# **Reference manual** Guidelines for otter spraint based diet analysis: searching, preservation, preparation and determination



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This manual is commissioned by:

Niewold, F.

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#### Abstract

This reference manual contains a simple standard method for finding and handling of otter (*Lutra lutra*) spraints for diet related research. The method describes, how to do spraint collection, how to preserve spraints, how to prepare spraints and how to determine to which species the prey remains belong. Also a description of the necessary materials is given. The reference manual also includes photographs of prey remains of several species that can be found in the otter spraints. The method is based on the analysis of 78 spraints from river and marsh habitats in the Netherlands. We anticipate that the manual will support and promote research on otter diet by amateurs interested in public science.

#### Preface

This reference manual is written as a thesis project of Hermsen, J. and Maarseveen, A., van for the BSc Animal Management, major Wildlife Management at the Van Hall Larenstein University of Applied Sciences in Leeuwarden. The project was supervised by Dr Strijkstra, A., and Mrs Heijer, M., den The research was commissioned by Drs. Niewold, F. J. J. of the Niewold Wildlife Infocentre and aimed for production of a realistic reference manual for prey remains in otter spraints, including a simple but effective standard method for the research on otter diet for amateur research in the Netherlands.

Leeuwarden, January 2012

Jessica Hermsen and Aloïse van Maarseveen

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# 1. Introduction

Not many diet studies on the Dutch otter (*Lutra lutra*) population have been done. Here for there is no standard method on how to do this research. The benefit of an easy standard method and reference manual may be that it promotes more research on the diet of this species by amateurs. This is the target group of this reference manual.

The method is based on information from literature study on otter diet studies, done in other countries, which was combined with expert opinions and our own practical experience on spraint analysis.

The manual explains the necessary steps for easy and efficient research on the otter's diet based on spraints.

It starts off with necessary preparation for fieldwork and the materials needed. Furthermore a guide on how to do the field work is given, followed by a full explanation of how to gather, store, prepare and analyse the spraints for prey remains, and how to examine prey remains to determine the species.

The manual includes reference photographs, which can be used to compare prey remains too. The photographs are all made on prey remains found in otter spraints from otter of river and marsh habitats on several Dutch locations, and from boiled fish.

# 2. Terminology

Some terms are used which could be interpreted in different ways. To prevent confusion, some of these terms are defined below.

Term	Correct interpretation
Reference	A manual that contains pictures, photographs and drawing of the identifiable parts
manual	in the spraints. Prey remain in following research can be compared with these
	images in the manual.
Spraint	A small faecal secretion from an otter which contains prey remains, to mark their
	territory.
Prey remains	The (mostly hard) indigestible parts in a spraint, such as bones, fish scales,
	jawbones, vertebrae, hair, feathers.
Determination	The identification of a prey part, to its species.
Detergent	Washing fluid with in this case a solution of Sodium Carbonate, the product that
	is used for cleaning up spraints.

Table 1; Terminology, the right way of interpretation of several terms used in this refere	nce manual.
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### 3. Standard method

This method is set up to make a study on the diet of the otter less time consuming.

#### **3.1 Material**

The following material is preferred in order to perform an optimal research (see table 2).

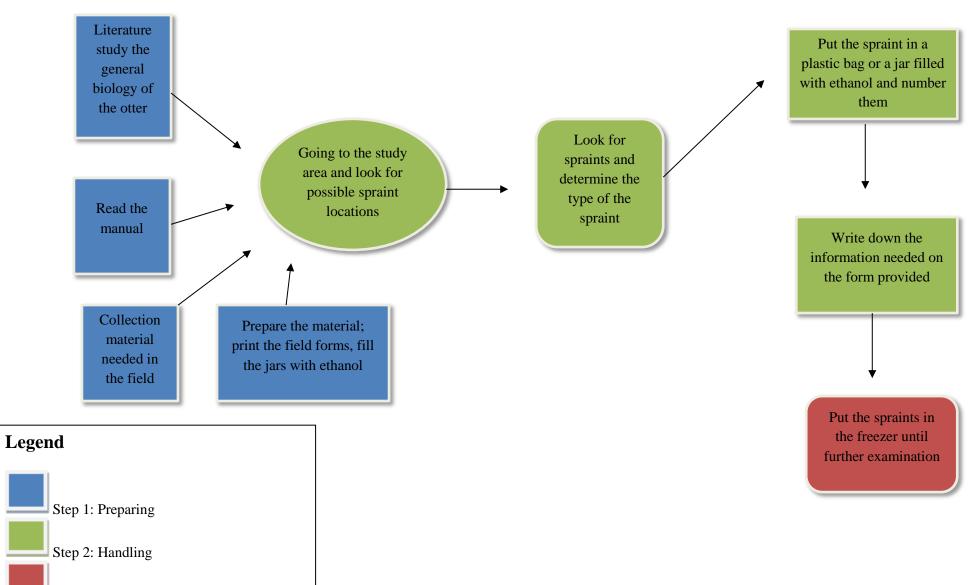
Table 2 Materials needed to perform this research	about the diet of the Eurasian otter
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Specific for this research	General materials
Sieve 0.6mm	Measuring cup and stirrer
Sodium carbonate Na <sub>2</sub> CO <sub>3</sub> , 0.07%	Cutting board
Tissues	Waterproof marker and pen
Petri- dishes	Computer and printer
Microscope, (used for this reference manual;	Drawing material; pencil
Olympus CHS)	
Microscope slides, cover slips and demineralised	Disposable gloves
water	
Measuring scale (with accuracy of 0.1 gram)	Beaker
Cooking plate	Knife
Binocular-camera (used for this reference	Stickers, that fit on the jars filled with ethanol
manual; Olympus SZ-CTV)	
Tweezers	White paper sheets; to empty the sieve with the
	prey remains on
Spraints	Freezer

