MIKOTOXIN-ALBUMIN KÖLCSÖNHATÁSOK VIZSGÁLATA ÉS JELENTŐSÉGÜK

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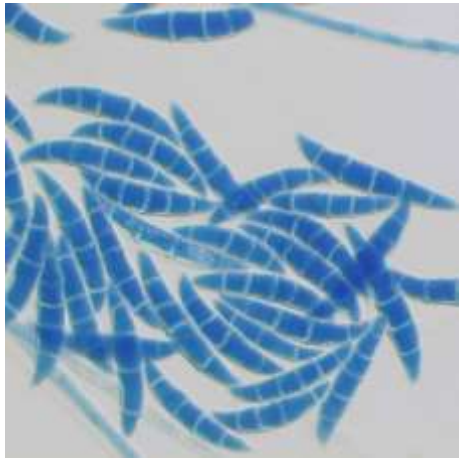
PTE GYTK Gyógyszerhatástani Tanszék, Pécs

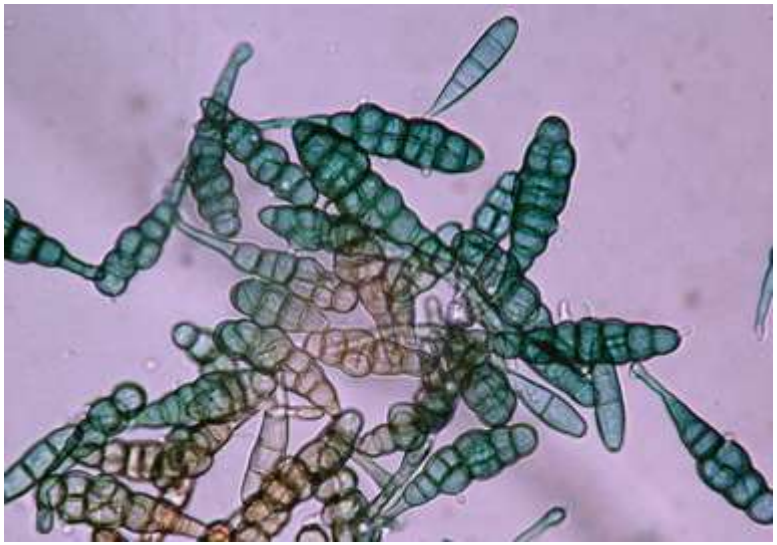
TOX'2019

Tudományos konferencia

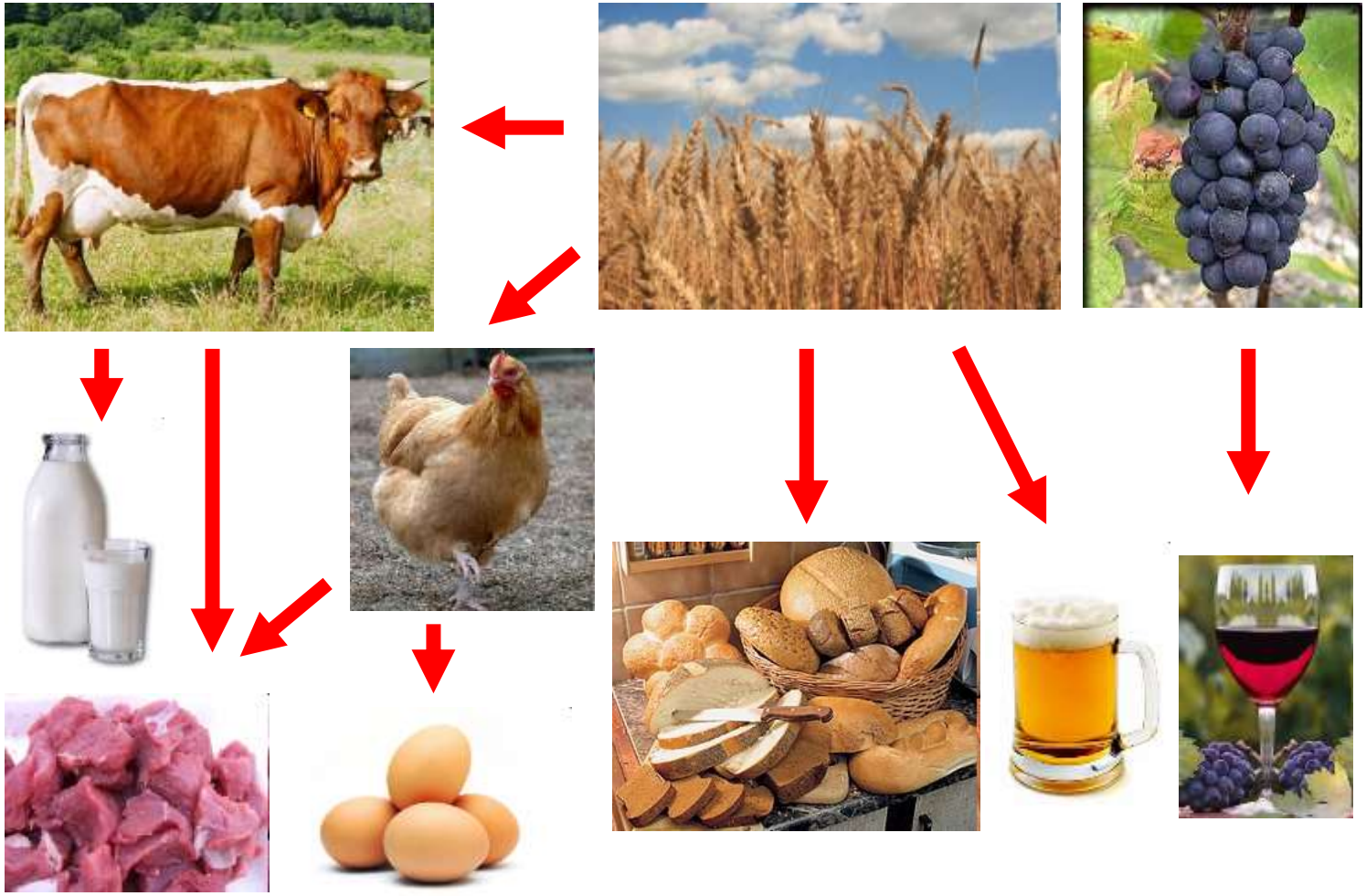
2019. 10. 10.

