RESEARCH ARTICLE

Robert W. Elner · Peter G. Beninger
Daniel L. Jackson · Tracy M. Potter

Evidence of a new feeding mode in western sandpiper (*Calidris mauri*) and dunlin (*Calidris alpina*) based on bill and tongue morphology and ultrastructure

Abstract Sandpipers of the genus *Calidris* tend to have similar body shapes and use narrow, tubular bills to feed on invertebrates in aquatic habitats over an extensive migratory range. Highly plastic foraging behaviours have been displayed but the associations between diet and feeding mechanics are less well understood. Here, scanning electron microscopy was used to relate the ultrastructure of the bills and tongues of two sympatric congeners, western sandpiper (*C. mauri*) and dunlin (*C. alpina*), to function and elucidate the sensory and mechanical basis to feeding. The morphology of the relatively larger, more robust bill of the dunlin suggests specialization for probing although both species are known to feed on epifaunal and infaunal prey. In general, external and internal bill features appear similar for the congeners and the microstructure of taste bud and salivary gland complexes in their mouth cavities correspond to descriptions for other birds. However, the tongues of the two species are remarkable for their distinctive microstructural details as well as copious mucus and extraneous material. In particular, each species has unique tongue papillae, and the keratinized lateral spines along the edges and at the tips of the western sandpiper tongue are markedly longer and denser than for those of the dunlin. Based on the unique features of the tongues and associated observations, inter-species differences in foraging performance can be inferred. Further, evidence suggests that the western sandpiper, and, to a lesser extent, the dunlin, deposit feed on surficial biofilm materials, as opposed to incidentally imbibing such materials while targeting epifaunal prey. Commensurate with the evidence, a novel functional and mechanistic case for unfiltered biofilm grazing in birds is proposed.

R. W. Elner (✉)
Canadian Wildlife Service, Pacific Wildlife Research Centre,
Environment Canada, RR#1, 5421 Robertson Road,
Delta, BC, V4K 3N2, Canada
E-mail: Bob.Elner@ec.gc.ca

P. G. Beninger
Laboratoire de Biologie Marine, Faculté des Sciences,
Université de Nantes, 44322 Nantes, Cedex 3, France

D. L. Jackson
Department of Biology, Life Sciences Centre,
Dalhousie University, 1355 Oxford Street,
Halifax, NS, B3H 4J1, Canada

T. M. Potter
Département de Biologie, Université de Moncton,
Moncton, NB, E1A 3E9, Canada

Introduction

Sandpipers, which are waders of the genus *Calidris*, feed in wetlands and shallow, soft-bottom aquatic habitats of their breeding grounds in sub-polar latitudes and overwintering areas along temperate to tropical coastlines (Alerstam 1990; Wilson 1994). Identified components of natural diets of calidrids encompass a variety of suspended, epibenthic, and infaunal invertebrate taxa that are reflective of local prey abundances (Holmes 1966; Baker 1977; Senner et al. 1989; Piersma 1994; Skagen and Oman 1996). Body mass of individual calidrids is small (usually between 15 and 50 g) and metabolic rates high compared to other non-passerine birds (Castro et al. 1992). When massive flocks of sandpipers concentrate onto restricted feeding areas, such as temperate stopover sites during migration to the breeding grounds, they remove a large biomass of invertebrate prey over a short time (Schneider 1983; Quammen 1984). Thus, pulses of shorebird predation can be critical elements of tidal flat ecology (Schneider and Harrington 1981; Piersma 1994; Wilson and Parker 1996) and have direct and indirect effects on intertidal community structure, both biotic and physical (Furness et al. 1986; Daborn et al. 1993; Erwin 1996; Piersma and Koolhaas 1997).

Sandpipers use three known foraging techniques (Zweers et al. 1994; Zweers and Vanden Berge 1997a): pecking surface prey, probing for infauna, and surface-tension feeding on prey suspended in the water column. Various studies have linked these respective behaviours with functional morphology (Gerritsen and van Heezik 1985; Zweers and Berkhoudt 1991; Zweers et al. 1994; Zweers and Vanden Berge 1997b). However, elucidation of the mechanics of feeding appears complex (Rubega 2002), particularly for specialists on infaunal (Gerritsen et al. 1983; van Heezik et al. 1983; Gerritsen and Meiboom 1986; Zweers and Gerritsen 1997; Piersma et al. 1998) and suspended (Rubega 1996, 1997) prey. Also, while shorebird predation in diverse habitats and on various types of macrofauna is well established (Holmes 1966; Baker 1977; Piersma 1994; Skagen and Oman 1996), the potential range of shorebird food and trophic sources has not been thoroughly investigated. Although there is experimental evidence that western sandpiper, *Calidris mauri*, include epifaunal meiofauna in their diet (Sutherland et al. 2000), studies to determine the functional basis for microphagy are lacking. In contrast, filter-feeding mechanisms to sieve food, including material in the meiofaunal size range ($< < 0.5$ mm in shortest dimension), are well explained for dabbling ducks, flamingos, and some seabirds (Jenkin 1957; Kooloos et al. 1989; Zweers et al. 1994, 1995).

As the sole feeding structures for shorebirds, the bill and tongue should lay the foundation for possible functional correlates. While there are studies on gross anatomy of bills for various *Calidris* species (Hoerschelmann 1970, 1972; Burton 1974; Gerritsen and Sevenster 1985; Gerritsen 1988) and some sensory structures (Bolze 1968; Gerritsen et al. 1983; Zweers and Gerritsen 1997; Piersma et al. 1998), comprehensive topological examinations are few (Rubega 1996) and there is no detailed exploration of the tongue for any shorebird. In the following study, we present the first descriptions based on the use of the scanning electron microscope (SEM) of feeding structures for two calidridine species, western sandpiper (*C. mauri*) and dunlin (*C. alpina*). Dunlin are a common overwintering shorebird on the tidal flats of the Fraser River estuary, British Columbia, and hundreds of thousands of migrating western sandpiper utilize the same habitat as a stop-over site (Butler and Campbell 1987). Taken together with life history and ecological understanding of both species, plus evidence for microphagous prey depletion (Sutherland et al. 2000), our work provides a framework for elucidating a hitherto undescribed feeding mode.

Materials and methods

Origin of specimens

Western sandpiper and dunlin were collected by shotgun on the intertidal flats of Boundary Bay and Roberts Bank, off Westham Island, southwest British Columbia

(Table 1). Each bird was weighed, the bill (exposed culmen) length and tarsus length measured, and maturity (either juvenile or adult) assessed on the basis of plumage characteristics (Wilson 1994). As both species exhibit sexual dimorphism in bill length (Page and Fearis 1971; Page 1974), only adult females were used as a precaution to avoid potential confusion from either age- or sex-related differences in morphology and ultrastructure. However, subsequently, we detected no such differences in examinations under light microscopy (D.L. Jackson and R.W. Elner, unpublished).

Light microscopy

Specimens for examination were stored frozen until required. We made observations of the (thawed) bills with a Nikon SMZ-1500 stereomicroscope, fitted with either an HR Plan Apo 0.5x objective (for larger specimens) or an HR Plan Apo 1x objective. A combination of transmitted light from the base stand and epi-illumination from a fibre-optic light ring was used to illuminate the specimens. We took digital images using a Nikon DXM-1200 high-resolution microscope camera, operating at a pixel resolution of 1280×1024. Additional image manipulations were performed using Adobe Photoshop, and we used Optimas version 6.1 for image analysis and measurements.