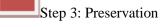
The determination books in table 3 were used during this research.

Table 3 Determination books needed to perform this research about the diet of the Eurasian otter in several study sites in the Netherlands; Wieden, Weerribben, Lindevallei, Rottige Meenthe, Doesburg, Giesbeek, and one in Germany; Peene.

Determination book; title	Author	ISBN
A guide to the identification of prey	Conroy, J.W.H. et al. 2005	0906282551
remains in otter spraint 3rd edition		
Atlas van schubben en andere	Steinmetz, B. & Muller, R.	9789080012042
beenachtige structuren van niet-	1991	
zalmachtige zoetwatervissen,		
Organisatie ter verbetering van de		
Binnenvisserij;		
The Tailless Batrachians of Europe	Boulenger, G.A. 1897	9780548217207
Voedselecologie van de 'verwilderde'	Honstede, B. van & Schut, G.	594000
kat	2010	



#### 3.2 Flow chart; Field work; The steps to follow when you are going to search for spraints in the field



#### Field manual

This field manual contains guidance about how to collect spraints.

#### **Preparation for a field trip**

Collect all the materials mentioned in table 4. Make sure the stickers will fit on the jars and the jars are pre-filled with ethanol.

Print and cut out the labels designed in appendix 1 according to the amount of spraints that you anticipate to collect. These labels contain all important information that needs to be written down during field work.

Print some extra forms, don't cut them; these will be used for the fresh spraints that are stored in jars with ethanol. You can number the jars with a pencil; these numbers refer to the ones you write down on your form.

Collect information on the research area and get a detailed map Google Earth, or 1:25000 map). Make an appointment with a specialist in this certain area to get information about the area and the otters that may live here.

Before starting check the weather conditions, this will influence the quality of the spraints. Rain; the spraints might get soak or flushed away. Cold weather close to freezing; DNA may be preserved for a longer time. Hot weather (20C+); the spraint will dry faster.

Table 4 Material list with the materials needed to perform this research about the diet of the Eurasian otter

Kind of material d of		
Clipboard		
Eraser		
GPS		
Pencil		
Plastic bags to collect the spraints		
Plastic jars (with screw caps) 150 ml		
Print paper (to make labels)		
Stickers to put on the jars		
Ethanol 100%, 70%		

#### Fieldwork

Otters have typical places to spraint, where the water is entered or exited and on conspicuous spots around passages where the otter often walks through. Places to look for spraints are indicated in table 5 and illustrated with photographs. You might also invite sprainting by creating a spot.

Table 5 Places to look for otter spraints in the field, with their explanation.

Place	Explanation
Otter passages (figure 1 and 2, appendix 2)	A small hole in the scrubs with a section of about
	30 cm
Otter path (figure 1, appendix 2)	A clear path, where the vegetation has often been
	walked on, comes out/ in these holes and goes to
	another place. Often from one water place to the
	other
Water edge	Look for spots at the water edge where the
	vegetation is flat (walked on) or brown/ yellow
	(urinated) or where there is an object available to
	spraint on, like a stone. Drove outs for game are a
	good spot to look.

Create a spot	Create a spot where otters often come, by offering
	them a tree trunk or another object

Look for traces of the otter at the place where the spraint was found (see figure 3). This will increase the determination reliability that the spraint originates from an otter.

Write down where the spraint was found (shade, sun, under a bridge, etc.) as this might have influenced the quality of the spraint and must be taken into account for further research.

There are different types of spraints, (table 6). The type needs to be determined during collection to know if the spraint contains DNA. Jellies contain most DNA, as a jelly is epithelia mucus from the intestine. Fresh spraints contain usable DNA as well.

