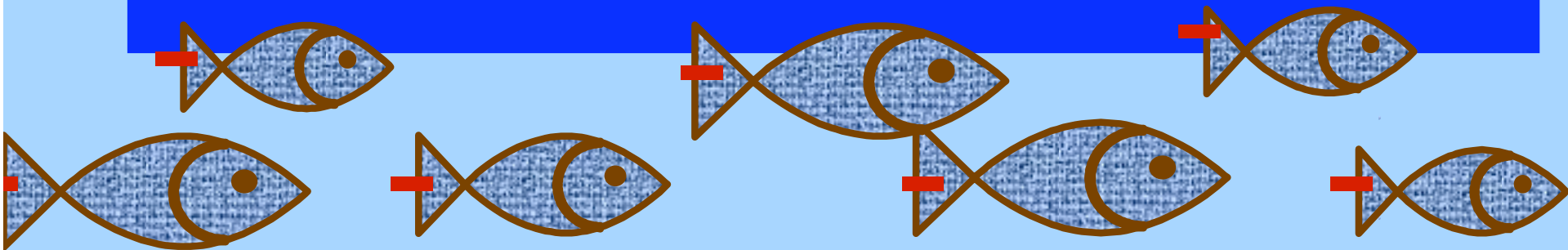


# An overview of existing guidelines for handling, bleeding, administration and identification techniques



Penny Hawkins, Research Animals  
Department, Science Group, RSPCA

# “Wish list” from the 2005 meeting

- Species-specific guidelines for the care and use of fish
- Easy access to the latest knowledge
  - ▶ because regulations and guidelines can change
  - ▶ a closed discussion group would help
- Guidance on best practice, not a standard set of techniques
- Education for scientists on information retrieval

# Views on guidelines in 2005

- Lots of guidelines on procedures; can be difficult to access
  - ▶ Need to “pull them together”
- Identification - best practice varies with circumstances, guidance would be helpful
  - ▶ “Marking methods” table should be produced
- Blood sampling - acceptable volumes based on total blood volume needed for each species
- Methods should be proven before changing practice

# Scope of the talk

- Many regulations and guidelines require refinement
  - ▶ Norwegian Animal Welfare Act
  - ▶ European Commission Directive 86/609
  - ▶ UK Animals (Scientific Procedures) Act 1986
- General requirement to use least painful/stressful technique possible
- This talk only covers guidelines specific to fish

# Outline of the talk

- How the search for guidelines on handling, sampling, administration, identification was done
- Current guidelines
- What they say
- How they relate to current views on good practice and the 2005 “wish list”
- What still needs to be addressed

# Guideline search

- CCAC
- Norwegian School of Veterinary Science
- NC3Rs
- USDA Animal Welfare Information Centre
- COST Action B24
- Johns Hopkins Altweb
- [www.fishwelfare.net](http://www.fishwelfare.net)
- Animal Welfare Institute
- Concerted Action for Tagging of Fishes
- Current legislation on animal experimentation

# What do we have?

- 1 FISHBIO [www.fishmarking.com](http://www.fishmarking.com) (2007)
- 2 Council of Europe Convention ETS123  
Appendix A (2006)
- 3 CCAC Guidelines on: The Care and Use of  
Fish in Research, Teaching and Testing  
(2005)

# What do we have (2)?

## 4 UK Home Office: Focus on Fish (2005)

- ▶ <http://scienceandresearch.homeoffice.gov.uk/animal-research/>  
see Animals (Scientific Procedures) Inspectorate  
Annual Report for 2005

## 5 Guidelines for the Use of Fishes in Research (2004)

- ▶ American Fisheries Society, American Institute of Fishery Research Biologists, American Society of Ichthyologists and Herpetologists



## What do we have (3)?

6 CATAG (2002) Improvements in Tagging Methods for Stock Assessment and Research in Fisheries

▶ [www.hafro.is/catag/](http://www.hafro.is/catag/)

7 The Laboratory Fish (2000)

8 UFAW Handbook on the Care and Management of Laboratory Animals, 7th edn, Volume 2 (1999)

▶ UFAW is hoping 8th will be published in 2010

# What should they encompass?

- Welfare principles
  - ▶ Impact on the fish - stress, welfare
  - ▶ Potential for physical damage, health issues
  - ▶ ... with BVAAWF/FRAME/RSPCA/UFAW Joint Working Group on Refinement approach in mind
- And from 2005 “wish list”...
  - ▶ Species-specific guidance
  - ▶ Proven methods
  - ▶ Guidance on identifying and implementing best practice

# Handling

- ETS123, Home Office, UFAW
  - ▶ Mention stress, damage, provide minimal guidance
  - ▶ ETS123, Home Office: anaesthesia
  - ▶ UFAW: “wet transfer”
- AFS
  - ▶ Stress, damage, more guidance
  - ▶ Eliminate rough handling, abrasion, sudden changes in water quality
  - ▶ Wet transfer, anaesthesia (to reduce metabolic rate)

# CCAC on handling

- Stress, damage
- Staff training
- Avoid and minimise handling
- Use of anaesthetics, sedatives
- Minimise visual stimulation to fish
- Minimise time out of water, up to 30 seconds
- No species-specific guidelines

# Blood sampling

- UFAW: freshwater fish
  - ▶ Anaesthesia or restraint, restrainer design
  - ▶ Recommends needle size
  - ▶ 0.5 ml from 150 g fish
  - ▶ Caudal vessels or cardiac puncture - with recovery?
- UFAW: marine fish
  - ▶ Blood volume is 3 - 5 % body mass
  - ▶ Cardiac puncture without recovery
  - ▶ Cutaneous vein or suborbital sinus

