

Monitoring and assessing welfare: Score sheets for *Nothobranchius furzeri*

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Introduction

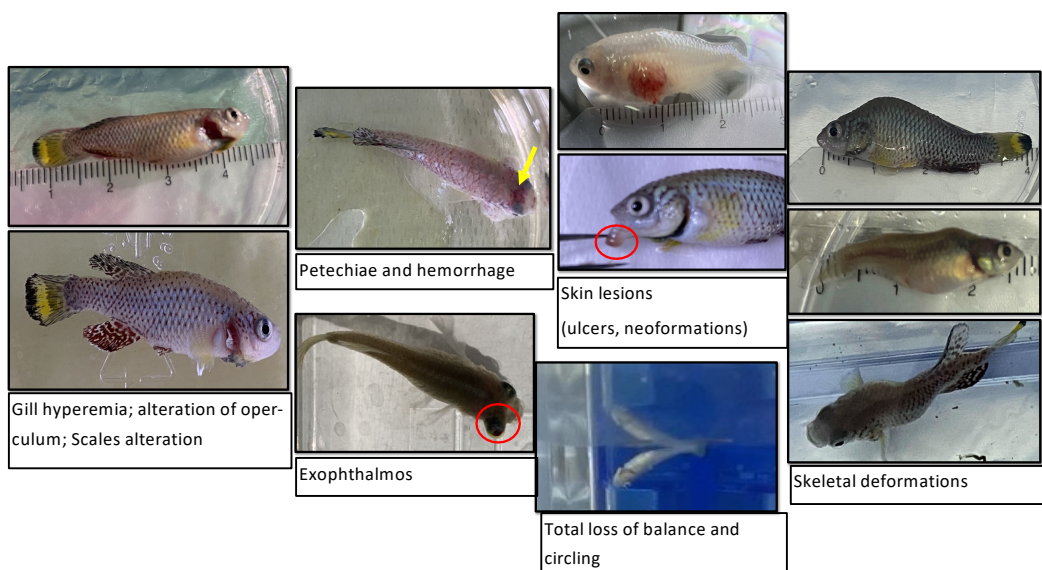
Animal based research needs to strictly adhere to the 3R principle (replace, refine, reduce) for the ethical justification of the use of animals in science as well as to ensure the highest quality of reproducible data.

The goal of our work was to determine a well-designed score sheet for the recently well-established animal model, the African turquoise killifish *Nothobranchius furzeri*, the shortest-lived vertebrate that can be bred in captivity[1].

Methods and discussions

We have identified four parameters that will serve to detect deviations from the normal state: behavior, swim, body condition and clinical signs and defined a numerical scoring from 0 to 3 in order to weigh each symptom. From symptoms sum up we established the interventions that are applied in the event of animal suffering, including humane endpoints [2].

Total score can vary from 0, no action, up to values higher than 8, that represent our humane endpoint implying the euthanasia of the fish. This simple scoring tool can be employed for long-term monitoring of individual animal welfare and integrated into routine management practices.



"ID" Date of birth	Symptom description	Date/Score
	Behavior 1 Normal 2 Aggression manifested towards the other sex 3 Social isolation 4 Food disinterest 2-Lethargy 5 Gasping	
	Swim 1 Normal 2 Partial or intermittent loss of balance (Vertical swim) 2-Circling 3 Surface swim/deep swim (belly slider) 4 Total loss of balance	
	Body Condition 1 Normal 2 Loss of 10% to 15% of weight 3 Loss of 15% to 20% of weight 4 Emaciation with loss >20 % of weight (head larger than the body, concave abdomen) 3-Obesity (head smaller than the body, convex abdomen)	
	Clinical signs 0- Normal 1- Color alteration 1 Gill hyperemia/alteration of operculum 1-Scales alteration 2 Hyperventilation 2-Cutaneous mucus hypersecretion 3-Petechiae and/or hemorrhage 3-Skeletal deformations 3-Exophthalmos 3-Skin lesions (ulcers, neoformations)	
	Total Scores	Action
0 Normal		No Action
1-4 Moderate alterations		Daily monitoring
5-8 Severe alterations		Monitoring twice a day
>8		Euthanasia

Conclusion

Future step will be to enrich this tool with age-related enhanced monitoring to define (a) the expected onset of one or more age-related adverse clinical signs (depending on the genetic background), (b) the age-related clinical signs that can cover possible pathological conditions, (c) the application of humane end points will help to reduce the numbers of animals maintained for longer than is scientifically justified.

Key References

- 1 Genade T, Benedetti M, Terzibasi E, et al. Annual fishes of the genus *Nothobranchius* as a model system for aging research. *Aging Cell* 2005; 4: 223-233.
- 2 Sneddon, L. U. Pain perception in fish: indicators and endpoints. *ILAR journal*, 2009; 50(4), 338-342.

Funding

This research was funded by the grant assigned of the University of Camerino, FAR 2019, Rector's decree 137/2020 and PRIN 2022LYPBCT *Notho-Diet: from chemosensory perception to husbandry standardization*