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Localised Human Impacts on the Harataonga Coastal Landscape, Great Barrier Island, Northern New Zealand

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ABSTRACT

Here we present results of analyses of sediment profiles and cores, and coprolites, from Harataonga Bay, Great Barrier Island. Using a range of analyses (sedimentological, plant microfossils, parasitological, microbial, and steroids and myoglobin) we concentrate on human impact and reconstruction of the geomorphology and vegetation of the near-shore environments. Two different sub-environments are represented: dunes and alluvial plain. Dune instability coincides with a major increase in disturbance-related plants (especially ground ferns) as a result of forest clearance. The present form of much of the Harataonga dunes and the swamp at the eastern end of the bay is directly a result of human impact, no earlier than 737 ± 178 ^{14}C yr BP. In the record from the alluvial plain of the main Harataonga watercourse, at the western end of the bay, it is difficult to clearly resolve sedimentary inputs that directly relate to human presence in this former tidal inlet that was open to storm surge and stream floods. The only exception is the slopewash materials forming the terrace surface, sediments of which bear pollen consistent with vegetation disturbance. The landforms are natural but the rate at which the tidal inlet was infilled to form a terrace was accelerated by human activity. The nature and timing of the localised human impacts at Harataonga are consistent with those observed elsewhere on Great Barrier Island and mainland New Zealand. Some of our techniques (e.g., bacteria, steroids) are newly applied to coprolites in New Zealand but none provided any useful information because of poor preservation.

Keywords: SEDIMENTOLOGY, PLANT MICROFOSSILS, COPROLITES, HARATAONGA BAY.

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INTRODUCTION

As part of a recent palaeo-environmental project at Harataonga Bay, Great Barrier Island, several sediment profiles and cores were examined. The record is assembled from a range of proxy indicators and is temporally constrained by tephrochronology and radiocarbon dating. Some of the results have been reported. One of the cores provided a perspective of the formation of the landscape since the mid-Holocene (Horrocks *et al*. 2002a; Nichol *et al*. 2007). The temporal and spatial scale for that perspective was broad, encompassing the last *c*. 6000 yr and incorporating geomorphic processes operating over several km^2. Also, some of the sediment profiles and other cores were used to study spatial variation in pollen and deforestation charcoal in relation to the *c*. 665 yr BP (cal. 1314 ± 12 AD, Hogg *et al.* 2003) Kaharoa tephra (Horrocks *et al*. 2002b). In addition, coprolites from one of the profiles provided evidence of prehistoric environments and diet (Horrocks *et al*. 2002a, Horrocks *et al*. 2004b).

In this paper we present the remainder of the sediment profile and core results from Harataonga Bay. We focus on a narrow temporal and spatial scale, concentrating on human impact and reconstructing the geomorphology and vegetation of the near-shore environments, areas of the catchment where human activity was presumably concentrated.

THE STUDY AREA AND SITES

Harataonga Bay is located on the central eastern coast of Great Barrier Island (Fig. 1). The bay is about 500 m from headland to headland and has a northerly aspect. The local watershed drains an area of 5.5 km^2 and comprises a single main watercourse that enters the sea at the western end of the beach. The sand dunes of the Harataonga shoreline rise to about 15 m above sea level and form an almost unbroken line along the length of the beach. The swamp system that formed behind these dunes is about 2 km long in an approximately west–east direction, extending inland more than 1 km at the western end. Most of the swamp system has been drained since European settlement and at present is pasture with abundant Cyperaceae.

Three sediment cores from Harataonga Bay (Cores 1, 2 and 3, Fig. 1), encompassing the last *c*. 6000 yr, incorporated gradual, catastrophic, and human-influenced sedimentation styles on the New Zealand coast and provided pollen records (Horrocks *et al.* 2002a, 2002b; Nichol *et al*. 2007). Postglacial sea-level rise, volcanism, possible tsunami (pre-settlement) and catchment clearance by early people were all evident in the sediment record, either as artefacts or indirect indicators. Nichol *et al*. (2007) argued that back-barrier wetlands formed behind stationary-type (aggraded) bay barriers (such as Harataonga) are of greater value (more sensitive) for longer-term environmental reconstruction than wetlands associated with prograded- and receded-type barriers, where the sediment record is typically less complete.

The longest pollen record (Core 1, Fig. 1) commenced *c*. 6000 yr ago in a freshwater environment with swamp forest composed mainly of *Laurelia*, *Leptospermum*, *Ascarina* and *Cyathea* (Horrocks *et al.* 2002a). *Dodonaea* and Cyperaceae grew on margins of this forest. Forest on the hills surrounding the wetland comprised mainly *Metrosideros,* with emergent *Dacrydium* and *Libocedrus*. *Ascarina*, *Rhopalostylis,* and *Cyathea* were a significant part of the under-storey of the hillside forest. Around the time of deposition of the *c*. 5550 cal yr BP Whakatane tephra, a freshwater lake developed at the site. Extensive Cyperaceae swamp developed on the fringes of the lake. Shortly after *c*. 2900 cal yr BP, *Dacrycarpus* briefly

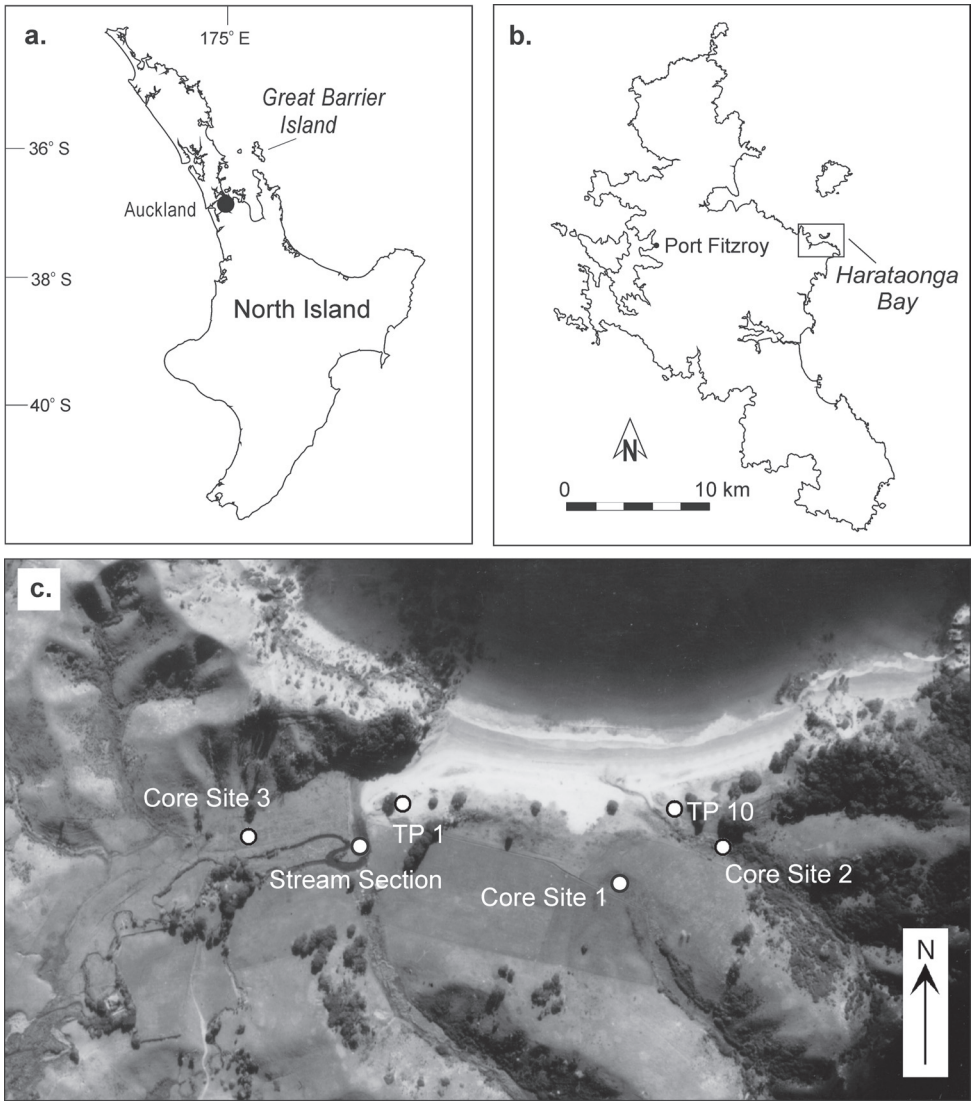

Figure 1: The location of Harataonga Bay and the sites discussed. a: Location of Great Barrier Island. b: Location of Harataonga Bay. c: 1942 aerial photograph of Harataonga Bay showing sites. Nowadays the hills surrounding the dunes and flats are more densely forested. Photo: Air Force Museum, Christchurch. Core 1 was analysed by Horrocks *et al*. 2002a and Nichol *et al*. 2007.

