Meeting the challenges of Dioxin analysis and more with GC-Orbitrap high mass resolution capabilities

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Abstract

Purpose: Evaluate the performance of gas chromatography (GC) Orbitrap high mass resolution spectrometry for trace analysis of polychlorinated dibenzo-p-dioxins/furans (PCDD/PCDF) at current maximum allowable limits in soil according to EPA 1613 method criteria[1]

Method summary: Sample analysis was carried out on the Thermo Scientific™ Orbitrap Exploris™ (OE) GC 240 mass spectrometer. A sample volume of 1.5 µL was injected for chromatographic separation using a TG-Dioxin (60 m × 0.25 mm, 0.25 µm) capillary GC column with a Thermo Scientific™ TRACE ™ 1610 GC. Ion source conditions were optimized to fulfill sensitivity criteria of EPA 1613. Performance was evaluated through analysis of a raw soil extract previously analyzed using a regulatory compliant method[2]. Quantification was performed using isotopic dilution in Thermo Scientific™ Chromeleon™ Chromatography Data System (CDS) software using the Thermo Scientific™ Dioxin Analyzer workflow .

Results: The Orbitrap GC mass spectrometer fulfilled the high sensitivity, mass accuracy and resolution criteria of EPA 1613 at femtogram (fg) levels within a raw soil extract. Combined with a large dynamic range (0.05 – 100 pg•µL^{-1}), the OE GC provides users with an alternative analytical approach to deliver sensitive and accurate analysis of PCDD/F while enabling laboratories to meet future analytical challenges with high resolution accurate mass data.

Introduction

Toxicity and exposure to PCDD/Fs have been a global issue for decades. Although globally restricted, accidental exposure can still occur due to their environmental persistence. Recent revisions to maximum allowable limits of PCDD/Fs in food (0.02 – 3.5 pg•g^{-1}) in Europe[3] place greater emphasis sensitivity and selectivity. Gas chromatography magnetic sector mass spectrometry has been the gold standard in the analysis of PCDD/F providing the required mass resolution (i.e., >10,000 resolving power at 10% valley) and accuracy (i.e., 5 pm) for global compliance, such as that laid out the U.S. Environmental Protection Agency (EPA) method 1613.[1] However, greater demands are being placed on analytical instrumentation where higher mass resolution and system flexibility is needed to increase scope into unknown chemical exposure analysis. Recent advances in mass spectrometry technology can provide greater mass resolution with sub ppm mass accuracy, providing alternative tools for PCDD/F analysis that still fulfill strict compliance criteria in accredited methodology.

Materials and methods

Standard preparation

Isotopically labelled calibration standards (EPA-1613) were purchased from Wellington Laboratories (Canada). Each standard was diluted by a factor of 2 to construct a 6-level calibration curve (Figure 1.).

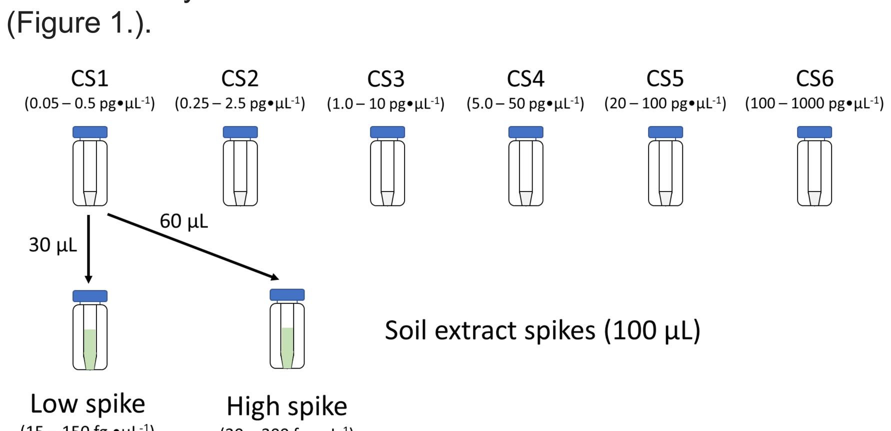


Figure 1. Overview of standard and sample preparation

Sample preparation

A two-gram soil sample was extracted in a bi-phasic mixture of acetonitrile/ hexane (4 ml:4 mL) followed by centrifugation at 3000 rpm. 100 µL of the hexane layer was evaporated to dryness and spiked with 15 µL of the lowest calibration standard and reconstituted in 100 µL nonane (Figure 1).