Scanning electron microscopy

We removed bills and tongues from specimens immediately after killing, because preliminary studies showed that tissue lysis of the tongue was extremely rapid, and fixed them in cold 3% glutaraldehyde-cacodylate buffer (pH 7.2) for 7 days. Bills were then rinsed for a further 4 days in sodium cacodylate buffer and dehydrated in

Table 1 Particulars of the six adult female western sandpiper (*Calidris mauri*) and six adult female dunlin (*C. alpina*) used for macrophotographs (no. 6) and scanning electron microscopy (nos. 1–5)

Species	Date	Location	Culmen length (mm)	Tarsus (mm)	Weight (g)
C. mauri					
1	4 May 1992	Roberts Bank	28.2	22.0	33.3
2	4 May 1992	Roberts Bank	27.6	23.3	38.1
3	4 May 1992	Roberts Bank	27.5	22.5	33.1
4	5 May 1992	Boundary Bay	25.0	22.7	32.3
5	5 May 1992	Boundary Bay	29.2	–	34.8
6	4 May 2000	Roberts Bank	28.8	–	27.3
C. alpina					
1	5 May 1992	Boundary Bay	41.9	29.1	58.9
2	5 May 1992	Boundary Bay	41.6	29.3	60.4
3	5 May 1992	Boundary Bay	39.3	27.7	52.9
4	5 May 1992	Boundary Bay	40.7	28.5	58.9
5	5 May 1992	Boundary Bay	42.6	27.9	56.2
6	4 May 2000	Roberts Bank	42.9	30.7	73.4

an ascending ethanol gradient. At this point, we used a small, soft paintbrush and ethanol rinse bottle to meticulously clean three of six bills of each species (including the tongues). We cut the bills transversally into three sections that could be fitted into the SEM specimen chamber. At the conclusion of the ethanol series, we immersed all bill sections in nitrogen-stored absolute ethanol and transferred them directly into a pre-refrigerated critical point dryer. Three bone-dry liquid CO_2 rinses were performed prior to critical point drying. The specimens were then mounted onto aluminum stubs and sputter coated with gold-palladium. We observed samples using a JEOL 5200 SEM.

Results

Western sandpiper

The three principal elements of the feeding apparatus are the maxilla, mandible, and tongue. In complete lateral and dorsal aspects (Fig. 1.1, 1.2) the external shape of the bill resembles a tapered forceps converging to a spatulate tip. Light microscopy of the tip of an excised tongue in water reveals that the edges are densely bristled (Fig. 1.3) and the texture of the dorsal surface appears "velvety" with a dense cover of mucus-laden fine material. SEM resolution of features on the maxilla, mandible, and tongue will be considered in succession.
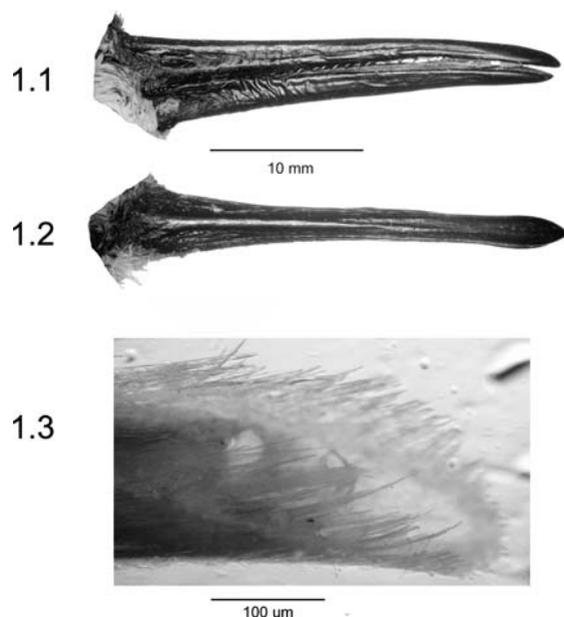
Fig. 1 Western sandpiper, *Calidris mauri*. Complete bill. *1.1* Side aspect, external surface of the maxilla and mandible (*scale bar* 10 mm). *1.2* Dorsal aspect, external surface of the maxilla (*scale bar* 10 mm). *1.3* Dorso-lateral aspect, distal section of tongue in water (*scale bar* 100 μm)

Maxilla

A smooth exterior and lateral border characterize the maxilla; internally, the border becomes progressively rolled near the tip and a median canal runs from the buccal region to the distal extremity, stopping approximately 0.5 mm from the tip (Fig. 2.1). A major feature of the maxilla is a prominent line of epidermal denticles or papillae (Fig. 2.2). Denticles appear within this canal as a single row approximately 3.5 mm from the tip. At approximately 7.5 mm from the tip, as the canal widens, some denticles become more massive, while others occur in pairs (Fig. 2.2, 2.3). Most denticles show evidence of various degrees of mechanical wear (Fig. 2.2, 2.3). The denticles are projections of the keratinized, squamous epidermis, which covers the maxilla (Fig. 2.4). All denticles are oriented obliquely towards the mouth.

Mandible

The interior of the mandible is completely filled by the tongue. Without the tongue, the cavity of the mandible appears featureless and the distal extremity similar to that of the maxilla (Fig. 2.5). Although flattened near the tip, the mandible contains a deep groove, the sublingual canal, for most of its length (Fig. 2.6). The canal is smooth, save for the presence of two types of circular openings: numerous small (15 μm diameter) holes presumed to harbour taste buds, surrounding less abundant larger (70 μm diameter) holes, presumed to be salivary gland openings (Fig. 2.7). The epidermis surrounding the openings is keratinized and squamous. A set of distally oriented peribuccal spines, ranging from 300 to 500 μm in length, project forwards from the buccal region (Fig. 2.8).

Tongue

The excised tongue presents as a thin (approximately 300 μm width in mid-length region) gutter-like structure (Fig. 3.1, 3.2, 3.3; note: the degree of tongue rolling is artefactual; for a fresh intact mandible, the tongue lies flat with its edges in close contact with the sides of the jaw). The dorsal surface is characterized by abundant, complex median papillae (Fig. 3.4), and distally oriented lateral spines, which appear approximately halfway along and become increasingly dense towards the tip (Fig. 3.1–3.6). These spines emerge as prolongations of the longitudinal keratin fibers of the dorso-lateral boundaries of the tongue and attain lengths of approximately 200 μm (Fig. 3.7). The papillae are irregularly shaped and covered with microvilli (Fig. 3.8). Numerous smooth discoid-shaped particles (approximately 8–10 μm longest axis), presumed to be clay, are evident amongst copious quantities of mucus on the spines and papillae of specimens with uncleaned tongues (Fig. 3.4–3.6).