invaded swamp forest and the site made the final transition to swamp. *Myrsine* and then *Hebe* shrubs invaded fringes of the swamp as the water table fell, possibly as a result of a change to drier conditions in the Late Holocene. Polynesian deforestation, as indicated by the presence of abundant charcoal and *Pteridium* spores, was recorded as occurring shortly after deposition of the *c*. 665 yr BP Kaharoa tephra. Pollen of the Polynesian introduced cultigen *Lagenaria siceraria* (gourd) was found in Core 2. For pollen records from

elsewhere on Great Barrier Island, see Horrocks *et al*. (1999; 2000a, 2000b) and Ogden *et al.* 2006).

Archaeological sites at Harataonga Bay were first recorded by Spring-Rice (1962), who also produced a preliminary report on their excavation (Spring-Rice 1963). Law (1972) completed the analysis and wrote the final report. Three sites were excavated: a beach midden (the Western Midden) thought to date from about the thirteenth century AD (on the basis of artefacts, faunal remains and obsidian hydration dating), a second beach midden (the Eastern Midden) and a fortified site (the Pā), both of which are later in time. When compared, the sites showed changes in the frequency of use of fish, birds, mammals and shellfish, and changes in the individual species used in each group. Changes in the raw material used for tools also occurred and the different nature of the sites showed changes in settlement pattern. The older of the two eastern sites (the Eastern Midden) subsequently provided a date of 441 ± 55 ^{14}C yr BP and the younger, fortified site provided dates of 247 ± 55 and 216 ± 55 ^{14}C yr BP (Law 1975). In more recent Harataonga studies, pollen of *Lagenaria siceraria* (gourd) and starch grains and xylem cells of *Ipomoea batatas* (kūmara or sweet potato), another Polynesian introduced cultigen, were identified in coprolites dated at least 467 ± 60 ^{14}C yr BP (Horrocks *et al.* 2002a; Horrocks *et al.* 2004a).

The five sites examined at Harataonga Bay in this study comprise two of the aforementioned core sites (2 and 3), a section of eroded stream terrace and two excavated test pits. Core Site 2, ~5 m from an active dune blow-out, is situated at the eastern end of the bay within a narrow (up to 30 m wide) *Leptospermum/Typha* swamp enclosed on its landward side by forested (regenerating *Leptospermum/Kunzea*) hill-slopes and by dunes on the seaward side (Fig. 1). An ephemeral stream flows through the swamp and discharges into the dunes. The core site is ~80 m landward of the beach. Much of the dune surface at the eastern end of the beach is vegetated, but evidence for past dune instability is clear, with the former blow-out surface and associated sand sheet deposited in the swamp margins. A ridge separates this waterway from the much larger wetland associated with the main stream. Test Pit 10 (TP 10) is situated on the dunes ~30 m seaward of Core Site 2 (Fig. 1), near Law's (1972, 1975) two later occupation sites, the Eastern Midden and the Pā.

Core Site 3 is on a drained swamp surface that forms an overbank (terrace) environment flanking the stream at the western end of the bay (Fig. 1). This site is in pasture with scattered, often dense Cyperaceae. Hill slopes nearby are covered in regenerating *Leptospermum/Kunzea* forest. The core was taken adjacent to an apparent Maori terrace located ~15 m distant. The core site is ~150 m to the east of the main stream channel but is within the flood zone and would have been within the reach of tidal flow prior to swamp formation. Today the site is ~400 m landward of the beach and well beyond the tidal limit. The section of eroded stream terrace (Stream Section) is located on the same terrace immediately west of the stream mouth, facing east (Fig. 1). Test Pit 1 (TP 1) is located on the western extremity of the dunes, ~380 m from Core Site 3 (Fig. 1), near Law's (1972, 1975) earlier occupation site, the Western Midden. The dunes at this end of the beach are more active than those at the eastern end, hence are less vegetated.

METHODS

Of five coprolites found in TP 1, four (F1, F6, F61 and F62) were examined for endoparasite eggs plus steroids and myoglobin. Samples F6 and F62 were also tested for originating species using the trisodium phosphate method (Bryant 1974), and examined for

bacteria. These analyses were carried out on solid coprolite pieces up to ~1 cm diameter that appeared clean of enclosing sediments.

The two sediment cores were collected in aluminium tubes (76 mm diam.) using a vibracoring system, achieving penetration to 2.56 m and 1.3 m at Core Sites 2 and 3, respectively. The facies in these cores were previously described and the pollen and tephra analysed by Horrocks *et al.* (2002b). In the laboratory, the cores were split lengthwise and sub-sampled at regular intervals. For the two test pits, a sub-sample was taken from bulk samples ("Layers"). TP 1 is not described in detail by us (Martin Jones, unpublished data). However, the depths below ground surface for the sampled layers of this profile are: 80 cm for Layer 3, 90 cm for Layer 5, ~95 cm for Layer 7, 101 cm for Layer 9, 116 cm for Layer 11, 121 cm for Layer 12a, 126 cm for Layer 12b and 131 cm for Layer 12c. The Stream Site was examined for stratigraphy but not sampled.

Core sub-samples were measured using a laser particle sizer (Galai™) that determines particle size based on the time of transition principle (Molinaroli *et al*. 2000). The Galai determines particle size assuming that larger particles produce a longer transition time across the laser path. In this study, 1 g of each sample was introduced to a solution of filtered water and Calgon to assist particle dispersion, with data collection set to the 99% confidence level. Results reported here are for grain size classes expressed as percent of total particle volume. Eight samples from the upper metre of Core 2 and 14 samples from the full depth of Core 3 were analysed.

The capacity for a sediment or soil to respond to a magnetic field is termed its magnetic susceptibility (a dimensionless quantity expressed in SI units) (Dearing 1994). For clastic sediments, such as those forming the dunes and wetlands at Harataonga Bay, variations in magnetic susceptibility values through a sediment sequence will correspond to changes in mineralogical composition. High values are typically given by sediments rich in magnetic minerals such as magnetite (iron sand), and low values represent sediment that is dominated by non-magnetic minerals such as quartz and feldspar (Thompson and Oldfield 1986). Changes in the concentration of magnetic minerals through the sediment column can therefore be used as indicators of changes in sedimentary processes. For example, wave swash action on a beach face and wind on dunes are highly efficient mechanisms for naturally concentrating heavy magnetic minerals, whereas fluvial action is less efficient at doing so (Li and Komar 1992). Cores 2 and 3 were analysed for down-core variations in magnetic susceptibility using a Bartington Instruments Core Scanner (80 mm diam. MS2C sensor). Measurements were taken at 2 cm intervals along each core. This technique does not account for variations in moisture content or mass of the sediment, and therefore provides an order-of-magnitude measure of variation in the relative concentration of magnetic minerals. We use it here to assist with separating the various sediment facies and for inferring sediment sources.

Organic content was determined by loss-on-ignition. Samples from TP 10 were dried overnight in a drying oven at 80°C, ground with mortar and pestle and weighed, then fired in a kiln oven for at least six hours at 420°C and reweighed.

Samples were prepared for pollen analysis by the standard acetylation and hydrofluoric acid method (Moore *et al*. 1991). Results for TP 10 are presented as percentage counts. All taxa are included in the pollen sum, which was at least 75 pollen grains and fern spores; this low figure is due to sparse pollen in some samples. Absolute counts of pollen were made on a section (45 cm in length) in the lower part of Core 2 (at least 255 pollen grains and spores counted for each sample). The percentage pollen count (Horrocks *et al.* 2002b) suggested that this part of the profile encompassed an early stage of human impact at the

site, so we thought that absolute counts might provide insight into the degree of pollen preservation and reworking of sediments during this period. Further samples were thus collected from the core, and at a much higher sampling resolution (mostly every 1 cm). Tablets containing a known quantity of exotic *Lycopodium* spores were added to samples to allow absolute counts. These are given as pollen concentration (i.e., grains/g) rather than pollen accumulation (i.e., grains/area/unit of time), with weight of each sample being measured after initial deflocculation with 10% KOH and sieving (Moore *et al*. 1991).

