# The analysis of faeces and regurgitated pellets for determining prey size: problems and bias illustrated for Green Sandpipers *Tringa ochropus* feeding on *Gammarus*

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A technique is described for assessing the size of *Gammarus pulex* in wader diets by measuring the size of their gnathopod claws. The claws can be found in both faeces and regurgitated pellets, thus the technique provides a method to investigate prey size selection by birds that produce either or both of these waste-products. To illustrate the use of this technique, the diet of a small population of passage and over-wintering Green Sandpipers *Tringa ochropus* was investigated at a cress-bed site in southern Britain. Significant differences were found between the size of *Gammarus* in the diet and the size available at the study site. Also, there were significant differences in the size and abundance of gnathopod claws in regurgitated pellets and in faeces. Thus knowing the ratio between numbers of pellets and faeces produced, it is possible to develop the method to reconstruct the prey size distribution in the diet.

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## INTRODUCTION

There have been many studies of wader diet. Most have concentrated on the type and proportion of prey taken (*e.g.* Dit Durell & Tyler 1988), but there have been investigations of the size of prey taken in relation to that available (*e.g.* Green & Tyler 1989; Worrall 1984). The size of prey taken has important implications for the foraging success of the bird, the rate of food intake and the effects on prey populations.

Green Sandpipers *Tringa ochropus* feed mainly on freshwater invertebrates and a wide variety of prey are taken with the diet tending to vary from site to site (Cramp & Simmons 1983). Ormerod & Tyler (1988) found that the shrimp *Gammarus*, mayfly (Ephemeroptera) and caddis larvae (Trichoptera) were the most important components of the winter diet of Green Sandpiper in calcareous channels (Morocco) and estuarine pools (Wales), but mosquito larvae were taken from a sewage pool (Ethiopia) and annelids from an irrigation pool (Morocco). In winter the bird is found in all types of wetland, but mainly favours small enclosed waters or water-edge habitats with protective vegetation, and is usually solitary or in small flocks (Cramp & Simmons 1983).

In Britain, the Green Sandpiper is mainly an autumn passage migrant but it is also recorded over-wintering in low numbers. In Hertfordshire in southern Britain, it tends to favour flooded gravel workings, water cress beds *Rorippa nasturtium-aquaticum* and stream margins (Smith *et al.* 1992). At water cress beds and stream margins, the freshwater shrimp *Gammarus pulex* (Gammaridae, Crustacean) is observed to form the major part of the diet of the birds (K.W. Smith, pers. comm.).

This study was undertaken to investigate ways of determining the size distribution of *G. pulex* taken by Green Sandpipers on a water cress bed, as part of a wider study of the feeding ecology. It was attempted to reconstruct the diet of the Sandpiper by examining the regurgitated pellets and faeces, and observing the feeding behaviour. It was considered that if Green Sandpipers showed any size preference for *G. pulex* it might be possible to assess the value of the habitat for the Sandpipers.

## METHODS

The study site was Lemsford Springs, a Nature Reserve of 7 ha adjacent to the River Lee, near Welwyn Garden City, Hertfordshire, England (51$^0$47'N, 0$^0$14'W). The site is an old water cress bed which was in commercial operation until 1970. The lagoon beds are spring fed from a number of bore holes and the water always stays above 0$^0$ C, so that even in the coldest weather most of the water remains ice-free. The water cress is raked into piles in autumn and left to rot overwinter. The rotting water cress and leaves from the surrounding trees provide a rich food source for *Gammarus pulex*. The shallow open water creates good feeding habitat for birds such as Snipe *Gallinago gallinago* and Green Sandpiper. The reserve has held over ten Green Sandpipers in some winters which is a high proportion of the wintering birds in Hertfordshire (Gladwin & Sage 1986). In recent years, the

number of birds wintering at Lemsford Springs has declined but for unknown reasons (K.W. Smith, pers. comm.).

The population of *G. pulex*, the only *Gammarus* species found in the cress beds, was sampled in November and December 1993 and February 1994. A rigid plastic tube of 232 mm diameter was inserted into the water and pushed through the substrate. The substrate within the tube was stirred up and *G. pulex* were removed using a fine sieve, the process being continued until no more were found. This was repeated four times. *G. pulex* were stored in 70% alcohol. The lengths of all *Gammarus*, with straightened back, were measured from head to telson (base of antenna to base of tail) under a microscope (Figure1a).