Spraint	Figure	Explanation
Jelly	Figure 7 and 8, appendix 2	Mucus, yellow, white or green
Fresh spraint	Figure 6, appendix 2	Wet, strong smell, soft when you poke in it
		(with a stick)
Old spraint	Figure 5, appendix 2	Dry, smell is not so strong, hard

Table 6 Spraint types with figure numbers for examples, and their explanation.

When the origin of the spraint is unclear the smell may be helpful. Otter spraints have a strong fish smell. This combined with knowledge about the area, the otter and traces found at the location will determine if it safe to assume that it is an otter spraint.

Old spraints are collected in a plastic bag. The information on the label should be filled out as complete as possible and is put in second plastic bag together with plastic bag with the spraint This prevents the label from getting wet and unreadable. Make sure to use capable letters, this makes sure that everyone can read it. Process the data on a computer as soon as possible after the fieldtrip. Fresh spraints can be used for DNA research and when used for that potential purpose, need to be stored in 70% ethanol. Number the jars with the fresh spraints and link these to a paper or digital archive, i.e. a form or a date file.

#### **Finishing spraint collection**

The collected spraints need to be stored frozen at -20C until they are used for further examination.

Date:	Number:	Date:	Number:
Place:		Place:	
Coordinate:		Coordinate:	
Weather conditions:		Weather conditions:	
Info spraint location:		Info spraint location:	
Amount of spraints found at location:		Amount of spraints found at location:	
Info spraint:		Info spraint:	
Info otter:		Info otter:	
Date:	Number:	Date:	Number:
Date: Place:	Number:	Date: Place:	Number:
	Number:		Number:
Place:	Number:	Place:	Number:
Place: Coordinate:	Number:	Place: Coordinate:	Number:
Place: Coordinate: Weather conditions:	Number:	Place: Coordinate: Weather conditions:	Number:
Place: Coordinate: Weather conditions: Info spraint location:	Number:	Place: Coordinate: Weather conditions: Info spraint location:	Number:
Place: Coordinate: Weather conditions: Info spraint location: Amount of spraints found at location:	Number:	Place: Coordinate: Weather conditions: Info spraint location: Amount of spraints found at location:	Number:



Figure 1 Passage and path of the otter (photographed at Wieden/Weerribben on 22 November 2011)



Figure 2 Otters passage (photographed at Wieden/Weerribben on 22 November 2011)



Figure 3 Scratch traces of an otter (photographed at Wieden/Weerribben on 22 November 2011)



Figure 4 Otter spraint (photographed at Wieden/Weerribben on 22 November 2011)



Figure 5 Otter spraint (photographed at Wieden/Weerribben on 22 November 2011)



Figure 6 Otter spraint (photographed at Wieden/Weerribben on 22 November 2011)



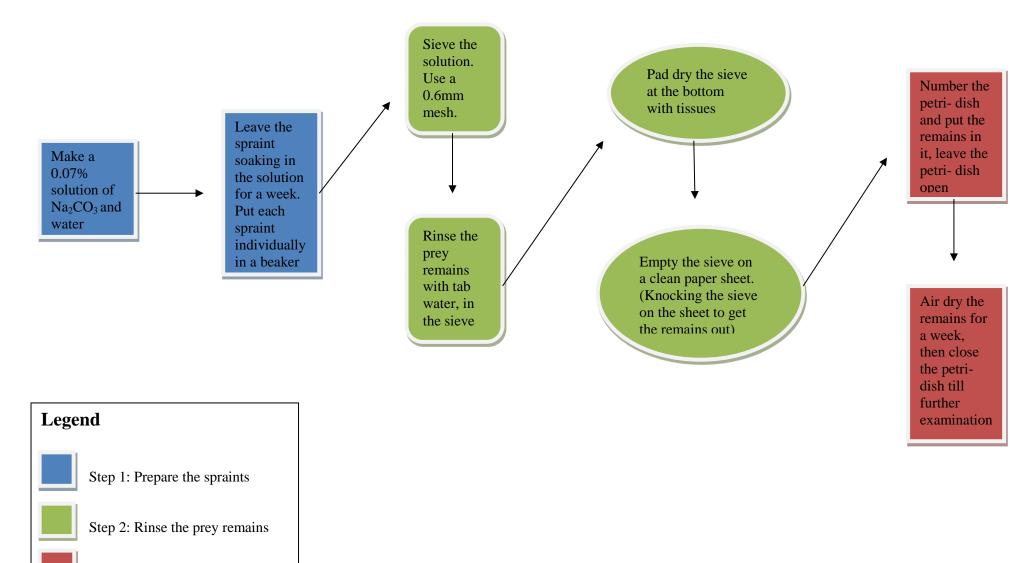
Figure 7 Otter spraint, jelly (photographed at Wieden/Weerribben on 22 November 2011)



Figure 8 Otter spraint, jelly, photographed by Bosma, H. (2011)

# **3.3 Spraint preservation**

The spraints are collected in plastic bags with tags about the study area and deep frozen at -20 degrees Celsius until examination for subsequent analysis (Colares, E., *et al.* 2000, Georgiev, D., 2004, de la Hey, D., 2008 & McMahon, J., *et al.* 2006).