A conventional radiocarbon age determination was carried out on a sample from TP 10 by the Waikato Radiocarbon Dating Laboratory, University of Waikato, Hamilton (Table 1). AMS radiocarbon age determinations were carried out on samples from the cores by the Rafter Radiocarbon Laboratory, Institute of Geological and Nuclear Sciences, Wellington.

TABLE 1
Radiocarbon dates (AMS) of plant material from Test Pit 10 and Cores 2 and 3

TP/ Core	Lab. no	Depth (cm)	Fraction	^{14}C BP	Cal yr AD (2 sigma)	δ^{13}C
10	WK-11827	180	twig	737±178	1161–1427	−21.6
2	NZA-12665	70	5 *Persicaria decipiens* nuts	646±45	1313–1402	−27.6
2	NZA-12666	100	1 Cyperaceae nut	1534±50	435–656	−27.0
2	NZA-12664	141.5	1 Cyperaceae nut	934±50	1030–1229	−13.2
2	NZA-12667	192	1 Cyperaceae nut	1846±60	86–387	−14.6
2*	NZA-12126	195	undif. small seeds	296±65	1459–1950	−17.0
3	NZA-12668	33	undif. small seeds	1853±75	67–406	−31.6
3*	NZA-12127	125	undif. small seeds	844±55	1053-1296	−26.4

* from Horrocks *et al*. 2002b

Parasitology of the coprolites was carried out by the Institute of Veterinary, Animal and Biomedical Sciences, Massey University. Faecal float procedures outlined in the Manual of Veterinary Parasitological Laboratory Techniques (1986) and Sloss *et al*. (1994) were used. For Coprolites F6, F61 and F62, 1 g was mixed thoroughly with a 33% solution of zinc sulphate (for density separation) then passed through a sieve into a universal bottle. For F1, 0.35 g was used. Additional 33% zinc sulphate was added to the bottle with a Pasteur pipette until a convex meniscus formed at the top. A glass coverslip was then placed on the meniscus. After 20 minutes, the coverslip was placed on a slide and examined at 100x magnification for endoparasite eggs, which would have adhered to the surface tension layer on the coverslip. Parasite eggs in coprolites may come from parasites of the depositing individuals or from parasites of animals that have been eaten by the individuals. As parasites often show high host specificity, this kind of analysis can not only show the type of parasite, but may also offer clues as to what types of animals were part of the diet.

Analysis of bacteria in the coprolites was also carried out by the Institute of Veterinary, Animal and Biomedical Sciences, Massey University. Bacteria were isolated from the coprolites by inoculating each specimen on to the following media:

2 x blood agar (1 incubated in O_2 and the other in CO_2)
chocolate agar
brain-heart infusion broth
cooked meat broth
selenite broth

Each of the broth cultures was sub-cultured after one day and incubated in O_2, CO_2 and anaerobic conditions. A pure culture of each organism was obtained and frozen at −70°C until further analysis. Subsequent tests for the identification of bacteria were carried out using methods given in Quinn *et al*. (1994) and McFaddin (2000). This type of analysis can provide information relating to the health of the individual. The presence of pathogenic bacteria in coprolites is an indication of health. For example, although some species of *Clostridium* are normal symbionts in human digestive tracts, a few species such as gangrene and botulism are known to be pathogenic.

The coprolites were tested for originating species using the trisodium phosphate method (Bryant 1974). Samples were placed in airtight containers in a 0.5% solution of this chemical and left for *c*. 3.5 weeks. Carnivore coprolites usually turn trisodium phosphate white, pale brown or yellow brown and have a musty odour. Herbivore coprolites turn the solution pale yellow to light brown and are musty. Human coprolites turn the solution dark brown or black, change it from translucent to opaque, and unfortunately have the original intense odour. Presence of surface scum indicates that meat was part of the diet.

Full details of the analysis of steroids and human myoglobin in the coprolites are given in Appendix 1. Concentrations of steroid hormones produced by the reproductive system, especially testosterone and estradiol, can be measured in faecal samples to provide information on the sex of the individual (e.g., Cockrem and Rounce 1994, 1995). Faecal steroids have previously been detected in coprolites (Bercovitz and Degraff 1989). Human myoglobin has recently been detected in coprolites from a site in the United States (Marlar *et al*. 2000), with the presence of human myoglobin taken to be evidence of cannibalism.

RESULTS

STREAM SECTION

This profile covers the period from an undetermined pre-settlement time to the present. The bank section is 0.8 m high, fully exposing the terrace stratigraphy from the stream bed to the terrace surface (Fig. 2). The base of the section comprises a bed of pebbly sand of unknown thickness. This is overlain by a 10-cm-thick bed of silty clay mixed with very fine sand. Resting above this is a 17-cm-thick bed of pumice mixed with medium sand. Pumice clasts range from granule to pebble size and are weathered. Above the pumice layer is a 25-cm-thick bed of fine to medium sand. An input of silt and clay forms the upper 29 cm of the terrace profile.

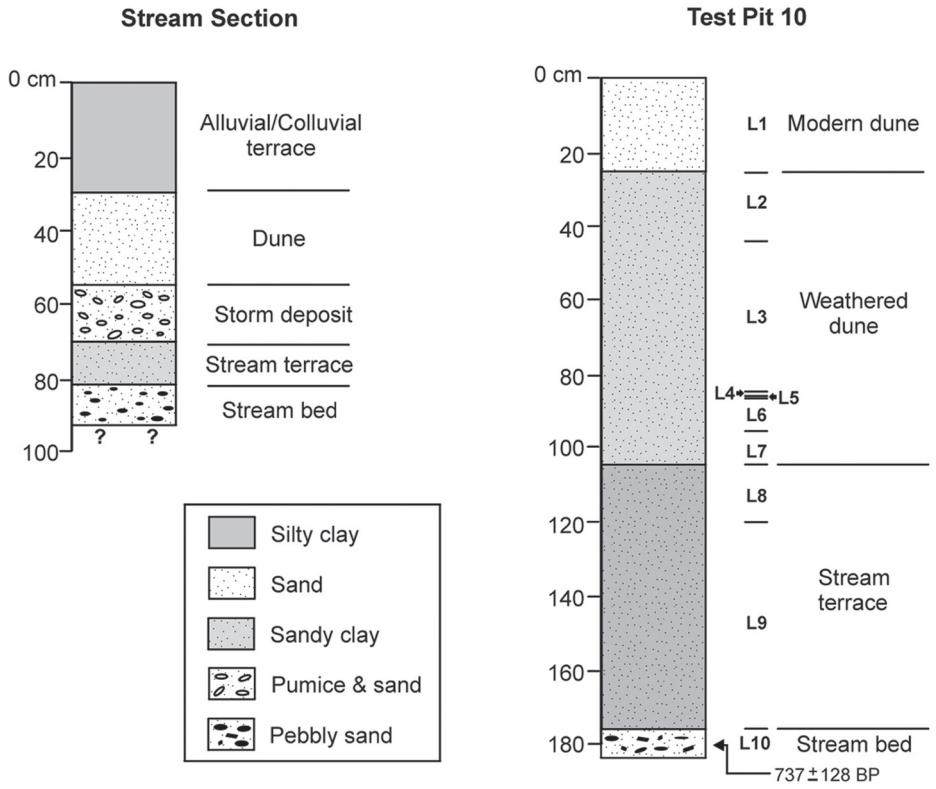

Figure 2: Graphic logs of TP 10 and Stream Section, Harataonga Bay.

TEST PIT 1

This profile comprises dune sand with organic habitation layers (Martin Jones, unpublished data). The pollen sum throughout the profile is dominated by monolete fern spores (Fig. 3). *Cyathea* tree fern spores record moderate levels in the lower two samples but this declines with decreasing depth. Putative truffle (hypogeous Ascomycotina) spores are recorded in all samples in small amounts. All other taxa record very low values, except in the uppermost sample where bracken has a moderately high value.