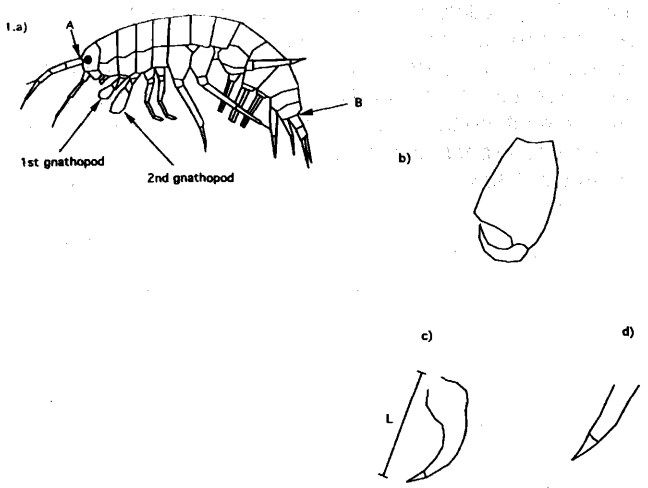


Figure 1a). *Gammarus pulex*, showing the gnathopods (length of *G. pulex* measured from A to B when back straightened). 1b) Second gnathopod. 1c) Claw of gnathopods (L = length measurement of gnathopod claw). 1d) Claw of other appendage.

On the water cress beds Green Sandpipers have a distinct feeding cycle (K.W. Smith, pers. comm., based on observations of > 20 feeding cycles in winters 1993-4 and 1994-5). Birds actively feed for 15-20 minutes, after which they stop feeding and move onto an area of drier ground (usually vegetation, occasionally wood). They rest for on average 7 minutes (range 4-10) and then regurgitate a pellet by contraction of the gullet. The pellet was flicked out of the bill and soon afterwards the bird resumes feeding. The birds also produced faeces on average every 7 minutes (range 3 to 14). Thus birds produced approximately 3.5 faecal droppings for each pellet. In December 1993 birds were watched on the Reserve from a hide and the positions of regurgitated pellets and faeces deposition were noted. To avoid disturbance to feeding birds, pellets and faeces were collected the following night. Due to the liquid nature of faeces, some could not be collected.

Ormerod & Tyler (1988) describe a method of estimating the size of *G. pulex* consumed by Green Sandpipers, by measuring the size of the second gnathopod of found in regurgitated pellets. An attempt was made to repeat this technique, but the second gnathopod was rarely found whole in pellets of Green Sandpipers feeding at Lemsford Springs. A revised method was therefore needed.

The claws on the gnathopod were found to be distinctive. Other claws found on *G. pulex* were examined and it was felt that there was a clear anatomical difference between the gnathopod claws and those from elsewhere on the body (Figure 1c, d). The claws on the gnathopods were dissected from *G. pulex* of a wide range of body lengths and the length of the claws were measured under x100 microscope using an eyepiece graticule. It was not possible to reliably distinguish between the claws from the first and second gnathopods, thus the lengths of each of the four gnathopod claws of 46 *G. pulex* were used to establish a claw length:body length relationship. To establish a body length:weight relationship, 47 *G. pulex* were dried for three days at 70° C and weighed on a microbalance.

A major advantage of this method was that it was possible to examine the faeces as well as pellets for the presence of gnathopod claws, whereas whole gnathopods were never found in the faeces.

Half of a pellet, by weight, was placed in a petrie dish and deflocculated using 2M sodium hydroxide overnight prior to examination. The sample was scanned at x30 magnification and all claws removed to a microscope slide for measurement. The scanning of a pellet was stopped if no claws were found in 10 minutes of searching. Faeces were analysed in a similar way except a whole faecal dropping was placed in a petrie dish.

## RESULTS

The winter population density of *G. pulex* in the lagoons was 4184.m$^{-2}$ (SE 474) in November 1993, 4118.m$^{-2}$ (SE 1170) in December 1993 and 6111.m$^{-2}$ (SE 1514) in February 1994. The largest numbers of *G. pulex* were of the smallest length class (< 4mm), with 63%, 52% and 44% of the total population respectively on the three sampling occasions. The numbers of *G. pulex* decline with each larger size class. The largest size class (>10 mm) formed only 7.1%, 2.9% and 2.3% of the population respectively on the three sampling occasions.

