

Overview of technologies for welfare indicators monitoring available at the TNA infrastructures

+

Existing and emerging technologies available for welfare indicators monitoring

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This document is an overview of the technologies for welfare indicators monitoring available for TNA users at selected TNA infrasctructres. The TNA applicants who are interested in the welfare monitoring can use this document to orient in the available technologies to support the planning of the TNA project. The list bellow provides basic information about available technologies and the description of use cases. To get more information about the technology and its availability for TNA access contact the contact person.

The technologies described bellow the list is the overiview of the technologies usable for welfare indicators monitoring.

Institute	Wageningen University	
Contact person		
	name	Roel Maas
	email	Roel. Maas@wur.nl
Technology name		era's and Noldus software (we have more camera's I 16 at the same time)
Short technology description or link	Video camara'	S
Welfare indicators	Swimming act	ivity/speed, behaviour
Fish species	Seabass, rainb	ow trout, tilapia,
Type of water	Salt and fresh	water (cold and warm)
Cultivation unit	Metabolic Research Unit, Circular tanks replicated RAS	
Advantages of the technology	Provides info on behaviour and in the MRU allows you to link behaviour with the energy expenditure (see link 1).	
Disadvantages of the technology	Labour to analyse video recordings	
Use case		
	use case 1	Effects of dissolved carbon dioxide on energy metabolism and stress responses in European seabass (<i>Dicentrarchus labrax</i>)
	use case 2	
	use case 3	
Links to existing studies		
	link 1	https://onlinelibrary.wiley.com/doi/10.1111/j.1365- 2109.2012.03142.x
	link 2	



	link 3	
Is the technology available for TNA users?	yes	
	restrictions	Max. 16 camera's can be recorded
	conditions	Metabolic chambers,

Institute	Wageningen Reserach	
Contact person		
	name	Wout Abbink
	email	wout.abbink@wur.nl
Technology name	acceltags	
Short technology description or link	accelerometer swimming beh	sensors that can be placed in the fish to monitor avior
Welfare indicators	activity, behav	iour
Fish species	multiple specie	es, as long as the fish have the right seize
Cultivation unit	RAS	
Advantages of the technology	monitoring of fish behaviour and swimming	
Disadvantages of the technology	fish needs surgery to insert the sensor	
Use case		
	use case 1	monitoring of fish behaviour
	use case 2	monitoring of fish swimming performance
	use case 3	
Links to existing studies		
	link 1	
	link 2	_
	link 3	



Is the technology		
available for TNA users?	yes	
		needs to be requested and discussed in advance,
	conditions	but it should be possible to arrange

Institute	UL	
Contact person		
·	name	Sylvain MILLA
	email	Sylvain.Milla@univ-lorraine.fr
Technology name	Fish videotrack	king and evaluation of behaviour and personnalities
Short technology		g of the behaviour (10.1093/cz/zow048 or
description or link		anim.2017.05.012 or 0.1007/s10499-019-00343-z)
		haviour (cross maze test), depth distribution,
		nming activity, distances between fish, number of
Welfare indicators	contacts, cannibalism, aggressivness	
Fish species	Eurasian perch, pikeperch, trout, tench, sturgeon, carps	
Type of water	fresh water	
Cultivation unit	tanks 0.5-3m2 or aquariums 100L	
Advantages of the		
technology	provides information about 2D position in the tank	
Disadvantages of the	need to be calibrated for new species (depth calibration, biomass	
technology	estimation calibration)	
Use case		
	use case 1	Understanding the cannibalistic activities
	use case 2	Observation of abnormal behaviour
	use case 3	Analyzing different fish personnalities
Links to existing studies		
	link 1	https://hal.archives-ouvertes.fr/hal-01611451



	link 2	https://cdnsciencepub.com/doi/abs/10.1139/cjfas- 2017-0037
	link 3	https://link.springer.com/article/10.1007/s10499- 019-00343-z
Is the technology available for TNA users?	yes	
	restrictions	Only 3 recording systems are available
	conditions	

	_	University of Agriculture and Life Sciences, Research Centre of	
Institute	Fisheries an	d Aquaculture	
Contact			
person			
	name	László Ardó	
	email	ardo.laszlo@uni-mate.hu	
Technology name	Recirculatio	n system and laboratory of fish immunology and physiology	
Short technology description or link	laboratory f	n system for rearing and keeping freshwater fish species, and a for measuring stress parameters from blood.	
Welfare indicators	Input indicators (except for lighting and noise), mortality, all individual external and blood indicators		
Fish species	common carp, rainbow trout		
Type of water	fresh water		
Cultivation unit	tanks 1-3 m	3	
Advantages of the technology		measurements of input OWIs and output group level, individual diblood OWIs	



Disadvantage s of the technology	Not suitable for monitoring behaviour, distribution, etc.		
Use case			
	use case 1	Measuring stress and welfare indicators in experimental conditions	
	use case 2		
	use case 3		
Links to existing studies			
	link 1	https://www.sciencedirect.com/science/article/pii/S2352513421002 350	
	link 2	https://link.springer.com/article/10.1007/s10499-018-0250-6	
	link 3		
Is the technology available for TNA users?	yes		
	restriction		
	S	technical assistance is provided by the institute	
	conditions		

Institute	Hellenic Centr	Hellenic Centre of Marine Research (HCMR)	
Contact person			
	name	Nikos Papandroulakis/ Dimitris Voskakis	
	email	npap@hcmr.gr/ d.voskakis@hcmr.gr	
Technology name	Stereo Camera	Stereo Cameras in Cages	
Short technology			
description or link	System for fisl	System for fish length estimation	
Welfare indicators	Biomass/Grov	Biomass/Growth	
Fish species	European seal	European seabass, Gilthead seabream, and potentially others	