Mikotoxinok



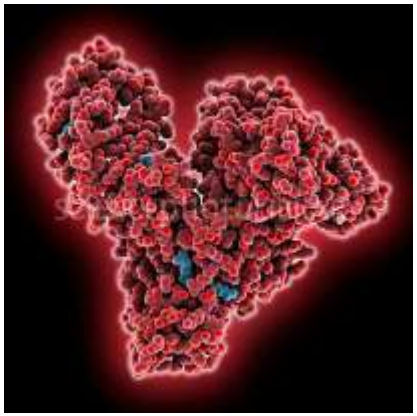


Mikotoxinok

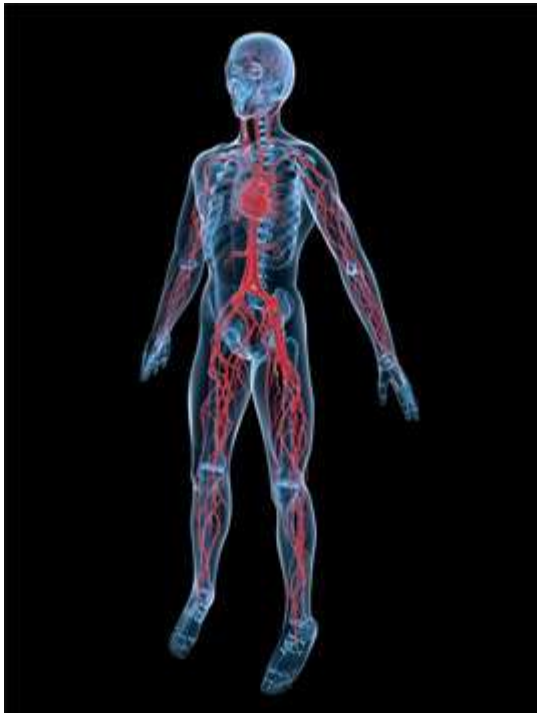


Humán szérum albumin (HSA)

- 66,5 kDa méretű makromolekula (585 aminosav)
- Vízoldható
- Plazmakoncentráció: 35-50 g/l
- Szintézis: májban (napi 10-15 g)
- Felezési idő a plazmában: ~ 3 hét
- Ellenálló: stabil pH 4 és 9 között, oldódik 40%-os etanolban, még 60°C-os hőmérsékletet is hosszabb ideig elvisel



HSA



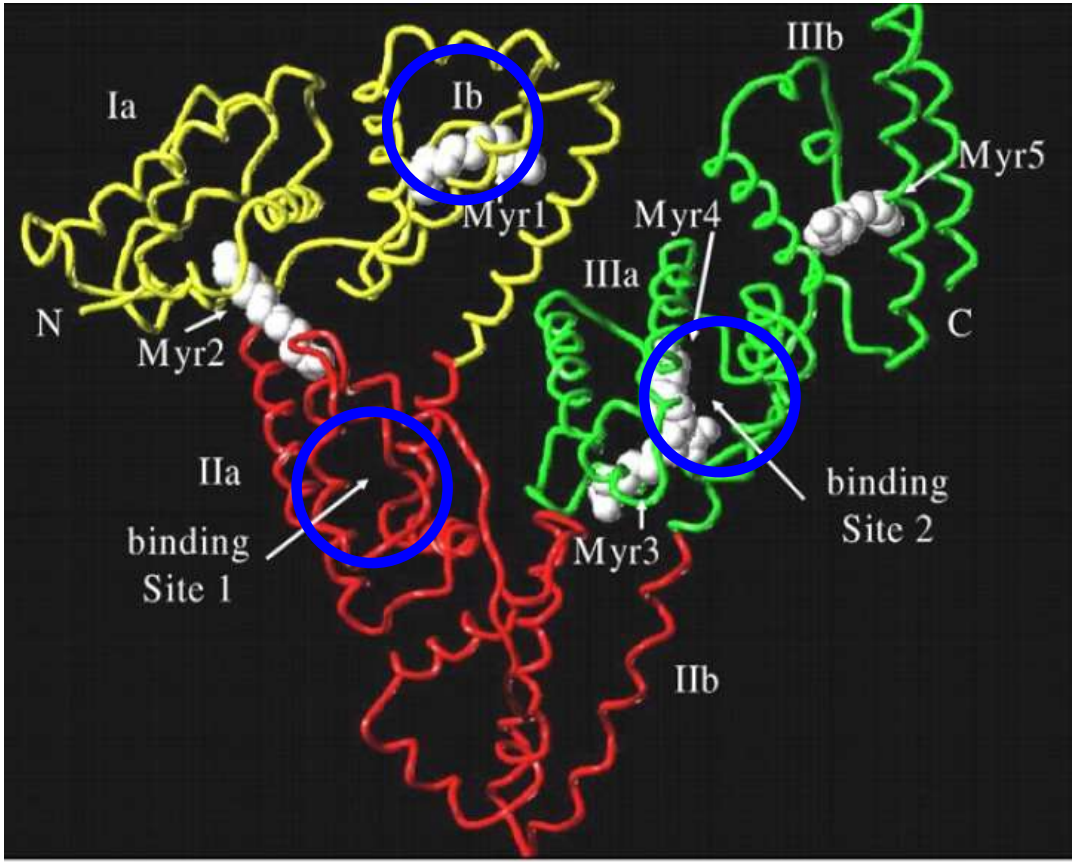
- Onkotikus nyomás fenntartása
- Puffer
- Antioxidáns
- (Pszeudo)-enzimatikus tulajdonság
- **„Transzport”**

Magas plazmafehérje kötődés

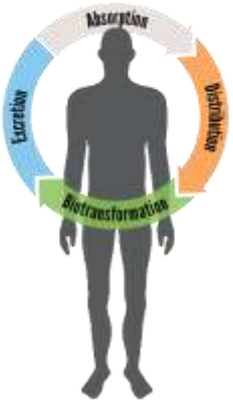
lassabb szöveti felvétel

lassabb elimináció

HSA - kötőhelyek



Miért érdekesek a mikotoxin-albumin kölcsönhatások?