◀
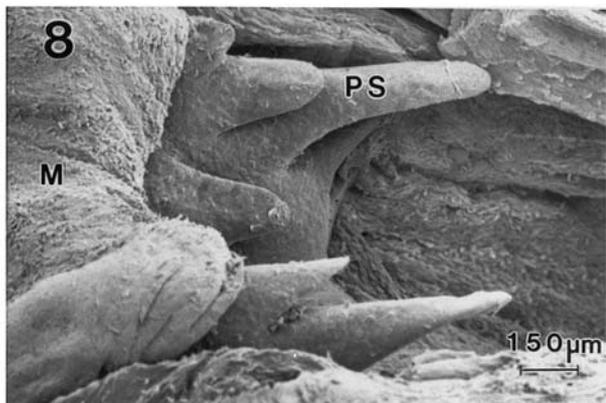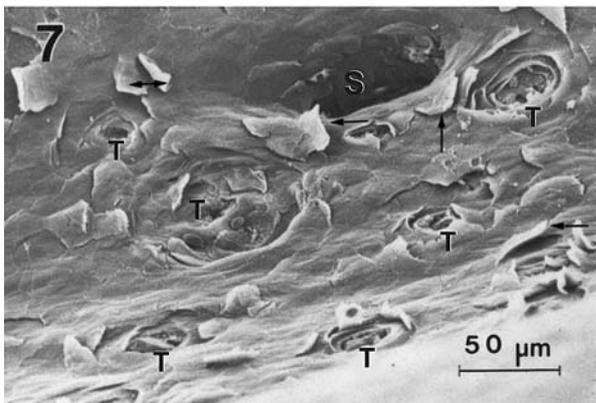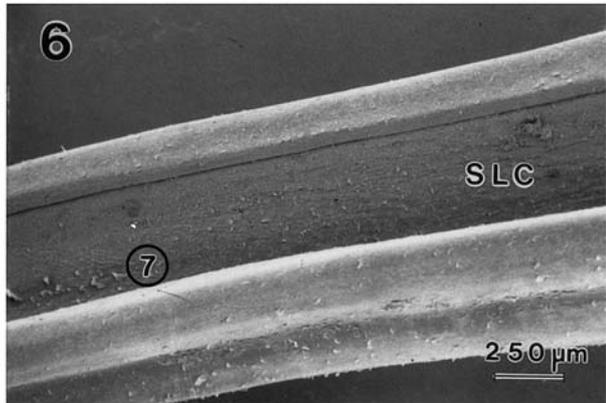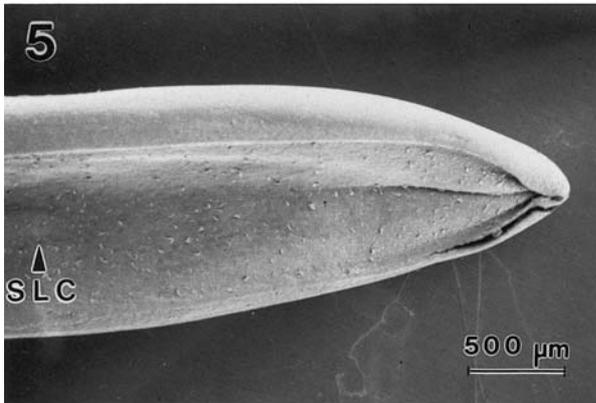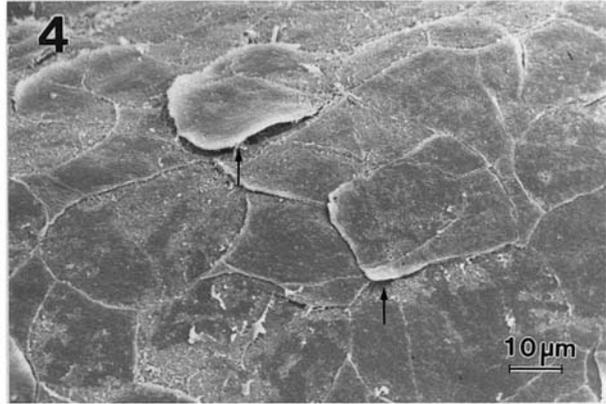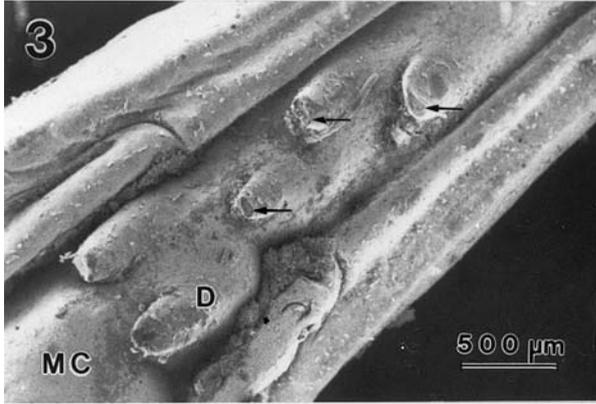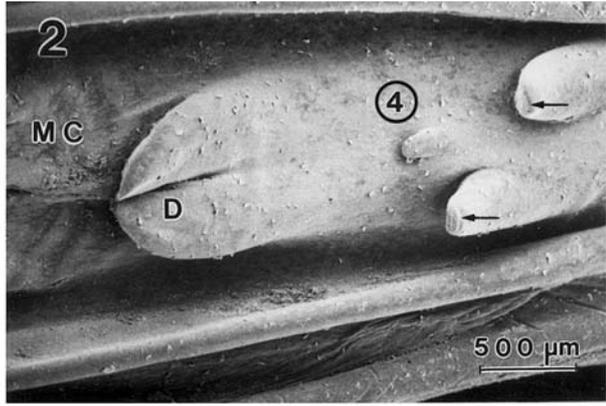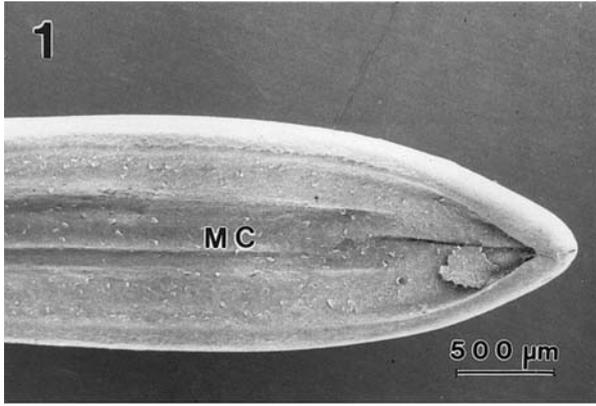
Fig. 2 Western sandpiper, *C. mauri*. Scanning electron micrographs of maxilla and mandible (note: tongue excised). *1* Maxilla tip, internal face. *MC* median canal. *2* Maxillary denticles (*D*) approximately mid-length along the median canal (*MC*). *Arrows* indicate mechanical wear. *Circled 4* Region observed in *image 4*. *3* Denticles of another specimen, showing extensive mechanical wear (*arrows*). *4* Desquamating keratinized layers (*arrows*) of underlying maxillary epidermis. *5* Mandible tip, inner surface. Distal extremity of attenuated sublingual canal (*SLC*). *6* Mandible, mid-region, showing pronounced sublingual canal (*SLC*). *Circled 7* Region observed in *image 7*. *7* Detail of surface of sublingual canal, showing orifices, which may be opening of salivary glands (*S*), surrounded by taste buds (*T*). *Arrows* indicate desquamating keratinized layers, also characteristic of outside surface of both mandible and maxilla. *8* Detail of buccal region, showing mouth (*M*) and anteriorly directed peribuccal spines (*PS*)

Dunlin

External dorsal and lateral aspects of the bill are similar to those for western sandpiper but resemble a longer, more robust forceps converging to a wedge-like tip (Fig. 4.1, 4.2).

Maxilla and mandible

The dorsal surface of the maxilla (Fig. 5.1, 5.2) and the ventral surface of the mandible are smooth, with no discernible sensory structures. The external surfaces are composed of smooth keratin layers (Fig. 5.3).

The inner surface of the maxilla is coated in mucus and adhering particles (Fig. 5.4). The particles (8–10 µm longest axis) resemble those noted for western sandpiper (Fig. 3.5, 3.6) and are likely clay that was not removed during the successive solvent baths prior to SEM examination. Their exogenous nature is further evidenced by the presence of a pennate diatom among them (Fig. 5.5). The distal extremity of the mandible inner surface has a lingual groove, as in the western sandpiper, as well as a rolled lateral margin (Fig. 5.6). The lateral margin presents knobby processes that appear worn towards the exterior (Fig. 5.7). As was the case for the inner surface of the maxilla, the dorsal surface of the mandible, outside the lateral margin (Fig. 6.1), is coated in mucus and adhering particles (Fig. 6.2).

Tongue

The tongue occupies most of the otherwise featureless mandibular cavity. The keratinous lateral spines at the distal extremity of the tongue (Fig. 6.3) are fewer and shorter (up to approximately 100 µm length) than for western sandpiper. They show breakage and wear (Fig. 6.4). As was observed for western sandpipers, specimens with uncleaned tongues present copious quantities of mucus and other material on and around the spines and papillae. The posterior two-thirds of the tongue surface is covered with double-stalked papillae (Fig. 6.5, 6.6).