Type of water	salt water		
Cultivation unit	Cages 4*4*6 m	Cages 4*4*6 m	
Advantages of the technology	Non invasive, f	ast processing	
Disadvantages of the technology	Needs adjustm	Needs adjustments for different species, Calibration equipment	
Use case			
	use case 1	Biomass estimation in European seabass	
	use case 2	Biomass estimation in Gilthead seabream	
	use case 3		
Links to existing studies			
	link 1		
	link 2		
	link 3		
Is the technology			
available for TNA users?	yes/no		
	restrictions		
	conditions		

Institute	Norwegian University of Science and Technology		
Contact			
person			
	name	Martin Føre	
	email	martin.fore@ntnu.no	
Technology			
name	Biosensors/	'Acoustic telemetry	
	Well established method for monitoring aquatic organisms where individual		
	animals are equipped with electronic devices that contain sensors for measuring		
	some property in or near the fish. These devices can either store the resulting data		
Short	internally for	or later download (logging tags) or transfer the data wirlessly to the	
technology	user through radio or acoustic signals (transmitter tags). There exist several		
description	companies delivering such technology, e.g.: https://www.thelmabiotel.com/,		
or link	https://ww	w.innovasea.com/fish-tracking/, https://www.star-oddi.com/	



1	Depends or	chosen sensors. Commercial solutions may be used to monitor several	
	such properties, including ventilation rate, swimming behaviour, exploratory		
Welfare	behaviour, feeding behaviour, depth distribution, heart rate, temperature,		
indicators	oxygen, Co2		
Fish species	Atlantic sal	mon, Rainbow trout, Brown trout, Seabass, Sea bream.	
Type of			
water	salt, brackis	sh and fresh water	
Cultivation	From swim	tunnels, through tanks on land, to full-scale sea-cages (choice between	
unit	logging and	transmitting tags is often based on this)	
Advantages			
of the		dual data histories for fish carrying tags, not dependent on observing	
technology	the animal	directly to collect data	
Disadvantag			
es of the		be handled for deployment, may have negative effects on the fish in	
technology	snort and id	ong term. Relatively expensive method per fish monitored.	
Use case			
		monitor fish behaviour in full scale during critical situations such as	
		crowding, when other equipment is difficult to keep installed at the	
	use case 1	site	
		monitor indicators believed to be closely connected with the	
		physiological response of the fish, such as heart rate, during	
	use case 2	potentially stressful lab trials	
	use case 3	,	
Links to			
existing			
studies			
		https://www.sciencedirect.com/science/article/pii/S0044848617324	
	link 1	407	
		https://www.sciencedirect.com/science/article/abs/pii/S0168169911	
	link 2	000536	
	link 3	https://link.coringer.com/article/10.1196/c40317.030.00336.0	
	IIIIK 3	https://link.springer.com/article/10.1186/s40317-020-00226-8	
Is the	a a at l		
technology	partly		



available for TNA users?		
	restrictio	Both logging and transmitter tags are consumables that need to be bought on a per experiment basis. Several partners in AE3 have
	ns	acoustic receivers that can be used at their respective sites
		If using transmitter tags, receiver units need to be prepared (i.e. equipped with full batteries, and empty memory) and deployed at a
	condition	strategic place relative to the fish. Equipping fish with tags requires
	S	surgical competence

Institute	SINTEF Ocean		
Contact person			
	name	Eirik Spjøtvold	
	email	eirik.spjotvold@sintef.no	
Technology name	Echo Sounder I	EK15	
Short technology description or link	Single Beam sonar. Reflection of soundwave.Trandsducer 200-28CM. https://www.simrad.online/ek15/sales/ek15_ds_en_a4.pdf		
Welfare indicators	Fish density and position in water column		
Fish species	Atlantic salmon		
Type of water	salt and fresh water		
Cultivation unit	Sea cage and tanks		



Advantages of the technology	non-invasive technology, independent of light conditions		
Disadvantages of the technology	limited angle of sight (26 degree)		
Use case			
	use case 1	Mapping of fish distribution in cages	
	use case 2	Mapping of fish distribution in tanks	
	use case 3		
Links to existing studies			
	link 1	doi.org/10.1016/j.apor.2020.102113	
	link 2	https://doi.org/10.1080/00221686.2015.1029016	
	link 3		
Is the technology available for TNA users?	yes		
	restrictions		
	conditions		

Institute	Consejo Superior de Investigaciones Científicas (CSIC)		
Contact			
person			
	name	name Jaume Pérez-Sánchez	
	email <u>jaime.perez.sanchez@csic.es</u>		
Technology			
name	Microbiota composition as a welfare indicator		



Short technology description or link Welfare	Microbiota is key for many host functions, such as digestion, nutrient metabolism, disease resistance or immune function. Thus, determination of microbiota composition can be indicative of welfare status, and manipulation of microbiome can enhance fish performance in terms of growth, health and welfare. For this reason, the combined determination of microbial population in water and fish mucosa (gut, gills, skin) can become a reliable welfare indicator. Technology is based on DNA purification, high-throughput sequencing and subsequent bioinformatic analysis for taxonomic assignation. This approach leads to further metagenome prediction and pathway analysis of the most relevant Operational Taxonomical Units (OTUs)		
indicators	Microbiota		
Fish species	Sea bream,	sea bass, meagre, flatfish	
Cultivation unit	Tanks, sea	cages	
Advantages of the technology Disadvantages of the technology	Early and even predictive indicator of disturbances due to challenging events High cost. Routine analysis with small-scale analytical tools is currently being considered to overcome this limitation		
Use case			
	use case 1	Targeting the core microbiota of fish mucosa in response to aquaculture stressors	
	use case 2	Correlation of microbiota patterns with other host welfare indicators	
	use case 3	Assessment of functional microbiome changes in fish facing a wide range of aquaculture stressors	
	use case 4	Novel insights in the modelling of welfare indicators in response to early life programming	
Links to existing studies			
	link 1	https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5745981	



	link 2	https://www.frontiersin.org/articles/10.3389/fmicb.2019.02512
	link 3	https://link.springer.com/article/10.1186/s40168-020-00922-w
	link 4	https://www.frontiersin.org/articles/10.3389/fmars.2021.659519
Is the technology available for TNA users?	yes	
	conditions	For IATS-ANA (analytical) TNAs, tissue mucus scrapping procedure by external users must be previously accorded with the analytical infrastructure