Instrumental parameters

Table 1. GC injection, oven temperature program and mass spectrometer parameters

Trace 1610 GC system parameters	
Injector	iConnect™SSL
Injection volume (µL)	1.5
Liner	Thermo Scientific™ Straight liner with quartz wool
Injection mode	Splitless (split flow 120 mL·min^{-1} after 2 min)
Split flow (mL·min^{-1})	120
Injector temperature (°C)	280
Carrier gas, (mL·min^{-1})	He, 1.2 (constant flow)
Oven temperature Program	
Initial temperature (°C)	120
Hold time (min)	2
Rate 1 (°C·min^{-1})	25
Temperature 1 (°C)	250
Hold time 1 (min)	0
Rate 2 (°C·min^{-1})	2.5
Temperature 2 (°C)	285
Hold time 2 (min)	0
Rate 3 (°C·min^{-1})	10
Temperature 3 (°C)	320
Hold time 3 (min)	15
Total run time (min)	44.7
Orbitrap Exploris GC-MS parameters	
Transfer line (°C)	300
Extractabrite™ ion source temperature (°C)	350
Electron energy (eV)	40
Acquisition mode	tSIM
Isolation window width (m/z)	10
Resolving power (at 200 m/z)	60,000
Emission current (µA)	50
C-Trap offset (V)	-5
Mass accuracy on lock mass	5ppm
Internal lock mass calibration (column bleed, m/z)	207.0324, 281.0511, 355.0699

2, 3, 7, 8-TCDD

One software for analysis, quantification and reporting

The strict performance criteria listed in regulatory methods for PCDD/Fs are integrated into the Dioxin Analyzer workflow within Chromeleon CDS 7.3.2 software. Analysis performance (i.e., ion ratio and relative response factor) are displayed as interactive graphics for easy evaluation (Figure 1.) Quantification using isotopic dilution together with custom Dioxin reporting templates are directly implemented within the Chromeleon CDS software, eliminating the need to export data for processing or report construction.