Discussion

Bills of calidridine sandpipers present a common morphology: a "forceps" formed by a slender maxilla and mandible, composed of keratin over an endoderm and bone matrix, converging to a blunt tip (Burton 1974). The bills exhibit distal rhynchokinesis whereby the highly flexible maxilla and mandible tips can be flexed rapidly outwards and closed while the remainder of the bill remains shut (Burton 1974; Zusi 1984). Apart from size differences, the gross external and internal features of western sandpiper and dunlin bills in the present study appear similar and in accord with previous morphological work on these and other shorebirds (Burton 1974; Gerritsen 1988; Rubega 1996). On the macroscale, for each species, a line of prominent denticles runs along the roof of the maxilla and a conspicuous tongue fills most of the mandible. Microscopic details, such as small pores arranged in a ring around a larger opening (Fig. 2.7), appear identical to structures identified as taste buds surrounding a salivary gland in the mandible of chicken (Berkhoudt 1985). However, while the oral cavities and associated structures seem similar in both sandpiper species, details of the tongues are novel and remarkably distinctive. The tongue of the western sandpiper is coated with unique microvillar papillae, and a dense fringe of long, fine keratinous spines, or bristles, emanate from the lateral margins, especially around the tip. In contrast, the dunlin tongue is less spinous and the papillae are double-stalked, these distinctions being so marked as to suggest a functional difference in feeding between the congeners. High densities of the peculiar papillae seen in the two species are likely responsible for the "velvety" textures of their tongues but have, hitherto, not been described. Although their function, whether sensory or mechanical, remains unknown there is a superficial resemblance to the papillated epithelium of mammalian tongues that contains both taste and salivary glands (Leake 1975).

Calidridine sandpipers display broad adaptive radiation in feeding behaviour (Zweers and Vanden Berge 1997b). All retain the archetypal pecking mode (Zweers et al. 1994) but, in addition, most species can forage by probing (Zweers 1985; Zweers and Berkhoudt 1991; Zweers et al. 1997; Sutherland et al. 2000) and/or surface-tension feeding (Rubega 1997). However, commonalties notwithstanding, calidridine bills exhibit subtle morphological differences in length, shape, and internal structure between species (Burton 1974). Thus, while bill morphology can be seen as a functional effector of plasticity in shorebird feeding behaviour (Gerritsen and van Heezik 1985; Zweers and Gerritsen 1997; Rubega 2002), each species has unique morphological distinctions suited to a particular specialization within a wider opportunistic foraging repertoire (Recher 1966; Skagen and Oman 1996). For example, both western sandpiper and dunlin feed on epifauna and infauna (Couch 1966; Senner et al. 1989; Sutherland et al.

probing puts the slender bills of *Calidris* species under considerable strain; thus, as seen in dunlin, compensating morphological features would include an increase in
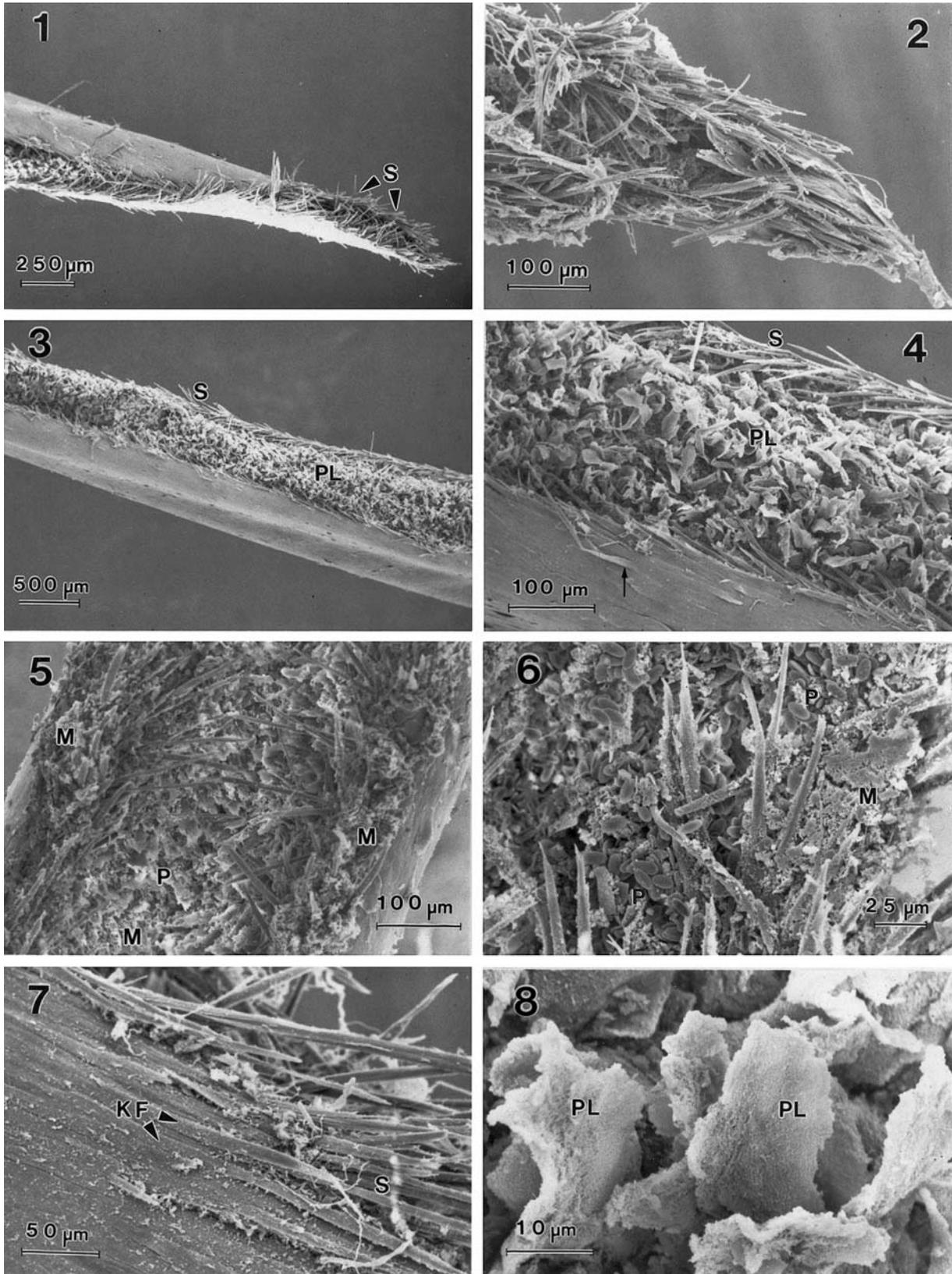
2000) but the relatively longer, more robust bill of dunlin seems better adapted to probe feeding (Gerritsen 1988; Figs. 1, 4). Gerritsen and Sevenster (1985) argue that

◄

Fig. 3 Western sandpiper, *C. mauri*. Scanning electron micrographs of excised tongue. *1* Distal extremity, showing dense border of spines (*S*). Degree of tongue rolling is artefactual. *2* Detail of tongue tip, showing dense covering of fibrous spines and mucus. *3* Mid-region, characterized by fewer spines (*S*) along the lateral border, and a very dense papillary (*PL*) region. *4* Detail of papillary (*PL*) region on a cleaned tongue. Note lateral spine originating from keratinous lateral border (*arrow*). *5, 6* Details of mid-region on an uncleaned tongue. Note copious amounts of mucus (*M*, dehydrated during SEM processing) adhering to papillae and spines as well as particles (*P*). *7* Detail of tongue lateral border, showing derivation of spines (*S*) from keratin fibers (*KF*). *8* Detail of papillae (*PL*) from cleaned tongue, showing irregular shape and surface covered with microvilli

Fig. 4 Dunlin, *Calidris alpina*. Complete bill. *4.1* Side aspect, external surface of the maxilla and mandible (*scale bar* 10 mm). *4.2* Dorsal aspect, external surface of the maxilla (*scale bar* 10 mm)

defense against abrasion (squamous keratin), a straight bill, and mechanical adaptations, such as a wedge-like bill tip, which facilitate sediment penetration while minimizing wear (Zweers and Gerritsen 1997).