TEST PIT 10

This profile covers the period from 737 ± 178 ^{14}C yr BP to the present, and comprises 10 layers (Fig. 2). The excavation was placed on the lower part of the dune and attained a depth of 1.85 m. The base of the trench exposed firm grey clay of unknown thickness that is overlain by an 8-cm-thick bed of poorly sorted angular pebbles mixed with wood fragments and other organic detritus. A twig in this deposit provided the aforementioned age determination. This deposit is overlain by a 72-cm-thick bed of grey clayey sand that includes roots and localised, iron-stained (oxidised) mottles. An 80-cm-thick bed of fine to

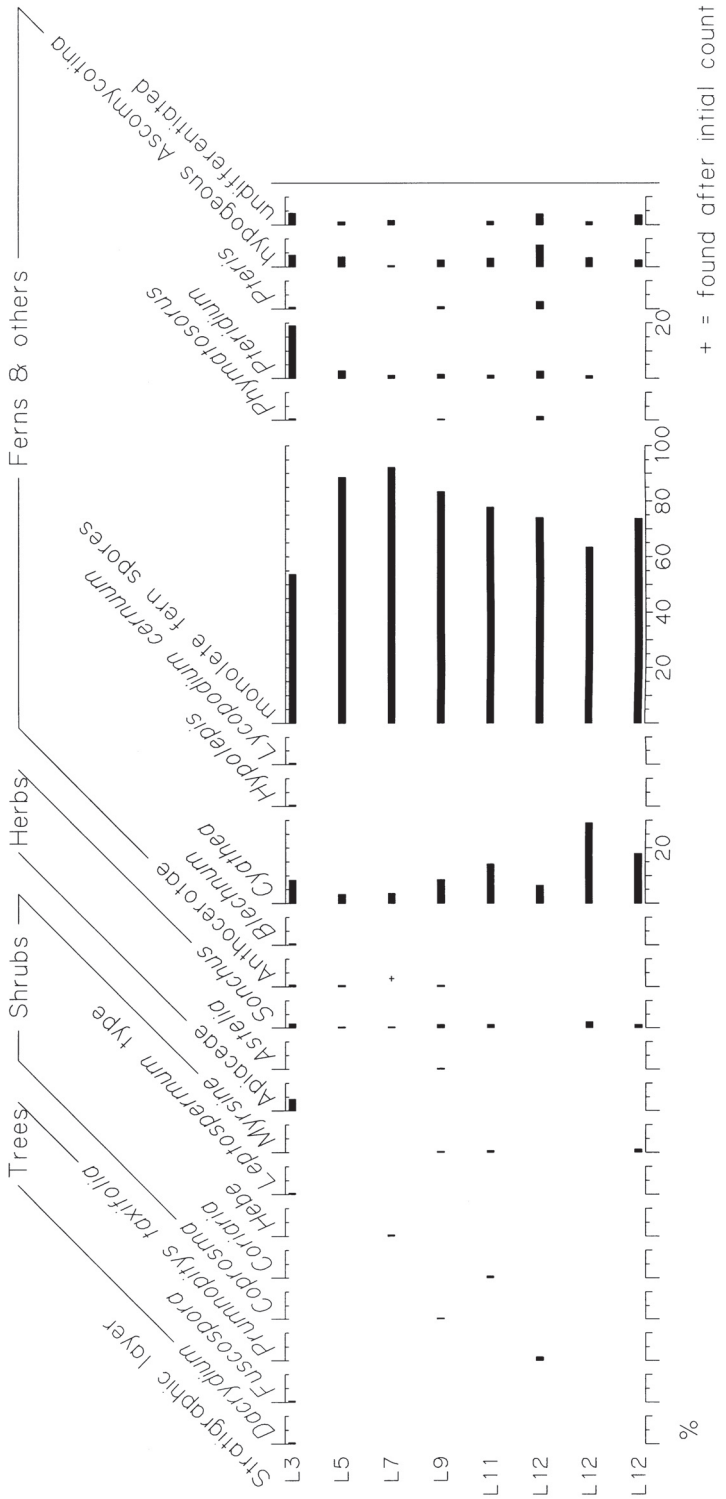

Figure 3: Pollen diagram of TP 1, Harataonga Bay.

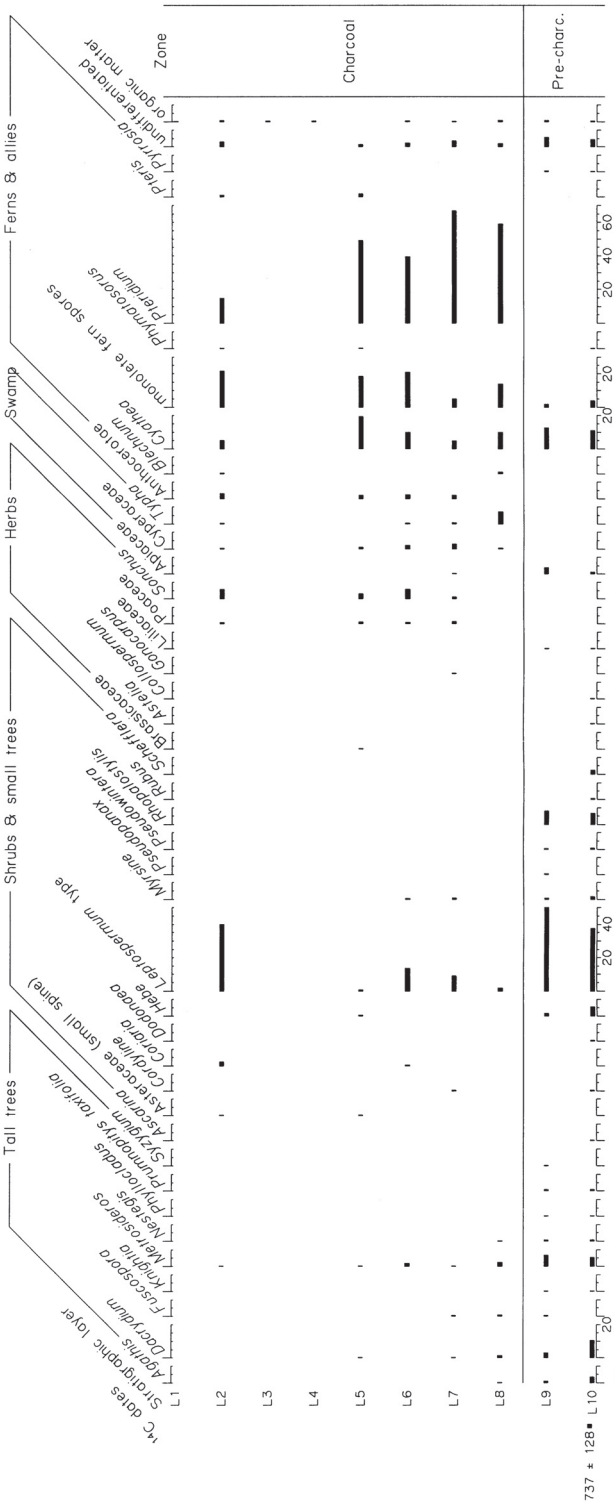

Figure 4: Pollen diagram of TP 10, Harataonga Bay.

medium sand in turn overlies this clayey sand and displays a moderate to strong degree of weathering in the form of oxidation mottles and a weakly-cemented iron pan at 100 cm depth. The sand also contains a 1-cm-thick deposit of very fine sand and silt, at 86 cm depth. The upper 18 cm of the sand is stained grey-brown with fine organic material. The surface layer in this section is 25 cm thick and comprises modern, non-weathered dune sand. Organic matter levels, measured in all layers except Layer 5, are uniformly low throughout the profile.

Samples from Layers 1, 3 and 4 contained insufficient pollen for analysis. The pollen profile is separated into two zones on the basis of absence/presence of microscopic charcoal (Fig. 4).

