#### Version 1.1

Introduction	RATIONALE	PREPAREDNESS	BIOLOGICAL ADVICE	IMPACT ASSESSMENT	LIBRARY	WEB LINKS	TECHNICAL DOCUMENTS	SHOPPING LISTS
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# HANDBOOK ON OIL IMPACT ASSESSMENT

# 4.0 SPILL RESPONSE

# 4.1 Assessing the damage Technical document

# Ageing and sexing manual for stranded seabirds

This technical document provides guidelines for ageing seabirds during autopsies. For external characteristics of sex and age, do consult the specific technical documents for each of the seabird families or general field guides.

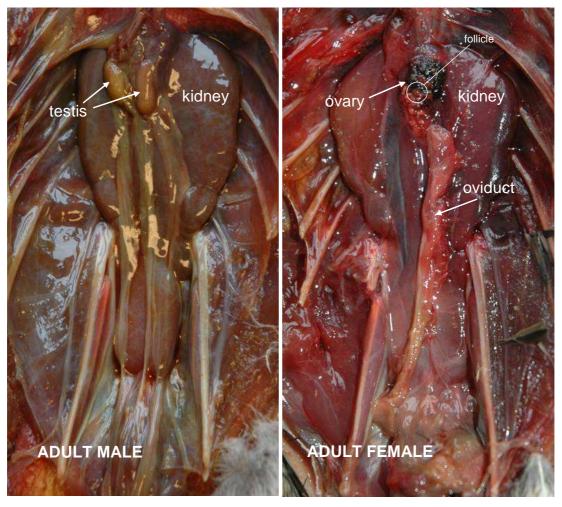
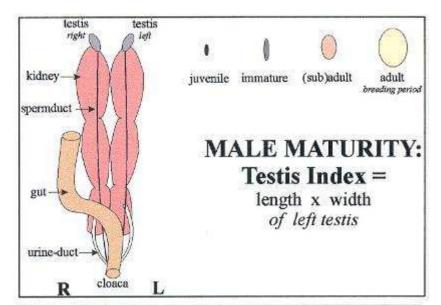


Figure 1. Urogenital system of (a) adult male and (b) adult female Common Guillemot *Uria aalge* in the non-breeding season. In males, length and width (0.1mm) of the largest (left) testis is measured. For orientation, while looking into the abdominal cavity, the left testis is seen on the right side! In females, the diameter of the largest visible follicle is measured, while the shape and development of the oviduct is described (Fig. 3).

**Sexing birds** Male birds have paired testes within the abdominal cavity, anterior and ventral to the lobes of the kidneys (Fig. 1a), while the reproductive system in female birds is reduced to a left ovary and oviduct, just anterior and medial to the left kidney (Fig. 1b). For orientation, while looking into the abdominal cavity during a standard autopsy, the left testis is seen on the right side. Similarly, ovary and oviduct are seen on the right.



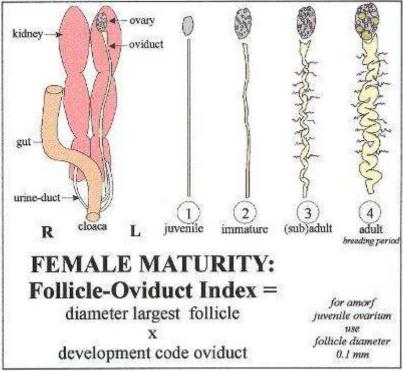


Figure 2. Testis index suggested by Van Franeker (2004) based on Northern Fulmar Fulmarus glacialis dissections.

Juveniles have an often dark, short and thin testis, mature birds have rounded testis, considerably swollen in the breeding season.

In other seabirds, testis shapes may be slightly different, but the general plan of development is the same as in the illustrated case. In guillemots, for example, juvenile testis are rather long and thin (often 12x0.5 mm or similar), often bicoloured, and sometimes with a clear nick in the top. As in fulmars, the adult male preparing for breeding will develop a white-bean type testis as illustrated.

Figure 3. Follicle-Oviduct index suggested by Van Franeker (2004) based on Northern Fulmar dissections.

The ovary of juveniles has no structure (amorf). Small grains (tiny follicles) are visible in young immature birds, whereas older birds develop clearly visible follicles of different sizes. All the examples drawn here are outside the breeding season, or just prior to nesting. During egg-laying, some follicles will greatly increase in size (finally reaching the actual egg size), and the oviduct is greatly enlarged.