Western sandpiper can feed on meio- as well as macrofaunal crustaceans (Sutherland et al. 2000). Mean surface pecking rates of approximately 90 pecks per minute yielding over 100 meiofaunal harpacticoid copepods per bird per minute have been recorded (Sutherland et al. 2000), implying that more than one prey can be removed per "peck". Small sandpipers are not considered predominantly sight-feeding species and reportedly forage by both day and night using visual and tactile cues (Robert et al. 1989; Mouritsen 1993; Dodd and Colwell 1996). However, neither visual nor tactile cue-based pecking mechanisms readily account for a high consumption rate on meiofaunal prey that blend into the soft homogenous topography of intertidal mudflats. Hitherto, biofilm, the medium with which meiofauna are associated, has only been linked indirectly to shorebirds through studies of sediment stability, amphipod grazing on biofilm producing diatoms, and semipalmated sandpiper predation on amphipods in the Bay of Fundy (Daborn et al. 1993; Wilson and Parker 1996). A biofilm community consists of a densely packed mat (10 μm to approximately 2 mm) of microbes, organic detritus, and sediment particles in a mucilaginous matrix of largely extracellular polymeric substances (EPS), in both dissolved and particulate form, together with a variety of non-carbohydrate components (including amino acids, amino sugars, proteins and uronic acids), secreted by benthic diatoms and bacteria (Hoagland et al. 1993; Westall and Rincé 1994; Decho and Herndl 1995). EPS is dominated by acid mucopolysaccharides (MPS), a class of molecule characterized by high viscosity, allowing invertebrate grazers to remove biofilm, and low solubility, allowing biofilm to form and persist even in rain or when covered by the tide. Decho (1990), Decho and Lopez (1993), and Westall and Rincé (1994) provide reviews of the physical, chemical, and ecological properties of biofilm and microbial mats. Considering biofilm as a food resource brings a new perspective to calidrid foraging.

Based on our observations on sandpiper feeding apparatus (in particular, the tongues) and evidence from other studies, we suggest that findings by Sutherland et al. (2000) are more consistent with directed, unfiltered deposit feeding, or grazing, on surficial biofilm material than targeted feeding on meiofauna. Accordingly, we propose biofilm grazing as a fourth feeding mode for, at least, western sandpiper. Further evidence for both the mechanism and the unsorted nature of the natural diet, respectively, are the quantities of mucus observed and the profusion of sediment on the uncleaned tongues of both western sandpiper (Fig. 3.6) and dunlin (Fig. 5.4), including a diatom (Fig. 5.5). Wear on structures, such as the distal-most denticles of the maxilla for western sandpiper (Fig. 2.2, 2.3) and tongue spines (Fig. 6.4) and mandible margin (Fig. 5.7) for dunlin, could be associated with abrasion from sediment intake as coincidental to either biofilm grazing or other forms of feeding. Sediment is a common and substantial component in stomachs and digestive tracts of both species (Couch 1966; R.W. Elner, unpublished) and Stein (2002) determined that over 50% of the dry mass of faeces collected from western sandpipers on migration consisted of inorganic material. Overall, the quantity of sediment manifested internally argues against such material being ingested coincidentally with either targeted pecking on epifauna or probing for infauna but seems consistent with unfiltered grazing on biofilm.

Notwithstanding experimental testing remains to be conducted, we suggest that there could be parallels between biofilm grazing and descriptions of nectar feeding by birds (Paton and Collins 1989). In particular, there appear to be similarities between our observations on calidrids and the brush-tipped tongues and slim bill shape described for honeyeaters. The food of these latter birds, thin films of nectar spread over large areas (Paton 1980; Paton and Collins 1989), might be considered a foraging challenge analogous to biofilm. Our proposed mechanism starts with rapid separation of the spatulate bill tips by distal rhynchokinesis (Burton 1974; Zusi 1984; Gerritson 1988) and "dabbing" of the exposed
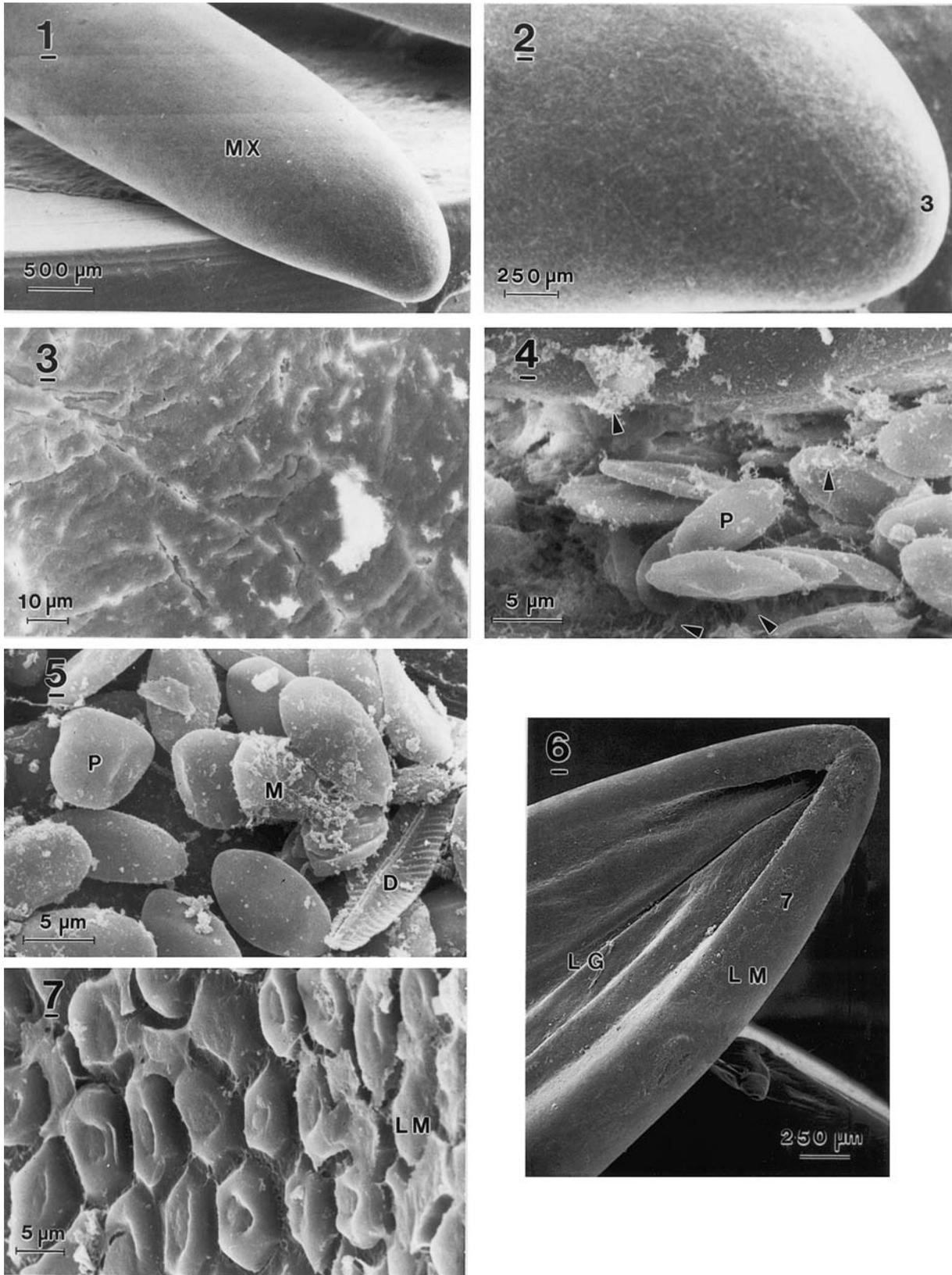
spinous mop at the tongue tip (Fig. 1.3) onto the biofilm surface. The conspicuous spines on the distal extremity of the tongue of both calidrine species, especially the western sandpiper, likely either brush up or adsorb substantial quantities of material (see below). The mucilaginous biofilm (Decho 1994) plus associated

