

Supplementary Material

# Fungicide Tebuconazole Influences the Structure of Human Serum Albumin Molecule

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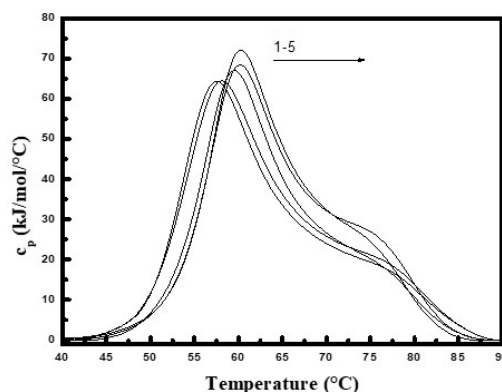
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**Table S1.** Peak position and intensity of fluorescence parameters for the interaction between HSA and TB.

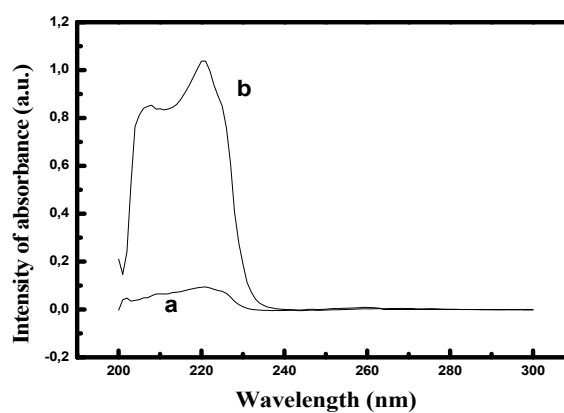
| Complex        | Peak | Peak Position<br>[ $\lambda_{ex}/\lambda_{em}$ nm/nm] | Intensity of<br>Fluorescence |
|----------------|------|-------------------------------------------------------|------------------------------|
| HSA            | a    | 250/250 → 310/310                                     | 221.58 → 830.90              |
|                | b    | 240/480                                               | 83.44                        |
|                | 1    | 275/340                                               | 674.46                       |
|                | 2    | 230/330                                               | 110.54                       |
|                | a    | 250/250 → 310/310                                     | 1015.86 → 1015.88            |
| TB/HSA<br>16/1 | b    | 240/480                                               | 402.56                       |
|                | 1    | 275/340                                               | 628.73                       |
|                | 2    | 230/330                                               | 106.32                       |

**Table S2.** Association constants of competitive experiments for the interaction between TB and SA.

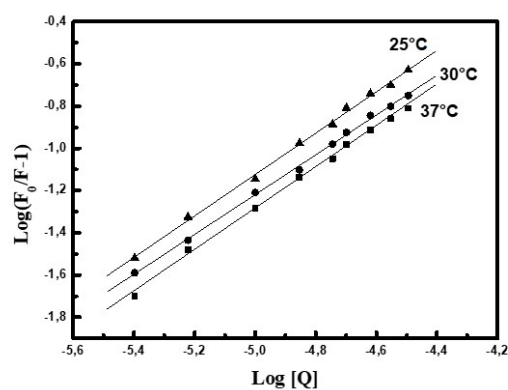
| Site Markers | $K_A$ (L/mol)               | $n$             |
|--------------|-----------------------------|-----------------|
| Blank        | $8.51 \times 10^3 \pm 0.09$ | $1.01 \pm 0.02$ |
| KTF          | $0.32 \times 10^3 \pm 0.09$ | $0.72 \pm 0.02$ |
| IBF          | $6.03 \times 10^3 \pm 0.09$ | $0.93 \pm 0.02$ |



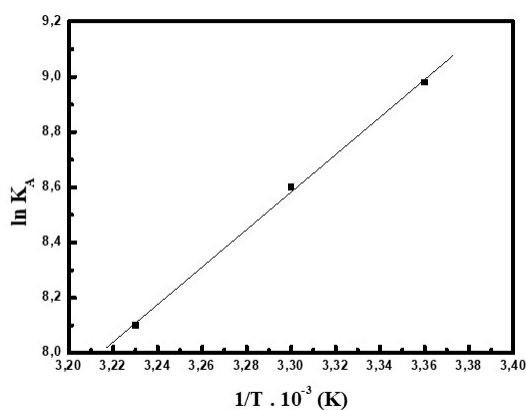
**Figure S1.** DSC curves of HSA: (1) DSC curve of HSA,  $c(\text{HSA}) = 3 \times 10^{-5} \text{ mol/L}$ ; (2–5): TB/HSA,  $3 \times 10^{-5} \text{ mol/L}$  HSA in the presence of  $3 \times 10^{-5}$ ;  $15 \cdot 10^{-5}$ ;  $21 \times 10^{-5}$ ;  $3 \times 10^{-4} \text{ mol/L}$  TB.