Institute	Consejo Su	perior de Investigaciones Científicas (CSIC)		
Contact				
person				
	name	Jaume Pérez-Sánchez		
	email	jaime.perez.sanchez@csic.es		
Technology				
name	AEFishBIT			
	A tri-axial accelerometer placed in the operculum of fish that reads			
	simultaneously data of physical activity and respiratory frequency processed by			
	on-board algorithms working in stand-alone mode. The patented smart device			
	has been tested and validated in a swim chamber respirometer and culture tanks			
	•	(TRL level 7). Field tests with free-swimming fish facing a wide array of biotic and		
	abiotic factors have highlighted the association of AEFishBIT measurements with			
	differences in growth performance, stress, and disease resilience. This can be			
Short		lective breeding and for fine adjustment of environmental conditions		
technology	•	h global changes. video link: https://vimeo.com/325943543; Output		
description		//aquaexcel2020.eu/sites/default/files/inline-		
or link	files/Catalo	gue%203%20CSIC_4_v1_0.pdf		



Welfare indicators	Ventilation rate, swimming behaviour, feeding behaviour, aggresive behaviour, exploratory behaviour		
Fish species	Sea bream, sea bass		
Cultivation unit	Tanks		
Advantages of the technology	Miniaturized device (less than 1 g) that can be applied to individuals from 50 g of body weight. Attachment is external, with very short recovery time. Data post-processing is not required		
Disadvantage s of the technology	Proper atta	chment procedure to operculum must be validated for new fish	
Use case			
	use case	Behavioural discrimination of proactive and reactive individuals after exposure to stressful events	
	use case 2	Temporal patterns of adaptation to new culture conditions	
	use case 3 Health condition assessment based on individual and group behaviour criteria use case 4 Use of behaviour patterns as a new criteria of quality certification of aquafeeds		
Links to existing studies			
	link 1	https://www.frontiersin.org/articles/10.3389/fphys.2019.00667	
	link 2	https://www.sciencedirect.com/science/article/pii/S0168169919325 61X	
	link 3	https://www.sciencedirect.com/science/article/pii/S0044848621002 714	
	https://www.sciencedirect.com/science/article/pii/S235251 link 4 612		
Is the technology	yes		



available for TNA users?		
	condition s	AEFishBIT can be implanted in fish species offered in the infrastructure catalogue

Institute	University of South Bohemia		
Contact			
person			
	name	Petr Cisar	
	email	<u>cisar@frov.jcu.cz</u>	
Technology			
name	Infrared ref	lection system	
Short	System for t	fish swimming monitoring in tanks. The system provides the	
technology		about 3D fish position, short term track of the fish and long-term	
description		about fish school swimming behaviour.	
or link		org/10.1016/j.aquaeng.2015.09.002	
Welfare	1 77		
indicators	swimming behaviour, growth, feeding behaviour, depth distribution		
Fish species	Atlantic Salmon, Sea bass, Common Carp		
Type of			
water	satl and fresh water		
Cultivation			
unit	tanks 1-2m2		
Advantages			
of the			
technology	provides information about 3D position in the tank		
Disadvantage s of the			
	need to be calibrated for new species (depth calibration, biomass estimation calibration)		
technology	calibration)		
Use case			
		estimation of the fish biomass during the time without fish	
	use case 1	manipulation	



	use case 2	detection of abnormal behaviour based on the changes of school behaviour
	use case 3	
Links to existing studies		
	link 1	https://www.sciencedirect.com/science/article/pii/S0168169917310 001
	limb 2	https://www.sciencedirect.com/science/article/pii/S0168169915003
	link 2	<u>968</u>
	link 3	
Is the technology available for TNA users?	yes	
	restriction s	3 devices available, the technical asistance is provided by the institute
	conditions	the technology can be instaled at any tank of the institute



Institute	INRAE - PEIMA		
Contact person			
	name	Violaine Colson	
	email	violaine.colson@inrae.fr	
Technology name	video recorder	and cameras	
	12 video camer	ras positioned above 12 tanks: domes FULL HD	
Short technology		IR 30m, 105° + video recorder for 16 cameras	
description or link		rovider: Adrien Alarme. Www.adrienalarme.com	
Welfare indicators		aviour, feeding behaviour, group distribution	
Fish species	Rainbow trout		
Type of water	fresh water		
Cultivation unit	tanks 0.5-2m2		
Advantages of the	Recent system.	. Provides information about fish school	
technology	swimming beh		
Disadvantages of the	the same time. No possibility to equip another site with this		
technology	system		
Use case			
		detection of abnormal behaviour based on the	
	use case 1	changes of school behaviour	
		Observation of any fish group behaviour	
	use case 2	(feeding, aggression)	
	use case 3		
Links to existing studies			
		https://www.sciencedirect.com/science/article/	
	link 1	abs/pii/S0168159118301114?via%3Dihub	
	link 2		
	link 3		
Is the technology available			
for TNA users?	yes		
	restrictions	12 tanks video-equipped	
		the technology cannot be installed in another	
	conditions	site	



Existing and emerging technologies available for welfare indicators monitoring

1. Objective

The main objective of this deliverable is to make an overview of existing and emerging technologies available for monitoring fish welfare in tanks and sea cages. The overview serves as a sorted list of technologies suitable for experimental work planned in WP6 of the project.

