Article

Zearalenone-14-Glucoside Is Hydrolyzed to Zearalenone by β-Glucosidase in Extracellular Matrix to Exert Intracellular Toxicity in KGN cells

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1. Materials and Methods

1.1. Accuracy and precision

Both accuracy and precision were assessed using spiked samples. The accuracy of the proposed method was evaluated by the recoveries ($n = 6$), which were calculated using the following equation: % Recovery= (Area spiked sample – Area unspiked sample)/ Area standard×100%. Intra-day ($n = 6$) and inter day precision ($n = 6$) for each mycotoxins were assessed on one day and on three continuous days and were evaluated based on relative standard deviation (RSD) values.

2. Results

Table S1. Optimized MS/MS Parameter of ZEN, Z14G, α-ZEL and β-ZEL.

Mycotoxin	Ionization Mode	Q1	Species	Q2	Rt	DP	CE	CXP
ZEN	ESI⁻	317.0	[M−H]⁻	175.1* 131.1	5.1	−100	−42 −40	−7 −25
Z14G	ESI⁺	479.2	[M−H]⁺	283.4* 319.2	2.7	110	20 20	15 18
α-ZEL	ESI⁻	319.3	[M−H]⁻	159.9* 174.3	4.4	−140	−40 −34	−13 −8
β-ZEL	ESI⁻	319.2	[M−H]⁻	275.3* 301.3	3.9	−95	−34 −32	−9 −10

Q1:Precursor ion; Q2:Product ions; Rt: Retention time; DP: Declustering potential; CE: Collision energy; CXP: cell exit potential; *, quantitative ion.

Table S2. Accuracy and Precision of ZEN,Z14G, α-ZEL and β-ZEL.

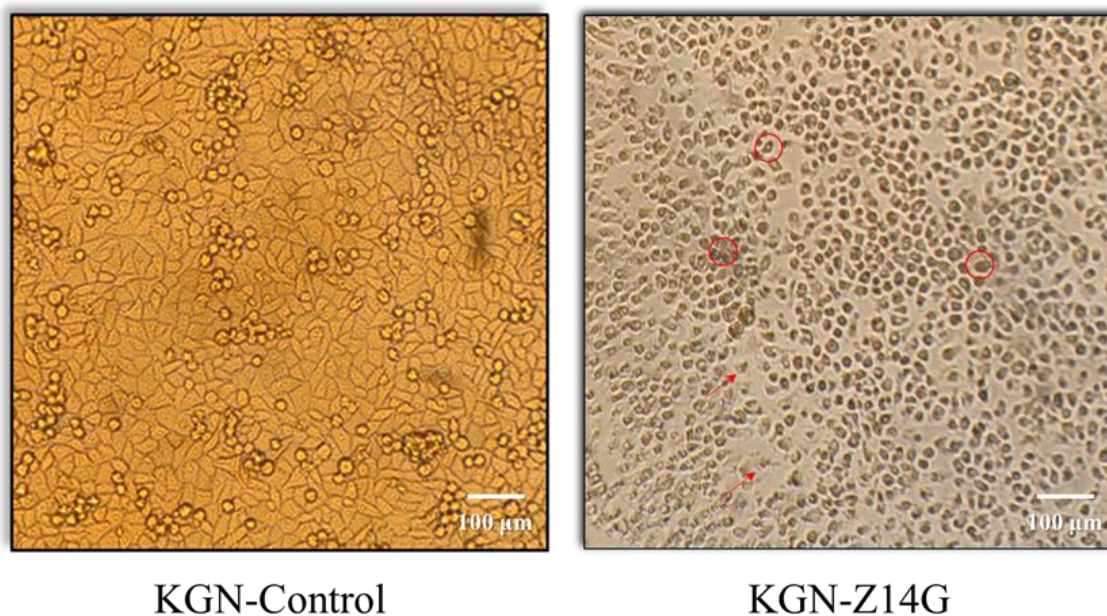| Mycotoxins	Intra-day Precision (RSD%, $n = 6$)	Inter-day Precision (RSD%, $n = 6$)	Mean Recovery(%) ($n = 6$)	
			Intracellular	Extracellular
ZEN	1.29	0.96	102.13	91.41
Z14G	1.35	0.55	100.81	100.73
α-ZEL	0.86	1.11	88.03	92.59
β-ZEL	0.91	0.98	109.83	85.58

KGN-Control

KGN-Z14G

Figure S1. Cell Morphology of KGN after Administration of Z14G (420 μM, 24 h) (100×), red circles represent cell shrinkage and red arrows represent cell fragmentation.