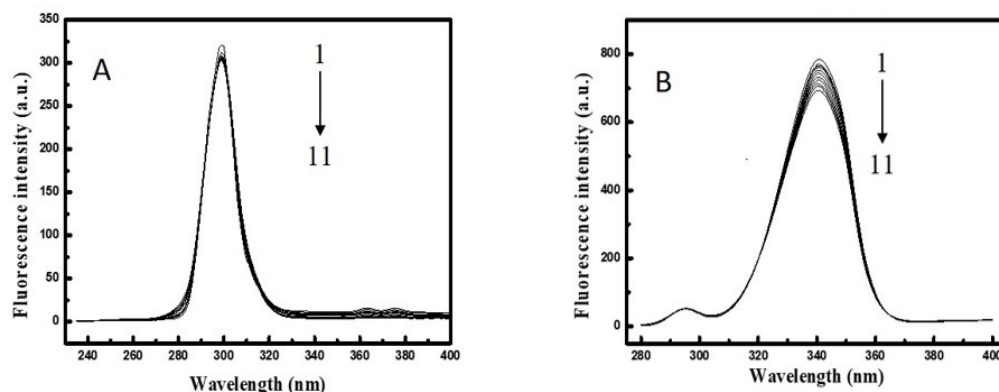
**Figure S2.** Absorption spectra of TB. (a)  $c(\text{TB}) = 2 \times 10^{-6} \text{ mol/L}$ ; (b)  $c(\text{TB}) = 32 \times 10^{-6} \text{ mol/L}$ ;  $\text{pH} = 7.4$ ;  $t = 25^\circ \text{C}$ .



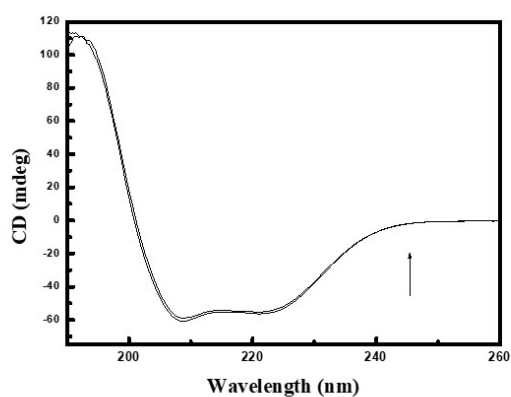
**Figure S3.** Hill plots of TB quenching effect on HSA fluorescence at the different temperatures.  $c(\text{HSA}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 0\text{--}32 \times 10^{-6} \text{ mol/L}$ ;  $\lambda_{\text{exc}} = 295 \text{ nm}$ ;  $\lambda_{\text{em}} = 300\text{--}500 \text{ nm}$ ;  $\text{pH} = 7.4$ ;  $t = 25, 30$  and  $37^\circ \text{C}$ .



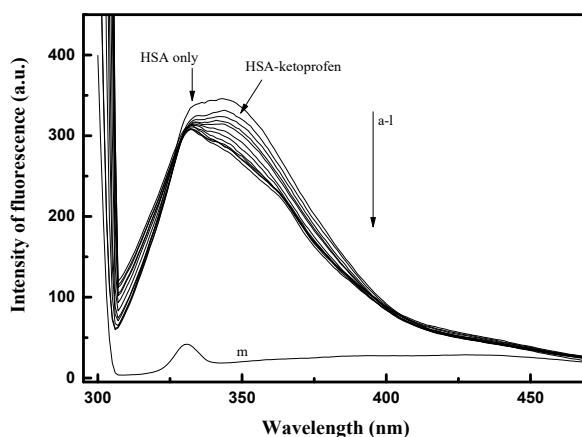
**Figure S4.** Van't Hoff plot for the interaction between TB and HSA.  $c(\text{HSA}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 0\text{--}32 \times 10^{-6} \text{ mol/L}$ ;  $\lambda_{\text{exc}} = 295 \text{ nm}$ ;  $\lambda_{\text{em}} = 300\text{--}500 \text{ nm}$ ;  $\text{pH} = 7.4$ .