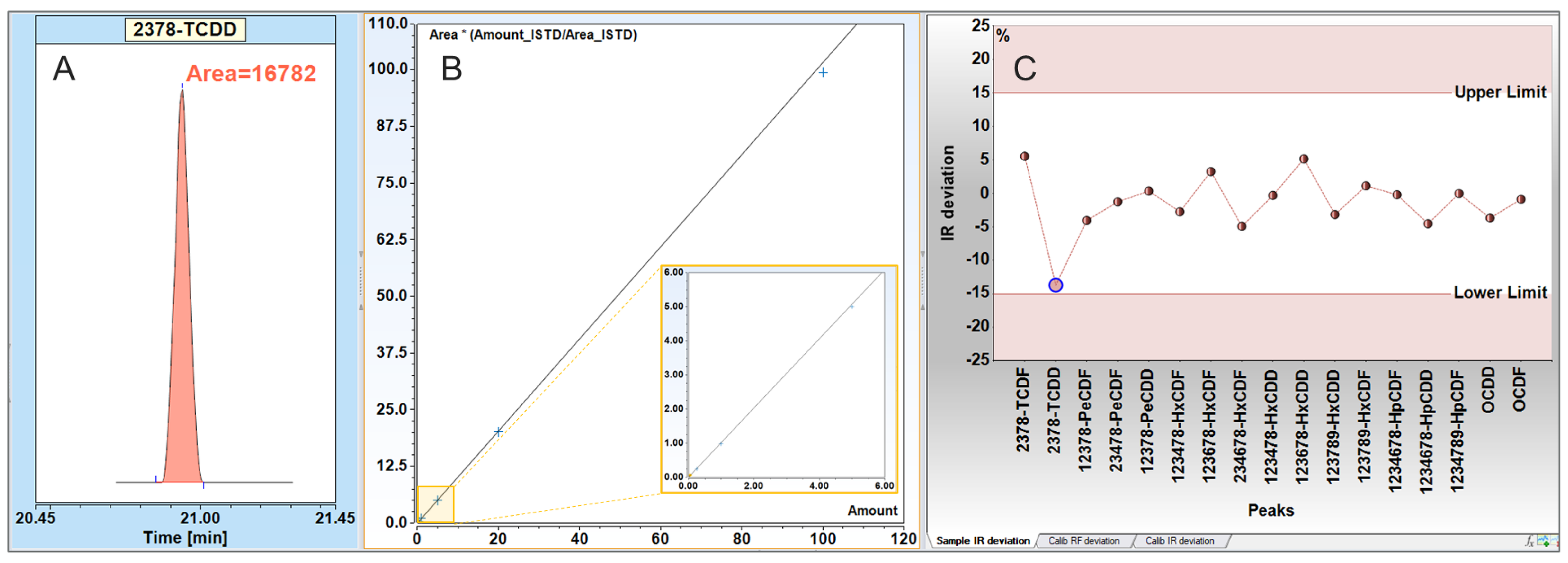


Figure 3. (A) Extracted mass of 2,3,7,8- tetrachlorodibenzodioxin (TCDD) (m/z 321.8930) at 0.05 pg•µL^{-1}; (B) calibration curve (0.05 – 100 pg•µL^{-1}; and (C) ion ratio deviation for all PCDD/F congeners at 0.05 pg•µL^{-1} calibration level

Source optimization for ultimate sensitivity

Criterion in EPA method 1613 list electron energy (eV) between 28 – 40 eV to avoid significant fragmentation of molecular or targeted ion of interest to obtain sensitivity required for dioxin analysis. The dioxin method template provides a default setting of 35 eV, although this parameter should be optimized/confirmed. The tune application built into the Chromeleon 7.3.2 CDS software allows the ion source optics and mass calibration at a user defined eV within minutes to allow for rapid optimization. Results of eV optimization for 5 pg•µL^{-1} standard of 2, 3, 7, 8 - TCDD can be seen in Figure 2a. where 40 eV was found to give the optimal response for the detection of dioxins in a spike soil extract with no clean-up (Figure 2b).