There were significant regression relationships between length of gnathopod claw and body length of *G. pulex*, and between *G. pulex* body length and dry weight (Figures 2 and 3). Careful examination of the sample of pellets and faeces produced by Green Sandpipers at Lemsford Springs revealed remains of no prey item other than *G. pulex*. These results confirm the impression gained from a brief examination of pellets by K.W. Smith (pers. comm.). Over 13 times more gnathopod claws were found in the regurgitated pellets than in the faeces (t-value 19.63, df = 14, $p < 0.0001$). Gnathopod claws from smaller *G. pulex* were evident in the faeces in comparison with the sizes of these claws from *G. pulex* in the pellets. The mean size of the claws in pellets was significantly over 50% larger that those found in faeces (Mann-Whitney U statistic=722, $p < 0.0001$) (Table 1).

Table 1. Mean number and lengths (µm) of gnathopod claws in a small sample of regurgitated pellets and faecal droppings of Green Sandpiper collected from Lemsford Springs Nature Reserve. Numbers are given per whole pellet or faecal dropping.

| | Mean no. claws | St. dev. | Range | Mean length claw (µm) | St. dev. | Range (µm) |
|---|---|---|---|---|---|---|
| Pellet (n=8) | 134 | 14 | 108-148 | 455 | 70 | 245-588 |
| Faecal dropping (n=8) | 10 | 4 | 2-15 | 294 | 63 | 175-511 |

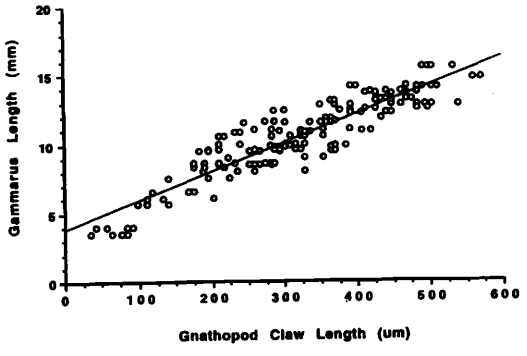


Figure 2. Relationship between length of gnathopod claw (µm) (X) and body length (mm) (Y) of *Gammarus pulex*. Linear regression: Y = 0.021X + 3.816, n = 171 claws, $r^2 = 0.82$, $p < 0.0001$.

Using the regression equation (Figure 2) to convert the measurements of gnathopod claws found in pellets and faeces to body length, Green Sandpipers were selecting prey in the upper part of the range present in the *G. pulex* population (Figure 4) (Mann-Whitney U-test, $p < 0.001$ for both pellets and faeces compared to population). No *G. pulex* smaller than 7.5 mm (3.0 mg) and 9.0 mm (4.2 mg) were consumed, from the evidence of faeces and pellets respectively (Figure 4). Only about 10% of the *G. pulex* population were over 7 mm in length and 5% over 9 mm in the water cress beds in winter.

In one feeding cycle, the Green Sandpipers at the Reserve on average produced one pellet and 3.5 faecal droppings. Combining the size ranges for the contents of one pellet and 3.5 faeces, the mean size of *G. pulex* taken by Green Sandpipers were 10.7 mm (equivalent to 5.8 mg dry weight). Approximately 5% of the Green Sandpiper diet consisted of *G. pulex* of less than 8 mm (under 3.4 mg) in size, 40% of 8.1 to 10 mm (3.5-4.9 mg) and 55% greater than 10.1mm (over 5.0 mg).

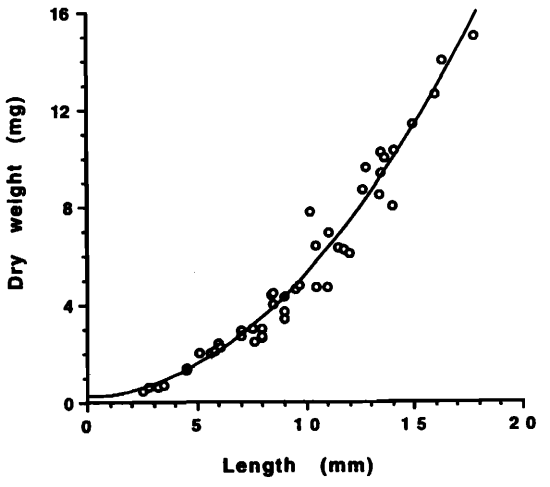


Figure 3. Relationship between body length (mm) (X) and dry weight (mg) (Y) of *Gammarus pulex*. Polynomial regression: $Y = 0.199 + 0.024X + 0.047X^2$, n = 47, $r^2 = 0.96$, $p < 0.0001$.