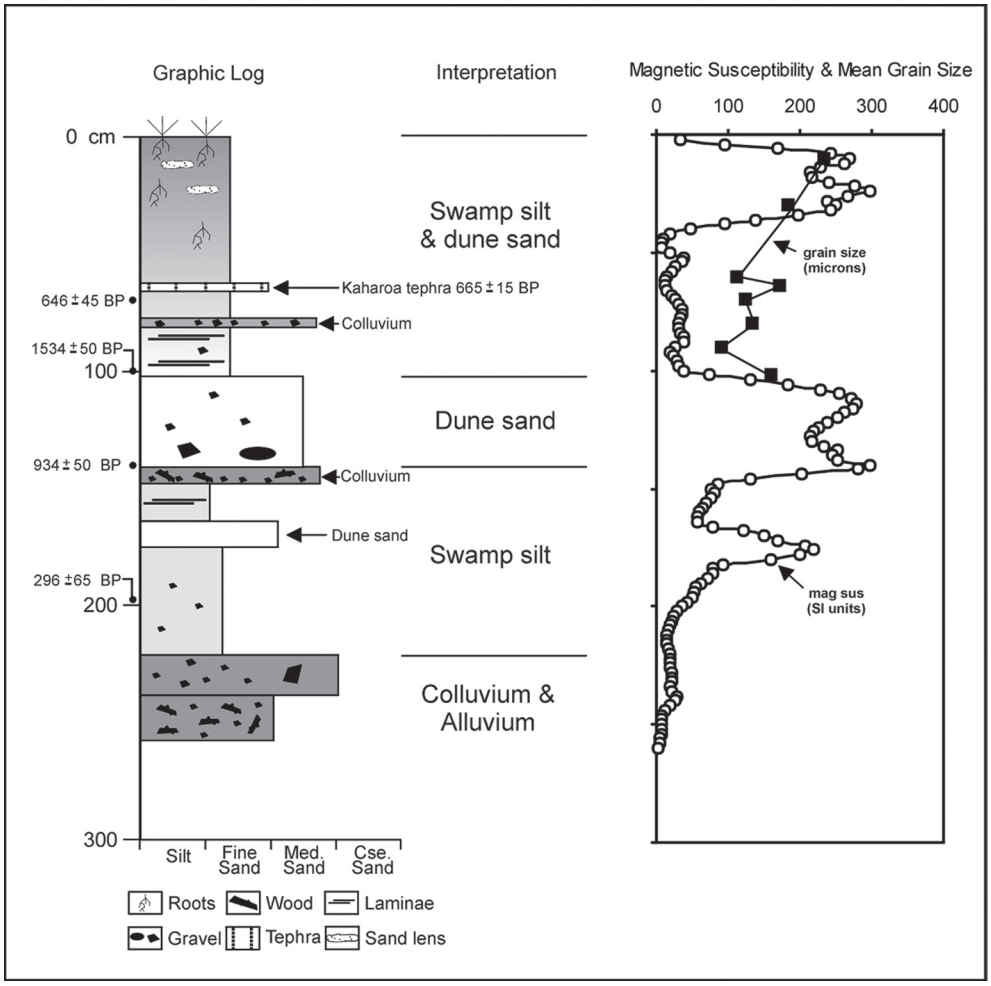

Figure 5: Graphic log, magnetic susceptibility and mean grain size of Core 2, Harataonga Bay.

Pre-charcoal Zone, Layers 10 and 9

The Pre-charcoal Zone is characterised by lack of charcoal, high values for *Leptospermum* type pollen, and significant, albeit low, values for the forest taxa *Agathis, Dacrydium*, *Metrosideros* and *Rhopalostylis*.

Charcoal Zone, Layers 8–1

This zone is characterised by the presence in all samples of abundant charcoal, a dramatic appearance of *Pteridium* spores and a decline in the forest taxa noted in the previous zone. *Leptospermum* type pollen values decline at the lower zone boundary and increase in the uppermost sample. Pollen and spores of the disturbance indicators *Coriaria,* Poaceae*, Sonchus* and Anthocerotae appear for the first time in this zone. Pollen of the swamp taxa Cyperaceae and *Typha* also appear for the first time.

CORE 2

Core 2 corresponds to the Charcoal Zone in the TP 10 pollen profile. It covers the period from an unknown date before deposition (or redeposition, see below) of the *c*. 665 yr BP Kaharoa tephra to an unknown, post-Kaharoa date. However, as European-introduced pollen types were not found in the uppermost sample (Horrocks *et al.* 2002b), the latter is presumably pre-European.

The core recovered a 2.56 m thick deposit of distinctly heterogeneous sediments ranging from fine-grained organic silts to sand and gravel (Fig. 5). These sediment types are preserved as clear beds through the core. The base of the core comprises a 36-cm-thick bed of poorly sorted, sandy silt mixed with coarse angular gravel and macroscopic organic remains. Magnetic susceptibility values through this bed are consistently below 30 and the lowest in the core. An abrupt transition at 2.2 m depth is defined by a decrease in the concentration of gravel and increase in the proportion of silt and very fine sand. Gravel fragments become rare and organic fragments are concentrated locally as thin laminae. This bed extends to 1.47 m depth, but is interrupted at 1.74 m by a 12-cm-thick interbed of well sorted fine sand. Magnetic susceptibility values through the silty sand range between 20 and 100, but peak abruptly at 220 within the sand interbed. A second interbed of well sorted sand mixed with silt and gravel is preserved between 1.03 m and 1.47 m depth. The gravel fraction includes well rounded and angular clasts up to 3 cm in length. This interbed also yielded high magnetic susceptibility values, ranging from 130 to 300. The upper metre of Core 2 comprises silty fine sand with organic fragments throughout that are preserved as matted silty peat beds below 50 cm depth. Within this silty peat, two well defined sandy interbeds are preserved, at 80 cm and 66 cm depth. The deeper interbed is 4 cm thick and consists of sand and angular gravel with a low magnetic susceptibility (<35). The upper interbed, comprising 665 yr BP Kaharoa tephra (Fig. 6), is a 4 cm deposit of fine silty sand that also yields low magnetic susceptibility values. Above this point, the susceptibility profile rises to maximum values of 300 within the upper 30 cm of the core, where fine sand is diffused through the silts and organics.

Results of pollen concentration analysis from high resolution sampling in the lower part of Core 2 are given in Figures 7 and 8. This analysis covers the lowermost part of the core, from 215 cm to 170 cm depth. Only taxa relevant to our discussion of the local environment and human impact on this are included. Total pollen concentration is very low in the

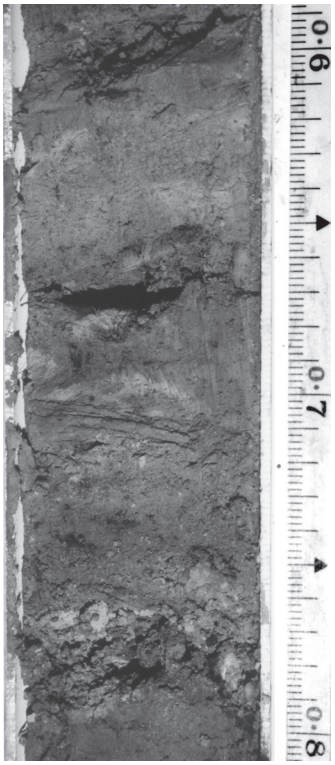

Figure 6: Kaharoa tephra in Core 2, Harataonga Bay, present as a light-coloured layer at 62–66 cm depth (scale in m).

lowermost samples (2000 grains/g) but starts increasing (up to 98,000 grains/g) midway through the profile. The profile is overwhelmingly dominated by spores of a few fern taxa, mainly *Cyathea* (up to 52,300 grains/g) and *Pteridium* (up to 41,300 grains/g), and also Anthocerotae (up to 10,700 grains/g) and monolete ferns (up to 8200 grains/g). *Cyathea* spores are more highly concentrated than *Pteridium* spores in the lower section of the core. The next most abundant group of taxa (Asteraceae, *Coprosma, Coriaria, Leptospermum, Gonocarpus,* Poaceae, *Sonchus*, Cyperaceae, *Typha, Hypolepis* and *Pteris*) record <5000 grains/g. Remaining taxa (*Cordyline, Schefflera, Syzygium, Acaena,* Brassicaceae, *Rumex, Phormium, Blechnum* and *Phylloglossum drummondii*) record < 250 grains/g.

