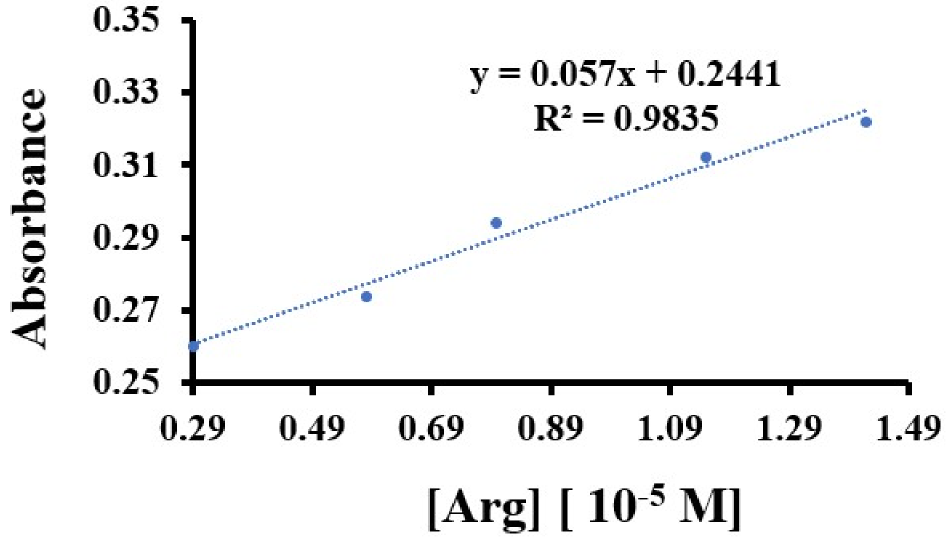


Figure S15: LOD plot of sensor BTAN upon aliquot addition of 2.7 x 10^{-5} M arginine spiked blood sample.

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1. M. Taleuzzaman, *O. M. C. I. J.*, 2018, **7**, OMCIJ.MS.ID.555722.
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