Supplementary Material

Toward the future of OECD/ISO biodegradability testing - new approaches and developments

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Table S1. Data about existing chemicals (modified after Wang et al., 2020)

Number of chemicals and mixtures of chemicals registered for production and use: - claimed confidential - ambiguously described	 > 350,000 > 50,000 up to 70,000
Total chemicals with CAS numbers - individual compounds - organics - mixtures, polymers, UVCBs	235,323 157,896 137,325 75,384

Table S2. Microbial toxicity tests

Test	Method	Test principle
Inhibition of heterotrophic respiration	OECD 209 ISO 8192	measurement of oxygen consumption due to heterotrophic respiration
Inhibition of nitrification inhibition	OECD 209 ISO 9509	measurement of oxygen consumption due to nitrification respiration (OECD 209) or ammonium oxidation, nitrite and nitrate formation (ISO 9509)
Luminescent bacteria test	ISO 11348 part 1 to 3	measurement of inhibition of light emission
Pseudomonas putida growth inhibition test	ISO 10712	measurement of growth inhibition of a growing pure culture of *Pseudomonas putida*
Growth inhibition test with activated sludge bacteria	ISO 15522	measurement of growth inhibition of activated sludge bacteria
Determination of the inhibition of the activity of anaerobic bacteria	OECD 224 ISO 13641	measurement of inhibition of biogas production

Table S3. Bin system of reference compounds (modified after Comber and Holt, 2007)

There are four bins for classification:
Bin 1: normally passes a RBT or modified RBT
Bin 2: normally passes an enhanced RBT, but currently fails any other RBT
Bin 3: normally fails any RBT
Bin 4: should never pass an enhanced or modified RBT

Classification	Compounds
Bin 1	Aniline, sodium benzoate, 1-octanol, anthraquinone, phenol
Bin 2	Diethylene glycole, 4-chloroaniline, 1,3,5-trimethylbenzene, Dinitrotoluene, 4-Fluorophenol
Bin 3	Di-isotridecyladipate, Cyclododecane, Cyclododeca-1,5,9- triene , Cyclododecane , Dibutylphenol
Bin 4	Musk xylene , Hexachlorobenzene, Benzo(a)pyrene, Hexachlorohexane

Table S4. Biodegradation of some commonly used reference compounds recommended by the OECD

Compound	Test system	Extent of degradation (%)	References
Benzoic acid, sodium benzoate	OECD 301 F	91 - 93	Reuschenbach et al., 2003; Gu et al., 2020
Aniline	OECD 301 A - F	75 - 98	Reuschenbach et al., 2003, Strotmann, et al., (2004), Gu et al., 2020
	MCTS*		Strotmann et al., 2004
	- DOC elimination	96	
	- oxygen consumption	82.5	
	- CO_2 production	83	
	CTS**		Strotmann et al., 1995
	- DOC elimination	> 95	
	- CO_2 production	78	
	OECD 302 B	98	Reuschenbach et al., 2003
Diethylene glycole	OECD 301 F	59 - 93	Reuschenbach et al, 2003; Gartiser et al., 2022
	CTS**		Strotmann et al., 1995, Gartiser et al., 2022
	- DOC elimination	> 95 - 98	
	- CO_2 production	90 - 99	
	OECD 302 B	70	Comber et al., (2010); Lapertot et al., 2006
Sodium acetate	OECD 301 F	> 95	Gu et al., 2020
Sodium benzoate	OECD 301 F	> 95	Gu et al., 2020
1-octanol***	OECD 310	85	Comber et al., 2010; Battersby et al., 1997

* multi component test system based on OECD 301 tests; ** combined test system based on OECD 301 A/B tests; *** water insoluble compound