CORE 3

Core 3 covers the period from an undetermined date during Polynesian occupation before deposition (or redeposition) of the *c*. 665 yr BP Kaharoa tephra (at 20–25 cm depth) to immediately after its deposition. The core provided a 1.3-m-thick sample of dominantly sandy sediment, divided into two beds (Fig. 9). The lower bed is 90 cm thick and is characterised by massive silty, fine to medium sand with local gravel clasts and concentrations of rafted organic material. Magnetic susceptibility values are highest in this bed, ranging from 120 to 270, and include two peaks associated with medium to coarse sand of low silt content (mean: 300–340 μm, 5% silt). A sharp surface at 40 cm depth defines

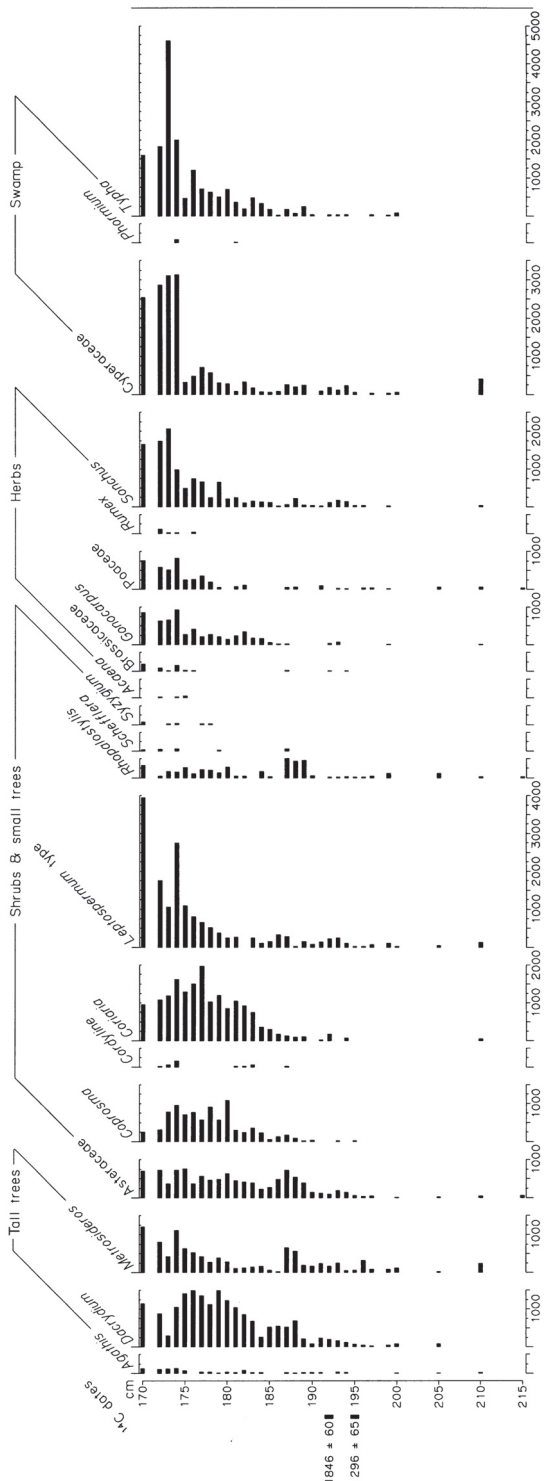

Figure 7: Pollen diagram (absolute counts) of lowermost section of Core 2 (part 1).

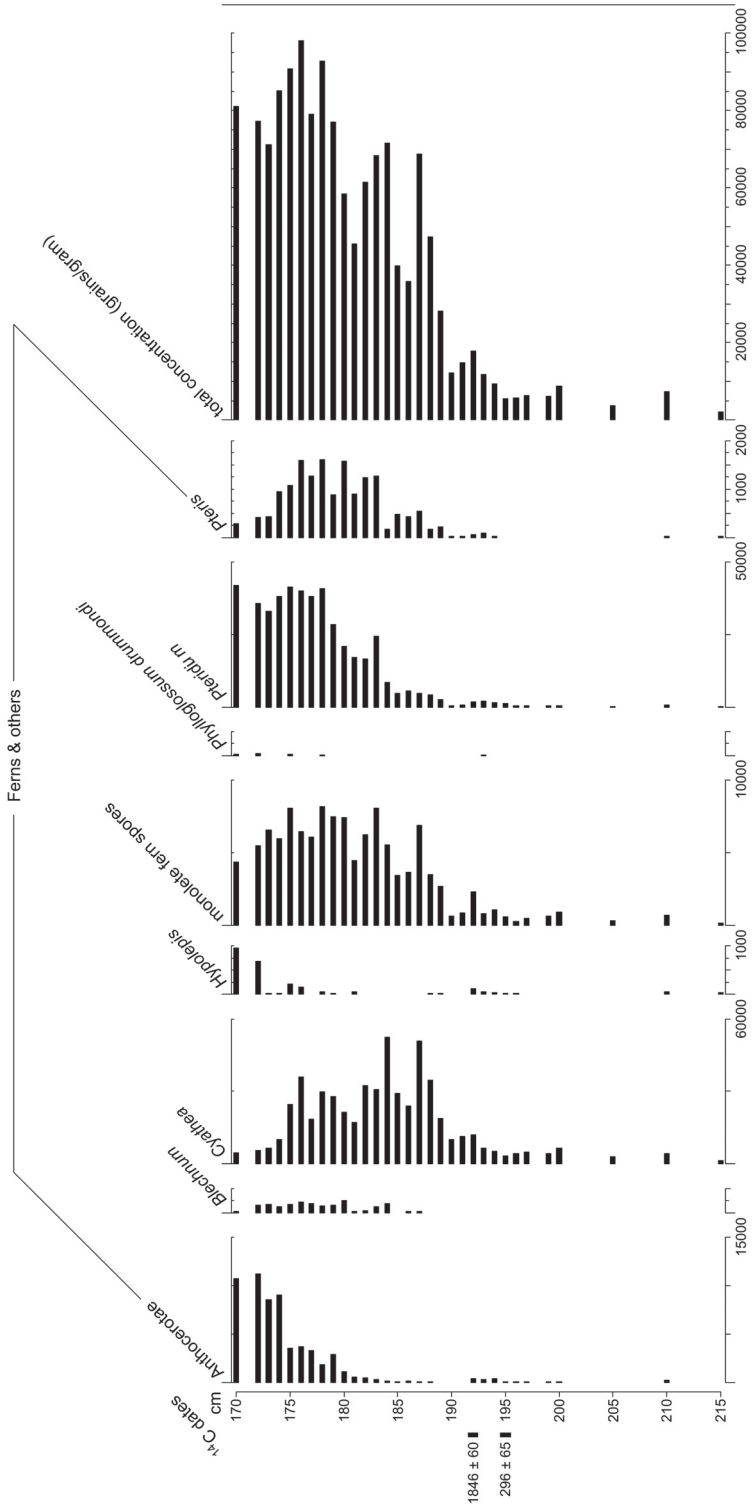

Figure 8: Pollen diagram (absolute counts) of lowermost section of Core 2, part 2.

the contact with the upper bed of this core. This upper bed comprises massive fine sand with higher silt content than the underlying sand (15–40% silt). Modern plant roots penetrate to 30 cm depth, with negligible gravel and rafted organic fragments. This bed also preserves isolated lenses of Kaharoa tephra 1–2 cm in diameter between 20 cm and 25 cm depth. The magnetic susceptibility profile of this bed decreases to <10 at the surface from a maximum of 90 at 40 cm depth. Microscopic charcoal was noted throughout the profile.