Oviduct development four-point scale:

- 1. thin and straight
- 2. thicker, straight
- 3. thicker still, slightly twisted
- 4. swollen and twisted

**Male birds** The (paired) testis develop with age, but there is seasonal growth as well. It is impossible to age a seabird with certainty solely on the basis of testis size, but a testis index (length x width; Fig. 2) is surely indicative of age. Testis size and exact shape are species specific, but the general plan of growth as illustrated in Fig. 2 holds for all species. Sausage-shaped, yellowish or whitish testis (Fig. 1a) are typical for adult males in the non-breeding season; a (white) bean shape is often seen immediately prior to or during breeding. Juvenile testis are thin (and can be long), often dark or bi-coloured. Juvenile testis can be very difficult to spot, particularly in smaller species. When in doubt between an ovary and a testis, note that testis are paired, even although the other one may be hidden in membrane, blood or other liquids. So, blot dry and thoroughly search the abdominal cavity for a second one.

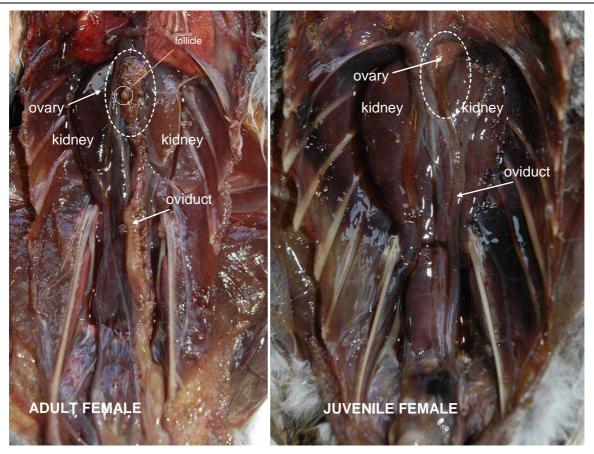


Figure 4. Urogenital system of (a) adult female Common Guillemot and (b) juvenile female Razorbill *Alca torda*. In older females, the diameter of the largest visible follicle is measured, while the shape and development of the oviduct is described as straight, swollen and straight, twisted, or swollen and twisted (Fig. 3). In juveniles, the amorf ovary is often difficult to detect, can be simply described as 'not structured' (i.e. no visible follicles), while the oviduct is thin and straight and very hard to distinguish from the ureters running from each of the kidneys to the cloaca.

**Female birds** In developing or adult females, the normally yellowish coloured ovary resembles a bunch of grapes with its many round follicles (Fig. 1b, 4a). The tubular structure running posteriorly from ovary to cloaca is the oviduct, which begins next to the ovary in a funnel-shaped structure called the *ostium*. The follicles differ in size and usually there are a few large (mature) follicles visible, next to a large number of very small follicles. We suggest to measure the diameter of the largest follicle seen during the autopsy, to evaluate the development of the oviduct on a four-point scale (thin and straight (1), slightly swollen and straight (2), slightly swollen and slightly twisted (3), swollen and twisted (4)), and to calculate a 'Follicle-Oviduct index' as suggested by Van Franeker (2004; Fig. 3). In juvenile birds, there will not be visible follicles, the oviduct is typically thin and straight (1). The ovary development will simply be recorded as 'not structured' in these cases, use 0.1 as a score to calculate the 'Follicle-Oviduct index' (cf. Van Franeker 2004).

**Bursa Fabricii** While the gonadal development gives an idea of the age of the bird, more foothold can be obtained by assessing the presence or absence of the bursa Fabricii, one of the glands in the endocrine system of birds. The bursa Fabricii (or Bursa of Fabricius) is visible as a pouch on the outer wall of the cloaca, and it is only found in very young birds, while it atrophies in sub-adults and is absent in mature birds. From frequent comparisons of the presence/absence and the size of the bursa with known age (ringing results) of individual birds, we are certain that all juvenile seabirds have a large bursa, young immatures (2<sup>nd</sup> year) may have a much reduced bursa, while old immatures (>3<sup>rd</sup> year) and adults normally have not a trace of a bursa. Hence, the presence of a large bursa in combination with non-developed gonads is a fine indication that the bird in question is a juvenile.

The bursa is involved with forming and stimulating cells of the immune system and it is assumed to secrete hormones that stimulate the production of antibodies to infections and the production of lymphocytes (a type of white blood cell) (Proctor & Lynch 1993).