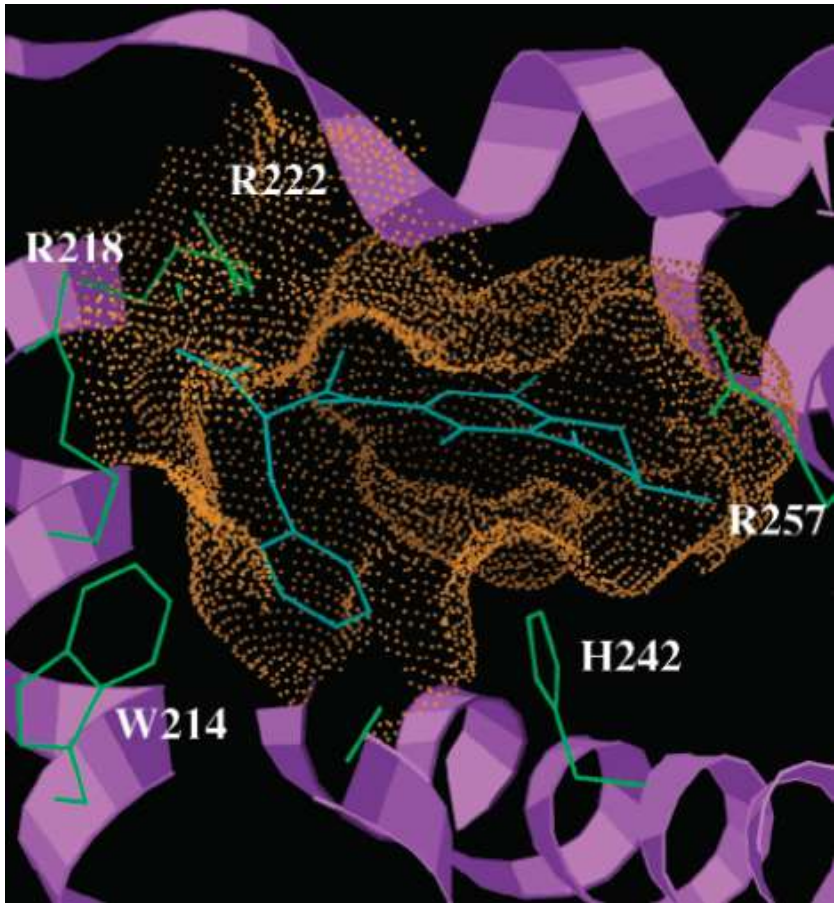


Toxikokinetikai tulajdonságok?

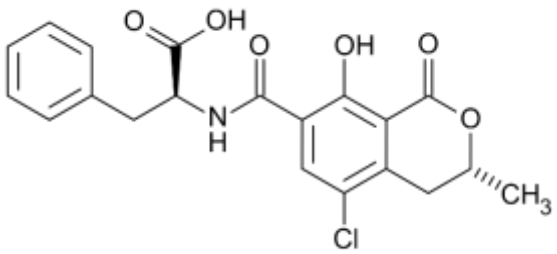
Leszorításos kölcsönhatások eredménye?

Faji eltérések?

Ochratoxin A (OTA)



Perry és mtsai., *J. Phys. Chem. B* 108 (**2004**) 43.



log*K* ~ 7,3

$t_{1/2}$ ~ 1 hónap

Miért lehet még érdekes az OTA-albumin kölcsönhatás?

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BSA-based sample clean-up columns for ochratoxin A determination in wine: Method development and validation

Tânia Leal, Luís Abrunhosa, Lucília Domingues, Armando Venâncio, Carla Oliveira*

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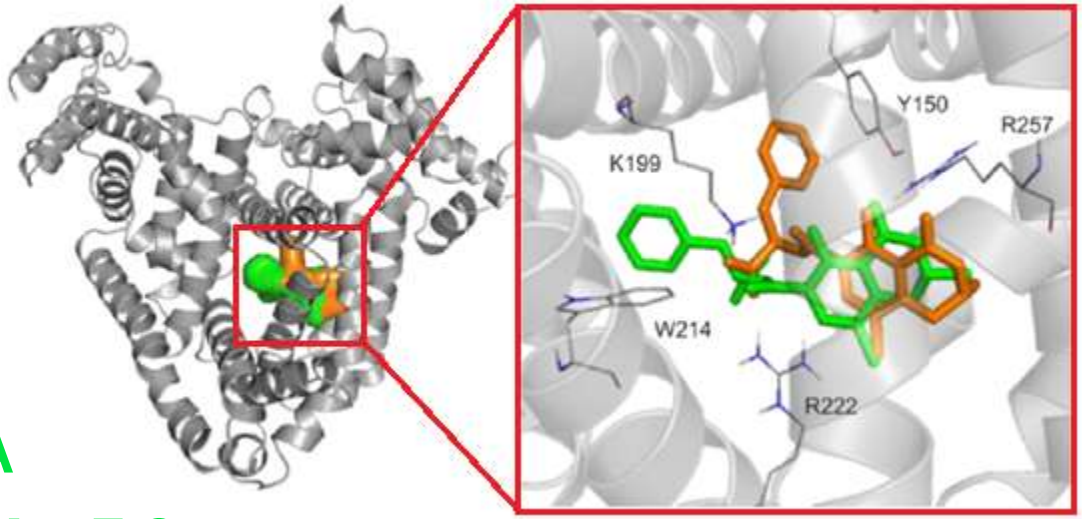
ARTICLE INFO

Keywords:
Ochratoxin A
Bovine serum albumin
Solid-phase extraction
Wine analysis
Method validation

ABSTRACT

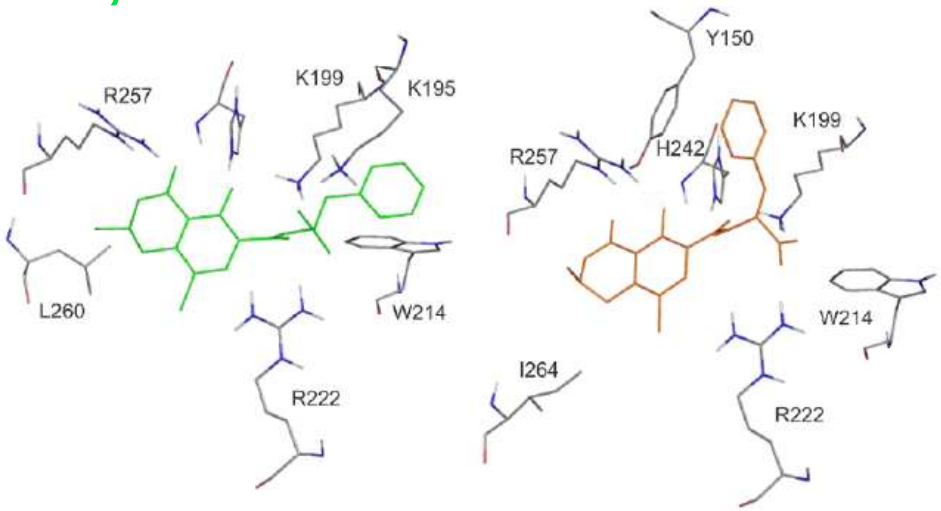
Analytical chromatographic techniques for mycotoxins control are well established, but they often depend on costly immunoaffinity sample clean-up. Serum albumins, particularly that from bovine origin (BSA), have stable binding affinity towards some mycotoxins, and can be cheaper alternative receptors for sample clean-up due to their wide availability. Thus, this work used BSA immobilized in agarose beads as a novel solid-phase extraction method for quantification of ochratoxin A (OTA) in wine. Constructed BSA-agarose columns could extract OTA efficiently from red wine after its dilution (4-fold) in 0.1 M Tris pH 8.0. The method was linear ($R^2 = 0.9999$) in the OTA concentration range studied (0.05 to 3.0 $\mu g\,L^{-1}$), with recovery rates above 98%. It also showed low detection (0.017 $\mu g\,L^{-1}$) and quantification (0.051 $\mu g\,L^{-1}$) limits. The efficacy of the BSA-based method was further validated by direct comparison with commercial immunoaffinity columns. Portuguese wines analyzed by both methods had agreeing results.