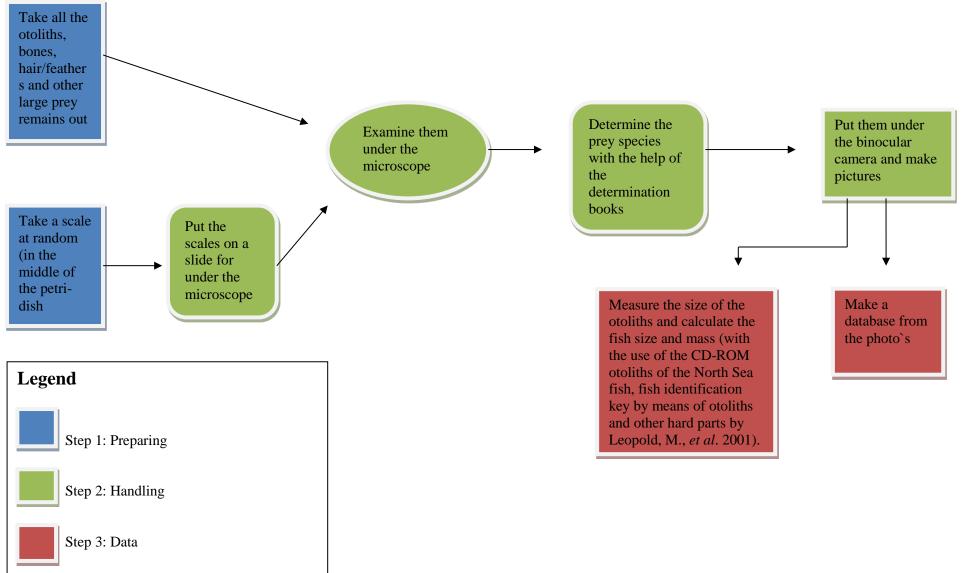


Step 3: Dry the prey remains

3.4 Flow chart; spraint preparation; the steps to follow when you clean the spraints (remove waste and mucus from the prey remains)

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#### 3.5 Flow chart; determination; the steps to follow when you want to determine the species of the prey remains



#### 4. Analysis

Determination material for the European perch, Bream, Silver bream, Pike, Tench, Rudd, Common roach, Common carp, Gudgeon, Round goby and the Kessler's goby is given. These species were encouraged after the analysis of 78 spraints of otters in Dutch waters.

#### 4.1 European perch

 Table 7 Determination material for the European perch (Perca fluvialtilis)

Scientific name	Perca fluvialtilis
Exterior	Figure 9 (Sportvisserij Nederland, 2012)
Scale	Figure 10
Otolith	Figure 11
Spine	Figure 12



Figure 9 *Perca fluvialtilis* (Sportvisserij, 2012); 1; The Perch has two separated dorsal fins of which only the first one has hard spines. 2; The Perch has a back spot on the back-side of the first dorsal fin. 3; The Perch has vertical dark ties across the body.



Figure 10 *Perca fluvialtilis* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4



Figure 11 *Perca fluvialtilis* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4



Figure 12 *Perca fluvialtilis* spine, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67

#### 4.2 Bream

 Table 8 Determination material for the Bream (Abramis brama)

Scientific name	Abramis brama
Exterior	Figure 13 (Sportvisserij Nederland, 2012)
Scale	Figure 14
Otolith	Figure 15
Jaw	Figure 16
Spine	Figure 17



Figure 13 *Abramis brama* (Sportvisserij, 2012); 1; Number of rows of scales above the side-line, counted from diagonal pointing arrow to the dorsal fin, contains 12-14 scales (excluded the one from the side-line itself). 2; The eye-diameter is smaller than the distance from the eye till the point of its mouth. 3; The mouth is pointing downwards and is far bulging.



Figure 14 *Abramis brama* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 15 *Abramis brama* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 16 *Abramis brama*, jaw, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.

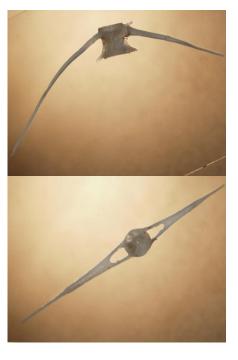


Figure 17 *Abramis brama* spine, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67. Upper photo; side view, bottom photo; front view.