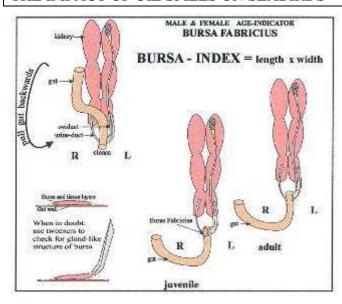


Figure 5. Bursa index suggested by Van Franeker (2004) based on Northern Fulmar *Fulmarus glacialis* dissections.

The bursa is most easily found when the large intestine (gut) is folded down during a standard autopsy, to inspect the region closest to the cloaca, and between the two ureters. In juvenile birds, the bursa is so large, that folding down the gut is not needed (Fig 6a), even although the presence will become more clear (Fig. 6b). To measure the size, however, the other organs should be moved' out of the way'. In immature birds, small bursa's might be easily overlooked if the gut is not folded down.

Note that in some species the bursa does not stand out as a clear pouch, because a strong membrane holds it tight against the outer wall of the cloaca. There is usually a colour difference between bursa and intestine, and tweezers may have to be used in case of doubt.

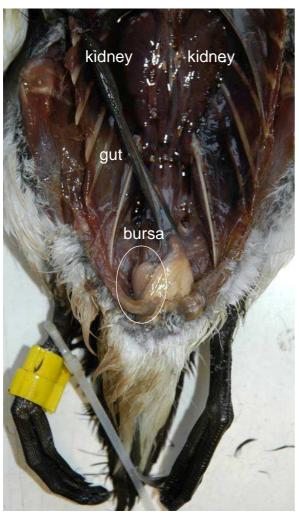




Figure 6. Urogenital system of juvenile female Razorbill *Alca torda* showing the position of the bursa Fabricii, (a) just visible slightly to the side of the cloacal exit of the large intestine, and (b) more clearly visible with the gut folded down. Note that the whitish, swollen appearance of the intestine is caused by excrements filling up the organ. This swelling should not be confused with a bursa (see also Fig. 7).

The bursa is shown in Figs 6 and 7. Note that assessing the presence is more easy than assessing the absence of this organ. Small bursa's may be hard to find and the large intestine must be manipulated such that the absence can be seen very clearly, such as in Fig. 7a. Length (base to top) and width of the bursa is measured (0.5 mm) without squeezing or stretching the organ.

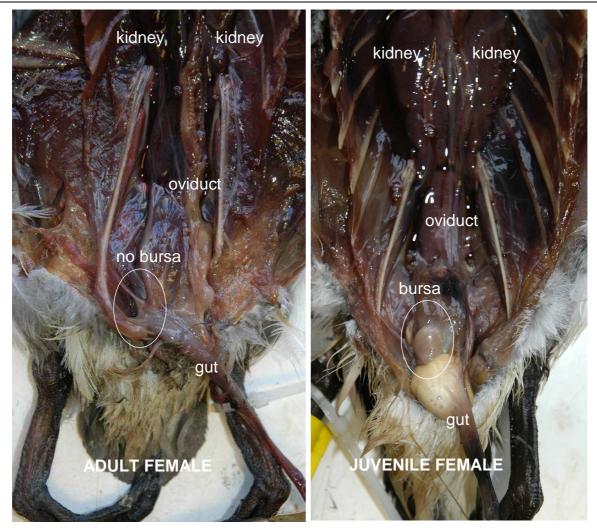


Figure 7. Urogenital system of (a) adult female Common Guillemot and (b) juvenile female Razorbill showing the position of the bursa Fabricii. The absence of a bursa can only be properly checked by folding down the large intestine. The hockeystick-shape structure in Fig, 7a is a rib, while one of the ureters is clearly visible. Any (tiny) bursa present should be visible as a pouch. Note the difference between an empty gut and the swollen appearance of a cloaca filled with excrements, just below the clearly visible bursa in the young Razorbill (Fig. 7b).

**Logging data** Autopsy forms supplied with this handbook were designed such that all data can be logged easily (Fig. 8). For testis in male birds, length and width can be entered, or a brief description (measurements are preferred), for females, the development of the oviduct can be described scoring 1, 2, 3, or 4 as described earlier, while an entry for the diameter of the largest follicle can be made if the ovary is indeed structured. And finally, the bursa can be coded as 'present' (+), 'absent' (-) or 'unknown' (?), plus an entry of the organ size or a brief description (again, measurements are preferred). Note that it is important to log bursa absence specifically, don't simply leave the box blank in case you checked but didn't locate the organ! Checking and confirming absence is different from *not* checking for the presence absence of the organ, and in later analysis there should not be any doubt.