Ochratoxin A



OTA

logK ~ 7,3

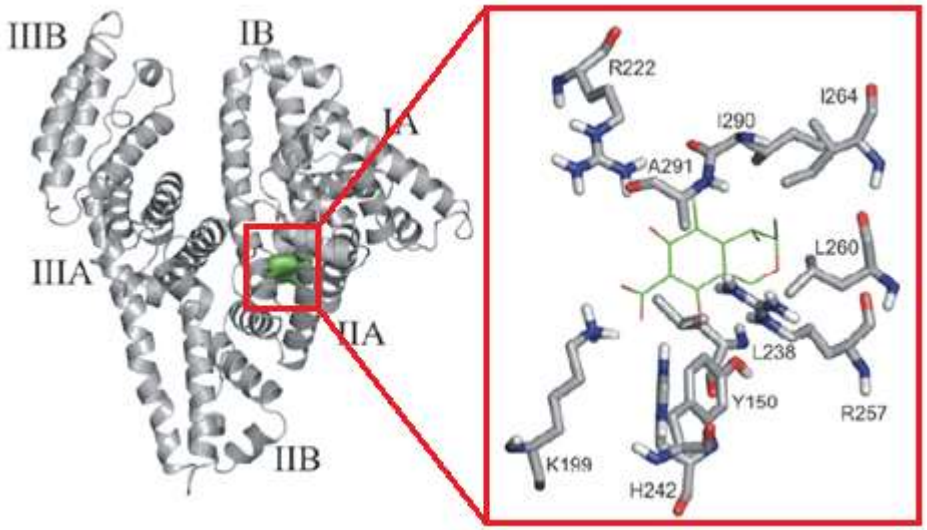


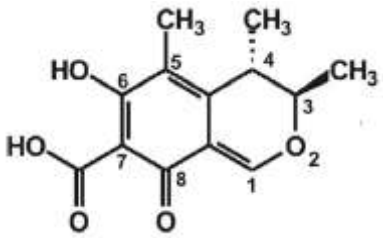
2'R-OTA

logK ~ 6,4

Faisal és mtsai., *Toxins* 10 (**2018**) 353.

Citrinin és dihidrocitrinon

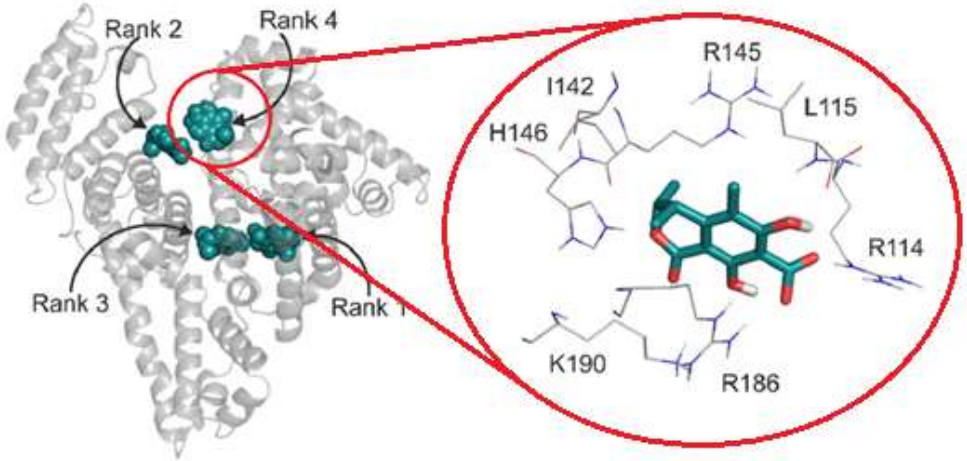


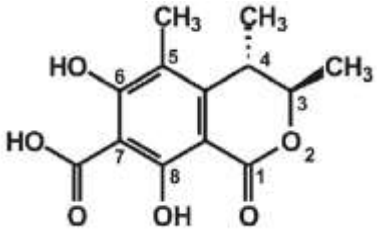


Citrinin

log*K* = 5,3

Poór és mtsai., *Toxins* 7 (**2015**) 5155.