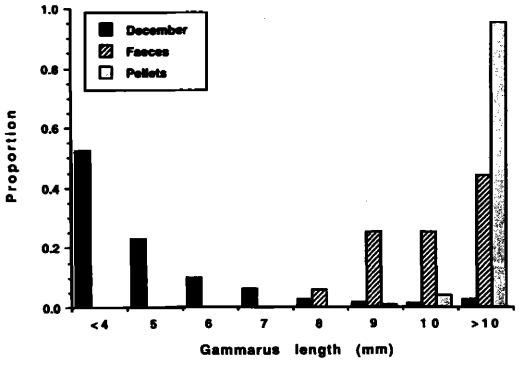


Figure 4. The proportion of different size classes (body length) of *Gammarus pulex* in the substrate in December and in the regurgitated pellets and faeces of Green Sandpipers. Samples from Lemsford Springs nature reserve December 1993.

In 3.5 faeces and one pellet, there were on average 169 gnathopod claws, or the remains of approximately 42 *G. pulex*. Thus in one feeding cycle a Green Sandpiper at Lemsford Springs consumed *G. pulex* of approximately 244 mg dry weight.

## DISCUSSION

The *Gammarus pulex* population consisted of a large proportion of individuals of less than 4 mm in length. This result is very similar to a study by Welton (1979) in which *G. pulex* of less than 4 mm formed 39-76% of the population in a chalk stream in southern Britain. The

density of *G. pulex* of over 4100.m$^{-2}$ in the shallow cress beds at Lemsford Springs was very high, over four times greater than in Welton's study. The site appeared to provide a superabundant food resource for Green Sandpipers.

The examination of pellets and faeces was very time consuming, therefore, only a small sample of each was analysed to illustrate the methodology described. No partly digested gnathopod claws were observed in samples totalling over 1,000 claws, therefore it is assumed that these parts of *G. pulex* are not digestible. Thus the size of the claws is thought to provide a good indication of the size of *G. pulex* in the diet of the Green Sandpipers at Lemsford Springs.

The birds consumed the larger *G. pulex* at the site, with about 76% of the diet consisting of *G. pulex* over 9 mm in length (4.2 mg dry weight). *G. pulex* over 9 mm had a density of about 240 individuals.m$^{-2}$ (about 6% of the December population). Thus despite the huge abundance of *G. pulex* at the site, the Green Sandpipers were taking a food resource that amounted only to about 1.3 g.m$^{-2}$ in December.

Ormerod & Tyler (1988) reported a very similar result with Green Sandpipers taking significantly larger *Gammarus zaddachi* than were present within their study sites. The birds selected prey of over 5 mg dry weight, and these sizes were reported to account for less than 20% of the *G. zaddachi* population, although no data were given on the density of prey.

About 13 times more gnathopod claws of *G. pulex* were present in a pellet than in a faecal dropping and these claws were not of equal size distribution. A different picture of the diet of these birds would be obtained if only faeces or pellets alone were investigated. The use of pellets alone would over-estimate prey size. A similar conclusion was reached by Dekinga & Piersma (1993) working with the Knot *Calidris canutus* feeding on molluscs. Thus care is needed in determining diet and prey size selection for birds that produce both pellets and faeces, such as the Green Sandpiper, Knot and Dunlin *Calidris alpina*, when feeding on prey that contains a significant amount of indigestible hard parts. For birds exhibiting this feeding behaviour, it is necessary to examine both faeces and pellets and, therefore, a method must be developed that will enable body remains of prey to be identified and their size assessed, from both of these products.

In principle, the measurement of prey fragments in pellets and faeces, combined with information on faeces and pellet production rates, would provide reasonable estimates of prey size. However, the ratio of items in pellets and faeces is likely to vary depending on the type of prey. The efficiency of detecting body fragments in faeces and pellets, and the likelihood of passage of items through the gut, may vary for different sized prey of the same species. The only way to evaluate this is to carry out calibration work with captive birds (*e.g.* Dekinga & Piersma 1993; Goss-Custard 1969; Johnstone *et al.* 1990) but this is unlikely to be possible with all wader species and the breadth of their diets. In many studies, prey size evaluation will probably depend on faeces and pellet analyses, and it is recommended that both of these products are always examined for prey fragments and methods devised to evaluate prey size from both these products.

From studies of prey size in faeces and pellets, combined with data on the rate of production of these waste products and the length of time spent feeding, it would be possible to estimate the dry mass or biomass of prey ingested per day by a wader. The lack of data on the number of feeding cycles per day of Green Sandpipers in this study prevents such an estimate.

## ACKNOWLEDGEMENTS

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