SEX AND AGE:	d testis:	×	mm	descr.: long	ort round //				
gonadal development:	♀ oviduct:	1	2	3 4	foll.	max:	mm ø / not structured (juv)		
bursa Fabricii:	+	-	?	size:	×	mm	descr.: large / mod. / small		

Figure 8. Detail of proposed autopsy form where data on gonadal development and presence / absence of the bursa Fabricii can be entered according to the ageing and sexing protocol in this manual. All autopsy forms associated with this handbook will have these or very similar boxes included. Accurate measurements are preferred over descriptions such as 'long thin' and 'large', 'moderate', and 'small', also because organ size is species specific.

Checking data entries During a routine autopsy, information on the age of birds may be derived from external characteristics (e.g. Northern Gannet *Morus bassanus*, gulls Laridae, auks), internal characteristics (e.g. petrels, shearwaters and storm-petrels), or a combination of both. Heavily polluted seabirds may have no external (plumage) characteristics available for inspection, or an incomplete subset only. When working on a bird, it is useful to check and cross-check entries *during* the actual work, to avoid surprises afterwards. Mistakes are easily made, age-characteristics are variable, and the size of flexible organs is nothing to fully rely on. When a bird shows clear juvenile plumage characteristics, while a bursa cannot be found, do check again (both). If true, do highlight an extraordinary entry on the datasheet with an exclamation mark. Similarly, fully developed ovaries, or bean shape testis but also a large bursa (or the reverse, juvenile-looking gonads but no bursa found): step back, check again, something may have gone wrong. Finally, and very importantly, the system needs further validation. *Any* ringed bird should be very carefully checked according to this protocol and the outcome should be compared with the ringing data. The authors would be very grateful if the outcome of such checks could be reported (contact address below).

**Expected size range** Judging the size of a flexible organ like a bursa Fabricii by eye is hard and one has to realise that a large bursa for a juvenile Little Auk *Alle alle* would be a very small bursa for a Common Guillemot *Uria aalge* (Fig. 9). Table 1 offers some guidance while listing measurements of a selection of immature and juvenile seabirds collected in winter. Camphuysen (1987) reported that slightly decomposed, severely oiled seabirds collected after a couple of days baking in the sun had swollen organs (intestines, gonads and bursa), making exact measurements pointless. Be aware of decomposition effects: fluffy organs cannot be measured. A bursa Fabricii in good condition is a fairly stiff pouch, flexible, but certainly not fluffy.

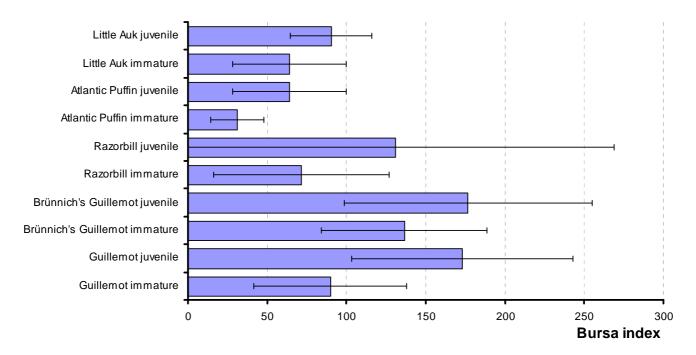


Figure 9. Bursa index (±SD) in immature and juvenile Atlantic Alcidae based on measurements at stranded, wintering seabirds (Nov-Mar) in the Southern North Sea and in Newfoundland (CJ Camphuysen unpubl. data). Note that the age (I for immature, J for juvenile) is based on interpretations during dissections following the standard ageing protocol described in this manual. Only a fraction of these birds could be confirmed with true age from ringing data!

Table 2 will offer some guidance by listing testis size of seabirds wintering in de southern North Sea. In all reported cases, only the left testis was measured (visible on the right side during the standard autopsy). Follicle size is so variable, that there is little point in producing a list of measured examples.