#### 4.3 Silver bream

 Table 9 Determination material for the Silver bream (Blicca bjoerkna)

Scientific name	Blicca bjoerkna
Exterior	Figure 18 (Sportvisserij Nederland, 2012)
Scale	Figure 19
Otolith	Figure 20
Spine	Figure 21
Jaw	Figure 22

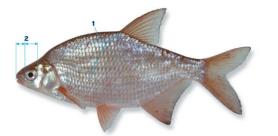


Figure 18 *Blicca bjoerkna* (Sportvisserij Nederland, 2012); 1; Number of rows of scales above the sideline, counted from the oblique pointed arrow to the dorsal fin, counts 8-10 (excluded is the scale on the side-line). 2; The eye-diameter is larger than the distance from the eye to the point of the mouth.



Figure 19 *Blicca bjoerkna* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 20 *Blicca bjoerkna* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 21 *Blicca bjoerkna* spine, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.



Figure 22, *Blicca bjoerkna* jaw, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.

#### 4.4 Pike

 Table 10 Determination material for the Pike (Esox lucius)

Scientific name	Esox lucius
Exterior	Figure 23 (Sportvisserij Nederland, 2012)
Scale	Figure 24
Otolith	Figure 25
Spine	Figure 26
Jaw	Figure 27

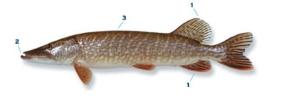


Figure 23 *Esox lucius* (Sportvisserij Nederland, 2012); 1; Anal fin and dorsal fin occurs far backwards on the body. 2; The head runs out into a flat, wide mouth. 3; All over the body there are gold-colored dots and stripes.



Figure 24 *Esox lucius* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 25 *Esox lucius* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 26 *Esox lucius* spine, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.



Figure 27 *Esox lucius* jaw, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.

#### 4.5 Tench

 Table 11 Determination material for the Tench (*Tinca tinca*)

Scientific name	Tinca tinca
Exterior	Figure 28 (Sportvisserij Nederland, 2012)
Scale	Figure 29
Otolith	Figure 30
Spine	Figure 31



Figure 28 *Tinca tinca* (Sportvisserij Nederland, 2012); 1; The iris is orange-colored. 2; The fins (point out is the dorsal fin) have a convex-shape. 3; There are two little mouth-wires present.



Figure 29 *Tinca tinca* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 30 *Tinca tinca* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 31 *Tinca tinca* spine, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.

#### 4.6 Rudd

Table 12 Determination material for the Rudd	(Scardinius	erythrophthalmus)
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Scientific name	Scardinius erythrophthalmus
Exterior	Figure 32 (Sportvisserij Nederland, 2012)
Scale	Figure 33
Otolith	Figure 34
Spine	Figure 35
Jaw	Figure 36

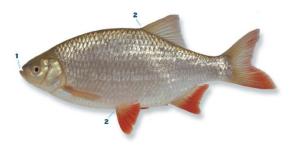


Figure 32 *Scardinius erythrophthalmus* (Sportvisserij Nederland, 2012); 1; The mouth is pointing upwards. 2; The front-side of the dorsal fin is clearly further to the tail of the fish than the pelvic fins.



Figure 33 *Scardinius erythrophthalmus* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 34 *Scardinius erythrophthalmus* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 36 *Scardinius erythrophthalmus* photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.



Figure 35 *Scardinius erythrophthalmus* spine, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 0.67.

#### 4.7 Common roach

Table 13 Determination material for the Common roach (Rutilus rutilus)

Scientific name	Rutilus rutilus
Exterior	Figure 37(Sportvisserij Nederland, 2012)
Scale	Figure 38
Otolith	Figure 39

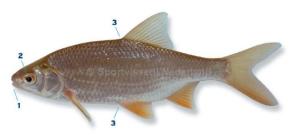


Figure 37 *Rutilus rutilus* (Sportvisserij Nederland, 2012); 1; The mouth is pointing forwards. 2; In the top of the eye, a red spot is shown. 3; The front-side of the dorsal fin is just as far from the tail as the pelvic fins.



Figure 38 *Rutilus rutilus* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 39 *Rutilus rutilus* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.

#### 4.8 Common carp

 Table 14 Determination material for the Common carp (Cyprinus carpio)

Scientific name	Cyprinus carpio
Exterior	Figure 40 (Sportvisserij Nederland, 2012)
Scale	Figure 41
Otolith	Figure 42



Figure 40 *Cyprinus carpio* (Sportvisserij Nederland, 2012); 1; There are four mouth-wires present, from which two in de corners of the mouth and two shorter ones on top of the upper lip. 2; The edge of the large dorsal fin is hollow incised. 3; The first fin rays of the dorsal fin is firm serrated.



