



# NAVIGATING THE COMPLEXITIES OF BIODEGRADATION TESTING FOR UVCBS: CHALLENGES, APPROACHES AND CASE-STUDY INSIGHTS Scan me

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# INTRODUCTION

Substances of Unknown or Variable Composition, Complex Reaction Products, or Biological Materials (UVCBs) represent a significant challenge in environmental risk assessment due to their complex and variable compositions. Unlike single chemicals, traditional testing methods may not adequately capture their biodegradation behavior. We present strategies for assessing UVCB biodegradation and highlight challenges in OECD 309 variability and reliability using a specific UVCB case study.

## TESTING STRATEGIES FOR UVCBS



## CONCLUSION

• Significant inter- and intra-study variability raises concerns about the robustness of the OECD 309 test.

- Balancing microbial biomass and test concentration is critical; further guidance is needed.
- Effective UVCB biodegradation assessment requires careful planning and a well-justified testing strategy: -Constituents identified & quantified  $\rightarrow$  Known constituent approach -Constituents too numerous or complex for full ID  $\rightarrow$  Block profiling approach -Constituents and blocks cannot be reliably resolved  $\rightarrow$  Whole substance approach

## CASE STUDY: KNOWN CONSTITUENT APPROACH



SAR	MODELIN	GOF	UVC	3 CON	NPON	NENT	S –	
							Se co	lected mponent
	Component	1	2	3	4	5	BIOWIN2: ) BIOWIN3: ) BIOWIN6: )	
	weight (%)	26	10	2	43	11	$> 0.5 \rightarrow 1$ ultimate 1 $> 0.5 \rightarrow 1$	
	Mol. Mass (g/mol)	299	342	386	491	682	biodegrades biodegradatio readily biode	
	BIOWIN 2	0.99	0.84	0.09	1.00	1	fast; < 0. on timefr gradable	
	BIOWIN 3	3.13	3.03	2.92	2.99	2.84	5 → doe ame ≥ m ;; < 0.5 →	
	<b>BIOWIN 6</b>	0.92	0.87	0.81	0.95	0.97	s not bio onths (< ≯ not rea	
	Log BCFmax	0.50	0.50	0.5	0.18	1.06	degrade fast 2.25 – 2.75) fily biodegra	
	Logkow	-0.15	-0.43	-0.70	1.17	2.49	adable	

#### Identify Constituents

Determine the specific chemical compounds present

Screen Constituents Use existing data and models to filter constituents

Select Worst-Case Conduct Biodegradation Choose constituents with high persistence Tests indicators

Perform tests to

assess

biodegradability

#### **UVCB** Test substance

- Organic compound
- 5 main constituents
- Ether and alcohol groups
- 1 3 benzene rings in all constituent

- All 5 constituents are predicted to be **readily biodegradable**;
- All 5 constituents are **not bioaccumulative** based on Logkow < 3
- The major constituent (43%) was selected as representative due to structural similarity among components.
- Its higher concentration and lower molecular weight makes it a plausible worst-case for bioaccumulation and environmental release.

## BIODEGRADATION TESTING (OECD 309) OF WORST-CASE COMPONENT

### Biodegradation of the main constituent measured at 10 µg/L and 100µg/L

Test 1	% of A Incuba substa	pplied R tion tim nce	adioad e (days	% of Appl Incubatio substance				
Treatment (µg/L)	0	14	28	41	60	Treatment (µg/L)	0	14
10-A	101	82.5	88.0	73.9	57.1	10-A	99	93
10-B	103	82.2	87.7	74.7	55.4	10-B	95	96
100-A	99.1	82.4	81.1	21.7	0.7	100-A	-	-
100-B	99.6	85.3	76.8	22.9	0.4	100-B	-	-

ied Radioactivity (AR) by • The test-item **did not degrade** significantly in time (days) for parent the first 28 days, indicating a long lag-phase

> • Significant higher degradation observed at 100 ug/L (Test 1 and 2)

 Significant inter-replicate variability observed in replicates of 100 ug/L (Test 2)

 Differences in biodegradation may stem from microbial community development and concentration-dependent adaptation.

# REFERENCES

• ECHA 2023. Guidance on Information Requirements and Chemical Safety Assessment. Chapter R.11: PBT/vPvB Assessment.

• Seller et al. 2020. Biotransformation of chemicals in water-sediment suspensions: influencing factors and implications for persistence assessment. Environmental Science & Technology Letters 7.11: 854-860. • Birch et al. 2023. Technical guidance on biodegradation testing of difficult substances and mixtures in surface water. MethodsX 10: 102138.

60

78

75

1.7

68

40

66

75

1.6

74

28

80

80

75

51

70

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90

62

70