◄

Fig. 5 Dunlin, *C. alpina*. Scanning electron micrographs of bill. *1* Maxilla (*MX*), external dorsal surface of distal region. *2* Maxilla dorsal surface, detail of distal extremity. Note smooth surface and apparent absence of sensory structures. *Number 3*, location of *micrograph 3*. *3* Maxilla dorsal surface, detail of keratin layers typical of entire surface. *4* Maxilla, inner surface, detail of discoid particles (*P*) (approximately 8–10 μm longest axis) and adherent mucus (*arrowheads*). *5* Maxilla, inner surface, detail of particles (*P*), mucus (*M*), and a co-incident pennate diatom (*D*), indicating exogenous origin of particles. *6* Mandible, inner surface showing lateral margin (*LM*) and lingual groove (*LG*). *Number 7* Location of *micrograph 7*. *7* Mandible, lateral margin, detail of knobby processes that show considerable wear towards the lateral margin (*LM*)

components would adhere both to the spines and to the mucus on the tongue. Closing of the bill tips may then act to procure an unsorted bolus of biofilm matrix onto the tongue tip. The subsequent mechanism for transporting the material is unknown and likely complex. One possible scenario might involve a continuous "conveyor-belt" of mucus and biofilm being "syringed" up the tongue and into the oesophagus by a sucking

Fig. 6 Dunlin, *C. alpina*. Scanning electron micrographs of mandible and excised tongue. *1* Mandible, inner surface at two-thirds length; *C* Artefact (feather). *Number 2* Location of *micrograph 2*. *2* Mandible, inner surface, detail of particles (*P*) and adhering mucus (*M*), which appears to be secreted from mandible inner surface. *Asterisks* show mucus strands adhering to both particles and mandible inner surface. *3* Tongue, inner surface, distal extremity, showing keratinous lateral spines (*S*). *Number 5* Location of *micrograph 5* approximately 400 μm towards mouth. *4* Tongue, inner surface, showing detail of keratinous spines (*S*). Note breakage and wear. *5* Tongue, inner surface, median region, showing double-stalked papillae (*PL*). *6* Detail of papillae (*PL*) showing ridged surface, and interspersed particles (*P*)
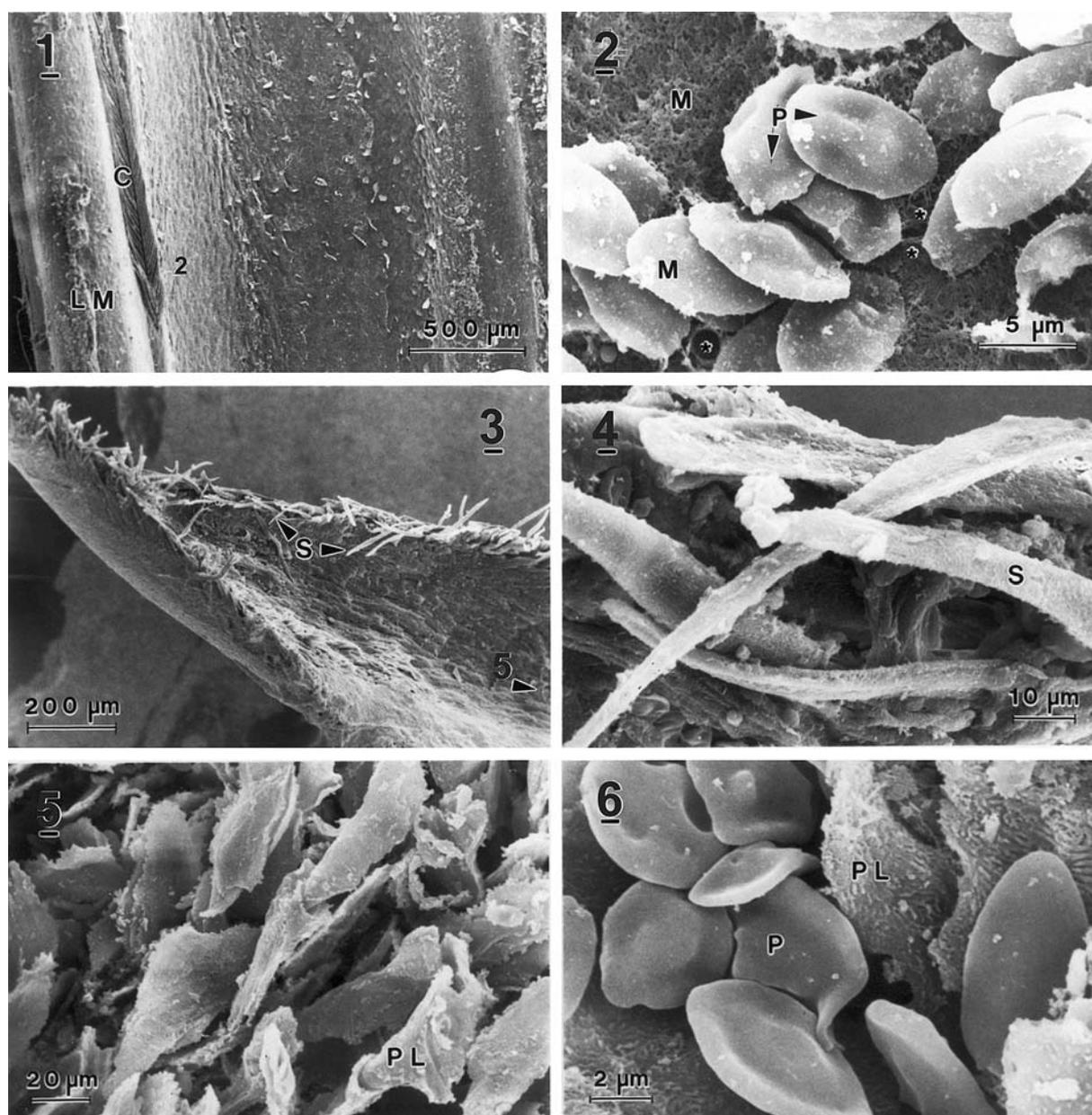
effect resulting from a rapid backwards and forwards tongue action (Burton 1974; Zweers 1985). En route up the bill, the backwardly directed maxillary denticles may function to prevent particle-laden mucus masses from slipping forwards out of the bill as well as passively scrape and channel material through the trough at the base of the tongue (Zweers 1985; Paton and Collins 1989) before being swallowed into the pharynx. Our proposed biofilm feeding system would necessarily involve copious quantities of mucus, as a capture and transport medium, originating from the sublingual salivary orifices on the mandible (Fig. 2.7) and, probably, from the tongue itself. For dunlin, considered a typical member of the subfamily Calidridinae, the principal two pairs of salivary glands in the floor of the buccal cavity are particularly prominent and highly developed compared to other waders (Burton 1974).