Figure 41 *Cyprinus carpio* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 42 *Cyprinus carpio* otolith, photographed with a binocularcamera (Olympus SZ-CTV) with an enlargement of 1.4.

### 4.9 Gudgeon

Table 15 Determination material for the Gudgeon (Gobio gobio)

Scientific name	Gobio gobio
Exterior	Figure 43 (Sportvisserij Nederland, 2012)
Scale	Figure 44
Otolith	Figure 45



Figure 43 *Gobio gobio gobio* (Sportvisserij Nederland, 2012); 1; The mouth is pointing downwards. 2; There are two mouth-wires present, one in each corner of the mouth.



Figure 44 *Gobio gobio gobio* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.



Figure 45 *Gobio gobio gobio* otolith, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.

#### 4.10 Round goby

 Table 16 Determination material for the Round goby (Neogobius melanostomus)

Scientific name	Neogobius melanostomus
Exterior	Figure 46 (Sportvisserij Nederland, 2012)
Scale	Figure 47



Figure 46 *Neogobius melanostomus* (Sportvisserij Nederland, 2012) 1; The eyes are highly placed into the head. 2; The pelvic fins are fused to a suction-plate, with which the fish can suck itself on a stone-based underground. 3; This species is yellow-grey-colored and has a checker-pattern. 4; In the back of the first dorsal fin, a clear black spot is shown. Males are black of colour during mating season.



Figure 47 *Neogobius melanostomus* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.

# 4.11 Kessler's goby

Tuble 17 Determination material for the Resider's goby (100gobias Resident)	
Scientific name	Neogobius kesslerii
Exterior	Figure 48 (Sportvisserij Nederland, 2012)
Scale	Figure 49

 Table 17 Determination material for the Kessler's goby (Neogobius kesslerii)



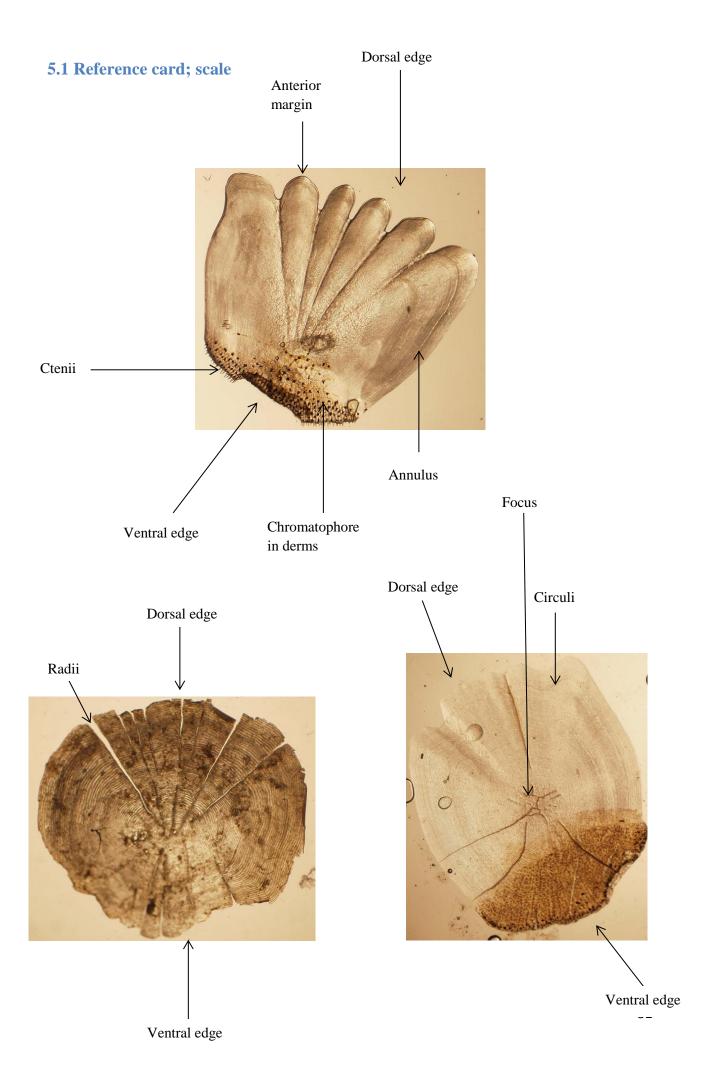
Figure 48 *Neogobius kesslerii* (Sportvisserij Nederland, 2012); 1; The eyes are close together and highly placed into the head. A big, wide head with swollen cheeks and lips, and a mouth pointing upwards. A neck with scales. 2; Pelvic fins are fused with which the fish can suck itself on a hard-based underground. 3; The body and the head are red-brown marbled. 4; The bases of both dorsal fins touch each other. Dorsal fins with horizontal red-brown ties on a lighter underground and without a black spot.