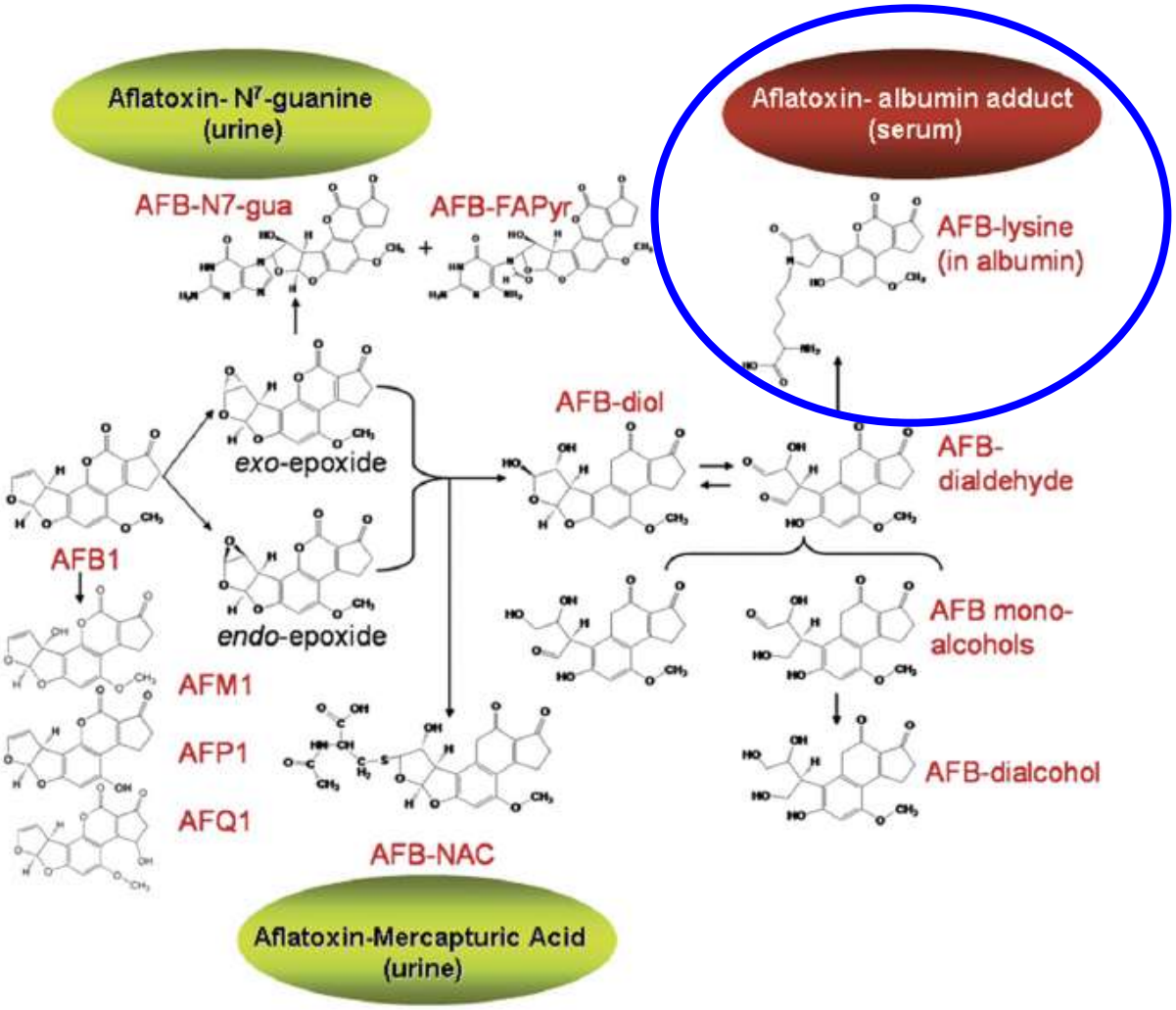


Dihidrocitrinon

log*K* = 5,5

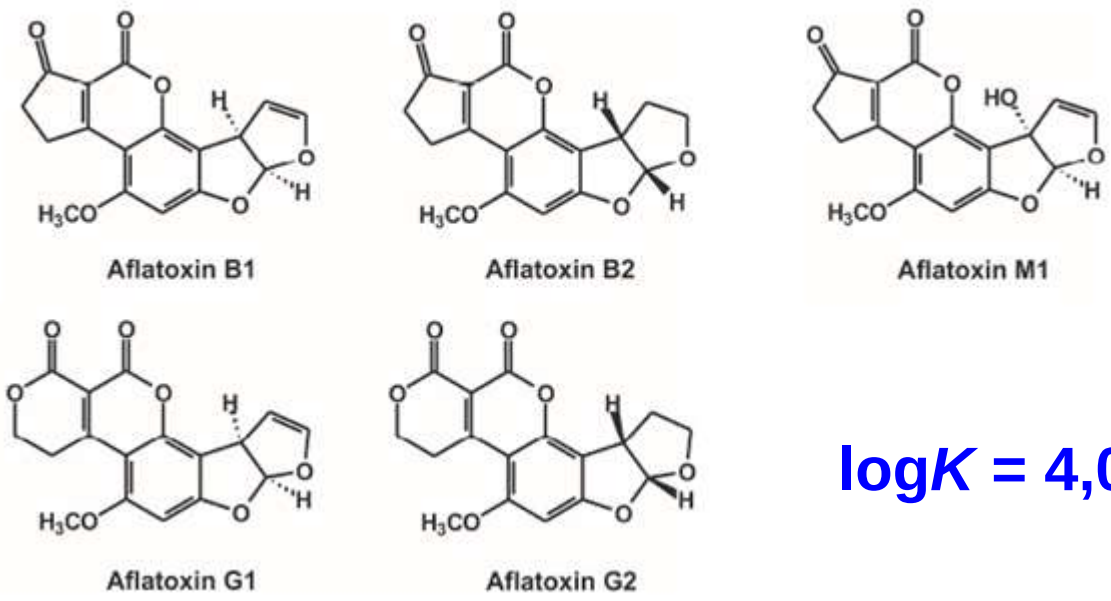
Faisal és mtsai., *Mycotoxin Res.* 35 (**2019**) 129.

Aflatoxinok

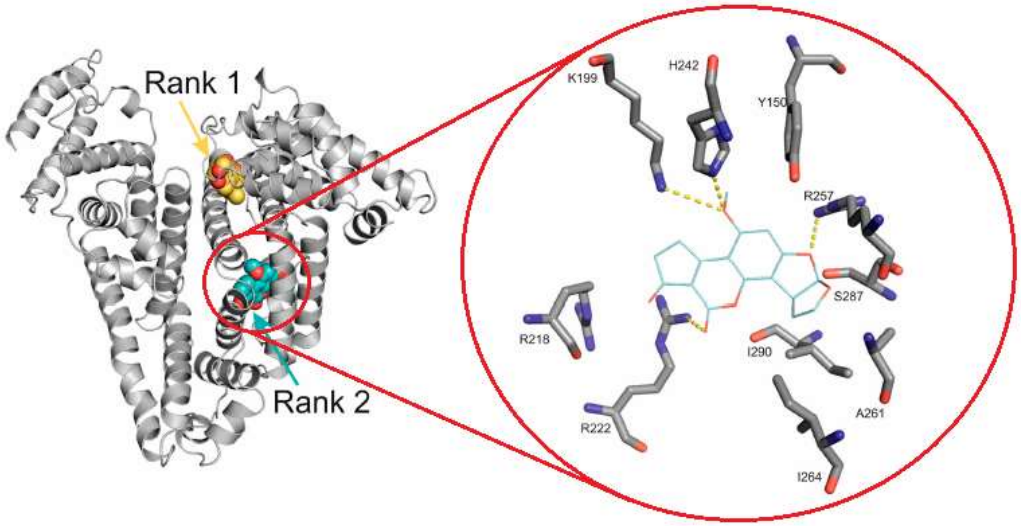


Kensler és mtsai., *Toxicol. Sci.* 120 (**2011**) S28.