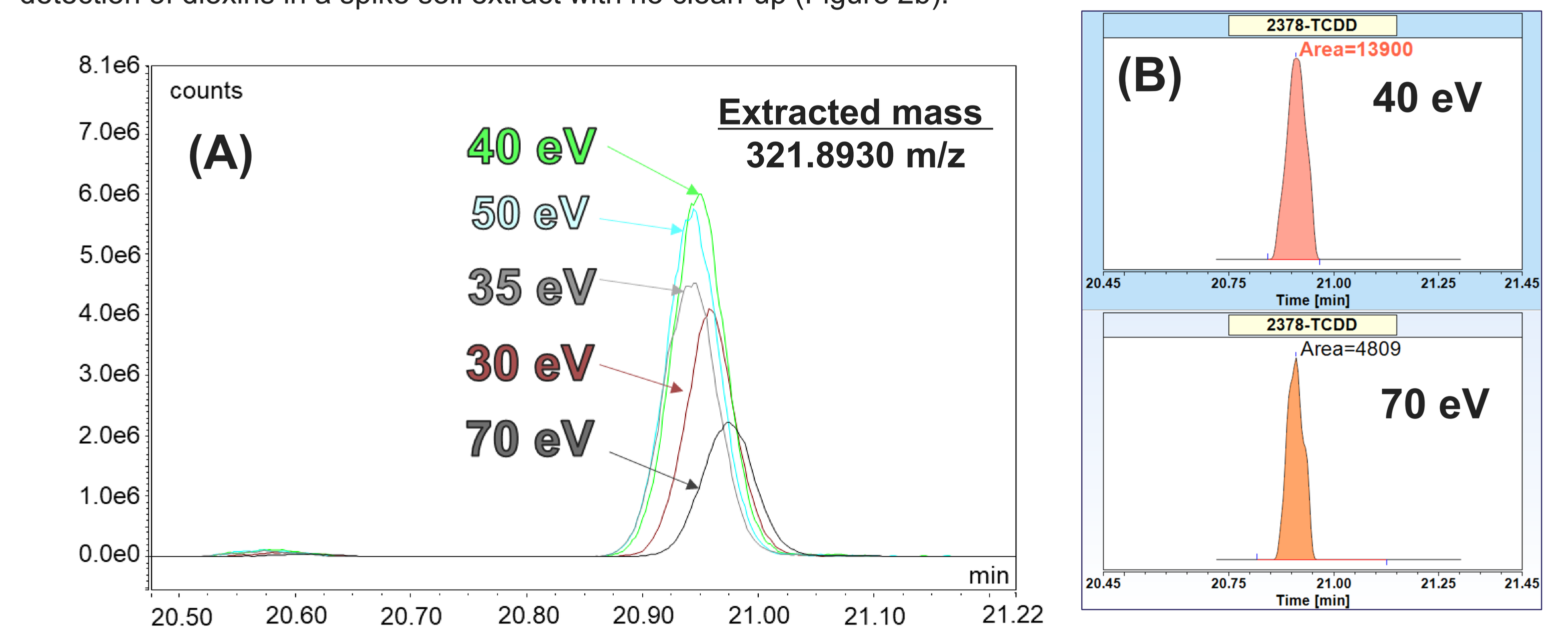


Figure 2. (a) Signal response of 5 pg•µL^{-1} 2 ,3, 7, 8- TCCD calibration standard at electron energies from 30 – 70 eV; (b) response (counts• sec) of 2,3,7,8-TCCD in spiked soil extract (45 fg on column at 40 and 70 eV)

Mass resolution for regulatory compliance

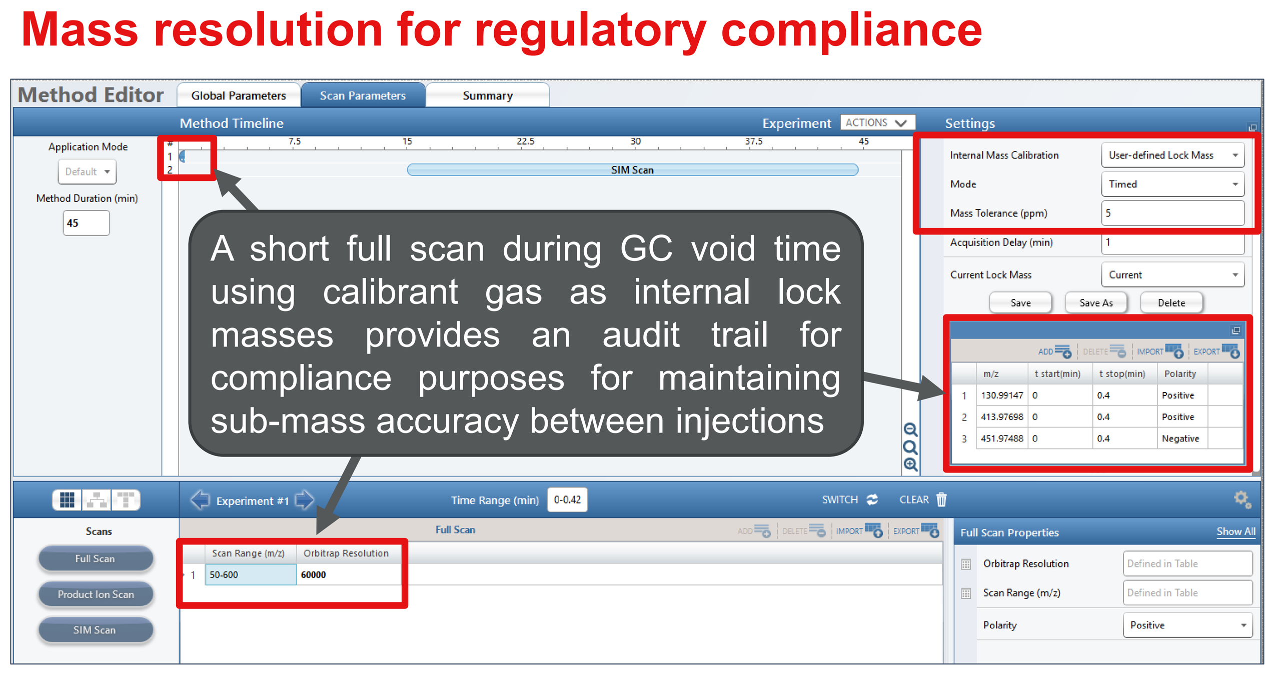


Figure 3. Full scan together with tSIM acquisition setup in method editor withing Chromeleon CDS software for full compliance for dioxin analysis