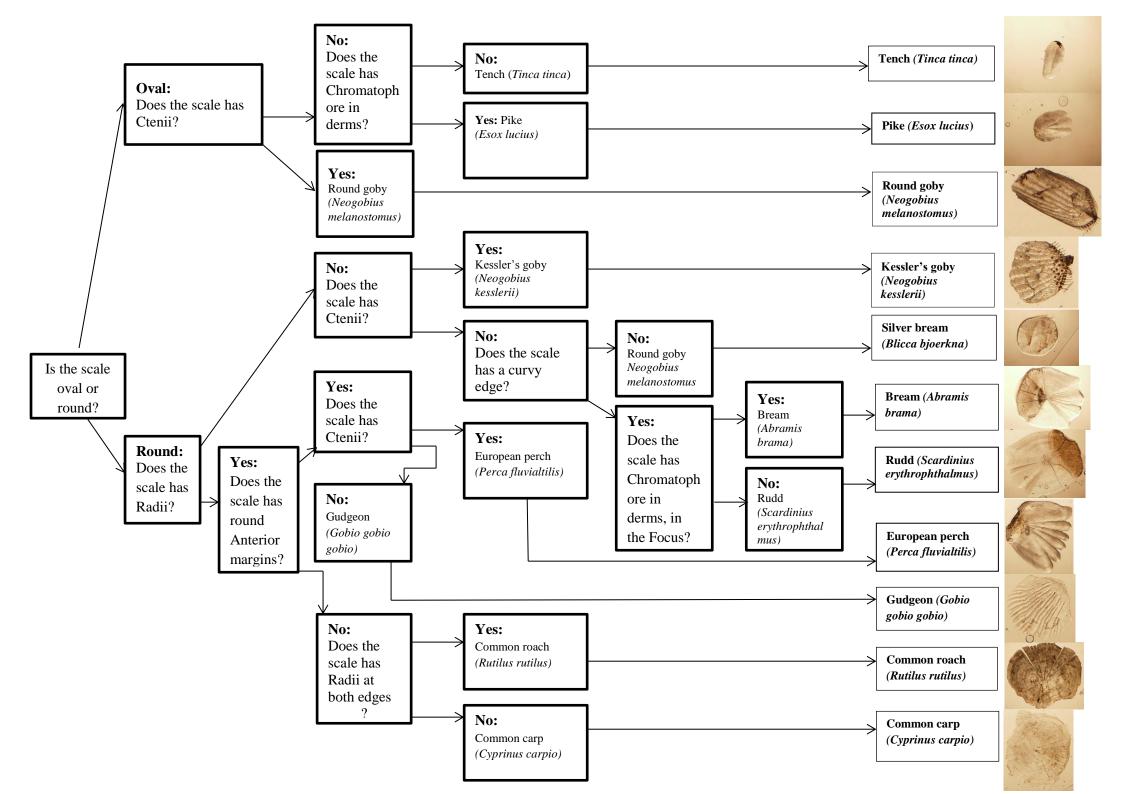


Figure 49 *Neogobius kesslerii* scale, photographed with a binocular-camera (Olympus SZ-CTV) with an enlargement of 1.4.