Aflatoxinok

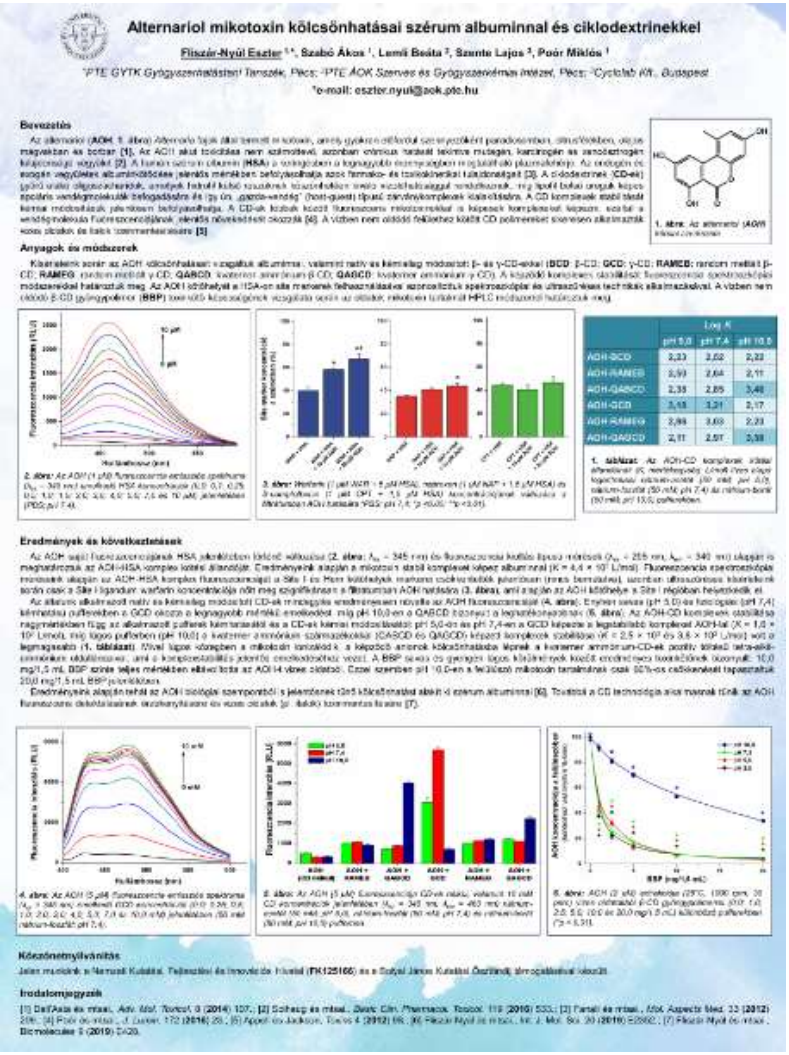


log*K* = 4,0-4,5



Poór és mtsai., *Toxins* 9 (**2017**) 339.

Alternariol (AOH)

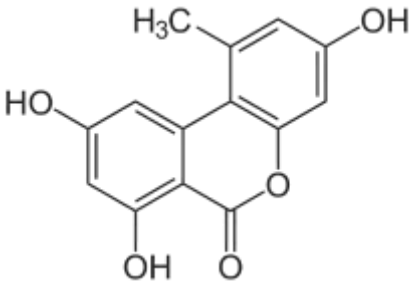


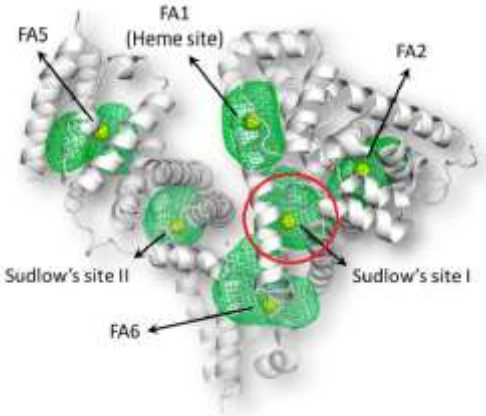
Alternariol mikotoxin kölcsönhatásai szérum albuminnal és ciklodextrinekkel

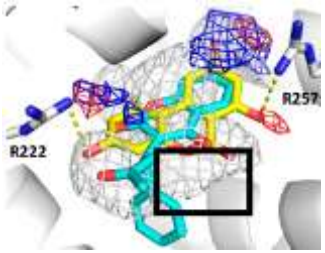
Fliszár-Nyúl Eszter [1,*], Szabó Ákos [1], Lemli Beáta [2], Szente Lajos [3], Poór Miklós [1]

[1]PTE GYTK Gyógyszerhatástani Tanszék, Pécs; [2]PTE ÁOK Szerves és Gyógyszerkémiai Intézet, Pécs; [3]Cyclolab Kft., Budapest

*e-mail: eszter.nyul@aok.pte.hu

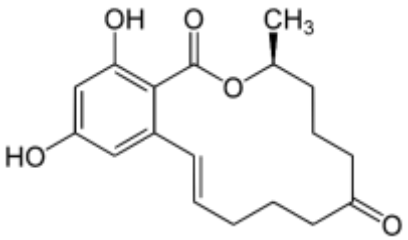


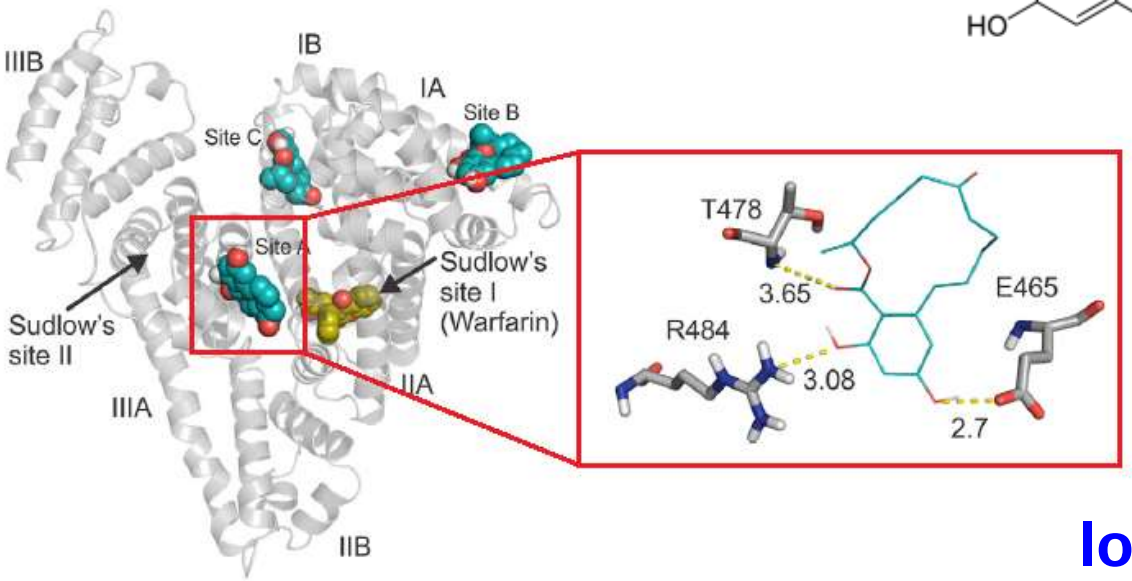




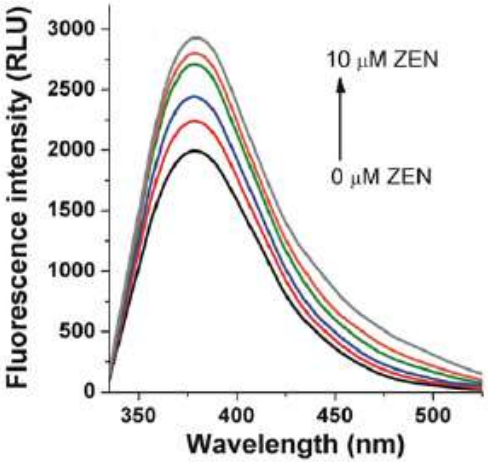
Fliszár-Nyúl és mtsai., *Int. J. Mol. Sci.* 20 (**2019**) 2352.