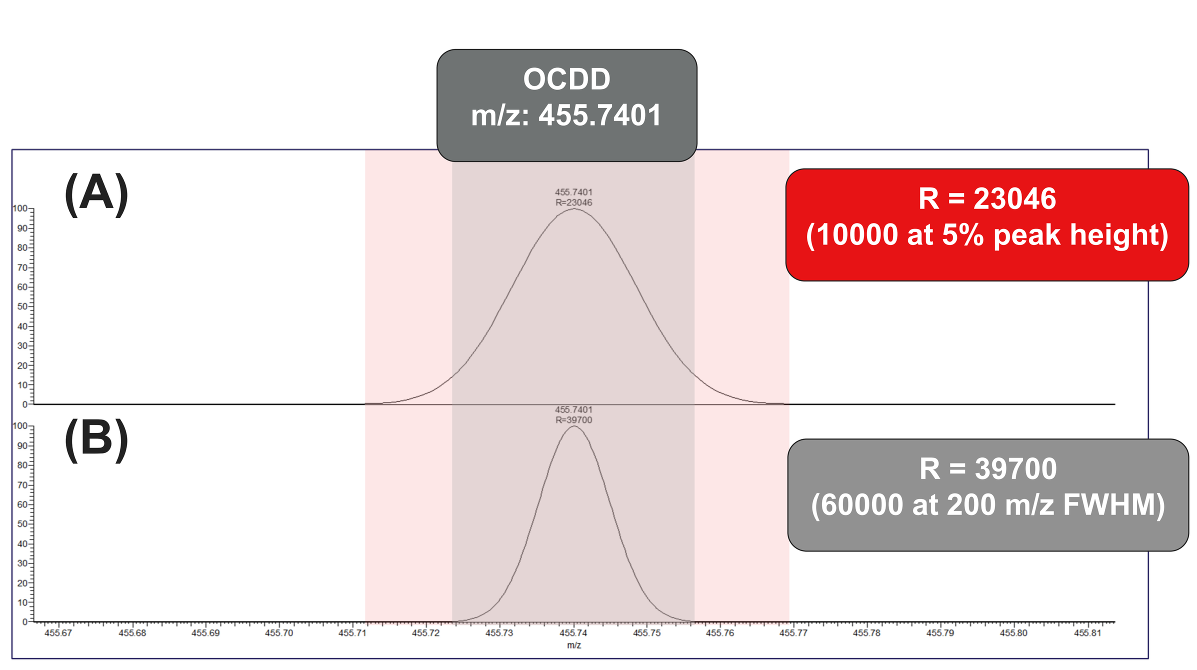
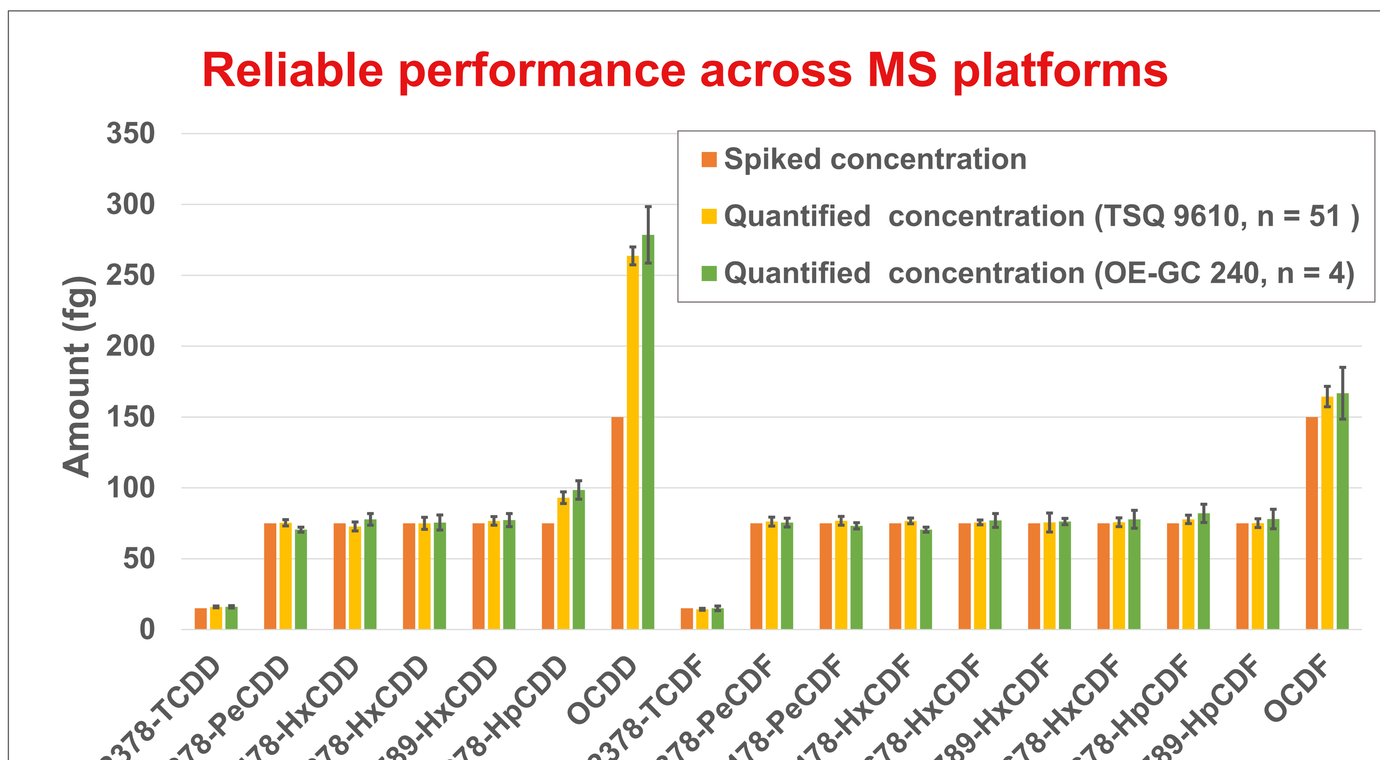