Whether biofilm grazing could entirely fuel the metabolic requirements of, say, a western sandpiper is presently unsubstantiated but indications are that such material could, at least, provide a valuable dietary supplement. Experiments by Decho and Moriarty (1990) demonstrated that harpacticoid copepods can feed on EPS-coated beads and suggest that microbial EPS attached to sediment is easily digested and a potential food source for various sediment-feeding and aggregate-feeding invertebrates. Field observations and video recordings (R.W. Elner, unpublished data, Fraser River delta) show that western sandpiper and dunlin peck opportunistically at visible epifaunal prey and probe for infaunal prey as they forage across the intertidal. However, for the most part, the birds characteristically and rapidly "dab", as opposed to peck, on the sediment surface. Also, western sandpipers, in particular, tend to follow the receding tide (Wilson 1994) to feed on wet sediment where a biofilm grazing mechanism would appear more feasible than on a dry surface. Thus, considering a mean surface feeding rate of 92.4 ($\pm$2.4 SD) pecks min^{-1} for western sandpiper on intertidal mudflats in the Fraser River estuary during spring migration (Sutherland et al. 2000) and the (probably conservative) inference that "dabbing", representing a biofilm grazing mode, occurs for 50% of the observed surface feeding episodes, birds might attain a mean "dabbing" rate of approximately 46 dabs min^{-1}. Further, an estimated mean pick-up performance of 0.00257 ($\pm$0.0031 SD) g material per swab ($n = 7$; estimate based on wet weight gain after the tip of a clean excised western sandpiper tongue was manually dabbed once onto a surface of fresh intertidal material from Roberts Bank; R.W. Elner, unpublished) generates an ingestion rate for wet, unsorted, biofilm and sediment, mass of 7.1 g h^{-1} bird^{-1}. Although energetic values for biofilm material are unreported, the estimated gross hourly ingestion rate approximates 20% of the total body weight of a western sandpiper on migration (Table 1). Even allowing for a high proportion of water and indigestibles, such a gross mass intake rate could potentially translate into a substantial dietary contribution.

Given broad similarities in bill shape and feeding habitats, extension of the diet base to biofilm is a consideration in other Calidridinae and, possibly, throughout the family (Scolopacidae). Historically, some field experiments designed to measure the "feeding footprint" of shorebirds have yielded ambiguous results (Sewell 1996) and identifiable prey remains have been lacking in analyses of stomachs from migrating shorebirds (Couch 1966; Sutherland et al. 2000). Although alternative methodological explanations are possible (Mercier and McNeil 1994), such enigmas could also be explained by failure to consider the full size spectrum of available prey (Sutherland et al. 2000), and, in particular, an ability to feed directly on biofilm. Further, heavy sediment ingestion, which may be a by-product to biofilm grazing, has been documented as an unexplained but widespread phenomenon in shorebirds (Hui and Beyer 1998). Biofilm and microbial mats are predictable resources on all intertidal sediment (Westall and Rincé 1994), and shorebirds might be expected to locate and discriminate between different qualities of feeding substrate through a well-developed sense of taste (Gerritsen et al. 1983). As documented here and elsewhere (Gerritsen et al. 1983), calidridine shorebirds have extensive batteries of tastebuds. Thus, biofilm may be viewed as a hedge to environmental uncertainty, particularly for shorebirds during migration over a large latitudinal range, in that such a food source would obviate the need for refined predation techniques adapted to local conditions of prey identity and availability. For dunlin, while the bill and tongue are so reminiscent of the western sandpiper that biofilm feeding appears as possible, the less well developed spines and longer bill may indicate relatively less efficiency in such a mode. We hypothesize a trade-off between the demands of probing and biofilm grazing and suggest that the latter should be more prevalent in shorter-billed birds given the postulated mechanics of transporting biofilm up a bill. Conversely, longer-billed birds would have an advantage in foraging on infaunal prey (Zweers and Gerritsen 1997).

In conclusion, our study suggests a hitherto unknown form of microphagy in birds. Invoking such a new mode is consistent with the data presented and provides a parsimonious explanation for various enigmas concerning shorebird feeding ecology. Nevertheless, we recognize that more research is required to elucidate foraging mechanisms in sandpipers and trust that the hypotheses arising from the present work may serve to focus future study. In particular, testing the plausibility of biofilm feeding will require a suite of definitive behavioural, sensory, mechanistic, and energetic studies. However, the existence of a vertebrate with a bulk-feeding mode that taps into the microbial loop should not be entirely unexpected given the general lack of understanding of feeding in birds (Rubega 2002). Hitherto, only invertebrates have been considered as consumers of biofilm (Taghon 1982; Baird and Thistle 1986; Decho and Moriarty 1990) and biofilm grazing by birds would add considerably to food web theory. Further,

the possibility of direct transfer of benthic, largely microbial-meiofaunal production to migratory shore-birds has novel implications for conservation as well as understanding of the ecology, physiology, and evolution of the species themselves.

References

Alerstam T (1990) Bird migration. Cambridge University Press, Cambridge

Baird BH, Thistle D (1986) Uptake of bacterial extracellular polymer by a deposit-feeding holothurian (*Isostichopus badionotus*). Mar Biol 92:183–187

Baker MC (1977) Shorebird food habits in the eastern Canadian Arctic. Condor 79:56–62

Berkhoudt H (1985) Structure and function of avian taste receptors. In: King AS, McLelland J (eds) Form and function in birds, vol 3. Academic Press, London, pp 463–495

Bolze VG (1968) Anordnung und Bau der Herbstschen Kõrperchen in Limicolenschnäbeln im Zusammenhang mit der Nahrungsfindung. Zool Anz 18:313–355

Burton PJK (1974) Feeding and the feeding apparatus in waders: a study of anatomy and adaptations in the Charadrii. British Museum (Natural History), London

Butler RW, Campbell RW (1987) The birds of the Fraser River delta: populations, ecology and international significance. Can Wildl Serv Occas Pap 65

Castro G, Myers JP, Ricklefs RE (1992) Ecology and energetics of sanderlings migrating to four latitudes. Ecology 73:833–844

Couch AB (1966) Feeding ecology of four species of sandpipers in western Washington. Master's thesis, University of Washington

Daborn GR, Amos CL, Brylinsky M, Christian H, Drapeau G, Faas RW, Grant J, Long B, Paterson DM, Perillo GME, Piccolo MC (1993) An ecological cascade effect: migratory birds affect stability of intertidal sediments. Limnol Oceanogr 38:225–231

Decho AW (1990) Microbial exopolymer secretions in ocean environments: their role(s) in food webs and marine processes. Oceanogr Mar Biol Annu Rev 28:73–153

Decho AW (1994) Molecular-scale events influencing the macro-scale cohesiveness of exopolymers. In: Krumbein WE, Paterson DM, Stal LJ (eds) Biostabilisation of sediments. BIS, Oldenberg, pp 135–148

Decho AW, Herndl GJ (1995) Microbial activities and the transformation of organic matter within mucilaginous material. Sci Total Environ 165:33–42

Decho AW, Lopez GR (1993) Exopolymer microenvironments of microbial flora: multiple and interactive effects on trophic relationships. Limnol Oceanogr 38:1633–1645

Decho AW, Moriarty DJW (1990) Bacterial expolymer utilization by a harpacticoid copepod: a methodology and results. Limnol Oceanogr 35:1039–1049

Dodd SL, Colwell MA (1996) Seasonal variation in diurnal and nocturnal distribution of nonbreeding shorebirds at North Humbolt Bay, California. Condor 98:196–207

Erwin RM (1996) Dependence of waterbirds and shorebirds on shallow-water habitats in the mid-Atlantic coastal region: an ecological profile and management recommendations. Estuaries 19:213–219

Furness RW, Galbraith H, Gibson IP, Metcalfe NB (1986) Recent changes in numbers of waders on the Clyde Estuary, and their significance for conservation. Proc R Soc Edinb 90B:171–184