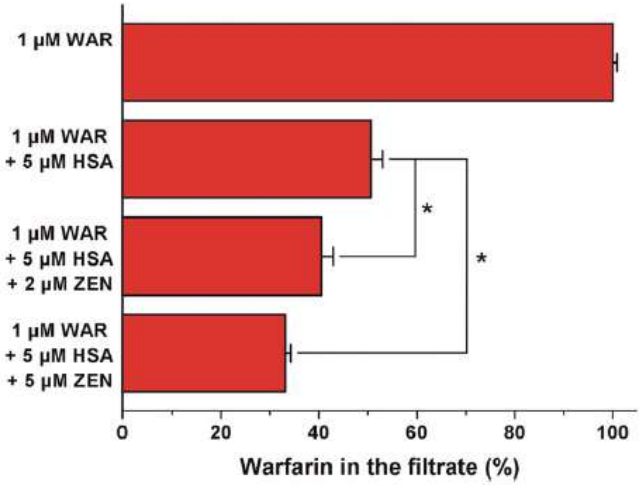
Zearalenon (ZEN)





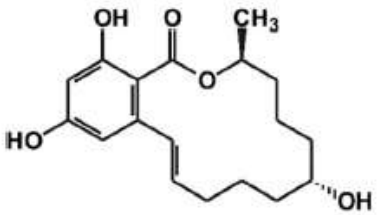
log*K* = 5,1



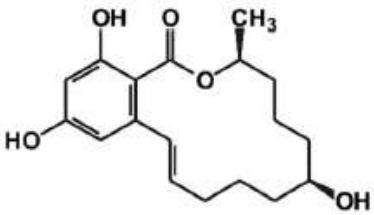


Poór és mtsai., *J. Photochem. Photobiol. B* 170 (**2017**) 16.

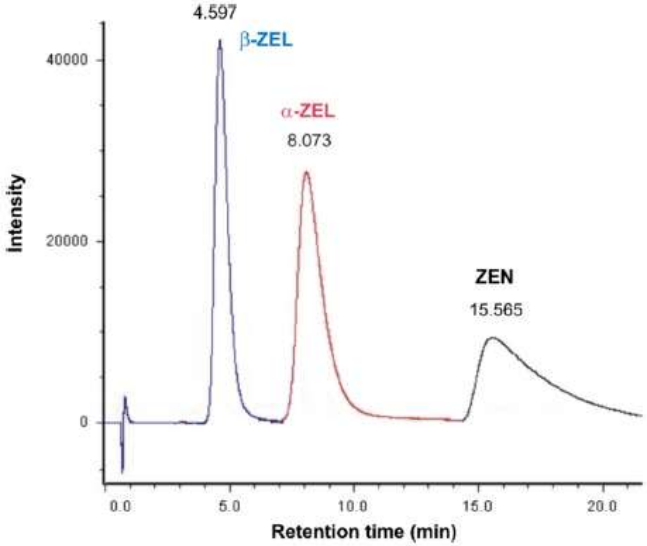
α- és β-zearalenol



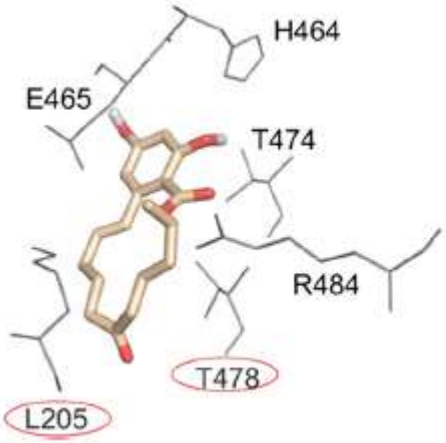
α-Zearalenol (α-ZEL)

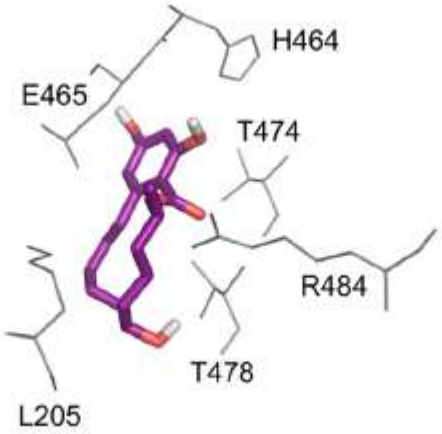


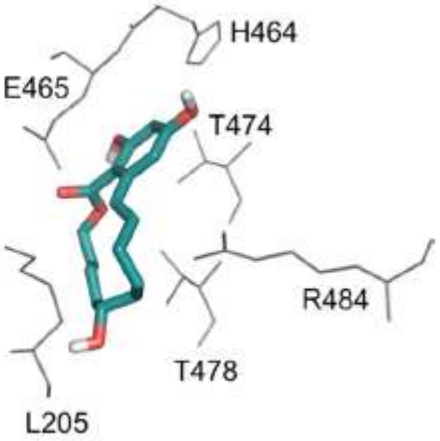
β-Zearalenol (β-ZEL)



ZEN **log*K* = 5,1** > **α-ZEL** **log*K* = 4,7** > **β-ZEL** **log*K* = 4,3**







Faisal és mtsai., *Mycotoxin Res.* 34 (**2018**) 269.