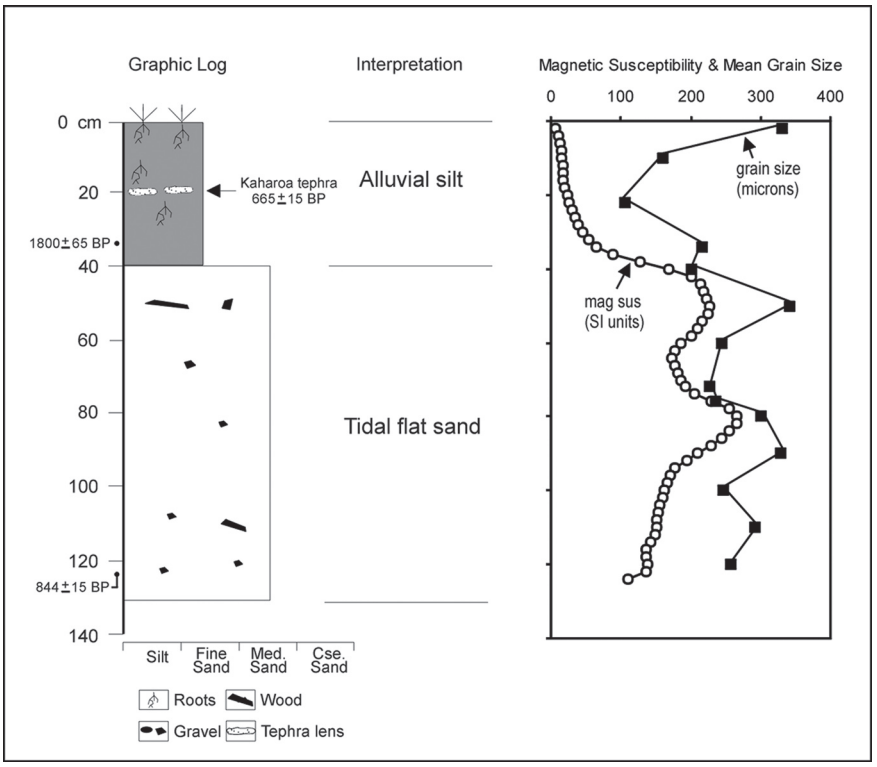

Figure 9: Graphic log, magnetic susceptibility and mean grain size of Core 3, Harataonga Bay.

COPROLITES

Coprolites F1, F6, F61 and F62 were found as two pairs directly on successive organic habitation surfaces in TP1. The first coprolite pair was on Layer 5 (see Methods section for layer depths), the other pair was on Layer 7. Samples of both pairs were 5 cm apart at their closest point. Coprolite 5 was found on a deeper organic habitation surface (Layer 11).

After soaking in the sodium triphosphate solution to test for originating species, the F6 solution had a musty odour and was pale yellow. The F62 solution was odourless and also pale yellow. Neither solution had developed surface scum or changed from translucent to opaque.

Endoparasite eggs were not detected in the coprolites. Bacteria isolated from the two coprolite samples analysed (F6 and F62) were as follows:

Pseudomonas fluorescens/*putida*
Pseudomonas spp.
Kurthia spp.
Bacillus spp.
Acinetobacter baumanii
Shingomonas paucimobilis
Brevibacillus brevis

Testosterone and estradiol were not detected in the coprolites (see Appendix 1 for details). Extracts of fresh human faeces did contain steroids and a test of parallelism indicates that the testosterone and estradiol assays were valid for the measurement of the steroids in the coprolites. The faecal extracts had to be diluted by factors of two or four before the serial dilutions were parallel to the standard curves. This indicates that a substance in the extracted sample cross-reacted with the steroid antibody but that at higher dilutions the substance did not interfere and the assays were valid for steroid measurements. The measured steroid concentrations in the fresh human samples were of the same order of magnitude as concentrations reported for testosterone in male (Strier *et al*. 1999) and estradiol in female (Ziegler *et al*. 1996) non-human primates. This result supports the suitability of our extraction and assay methods for the measurement of testosterone and estradiol in human faecal samples, and it is likely that if steroids were present in coprolites they would be detected by our assay methods.

DISCUSSION

The two profiles from the eastern end of the beach (TP 10 and Core 2) include the first signs of human impact in the near shore environments at Harataonga Bay. For the start of the observed sequence of environmental change, the date of 296 ± 65 ^{14}C yr BP of small seeds at the base of Core 2 (Fig. 5) is unreliable because its age and δ^{13}C value (−17.0) strongly suggest contamination with younger carbon (Chris Prior, pers. comm.). Likewise, the date of 1846 ± 60 ^{14}C yr BP of a Cyperaceae nut immediately above this is also unreliable because it is anomalously old, probably a result of reworking, as it coincides with presumably much later signals of large-scale deforestation (i.e., charcoal and *Pteridium* spores). We take the date of 737 ± 178 ^{14}C yr BP of the twig at the base of TP 10, in pre-charcoal deposits, as a reliable maximum age for the beginning of the observed sequence (Fig. 2). As we interpret the charcoal as a result of deforestation by early people, this date supports Law's (1972) suggested date of about the thirteenth century AD for the Western Midden at the opposite end of the beach and is consistent with the Core 1 pollen record (Horrocks *et al.* 2002a), in which abundant charcoal and *Pteridium* spores occur shortly after deposition of the *c*. 665 yr BP Kaharoa tephra. In a wider context, the new (and previous) Harataonga data are consistent with pollen records from elsewhere on Great Barrier Island (Horrocks *et al.* 1999; 2000a, 2000b; Ogden *et al.* 2006) and from radiocarbon data from archaeological sites throughout New Zealand indicating Polynesian settlement no earlier than *c*. 700 yr BP (Higham and Jones 2004).

The basal material in TP 10 is interpreted as a combination of colluvial and alluvial sediments deposited on the floor of a former stream. These deposits are most likely to be the lateral continuation of the modern stream (much smaller than the main watercourse at the opposite end of the beach), indicating stream migration in association with dune

formation. The pollen evidence suggests that *Leptospermum/Kunzea* scrub grew on the terrace flanking the stream (Fig. 4). The canopy of surrounding, largely undisturbed, forest comprised *Metrosideros* and emergent *Dacrydium* and *Agathis,* with *Cyathea* tree ferns and *Rhopalostylis* palms a significant part of the understorey.

The varied sediment texture of the deposit on top of the stream sediments in TP 10 (72 cm thick, Fig. 2), indicates mixing of material from a variety of sources, in this case marine (i.e., beach) and terrestrial. Given the proximity to a stream channel, this mixing would have occurred both in the stream and on the flanking terrace. At some undetermined time after 737 ± 178 ^{14}C yr BP, during terrace accretion, *Leptospermum/Kunzea* cover declined and large-scale burning of surrounding forest occurred (Fig. 4). Many of the pollen types recorded in the profiles indicate major anthropogenic disturbance to vegetation, with seral taxa now predominating. *Pteridium* fern became a major part of the hillside and possibly dune vegetation. Other disturbance-related plants, notably *Hypolepis* and *Pteris* ferns, and Poaceae and *Sonchus* herbs were also a feature of this new landscape. High values of Anthocerotae spores indicate large areas of disturbed soil.

The aforementioned disturbance marks the end of vertical accretion of the terrace surface, which was followed by a period of dune formation. This is evident in the 80 cm thick bed of fine to medium sand in TP 10 that overlies the sandy clay of the terrace (Fig. 2). The weathered sand indicates a fluctuating water table, with an iron pan forming at the saturated layer perched above the less permeable fluvial terrace sediments. Evidence for dune movement into the backswamp area is also preserved in Core 2, as discrete beds of wind-blown sand (Fig. 5). The appearance of Cyperaceae, *Typha* and *Phormium* pollen indicates development of swampy conditions, with the latter two suggesting eutrophication. *Cordyline* and *Syzygium* trees and *Gonocarpus* and Brassicaceae herbs were also part of the swamp vegetation. The transition to swamp required partial blocking of the stream by dunes. Such episodes of aeolian sand transport can be readily triggered by natural process such as storm events. Human disturbance of dune surfaces and vegetation cover is equally possible, and the presence of living surfaces buried within the dunes is compelling evidence that people were responsible for impeding the stream flow. The presence of *Lagenaria siceraria* (gourd) and *Ipomoea batatas* (kūmara) microfossils in Core 2 and the coprolites from TP 1 (Horrocks *et al.* 2002b, 2004b) indicates that these introduced cultigens were grown at Harataonga during Maori occupation and suggests forest clearance for agriculture. A kūmara storage pit in the Pā at the eastern end of Harataonga Bay is discussed by Law (1972, 1975).