Testis size varies between species and is roughly correlated with structural body size. Outside the breeding season, testis indices in auks vary between 6 (*Alle alle*) and 40 (*Uria aalge*), representing testis length variations between 5.5 and 12mm, and testis widt of less than 1 and 3.6mm. Some February or March auks may prepare for breeding, and a testis index of 300 (30x10) can be found in a large auks like the Common Guillemot. Non-breeding testis indices seldom exceed 100, even in large species like Northern Gannets *Morus bassanus*. The bean shape is typical for all studied seabirds preparing for breeding, and is typical for mature individuals. Since adult males are difficult to evaluate in terms of age, do cross-check for size and presence of bursa Fabricii!

Table 1. Size indications of bursa Fabricii based on measurements at stranded, wintering seabirds (Nov-Mar) in the Southern North Sea (CJ Camphuysen unpubl. data; cormorant data courtesy MF Leopold). Note that the age (I for immature, J for juvenile) is based on interpretations during dissections following the standard ageing protocol described in this manual. Only a fraction of these birds could be confirmed with true age from ringing data!

Species		Age	Length	SD	Min	Max	Width	SD	Min	Max	Index	SD	Sample
Great Northern Diver	Gavia immer	I	15.0		15	15	10.0		10	10	150.0		1
Little Grebe	Tachybaptus ruficollis	J	10.0		10	10	5.0		5	5	50.0		1
Great Crested Grebe	Podiceps cristatus	J	15.1	3.4	11	21	9.0	3.7	5	17	142.6	84.1	8
Slavonian Grebe	Podiceps auritus	J	5.0		5	5	4.0		4	4	20.0		1
Northern Fulmar	Fulmarus glacialis	J	15.4	4.2	10	21	10.9	2.5	7	15	169.9	68.0	11
Northern Gannet	Morus bassanus	1	12.0		12	12	8.0		8	8	96.0		1
Great Cormorant	Phalacrocorax carbo sinensis	J	28.6	7.1	20	42	13.3	3.7	7	21	383.9	147.8	9
European Shag	Phalacrocorax aristotelis	1	14.5		13	16	7.5		7	8	108.8		2
		J	18.0		18	18	9.0		9	9	162.0		1
Common Eider	Somateria mollissima	1	20.0		20	20	10.5		10	11	210.0		2
		J	16.0		16	16	6.0		6	6	96.0		1
Common Scoter	Melanitta nigra	1	22.0		22	22	9.0		9	9	198.0		1
		J	24.2	5.2	19	32	10.0	4.6	7	18	255.6	180.0	5
Pomarine Skua	Stercorarius pomarinus	1	6.5		5	8	5.0		5	5	32.5		2
		J	15.7	3.2	12	22	8.0	1.5	7	11	128.6	51.7	7
Great Skua	Stercorarius skua	J	23.0		18	28	12.0		9	15	276.0		2
Black-headed Gull	Larus ridibundus	J	10.0		10	10	10.0		10	10	100.0		1
Common Gull	Larus canus	1	5.0		5	5	6.0		6	6	30.0		1
Iceland Gull	Larus glaucoides	J	14.0		14	14	6.0		6	6	84.0		1
Glaucous Gull	Larus hyperboreus	1	9.0		9	9	6.0		6	6	54.0		1
Great Black-backed Gull	Larus marinus	J	12.7		11	15	10.3		7	15	130.9		3
Black-legged Kittiwake	Rissa tridactyla	1	9.0	2.6	7	12	6.0	2.0	4	8	56.7	34.4	3
		J	10.6	2.4	7	15	6.3	1.2	4	9	66.8	17.9	18
Common Guillemot	Uria aalge	1	12.4	3.8	4	20	6.8	2.6	1	16	89.8	48.3	57
		J	17.9	3.6	10	31	9.4	2.1	5	65	172.9	69.9	97
Brünnich's Guillemot	Uria Iomvia	1	16.5	3.7	10	20	8.0	1.6	6	10	136.5	52.1	8
		J	19.4	2.2	15	25	9.0	3.3	1	15	176.8	78.2	21
Razorbill	Alca torda	I	10.7	3.7	3	26	6.1	2.2	2	16	71.7	55.4	66
		J	14.5	3.0	10	22	8.8	8.9	3	69	130.8	138.0	50
Little Auk	Alle alle	I	7.0	2.3	4	9	4.2	1.3	3	6	31.2	16.8	5
		J	10.9	9.9	6	75	6.3	3.1	3	25	64.1	35.8	45
Atlantic Puffin	Fratercula arctica	I	9.6	2.3	7	13	4.8	0.4	4	5	64.1	35.8	5
		J	12.7	2.8	6	18	7.0	1.1	4	9	90.3	25.7	27

Table 2. Size indications (L= length, W = width, Index = LxW) of left testis based on measurements in stranded, wintering seabirds (Nov-Mar) in the Southern North Sea (CJ Camphuysen unpubl. data; cormorant data courtesy MF Leopold). Note that the age (A for adult, I for immature, J for juvenile) is based on interpretations during dissections following the standard ageing protocol described in this manual. Only a fraction of these birds could be confirmed with true age from ringing data!