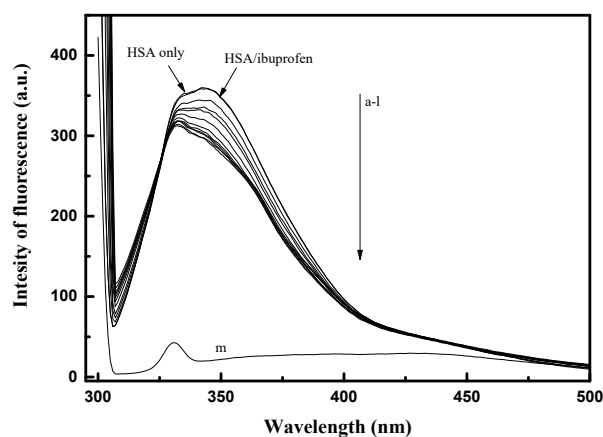
**Figure S5.** (A) Synchronous fluorescence spectra of HSA in the presence of different concentrations of TB;  $c(\text{HSA}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 0\text{--}32 \times 10^{-6} \text{ mol/L}$ ;  $\Delta\lambda = 15 \text{ nm}$ . (B) Synchronous fluorescence spectra of HSA in the presence of different concentrations of TB;  $c(\text{HSA}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 0\text{--}32 \times 10^{-6} \text{ mol/L}$ ;  $\Delta\lambda = 60 \text{ nm}$ .



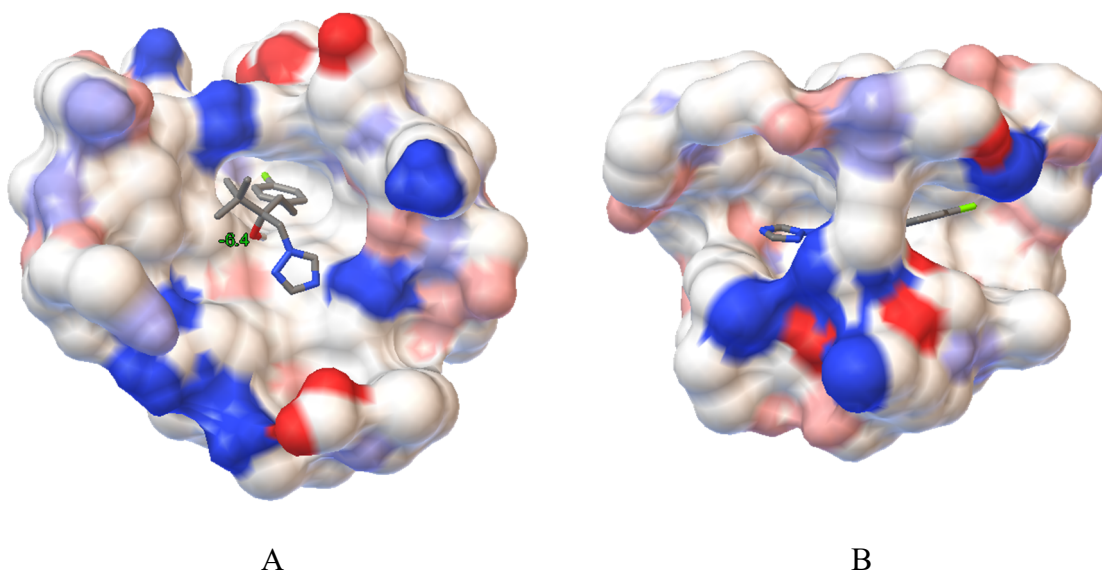
**Figure S6.** CD spectra of HSA and TB/HSA;  $c(\text{HSA}) = 3 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 15 \times 10^{-6} \text{ mol/L}$ ;  $\text{pH} = 7.4$ ;  $t = 25^\circ\text{C}$ .



**Figure S7.** Effect of site marker ketoprofen to the TB/HSA system;  $c(\text{KTF}) = c(\text{HSA}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 0\text{--}32 \times 10^{-6} \text{ mol/L}$ ; (a-l): TB/HSA: 0,2,4,6,10,14,18,20,24,28,32; (m): fluorescence spectrum of ketoprofen,  $c(\text{KTF}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $\text{pH} = 7.4$ ;  $t = 25^\circ\text{C}$ .



**Figure S8.** Effect of site marker ibuprofen to the TB/HSA system;  $c(\text{IBF}) = c(\text{HSA}) = 2 \times 10^{-6} \text{ mol/L}$ ;  $c(\text{TB}) = 0\text{--}32 \times 10^{-6} \text{ mol/L}$ ; (**a–l**): TB/HSA: 0,2,4,6,10,14,18,20,24,28,32; (**m**): fluorescence spectrum of ibuprofen,  $c(\text{IBF}) = 2 \times 10^{-6} \text{ mol/L}$ ; pH = 7.4;  $t = 25^\circ\text{C}$ .



**Figure S9.** Electrostatic potential of the ligand binding pocket in subdomain IIIA (site II) of HSA. The negative and positive electrostatic potentials are colored red and blue, respectively.