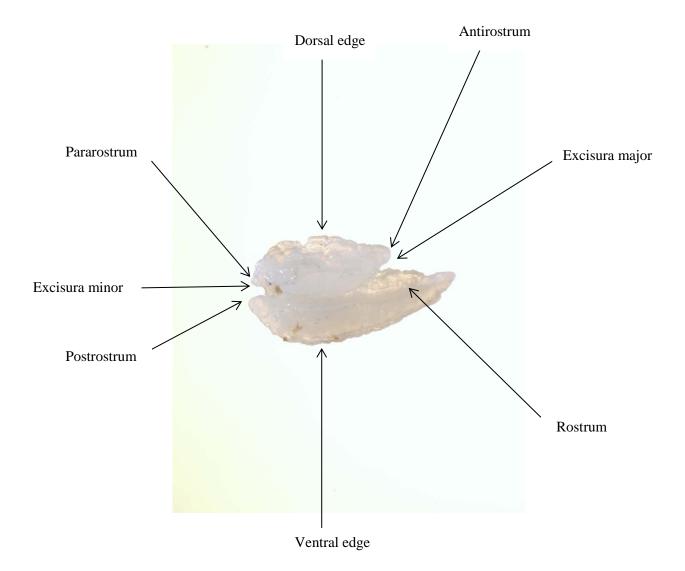
# **5. Reference cards**

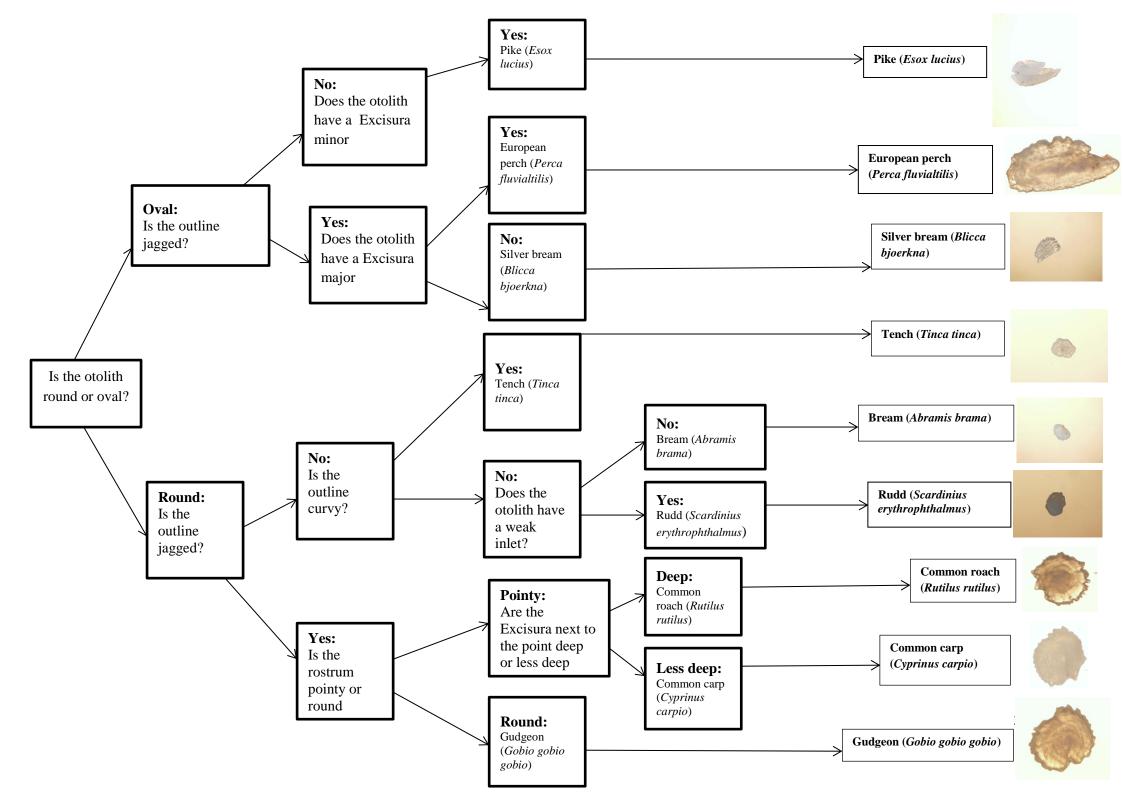
The following cards are developed to help determine the species from an otolith or scale found. By answering the questions you will find the species the scale or otolith belongs to.





# 5.2 Reference card; otolith





### Acknowledgements

For making of this reference manual we would like to thank Stijkstra, A. and Heijer, M., den for their guidance. We would like to thank Kijpers, H. and Ende, M., van den for helping us and sharing their knowledge and skills.

We would like to give out a special thanks to Bosma, H. and Leopold, M. for answering our questions which contributed to this manual.

The making of this reference manual is carried out at the van Hall Larenstein University of applied science, Leeuwarden. This University provided space and all the material needed for the research that needed to be done in advance in order to get the information and material needed to make a reference manual.

This was an assignment in order of the Niewold Wildlife Infocentre, we would like to thank Niewold, F. for giving us the opportunity of doing this, and for sharing information which contributed to the results of the research and this manual.

## References

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Georgiev, D.G. (2004) Diet of the otter (*Lutra lutra*) in different habitats of South-Eastern Bulgaria, IUCN otter specialist group bull. 23 (1): 5-11

Hey, D.C. de la (2008) The importance of birds in the diet of otter (*Lutra lutra*) on Shapwick Heath, Bioscience Horizons; volume 1, number 2; pp. 144, 145

McMahon, J. & McCafferty D. J. (2006) Distribution and diet of otters (*Lutra lutra*) in marine areas of Loch Lomond and The Trossachs National Park, Scotland, UK, McMahon & McCafferty / Lutra 2006 49 (1): 29-36; pp. 30

#### **CD-ROM;**

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