Species		Lft	Mean L	SD	Min	Max	Mean W	SD	Min	Max	Index	SD	Sample
Red-throated Diver	Gavia stellata	Α	11.8	2.2	9.0	14.0	4.6	1.0	3.8	6.0	52.2	3.4	4
Great Crested Grebe	Podiceps cristatus	Α	12.8	3.4	6.0	19.0	3.8	1.2	1.2	6.0	50.0	21.7	15
		J	11.8	1.8	9.0	14.0	3.1	0.8	2.0	4.0	36.5	8.9	9
Northern Fulmar	Fulmarus glacialis	Α	7.2	1.6	6.0	9.0	3.0	1.0	2.0	4.0	21.7	8.4	3
		J	4.6	0.4	4.0	5.0	1.5	0.7	0.5	2.5	6.9	3.6	5
Northern Gannet	Sula bassana	Α	12.0	0.2	12.0	12.0	8.0	1.5	6.5	9.5	96.0	19.0	3
Great Cormorant	Phalacrocorax carbo sinensis	J	22.8	3.6	19.4	27.3	3.9	0.7	3.3	5.0	88.6	19.5	5
Common Eider	Somat eria mollissima	Α	12.0	2.0	8.0	16.4	5.1	1.1	3.0	8.1	62.6	18.9	65
		1	10.7	1.8	7.1	14.4	3.9	1.3	1.8	7.2	43.1	18.8	50
		J	9.4	1.6	3.0	13.0	2.6	1.1	1.0	9.0	23.7	8.2	122
Pomarine Skua	Stercorarius pomarinus	J	6.5	0.5	6.0	7.0	1.3	0.3	1.0	1.5	8.3	2.3	3
Black-legged Kittiwake	Rissa tridactyla	Α	5.7	1.1	4.5	7.0	2.3	0.5	1.8	3.0	13.3	5.6	4
		1	5.0	1.0	4.0	6.0	1.5	0.9	0.5	2.0	7.5	4.8	3
		J	4.1	1.7	3.0	7.0	1.1	0.2	1.0	1.5	4.7	2.2	5
Common Guillemot	Uria aalge	Α	11.6	2.3	2.8	30.0	3.6	1.3	1.0	10.0	41.6	17.2	315
		1	10.6	1.8	6.0	14.0	1.9	1.0	0.5	7.0	20.6	12.0	94
		J	10.6	2.2	6.0	17.0	1.5	1.0	0.5	8.0	15.8	12.4	131
Brünnich's Guillemot	Uria lomvia	Α	10.8	1.1	10.0	12.0	2.6	1.3	1.5	4.0	27.4	11.9	5
		J	11.7	2.3	8.0	15.0	1.0	0.2	0.8	1.5	12.0	2.8	9
Razorbill	Alca torda	Α	9.0	1.6	5.0	14.0	3.1	1.1	1.0	6.0	28.6	11.9	155
		I	8.4	2.1	3.0	12.0	1.7	0.7	0.5	4.0	14.1	6.4	33
		J	7.3	2.3	3.8	12.0	1.4	0.5	1.0	2.5	9.7	4.5	17
Little Auk	Alle alle	Α	5.5	1.3	4.0	8.0	2.1	0.7	0.9	3.0	11.7	5.4	13
		J	5.8	1.7	3.5	8.0	1.1	0.2	0.5	1.5	6.3	2.7	16
Atlantic Puffin	Fratercula arctica	Α	7.4	2.6	5.0	12.0	3.5	1.0	2.0	5.0	27.5	17.0	7
		I	8.0	2.0	6.0	10.0	2.6	0.7	1.8	3.0	21.6	9.8	3
		J	6.5	2.3	4.0	10.0	1.6	0.6	1.0	3.0	10.9	5.8	11

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### Citation

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