Figure 4. Simulated mass resolution (R) of octachlorodibenzodioxin (OCDD) at (a) 10,000 at 5% peak height and (b) 60,000 at m/z 200 full width half maximum (FWHM)

A resolving power of ≥ 10,000 at 10% valley (or 5 % peak height) is required by EPA method 1613 is required for all PCDD/F target masses. Fulfillment of this criteria will be most critical at the high mass range. OCDD represents the heaviest target analyte with an exact mass of 455.7401. Simulation of the mass resolution (R) needed to fulfill criteria of EPA 1613 for OCDD is approximately 23,000 (Figure 4a). Analysis with the Orbitrap Exploris GC mass spectrometer at 60,000 resolution (at 200 m/z FWHM) provides a mass resolution of approximately 40,000 (Figure 4b) confirming full compliance with EPA method 1613.

Reliable performance across MS platforms



Results from low spike soil extract by the OE-GC MS were in excellent agreement to prior analysis on the Thermo Scientific™ TSQ™ 9610 GC-MS/MS system[3], showing reliable performance of the OE-GC MS for trace analysis of PCDD/Fs using high resolution accurate mass

Conclusions

- Femtogram sensitivity achieved for PCCD/F analysis with software-optimized optics at user-defined eV energies
- Mass resolution performance surpassing EPA 1613 requirements at 60,000 mass resolution settings (at 200 m/z FWHM)
- Accurate and precise performance at fg levels in environmental samples
- Ability to increase scope of contaminant analysis through full scan high resolution accurate mass data, providing a flexible system for laboratories to expand their analytical services to meet customer needs

References

1. Environmental Protection Agency (EPA) Method 1613. Tetra-Through Octa-Chlorinated dioxins and Furans by Isotope Dilution HRGC/HRMS. United States. Revision B, 1994. EPA 821-B-94-005.
2. Warner, N. A.; Ladak, A.; Cavagnino, D.; Kutscher, D. Thermo Scientific Technical Note 001594: Uncompromised sensitivity in polychlorinated dibenzo-p-dioxins/furan analysis using triple quadrupole GC-MS with cost-effective helium-saving technology (2022).
3. European Union Commission Regulation 2022/2002. Amending Regulation (EC) No 1881/2006 as regards maximum levels of dioxins and dioxin-like PCBs in certain foodstuffs. October 21, 2022.

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