2. Background

Fish welfare is a well-established term that resonates in the last years in the research but mainly in fish production. Related to the monitoring technologies, which is the focus of thi deliverable, we can use feeling-based welfare definition in which welfare links to the emotional (or emotion-like) states of the animal under review. Good welfare under these definitions typically requires a reduction in negative experiences (such as stress or fear) and an assurance of positive experiences (such as the presence of counterparts for members of social species). Good fish welfare is the key to sustainable fish production. AQUAEXCEL3.0 is a project with a focus on the improvement of aquaculture infrastructures improvement and the harmonization of the procedures through joint research activities. Therefore, WP6 deals with the harmonization of fish welfare indicators, monitoring of the indicators, and improvement of welfare by enrichment principles. Task 6.1. is defining welfare indicators and operational welfare indicators together with the recommended limits for several species. These indicators can serve as a measure of fish welfare only if they are easily measurable and interpretable by researchers and fish farm operators. Several technologies for welfare measures were developed and described in the literature, but still, there is no routinely used technology. Task 6.2 aims to test selected promising welfare monitoring technologies for different species and different infrastructures to demonstrate generality and usability under real conditions. To be able to choose the best technology, we need to have an overview of existing and emerging technologies suitable for welfare monitoring. This document provides the list of available technologies divided into several groups (based on the type of information produced by the measurement device) suitable for



monitoring at least some welfare indicators. The list excludes the basic technologies routinely used at the infrastructures for water quality monitoring (pH, temperature, etc.).

3. Technologies

The technologies were selected based on the literature search (keywords: fish; welfare; monitoring; technologies) and the questionnaire of the project filled by 11 infrastructures. In total, 22 research papers were used to make the list of technologies. There are several possibilities for how to group welfare monitoring technologies. First, we divided the technologies based on the type of information produced by the measuring device: water measurement, echo sounders, tags, video cameras, frames, and manual data collection. The second criteria is the availability of the determined indicator: online/offline. Online estimated welfare indicators can be used to make immediate action for welfare improvement. Offline estimated welfare indicators can be used for welfare documentation during the fish cultivation period only. In this review, we mainly focused on non-invasive technologies providing the data online. The reason is to be able to minimize fish stress caused by invasive technologies and the possibility of immediate action to improve fish welfare.

3.1. Water measurement

Water quality is strongly correlated to fish welfare even if it is an indirect indicator. Almost all infrastructures have the technology for online measuring basic water quality parameters (pH, oxygen, temperature. This technology provides information about immediate changes in water quality parameters and can be used for welfare monitoring. General recommendations for water quality parameters exist for the fish species, and the infrastructure keeps the parameters at defined levels. The technology for water quality measurement usually consists of the probes installed in the tank/cage measuring the data at a fixed point. Several advanced measurement devices exist to measure water quality during fish transfer and manipulation (https://sednatech.io/sensor-globeenglish/ or using wireless technology for real-time data collection (http://www.waterlinked.com/cagesense?hsLang=en).







Figure 1. Commercially available sensory and wireless network for data collections. Left – Globe sensor (https://www.sensorglobe.no/). Right – CageSense network of sensors (http://www.waterlinked.com/cagesense?hsLang=en)

One of the most interesting technologies in the area of water parameters measurement is cortisol measurement. Measurement of cortisol is considered a standard approach to assessing the effect of stress on fish. For example, measurement of plasma cortisol indicates well acute stress but does not necessarily reflect a state of chronic stress (Aerts et al., 2015). However, it has been used successfully by a number of scientists, from physiologists (Auperin et al., 1997) to behavioral ecology (Colson et al., 2015b), in a controlled environment as well as in the wild (Geffroy et al., 2017, Love et al., 2013) to assess the level of stress in fish. The relationship between stress and neurogenesis (Sadoul et al., 2018), growth (Sadoul & Vijayan, 2016), and sex determination or gender reassignment (Olivotto & Geffroy, 2017) are also well described and are subject to the role of cortisol in fish physiology and behavior. It is possible to measure cortisol directly in the water, but the usage is minimal. Cortisol extraction from tank water has been previously explored (Ruane et al., 2003, Ellis et al., 2007, Zuberi et al., 2014, Lower et al., 2005) . The advantage of cortisol in water measurement is that it is a completely non-invasive approach which makes the method useful in practice. It could be complicated to use the method in RAS systems where we face the issue of cortisol accumulation. The time of cortisol extraction and the costs are much higher than for other invasive methods. To measure individual fish cortisol release, sampled fish should be individually housed for the most precise measurements, and individual housing can be a stressful experience in itself. The usage of cortisol measurement in sea cages is not documented. More information about cortisol measurement is provided by Sadoul & Geffroy (2019).



The studies using the technology:

- Aerts, J., Metz, J. R., Ampe, B., Decostere, A., Flik, G., & Saeger, S. D. (2015). Scales tell a story on the stress history of fish. PLOS ONE, 10, e0123411. https://doi.org/10.1371/journal.pone.0123411
- Auperin, B., Baroiller, J. F., Ricordel, M. J., Fostier, A., & Prunet, P. (1997). Effect of confinement stress on circulating levels of growth hormone and two prolactins in freshwater-adapted tilapia (Oreochromis niloticus). General and Comparative Endocrinology, 108, 35–44. https://doi.org/10.1006/gcen.1997.6938.
- Colson, V., Valotaire, C., Geffroy, B., & Kiilerich, P. (2015b). Egg cortisol exposure enhances fearfulness in larvae and juvenile rainbow trout. Ethology, 121, 1191–1201.
- Ellis T, James JD, Sundh H, Fridell F, Sundell K, Scott AP. Non-invasive measurement of cortisol and melatonin in tanks stocked with seawater Atlantic salmon. Aquaculture. 2007;272(1–4):698-706. https://doi.org/10.1016/j.aquaculture.2007.07.219
- Geffroy, B., Sadoul, B., & Ellenberg, U. (2017). Physiological and behavioral consequences of human visitation. In D. T. Blumstein, B. Geffroy, D. S. M. Samia, & E. Bessa (Eds.), Ecotourism's promise and peril: a biological evaluation (pp. 9–27). Cham, Switzerland: Springer International Publishing. https://doi.org/10.1007/978-3-319-58331-0_2.
- Love, O. P., McGowan, P. O., & Sheriff, M. J. (2013). Maternal adversity and ecological stressors in natural populations: The role of stress axis programming in individuals, with implications for populations and communities. Functional Ecology, 27, 81–92. https://doi.org/10.1111/j.1365-2435.2012.02040.x.
- Lower N, Moore A, Scott AP, Ellis T, James JD, Russell IC. A non-invasive method to assess the impact of electronic tag insertion on stress levels in fishes. J Fish Biol. 2005;67(5):1202-1212. https://doi.org/10.1111/j.1095-8649.2005.00815.x
- Sadoul, B., Alfonso, S., Bessa, E., Bouchareb, A., Blondeau-Bidet, E., Clair, P., Geffroy, B. (2018). Enhanced brain expression of genes related to cell proliferation and neural differentiation is associated with cortisol receptor expression in fishes. General and Comparative Endocrinology, 267, 76–81. https://doi.org/10.1016/j.ygcen.2018.06.001.
- Sadoul, B, Geffroy, B. Measuring cortisol, the major stress hormone in fishes. J Fish Biol. 2019; 94: 540–555. https://doi.org/10.1111/jfb.13904