Gerritsen AFC (1988) Feeding techniques and the anatomy of the bill in sandpipers (*Calidris*). PhD thesis, Leiden

Gerritsen AFC, Heezik YM van (1985) Substrate preference and substrate relative to foraging behaviour in three *Calidris* species. Neth J Zool 35:671–692

Gerritsen AFC, Meiboom A (1986) The role of touch in prey density estimation by *Calidris alba*. Neth J Zool 36:530–562

Gerritsen AFC, Sevenster JG (1985) Foraging behaviour and bill anatomy in sandpipers. Fortschr Zool 30:237–240

Gerritsen AFC, Heezik YM van, Swennen C (1983) Chemoreception in two further *Calidris* species (*C. maritima* and *C. canutus*). Neth J Zool 33:485–496

Heezik YM van, Gerritsen AFC, Swennen C (1983) The influence of chemoreception on the foraging behaviour of two species of sandpiper, *Calidris alba* and *Calidris alpina*. Neth J Sea Res 17:47–56

Hoagland KD, Rosowski JR, Gretz MR, Roemer SC (1993) Diatom extracellular polymeric substances: function, fine structure, chemistry and physiology. J Phycol 29:537–566

Hoerschelmann H (1970) Schnabelform und Nahrungserwerb bei Schnepfenvõgeln (Charadriidae und Scolopacidae). Zool Anz 184:302–327

Hoerschelmann H (1972) Strukturen der Schnabelkammer bei Schnepfenvõgeln (Charadriidae und Scolopacidae). Z Wiss Zool 185:105–121

Holmes RT (1966) Feeding ecology of the red-backed sandpiper (*Calidris alpina*) in Arctic Alaska. Ecolocharacteristics 47:32–45

Hui CA, Beyer WN (1998) Sediment ingestion of two sympatric shorebird species. Sci Total Environ 224:227–233

Jenkin PM (1957) The filter feeding and food of flamingoes (Phoenicopteri). Philos Trans R Soc Lond B Biol Sci 240:401–493

Kooloos JGM, Kraaijeveld AR, Langenbach GEJ, Zweers GA (1989) Comparative mechanics of filter feeding in *Anas platyrhynchos*, *Anas clypeata* and *Aythya fuligula* (Aves, Anseriformes). Zoomorphology 108:269–290

Leake LD (1975) Comparative histology: an introduction to the microscopic structure of animals. Academic Press, London

Mercier F, McNeil R (1994) Seasonal variations in intertidal density of invertebrate prey in a tropical lagoon and effects of shorebird predation. Can J Zool 72:1755–1763

Mouritsen KN (1993) Diurnal and nocturnal prey detection by dunlins *Calidris alpina*. Bird Study 40:212–215

Page G (1974) Age, sex, molt and migration of dunlins at Bolinas Lagoon. West Birds 5:1–12

Page G, Fearis B (1971) Sexing western sandpipers by bill length. Bird-Banding 42:297–298

Paton DC (1980) The importance of manna, honeydew and lerp in the diets of honeyeaters. Emu 80:213–226

Paton DC, Collins BG (1989) Bills and tongues of nectar-feeding birds: a review of morphology, function and performance, with intercontinental comparisons. Aust J Ecol 14:473–506

Piersma T (1994) Close to the edge: energetic bottlenecks and the evolution of migratory pathways in knots. Uitgeverij Het Open Boek, Den Burg

Piersma T, Koolhaas A (1997) Shorebirds, shellfish(eries) and sediments around Griend, western Wadden Sea, 1988–1996. NIOZ Rapport 1997-7

Piersma T, Aelst R van, Kurk K, Berkhoudt H, Maas LRM (1998) A new pressure sensory mechanism for prey detection in birds: the use of principles of seabed dynamics. Proc R Soc Lond B 265:1377–1383

Quammen ML (1984) Predation by shorebirds, fish, and crabs on invertebrates in intertidal mudflats: an experimental test. Ecology 65:529–537

Recher HF (1966) Some aspects of the ecology of migrant shore-birds. Ecology 47:393–407

Robert M, McNeil R, Leduc A (1989) Conditions and significance of night feeding in shorebirds and other wader birds in a tropical lagoon. Auk 106:94–101

Rubega MA (1996) Sexual size dimorphism in red-necked phala-ropes and the functional significance of non-sexual bill structure variation for feeding performance. J Morphol 228:45–60

Rubega MA (1997) Surface tension prey transport in shorebirds: how widespread is it? Ibis 139:488–493

Rubega MA (2002) Feeding in birds: approaches and opportuni-ties. In: Schwenk K (ed) Feeding. Academic Press, New York, pp 395–408

Schneider DC (1983) The food and feeding of migratory shore-birds. Oceanus 26:38–43

Schneider DC, Harrington BA (1981) Timing of shorebird migra-tion in relation to prey depletion. Auk 98:801–811

Senner SE, Norton DW, West GC (1989) Feeding ecology of western sandpipers, *Calidris mauri*, and dunlins, *C. alpina*, during spring migration at Hartney Bay, Alaska. Can Field Nat 103:372–379

Sewell MA (1996) Detection of the impact of predation by migratory shorebirds: an experimental test in the Fraser River estuary, British Columbia (Canada). Mar Ecol Prog Ser 144:23–40

Skagan SK, Oman HD (1996) Dietary flexibility of shorebirds in the western hemisphere. Can Field Nat 110:419–444

Stein RW (2002) Busting a gut: age-related variation and seasonal modulation of digestive tract structure and function in the western sandpiper. Master's thesis, Simon Fraser University, Burnaby

Sutherland TF, Shepherd PCF, Elner RW (2000) Predation on meiofaunal and macrofaunal invertebrates by western sand-pipers (*Calidris mauri*): evidence for dual foraging modes. Mar Biol 137:983–993

Taghon GL (1982) Optimal foraging by deposit-feeding inverte-brates: Roles of particle size and organic coating. Oecologia 52:295–304

Westall F, Rincé Y (1994) Biofilms, microbial mats and microbe-particle interactions: electron microscope observations from diatomaceous sediments. Sedimentology 41:147–162

Wilson WH (1994) Western sandpiper (*Calidris mauri*). In: Poole A, Gill F (eds) The birds of North America, no. 90. The Academy of Natural Sciences, Philadelphia

Wilson WH, Parker K (1996) The life history of the amphipod, *Corophium volutator*: the effects of temperature and shorebird predation. J Exp Mar Biol Ecol 196:239–250

Zusi RL (1984) A functional and evolutionary analysis of rhyn-chokinesis in birds. Smithsonian Contrib Zool 395

Zweers GA (1985) Generalism and specialism in avian mouth and pharynx. Fortschr Zool 30:189–201

Zweers GA, Berkhoudt H (1991) Recognition of food in pecking, probing and filter-feeding birds. Acta XX Congressus Interna-tionalis Ornithologici (1990). New Zealand Ornithological Congress Trust Board, Wellington, New Zealand, pp 897–901

Zweers GA, Gerritsen AFC (1997) Transitions from pecking to probing mechanisms in waders. Neth J Zool 47:161–208

Zweers GA, Vanden Berge JC (1997a) Evolutionary transitions in the trophic system of the wader-waterfowl complex. Neth J Zool 47:255–287

Zweers GA, Vanden Berge JC (1997b) Birds at geological bound-aries. Zoology 100:183–202

Zweers GA, Berkhoudt H, Vanden Berge JC (1994) Behavioral mechanisms of avian feeding. Adv Comp Environ Physiol 18:241–279

Zweers G, de Jong F, Berkhoudt H, Vanden Berge JC (1995) Filter feeding in flamingos (*Phoenicopterus ruber*). Condor 97:297–324

Zweers GA, Vanden Berge JC, Berkhoudt H (1997) Evolutionary patterns of avian trophic diversification. Zoology 100:25–57