# Blood sampling

- AFS
  - ▶ Sterile conditions impossible in field
  - ▶ Mentions anaesthesia
  - ▶ Venous puncture, caudal bleed, cardiac puncture (with recovery?)
  - ▶ Cannulation
  - ▶ Amputate tail under terminal anaesthesia

# Blood sampling

- The Laboratory Fish
  - ▶ Much technical detail, *e.g.* anticoagulants, cannula materials
    - Caudal puncture
    - Tail amputation (without recovery)
    - Cardiac puncture (single collection)
    - Cannulae - dorsal or ventral aorta, caudal vein
  - ▶ Nothing about welfare

# Blood sampling

- CCAC
  - ▶ 1 ml kg<sup>-1</sup> in general (0.15 ml in 150 g fish)
  - ▶ Recover haematocrit before next sample - this is temperature dependent and species-specific
  - ▶ Sedation/anaesthesia
  - ▶ Staff training
  - ▶ Sterile equipment
  - ▶ Ventral tail vein, dorsal aorta, cardiac puncture (with recovery?)
  - ▶ Cannulation in teleosts over 150 g



# Blood sampling - summary

- Most mention anaesthesia
- Different levels of detail and consideration for welfare/refinement
- No guidance on choosing optimal route from welfare aspect
- No species-specific guidelines
- Cardiac puncture with recovery - not recommended for mammals; is it acceptable for fish?

# Administering substances

- UFAW: freshwater fish
  - ▶ Injections - sites, examples of needle gauge, volume
- The Laboratory Fish
  - ▶ Technical detail, *e.g.* chamber design, vehicles
    - Water-borne exposure
    - Oral administration; via feed or gavage
    - Injection: iv, ip, im
    - Implants; pellets, osmotic pumps
    - Topical exposure
  - ▶ Nothing on welfare

# Administering substances

- CCAC
  - ▶ Range of routes
    - Branchial diffusion
    - Oral gavage (1 ml per 100 g)
    - Injection - im, ip, iv
    - Implants
    - Windows, bioreactors
  - ▶ Practical guidance on how to get it in but less focus on fish's experience or choosing method
  - ▶ Further references recommended

# Administration summary

- Some practical guidance on protocols
- Overall, more technical than animal-centred guidance
- Very little on dose volumes
- Nothing species-specific
- No guidance on identifying best practice

# Identification

- ETS123
  - ▶ Anaesthesia, some guidance on least invasive
- AFS
  - ▶ Harms and benefits of marking
  - ▶ Users should review recent literature
  - ▶ Information on different techniques
- UFAW
  - ▶ Freshwater fish: mentions microchips; marine fish: stress of tagging, user should review literature

# Identification

- Home Office
  - ▶ List of methods
  - ▶ Dye/elastomer marking possibly causes least pain, suffering or distress
  - ▶ Fin removal “least refined”

# Identification

- CCAC
  - ▶ Principles for reducing stress during marking and subsequent identification; potential harms
  - ▶ Pilot studies for new methods; evaluation of welfare impact
  - ▶ Mentions tissue marking, tagging, genetic markers, internal tags and marks
  - ▶ Techniques that cause significant tissue injury should only be used if no alternative and justified to animal care committee

# Identification

- CATAG
  - ▶ Minimise pain, stress, adverse effects on health
  - ▶ Review data before tagging or do pilot studies
  - ▶ Fish taggers should all undergo training
  - ▶ Use of anaesthesia
  - ▶ Research needed into “anaesthetic” effects of low temperature
  - ▶ Pay attention to physiological impact of device and attachment methods



# Identification

- [www.fishmarking.com](http://www.fishmarking.com)
  - ▶ Definitions, advantages, disadvantages and other resources for long list of techniques
    - External attached tags; dye submersion; inoculation/tattoo/photonic; branding (non-chemical and chemical); fin clip; body cavity tag; coded wire tag (CWT); acoustic tags; visual implant/elastomeres/fluorescent tags; data storage tags; PIT tags; DNA; calcein binding to scales; oxytetracycline bath; otolith marking
  - ▶ Little on welfare; mentions tissue damage, behaviour, mortality

# Identification summary

- More on this than handling, administration or blood sampling procedures
- Information on techniques
- Information on harms
- Principles for refinement
- It would be nice if this were all in the same place - with guidance on preference from a welfare point of view!

# What has happened since 2005?

- Species-specific guidelines for the care and use of fish ✕
- Easy access to the latest knowledge ?
  - ▶ because regulations and guidelines can change
  - ▶ a closed discussion group would help
- Guidance on best practice ✕
- Education for scientists on information retrieval ?

# Guidelines since 2005

- Lots of guidelines on procedures; can be difficult to access
  - ▶ Need to “pull them together” X
- Identification - varies with circumstances, guidance would be helpful X
  - ▶ “Marking methods” table
- Blood sampling - acceptable volumes based on TBV for each species needed X
- Methods would need to be proven before changing practice X

## To sum up ...

- CCAC is best overall re coverage and welfare
- No species-specific guidelines for fish handling, blood sampling, administration of substances or identification
  - ▶ But what do we mean by “species-specific”? Most commonly used species?
- Guidance on identifying and implementing best practice urgently needed
  - ▶ Based on scientific evidence where it exists - otherwise need to consolidate good practice, continually monitor and evaluate

# Fin



Photo: Chris Latham, [www.flickr.com](http://www.flickr.com)