- Sadoul, B., Vijayan, M.M., 2016. 5 Stress and Growth, in: Schreck, L.T., Anthony P. Farrell and Colin J. Brauner Carl B. (Ed.), Fish physiology, biology of stress in fish (pp. 167–205). London, England: Academic Press.
- Olivotto, I., & Geffroy, B. (2017). Clownfish. In Marine ornamental species aquaculture (pp. 177–199). Hoboken, New Jersey: Wiley-Blackwell. https://doi.org/10.1002/9781119169147.ch12.
- Ruane NM, Komen H. Measuring cortisol in the water as an indicator of stress caused by increased loading density in common carp (Cyprinus carpio). Aquaculture. 2003;218(1–4):685-693. https://doi.org/10.1016/S0044-8486(02)00422-2
- Zuberi A, Brown C, Ali S. Effect of confinement on water-borne and whole body cortisol in wild and captive-reared rainbowfish (Melanoteania duboulayi). Int J Agric Biol. 2014;16(1):183-188.

SUMMARY:

Online/offline: more often online

Welfare indicators: water quality, cortisol level

Advantages: technically simple and robust, direct measurement of cortisol

Disadvantages: indirect measurement of fish welfare, difficult to study the welfare of individuals, cortisol – low sensitivity, and specific measurement conditions

Price: 30 – 2000Euro (depending on the number of water parameters and data transmission)

3.2. Tags

Tags (biosensors) are miniature sensors implanted to fish or attached to fish so that they can measure physiological parameters, acceleration, or depth. These devices can store measured data in the internal memory or transfer them online to a computer (acoustic tags).

These devices can collect a large amount of systematic and detailed data. However, the use can have short-term or long-term effects on the animal (Macaulay et al. 2021). In addition, placing the sensor

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can be a highly stressful procedure because it involves trapping, manipulation, anesthesia, and surgery. Marked fish usually have a higher mortality rate than unmarked fish (Macaulay et al. 2021).

The first group of tags is the biosensors monitoring the physiological parameters of the fish. Biosensors take advantage of molecular elements of biological processes such as enzymes and antibodies to identify target substances. When biocatalysts react with a target substance, they can serve as discriminating elements that can cause minor changes in parameters such as current, resistance, and heat due to the generation and consumption of chemical substances (Fig. 1). Biosensors can measure specific target substances easily and rapidly by detecting these small changes using signal conversion elements such as electrodes and optical devices, which transform the changes into electric signals. The overview of different biosensors is available in Endo & Wu (2019).

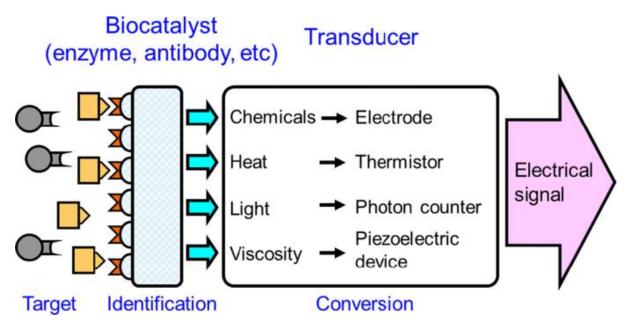


Figure 2. The principle of biosensor (https://link.springer.com/article/10.1007/s12562-019-01318-y)

One of the most relevant biosensors for fish welfare monitoring is the sensor to measure the level of glucose in the blood, which typically rises in response to a stressful event and can remain for more than one day (Jentoft et al., 2005). Wu et al. (2015) created an implantable biosensor for measurement that can transmit data in real-time. A $1.5 \times 1.5 \times 0.6$ cm biosensor (3 g without battery) is implanted in the interstitial glass of the eyeball because the glucose levels at this site are highly correlated with blood glucose. However, the sensor can only operate for 160 minutes in continuous monitoring mode.



The value correlates sufficiently with the blood glucose level measured. The glucose biosensor can be attached to the fish body with the electrode implanted into the fish's eye (Fig. 3).

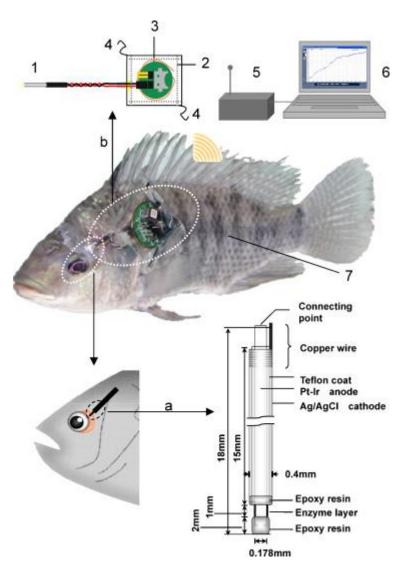


Figure 3. Wireless enzyme sensor system for real time monitoring of blood glucose concentrations in fish (https://www.sciencedirect.com/science/article/pii/S0956566308004764?via%3Dihub)



Another quantitative stress marker that can be measured using biosensors is the heart rate and its variability. Examples of such biosensors are DST milli-HRT biogeners or STAR-ODDI. These sensors have been tested in several studies on rainbow trout and Atlantic salmon (Brown et al., 2011, Føre et al., 2021). In these studies, the tags were implanted in the abdominal cavity, in close proximity to the pericardium. Biosensors were used for heart rate measurement for several weeks.

