COMMENTS ON THE PROPOSAL OF THE NEW ☒or REVISED☐ AOP

Title: AOP 263: Uncoupling of oxidative phosphorylation leading to growth inhibition via decreased cell proliferation

Comments submitted by (please fill in below -Name, Country, Organisation)

Dr. Michael Oelgeschläger
German National Coordinator Test Guideline Programme (Health)
German Federal Institute for Risk Assessment (BfR)
Max-Dohrn-Str. 8-10
D-10589 Berlin, Germany
Michael.Oelgeschlaeger@bfr.bund.de

National Experts or National Co-ordinators please complete the following:

Name of Expert:	Email:	Mailing Address,Tel/fax	Date comments were received
Tanja Burgdorf	Tanja.Burgdorf@bfr.bund.de	German Federal Institute for Risk Assessment (BfR) Department Experimental Toxicology and ZEBET Max-Dohrn-Straße 8-10 D-10589 Berlin Germany	
Judy Choi	Judy.Choi@bfr.bund.de	German Federal Institute for Risk Assessment (BfR) Department Pesticides Safety Max-Dohrn-Straße 8-10 D-10589 Berlin Germany	

Please include "GC" for General Comments, the paragraph number or line number. Please include only numbers in the left two columns.

Expert affiliation/ Country	Page #	Line #	Expert Comments	response
DE/BfR	GC		This AOP is very well-written, and we support the endorsement of this AOP for declassification and publication.	We thank German exerts for the good words.
DE/BfR	11 Event 1821		The effect of NEN and oxyclozanide as uncouplers on colon cancer metastasis in mice was investigated by Alasadi (2018) (https://www.nature.com/articles/s41419-017-0092-6). NEN or oxyclozanide either completely prevented or drastically reduced hepatic metastasis of colon cancer cells from spleen which could be mentioned here as well.	We have added these as indirect evidence to support Event 1821 and Alasadi et al., 2018 to the reference list on page 15. NEN and oxycloznide have also been added to the stressor list on page 8.
DE/BfR	15		The first few lines under Considerations for Potential Applications of the AOP (starting from "First, the AOP…" and ending in "…various invertebrates") describe growth as a recognised endpoint only for organisms in the environment. Growth is also relevant in many OECD test guidelines for mammalian toxicity (health effects).	We have added "mammals" and the corresponding

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			Since the authors mentioned the usefulness of the key events for toxicological studies, it would be good to also include mammals in this section. Please refer to our comment below for page 32 for examples of OECD test guidelines in mammals.	TG numbers to these sentences.
DE/BfR	32		As on page 15, the human health related TGs should be mentioned here as well. Currently, the list is very focused on the ecotoxicology assessment. Some examples for mammalian toxicity studies that could be added to the list: - Test No. 407: Repeated Dose 28-day Oral Toxicity Study in Rodents - Test No. 408: Repeated Dose 90-Day Oral Toxicity Study in Rodents - Test No. 416: Two-Generation Reproduction Toxicity - Test No. 422: Combined Repeated Dose Toxicity Study with the Reproduction/Developmental Toxicity Screening Test - Test No. 443: Extended One-Generation Reproductive Toxicity Study - Test No. 453: Combined Chronic Toxicity/Carcinogenicity Studies	We have added the mammalian TGs to the list.
DE/BfR	35		*whereas the same magnitude of effect required 1.6 µM* whereas the same magnitude of effect **for ATP reduction** required 1.6 µM	Revised accordingly.
DE/BfR	35		significant reduction in **ATP** required 20 µM FCCP in human RD cells	Revised accordingly.

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DE/BfR	40		thatare necessary -> that are necessary	Revised accordingly.
DE/BfR	40		led to 35% ATP depletion and 35% reduction check format	Format revised to make the sentence consecutive.
DE/BfR	43		significant growth inhibition please specify if this is related to growth of the zebrafish in general or if there were also effects on tissue and organs observed.	The sentence has been revised to provide more details about the growth inhibition effects.