Humán vs. egyéb albuminok

~ 25-30% eltérés a primer struktúrában!

```
                189                                                 232
Human           GKAS SAKQRLKCAS LQKFGERAFK AWAVARLSQR FPKAEFAEVS
Porcine         VLTS AAKQRLKCAS IQKFGERAFK AWSLARLSQR FPKADFTEIS
Bovine          VLAS SARQRLRCAS IQKFGERALK AWSVARLSQK FPKAEFVEVT
Rat             ALVA AVRQRMKCSS MQRFGERAFK AWAVARMSQR FPNAEFAEIT

            233                                                 282
Human       KLVTDLTKVH TECCHGDLLE CADDRADLAK YICENQDSIS SKLKECCEKP
Porcine     KIVTDLAKVH KECCHGDLLE CADDRADLAK YICENQDTIS TKLKECCDKP
Bovine      KLVTDLTKVH KECCHGDLLE CADDRADLAK YICDNQDTIS SKLKECCDKP
Rat         KLATDVTKIN KECCHGDLLE CADDRAELAK YMCENQATIS SKLQACCDKP

            283             299
Human       LLEKSHCIAE VENDEMP
Porcine     LLEKSHCIAE AKRDELP
Bovine      LLEKSHCIAE VEKDAIP
Rat         VLQKSQCLAE TEHDNIP
```

Faji eltérések

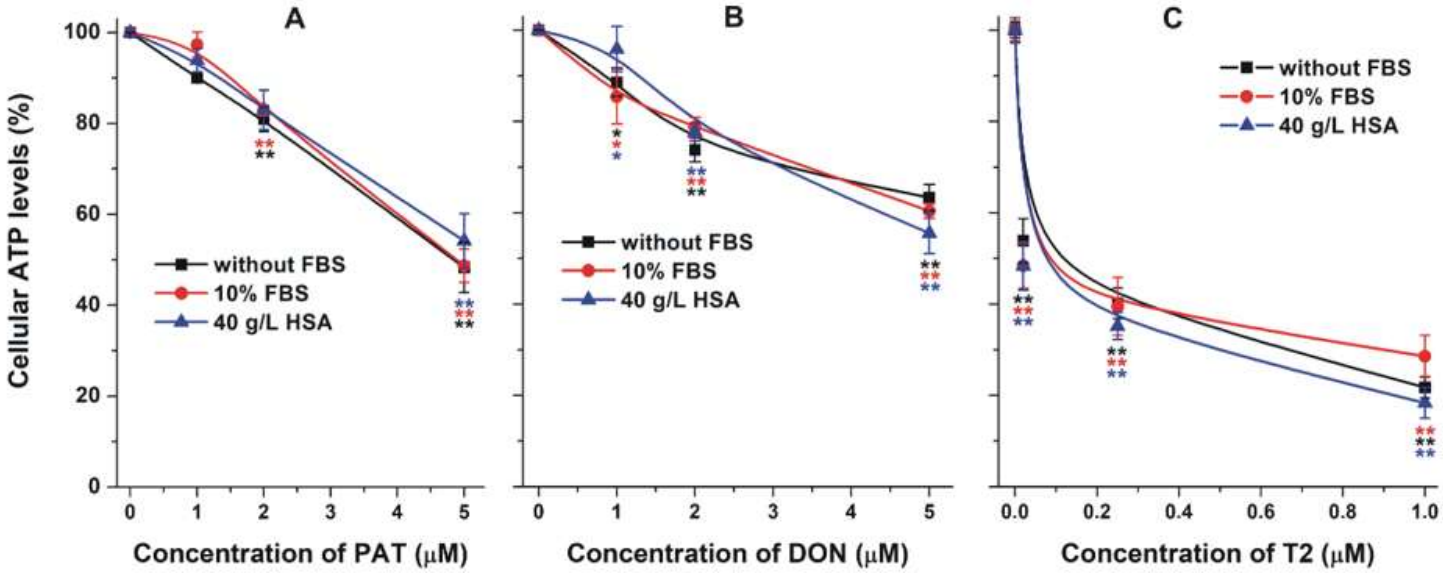
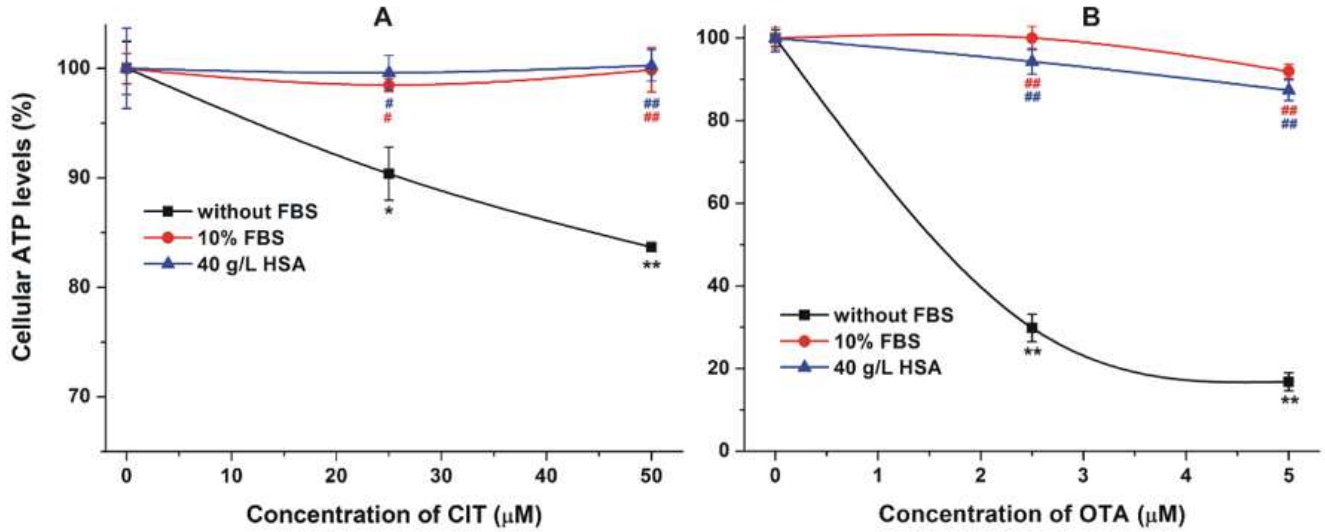
OTA: HSA > BSA (~10x)
> RSA (~30x)

Poór és mtsai.,
J. Lumin. 145 (**2014**) 767.

AOH: RSA > HSA (~8x)

Fliszár-Nyúl és mtsai.,
Int. J. Mol. Sci. 20 (**2019**) 2352.

	log*K*
ZEN-HSA	5.09
ZEN-BSA	4.78
ZEN-PSA	4.57
ZEN-RSA	5.42
α-ZEL-HSA	4.72
α-ZEL-BSA	4.46
α-ZEL-PSA	4.49
α-ZEL-RSA	5.43
β-ZEL-HSA	4.33
β-ZEL-BSA	4.37
β-ZEL-PSA	4.05
β-ZEL-RSA	5.43

Faisal és mtsai.,
Mycotoxin Res.
34 (**2018**) 269.

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