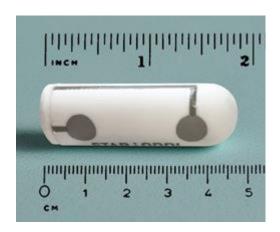


Figure 4. STAR ODDI data logger for heart rate, activity and temperature measurement (https://www.star-oddi.com/products/archival-tags/animal-heart-rate-activity-logger)

AEFishBIT (Martos-Sitcha et al., 2019) is an example of a biosensor that can be attached externally to the body of a fish without further implantation (only for selected species). The sensor is attached to the fish operculum with a clip and allows the measurement of acceleration in three axes. Two axes of the accelerometer (x, y) monitor locomotor activity through fish, and the third axis of accelerometer (z) monitors respiratory activity due to the location on the operculum.

The disadvantage of data loggers is an offline regime of data measurement which reduces their usability. The solution to this limitation is acoustic telemetry (Føre et al., 2018), which connects biosensors with a transmitter for data transmission. The receivers are generally immersed near or inside the tank/cage to capture and interpret the received signals into usable data, which is then stored or processed. There are problems with the implementation of a successful telemetry system, such as range limitation. Currently, the maximum range between acoustic sensors and their receivers is approximately 1 km. A new wireless system using Low Power Wide Area Networks (LPWAN) technology to overcome this limitation has recently been developed for use in suspended sea cages (Hassan et al., 2019). LPWANs are commonly used in Internet of Things technology. In this context, LPWAN nodes are added to the acoustic receivers to transmit the received signals to a single gateway.



It is then transferred to a personal computer, which serves as a server and user interface. This technology allows access to data in real-time from several sea cages.



Figure 5. Thelma Biotel acoustic tags (https://www.thelmabiotel.com/transmitter/)

The studies using the technology:

- Brown RS, Eppard MB, Murchie KJ, Nielsen JL, Cooke SJ. An introduction to the practical and ethical perspectives on the need to advance and standardize the intracoelomic surgical implantation of electronic tags in fish. Rev Fish Biol Fish. 2011;21(1):1-9. https://doi.org/10.1007/s11160-010-9183-5
- Endo, H., Wu, H. Biosensors for the assessment of fish health: a review. Fish Sci 85, 641–654 (2019). https://doi.org/10.1007/s12562-019-01318-y

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- Føre M, Svendsen E, Alfredsen JA, et al. Using acoustic telemetry to monitor the effects of crowding and delousing procedures on farmed Atlantic salmon (Salmo salar). Aquaculture. 2017;2018(495):757-765. https://doi.org/10.1016/j.aquaculture.2018.06
- Føre, M., Svendsen, E., Økland, F. et al. Heart rate and swimming activity as indicators of post-surgical recovery time of Atlantic salmon (Salmo salar). Anim Biotelemetry 9, 3 (2021). https://doi.org/10.1186/s40317-020-00226-8
- Hassan W, Føre M, Ulvund JB, Alfredsen JA. Internet of fish: Integration of acoustic telemetry with LPWAN for efficient real-time monitoring of fish in marine farms. Comput Electron Agric. 2019;163:104850. https://doi.org/10.1016/j.compag.2019.06.005
- Jentoft S, Aastveit AH, Torjesen PA, Andersen Ø. Effects of stress on growth, cortisol and glucose levels in non-domesticated Eurasian perch (Perca fluviatilis) and domesticated rainbow trout (Oncorhynchus mykiss). Comp Biochem Physiol A Mol Integr Physiol. 2005;141(3):353-358.
- Macaulay G, Warren-Myers F, Barrett LT, Oppedal F, Føre M, Dempster T. Tag use to monitor fish behaviour in aquaculture: a review of benefits, problems and solutions. Rev Aquac. 2021;13:1565-1582.
- Martos-Sitcha JA, Sosa J, Ramos VD, Bravo FJ, Carmona DC, Gomes HL, Calduch GJÀ, Cabruja E, Vega A, Ferrer MÁ, Lozano M, Montiel NJA, Afonso JM, Pérez SJ. Ultra-Low Power Sensor Devices for Monitoring Physical Activity and Respiratory Frequency in Farmed Fish. Frontiers in Physiology. 2019. DOI=10.3389/fphys.2019.00667
- Wu H, Aoki A, Arimoto T, et al. Fish stress become visible: a new attempt to use biosensor for real-time monitoring fish stress. Biosens Bioelectron. 2015;67:503-510.

SUMMARY:

Online/offline: data loggers – offline, acoustic tags -online

Data: depth, temperature, salinity, conductivity, acceleration (movement/respiratory activity), heart rate, glucose

Welfare indicators: glucose level, heart rate, swimming activity, respiratory activity

Advantages: independent of the water conditions, direct measurement of fish activity/physiological parameters

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Disadvantages: invasive, high price, only a few individuals are usually monitored, the limited size of the fish based on the tag size, the measurements are not usually taken continuously but in batches to save battery life, the fish must be caught to retrieve the tag

Price: 300 – 1500EUR (depending on the type and number of measurement parameters), acoustic tags need to use transceiver (8000EUR)

3.3. Echosounder

The echosounders are the technology that is completely non-invasive and enables the possibility of online (real-time) data measurement. The modern echosounder device needs a personal computer (PC) to analyze and visualize data; therefore, it comes with the software. The installed software in the PC can display the received signal graphically as an echogram. The echosounder mainly consists of four components: transducer, transmitter, receiver, and display unit. The transmitter generates an electrical pulse that is transformed to sound waves by the transducer and propagate through the water. The reflected signal from the targets in water are sampled by the transducer again, changed back to an electrical signal, and echograms are generated. However, because of its accuracy and noninvasive nature, acoustic echo-sounding is a popular choice for fish count estimation (Sthapit et al., 2020). In a typical echosounder, the transmitter emits a ping (acoustic signal), and the received echo signals from targets are analyzed. The academic as well as commercial use of acoustics for estimating fish abundance have been widespread (Lubis et al., 2017, Conti et al., 2006). Although fish have been the main focus, squids and other marine organisms have also been investigated. Modern echosounder implements echo integration to analyze fish and their behavior (Røttingen 2020). Even though the echo integration method has been extensively employed in the open ocean, however, when the method is employed in the farming cages, it offers new challenges (Sthapit et al., 2019). Firstly, the fish are closer to a transducer, therefore, classical Time Varying Gain (TVG) correction could be inappropriate. Secondly, the reverberation of the acoustical signal from the boundaries of the cage needs to be considered. Another problem that needs to be addressed is removing the cage signal from the received echo. Similarly, the stability of the transducer is another issue as the signal quality highly depends on its orientation. The fish position (depth) and distribution over the cage is usually the main parameter of investigation, which is useful for fish welfare monitoring. Behaviour-related welfare indicators are strongly dependent on the fish species, and it is difficult to define the baseline of "normal" fish behavior to detect the abnormalities.



The echo sounder is not limited just to the open water. The small echo sounders used by fishermen could be used in the limited space of fish tanks. There is no scientific study using sonar in a fish tank, but some experiments were already done (https://youtu.be/Wggk1y5erlU).

The echo sounders could also be used to estimate the number of fish and the biomass in sea cages. The first successful experiments were done in the project PerformFISH (https://thefishsite.com/articles/using-echo-sound-to-estimate-biomass-in-aquaculture) or by Sthapit (Sthapit 2020).

The studies using the technology:

- Conti S.G., Roux P., Fauvel C., Maurer B.D., Demer D.A. Acoustical monitoring of fish density, behavior, and growth rate in a tank. Aquaculture. 2006;251:314–323.
- Lubis M.Z., Manik H.M. Acoustic systems (split beam echo sounder) to determine abundance of fish in marine fisheries. J. Geosci. Eng. Environ. Technol. 2017;2:76–83. doi: 10.24273/jgeet.2017.2.1.38.
- Røttingen I. On the Relation between Echo Intensity and Fish Density. [(accessed on 26 October 2020)]; Available online: https://imr.brage.unit.no/imr-xmlui/bitstream/handle/11250/114400/sh_vol16_09(2)_1976.pdf?sequence=1.
- Sthapit P., Teekaraman Y., MinSeok K., Kim K. Algorithm to Estimation Fish Population using Echosounder in Fish Farming Net; Proceedings of the 2019 International Conference on Information and Communication Technology Convergence (ICTC); Jeju Island, Korea. 16–18 October 2019; pp. 587–590.
- Sthapit, P.; Kim, M.; Kang, D.; Kim, K. Development of Scientific Fishery Biomass Estimator: System Design and Prototyping. *Sensors* 2020, *20*, 6095. https://doi.org/10.3390/s20216095

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Online/offline: online/offline



Welfare indicators: fish swimming activity, grouping, position preference in sea cage, maybe fish biomass

Advantages: a global view of fish activity in sea cage, independent of the light conditions

Disadvantages: low resolution in comparison with camera system, impossible to track individuals, usually 2D information, synchronization of more echo sounders, complicated use in fish tank, limited to behavioural observations

Price: 100Euro – 3000Euro (depending on the resolution, number of sonar beams, and number of frequencies)

3.4. Video monitoring

Video monitoring is more and more used in aquaculture, which is caused by the decreasing price of video cameras, increasing quality of video records, and improvement of the methods of video processing. Many sea cages are equipped with feeding cameras, and the researchers try to find a way to automatize the video processing of fish behavior to extract information about fish swimming, growth, and fish appearance. All three parameters can be used for welfare estimation because it corresponds to the welfare indicators like behavior patterns (abnormal behavior), fish deformation or injuries, and abnormalities in fish growth. Different camera systems using different spectra were used in the research studies. The camera in the near-infrared spectrum was used to analyze behavior (Pautsina et al., 2015), fish biomass in a tank (Saberioon et al., 2018), and feeding behavior (Zhou et al., 2018). The camera in the visible spectrum is usually used for fish behavior monitoring in the sea cages (Pinkiewicz et al., 2011) and aquarium (Chunley et al., 2016). The 3D cameras and stereo vision principle are usually used for 3D fish position monitoring in tanks (Saberioon et al., 2016) and fish biomass estimation in sea cages (Muñoz et al., 2018). The video camera can be used for a variety of fish monitoring: fish swimming monitoring in 2D or 3D space, feeding behavior analysis, fish size measurement (see Fig. 6), but it requires a specific method for video data processing.





Ground truth Identification with FEI≤1.8

Figure 6. Example of automatic fish detection using a bottom-up video camera. The algorithm is for tuna detection is trained from manually labeled data (left), and the fish biomass and behavior are estimated from automatically detected individuals (right). https://www.sciencedirect.com/science/article/pii/S0168169916309395?via%3Dihub#f0005

The main difference between tags, echo sounders, and video cameras is the need for additional complicated data processing of the video records from the camera. The cameras are also limited to monitoring in the clear water during the daylight period. Artificial light as near-infrared light can be used to extend the monitoring to the night period of a day, but some fish species are sensitive to infrared spectra. The image processing methods can detect individual fish to estimate the trajectory of individuals or determine the fish shape or process the behavior pattern of the whole fish school. Modern image processing methods based on deep neural networks can detect individual fish and estimate fish breathing (Valenzuela et al., 2021) or fish mouth opening which is correlated with the fish welfare state. These studies are just experimental trials and are under development. Tags or sonar can be used in the same way for different species and different conditions. The flexible solutions based on the video cameras are usually designed for specific conditions and specific fish species and, therefore, are difficult to apply to other species and conditions. The main reason is the specificity of the data image processing methods.

There are few commercial systems using video cameras for biomass estimation and sea lice detection reporting welfare monitoring. For example, VAKI AQUACULTURE SYSTEMS LTD. offers Falcon camera system (https://www.aquafalcon.com/) for automatized sea lice detection.

The use of video cameras can be combined with the use of other devices. For example, Key et al. (2019) introduced the advantage of combination of sonar and optical camera for fish monitoring. They combined both data using neural network to monitor fish activity during the day and night period, see Fig. 7.





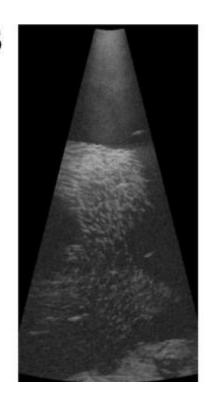


Figure 7. Combination of optical camera (A) and high-resolution sonar (B). https://www.sciencedirect.com/science/article/pii/S0144860919300640?via%3Dihub

The studies using the technology:

- Chunlei X., Tae-Soo Ch., Yuedan L., Jing Ch., JangMyung L. Posture tracking of multiple individual fish for behavioral monitoring with visual sensors, Ecological Informatics, Volume 36, 2016, Pages 190-198, ISSN 1574-9541, https://doi.org/10.1016/j.ecoinf.2016.07.004.
- Kei T, Kento S, Katsunori M, Koji T. Integration of sonar and optical camera images using deep neural network for fish monitoring. Aquacultural Engineering, Volume 86, 2019, 102000, ISSN 144-8609, https://doi.org/10.1016/j.aquaeng.2019.102000.
- Muñoz-Benavent P, Andreu-García G, Valiente-González JM, Atienza-Vanacloig V, Puig-Pons V, Espinosa V. Enhanced fish bending model for automatic tuna sizing using computer vision. Comput Electron Agric. 2018 ;150:52-61. https://doi.org/10.1016/j.compag.2018.04.005
- Pautsina A, Císař P, Štys D, Terjesen BF, Espmark ÅMO. Infrared reflection system for indoor 3D tracking of fish. Aquac Eng. 2015;69:7-17. https://doi.org/10.1016/j.aquaeng.2015.09.002



- Pinkiewicz TH, Purser GJ, Williams RN. A computer vision system to analyse the swimming behaviour of farmed fish in commercial aquaculture facilities: a case study using cage-held Atlantic salmon. Aquac Eng. 2011 ;45(1):20-27. https://doi.org/10.1016/j.aquaeng.2011.05.002
- Saberioon MM, Cisar P. Automated multiple fish tracking in three-dimension using a structured light sensor. Comput Electron Agric. 2016;121:215-221. https://doi.org/10.1016/j.compag.2015.12.014
- Saberioon M, Císař P. Automated within tank fish mass estimation using infrared reflection system. Comput Electron Agric. 2018;150:484-492. https://doi.org/10.1016/j.compag.2018.05.025
- Valenzuela A, Sibuet N, Hornero G, Casas O. Non-Contact Video-Based Assessment of the Respiratory Function Using a RGB-D Camera. *Sensors*. 2021; 21(16):5605. https://doi.org/10.3390/s21165605
- Zhou C, Xu D, Lin K, Sun C, Yang X. Intelligent feeding control methods in aquaculture with an emphasis on fish: a review. Rev Aquac. 2018;10(4):975-993. https://doi.org/10.1111/raq.12218

SUMMARY:

Online/offline: online

Welfare indicators: swimming activity, feeding, fish growth, fish deformation, fish injury

Advantages: high-quality data, individual monitoring, several types of parameters can be determined from a video signal

Disadvantages: water clarity, high demanding data processing, difficult to generalize the solution for more species and conditions

Price: 10 - 2000 EUR

3.5. Measuring frames

Measuring frame is a commercial solution specialized for fish biomass estimation only. The link between biomass and welfare is through the influence of fish growth by the stress. Several producers (VAKI AQUACULTURE SYSTEMS LTD.) offer the frames operating 24/7 online to estimate the biomass of the fish swimming through the frame. Difford (Difford et al., 2020) validated that the frames can



precisely estimate the fish biomass in the sea cage. Biomass estimation can be used as a welfare indicator to monitor fish growth online.

The studies using the technology:

Difford GF, Boison SA, Khaw HL, Gjerde B. Validating non-invasive growth measurements on individual Atlantic salmon in sea cages using diode frames. Computers and Electronics in Agriculture, Volume 173, 2020, 105411, ISSN 0168-1699, https://doi.org/10.1016/j.compag.2020.105411.

SUMMARY

Online/offline: online

Welfare indicators: biomass

Advantages: robust commercially available solution

Disadvantages: biomass estimation only, fish must swim through the frame

3.6. Applications

The alternative solution to the automatic systems of welfare monitoring is the support of user measurement/observation from the daily fish maintenance. The mobile application Fishwell (https://www.kyst.no/article/en-app-gjoer-velferdsovervaakningen-av-forsoeksfisk-mer-presis/) was developed to record operators observations of welfare indicators based on gill lid status, eyes, fins, etc. The user provides the score of the observation together with the image (taken by mobile phone), and the system is used as evidence of the fish welfare. The combination of human data collection followed by machine data processing is a promising direction of fish welfare monitoring. This solution is limited to the tanks only.

SUMMARY

Online/offline: online



Welfare indicators: any human can score

Advantages: a simple approach

Disadvantages: based on human observation

4. Conclusion

The overview of the technologies available for welfare indicators measurement was extracted from the literature review and the questionnaires delivered by project partners. The main six categories of measurement devices (principles) were described together with the links to the existing studies and a summary of the advantages/disadvantages of the technologies. The most promising technologies for operational welfare indicators measurement are the non-invasive technologies providing online data and information about fish swimming or appearance. There are several emerging video processing methods that could bring new possibilities of physiological fish parameters remotely by the video camera, but they are not commercially available now.

5. Glossary/Definitions

AQUAEXCEL3.0: AQUAculture Infrastructures for EXCELlence in European Fish Research