The 1-cm-thick deposit of very fine sand and silt at 86 cm depth within the sand layer in TP 10 (Layer 5, Fig. 2) at first glance appeared similar to layers of Kaharoa tephra found in the sediment cores from the catchment. However, subsequent analysis showed absence of glass shards in this particular layer. The humic-stained sand of the upper 18 cm of dune sand (Layer 2) indicates a soil humus horizon in the dune profile, which is probably due to stabilisation by *Leptospermum/Kunzea* scrub and ground ferns. The stabilised dune was subsequently buried by the modern dune (Layer 1).

In Core 2, the preservation of well defined interbeds of fine-grained sediments with coarse gravels and sands within the context of a backdune swamp is strong evidence for mixing of sediments derived from multiple sources (Fig. 5). As with TP 10, here the sources of clastic sediments are recognised as the immediate hill slopes that drain into the swamp and the beach-dune system a few metres to seaward. The former will supply sediment ranging from silts to gravel and organic fragments washed into the swamp as a mix of alluvial and colluvial materials. In contrast, the dunes will mostly supply well-sorted sand. Biogenic

sediments in the form of peat reflect the *in situ* product of wetland plant growth. In Core 2, therefore, the basal bed of silt and sand mixed with angular gravel is interpreted as colluvial sediment that formed the floor of the swamp with the overlying silt-dominated bed recording swamp filling via alluvial deposition to 147 cm depth. The higher concentration of *Cyathea* compared to *Pteridium* spores in the lower section (Fig. 8), consistent with higher *Cyathea* percentages (Horrocks *et al.* 2002b), is probably in part due to differential preservation in the highly oxidising conditions of the stream bed — fern spores are more resistant to decay (Wilmshurst and McGlone 2005). The interbed of well sorted sand with high magnetic susceptibility represents a temporary departure from low energy sedimentation (Fig. 5). The key to the interpretation of the origin of this sand is the magnetic susceptibility peak. We interpret this to represent an increase in the concentration of heavy minerals derived from the beach dunes. The well-sorted texture of the sand is also characteristic of a wind-blown deposit. Further, both the degree of sorting and magnetic susceptibility are in strong contrast to the enclosing alluvial-swamp sediments.

A second deposit of beach dune sand in Core 2 is recognised in the interbed from 1.03 to 1.47 m depth (Fig. 5). Supply of wind-blown beach dune sand is recorded in the high relative magnetic susceptibility values through this interval but the inclusion of large angular and rounded gravel clasts toward the base of this bed indicates some contribution from slopewash (colluvium). It is also possible that these gravels were placed in the swamp by people, as the *Lagenaria siceraria* pollen at the base of this interbed (Horrocks *et al.* 2002b) indicates their presence at this time. Indeed, disturbance of surrounding forest by human activity would probably have led to increased aeolian transport of dune sand into the back-swamp, and increased slopewash from the adjacent hillside.

The upper metre of Core 2 records a period of relatively low energy sedimentation in the swamp, dominated by the supply of alluvial silts, minor amounts of wind blown fine sand and peat production by local plants (Fig. 5). The only evidence for interruption to these depositional conditions is the two sand interbeds. The deeper interbed incorporating angular gravel and yielding a low magnetic susceptibility is therefore interpreted as a slopewash deposit possibly laid down during a storm. The second interbed is Kaharoa tephra, following deposition of which the swamp has continued to collect fine-grained sediments from the hillslopes and to produce peat. The up-profile increase in pollen concentration reflects better preservation of organic matter and/or a decrease in the sedimentation rate, with more pollen grains and spores contained in fewer deposited particles. The latter suggests decreased disturbance of vegetation on the surrounding hill slopes. The significant peak in magnetic susceptibility within the upper 30 cm of the core indicates that the input of fine dune sand has increased during post-Kaharoa time (Fig. 5).

Horrocks *et al.* (2002b) considered that the stratigraphic position of the 665 ± 15 ^{14}C yr BP Kaharoa tephra in Core 2 well above signals of human disturbance (charcoal, *Pteridium* spores and *Lagenaria* pollen) suggested human activity at Harataonga well before this date. However, the tephra layer had a variety of glass shards, was discontinuous and lacked biotite, very strongly suggesting reworking. Further, the sediments above 80 cm depth in this core, which include the tephra layer, appear mixed and cannot be identified as discrete beds as readily as the sediments below this depth. The inverted radiocarbon dates in the central section of the core attest to this (Fig. 5). On the other hand, the radiocarbon date immediately below the tephra layer (646 ± 45 ^{14}C yr BP) is consistent with the date of the eruption. Also, we looked for background microscopic occurrences of dispersed tephra glass from samples of sediment below the tephra layer (at depths of 50 cm, 55 cm, 70 cm, 75 cm, 125 cm and 190 cm), and found none. If the tephra layer is a secondary rather than primary

deposit, we would have expected to find glass in at least one of these samples. However, we emphasise that the occurrence of a variety of glass shards in the tephra layer does indicate reworking has occurred. We assume that the deposition of the tephra occurred at about the same time as or some time after the Kaharoa eruption. Although the abundance of Kaharoa glass in the analysed sample suggests deposition soon after the eruption, at this stage it is impossible to determine precisely when. Regardless, the radiocarbon and pollen evidence from TP 10 (Fig. 4) strongly suggests that people were not active in the bay before 737 ± 128 ^{14}C yr BP.

Profiles from the opposite, western end of the beach (Stream Section, TP 1 and Core 3) show similar processes of landscape development. The pebbly sand base of the Stream Section site (Fig. 2) represents the lateral continuation of the modern stream bed beneath the modern terrace. The bed of silty clay mixed with very fine sand directly above this is interpreted as a low levee bank deposit formed by fluvial processes. The formation of a levee above the old stream bed indicates local narrowing or migration of the channel. The pumice-sand bed above this represents a combination of sea-rafted pumice (source unknown) and beach sand, and therefore signals the first delivery of marine-derived sediments to the terrace. It is possible that this bed is a storm deposit, which has been preserved until now because of its previously sheltered location away from the active shoreline. The supply of marine sand continued with the subsequent deposition of the fine to medium sand above the pumice layer. The finer grain size and uniform texture of this deposit is evidence for deposition by aeolian mechanisms, forming a low dune alongside the stream. The elevation of this sand bed is considered too high for it to be a beach deposit. The supply of wind-blown sand to this site ended with the input of the silt and clay forming the upper layer of the terrace deposit. This material is clearly terrestrial in origin, and is most likely to be a combination of local slopewash material and overbank stream sediments. This mix is assumed to be a result of anthropogenic disturbance of the vegetation (repeated firing) on hill slopes of the catchment.

Nearby at TP 1 on the modern dune, successive organic-rich layers in the profile indicate alternating periods of dune stability and instability. The pollen evidence suggests that during the stable periods the dunes were covered in ground ferns (monolete), most likely *Hypolepis* and possibly *Paesia,* both colonists of disturbed ground (Fig. 3). (*Hypolepis* is identified in the top sample because the distinctive perine has not been lost.) *Cyathea* tree ferns were a significant part of surrounding vegetation. Low values for *Pteridium* spores in all but the uppermost sample suggest that most of the sequence represents a period before large-scale deforestation of the catchment. Alternatively this may simply reflect distance from extensive stands of *Pteridium* or over-representation of other ground ferns due to stands of high density.

Two kinds of sedimentation are recognised from the sequence recovered from the stream terrace at Core Site 3, further inland (Fig. 9). The lower sand-dominated bed is interpreted as a tidal flat deposit on the basis of the low silt content, presence of rafted organic material and relatively high magnetic susceptibility. Together, these properties indicate processes of sediment sorting that are characteristic of a tidal environment. Specifically, the regular flood and ebb of tidal currents across shallow intertidal surfaces will cause winnowing out of fine-grained particles (silt and clay), resulting in sand-dominated deposits. Rafting and deposition of organic detritus also occur under these conditions, with larger fragments placed on strandlines at the tidal limit and in shallow tidal pools. Local concentrations of magnetically susceptible minerals (iron sands) in this instance are associated with slightly coarser sands that together indicate marginally higher depositional energy, such as would develop during

a large spring tide and/or storm surge. In either event, these sands will have been sourced from the beach, where medium to coarse, heavy-mineral bearing sands are available.

The surficial bed in Core 3 forming the stream terrace represents the transition from a tidal to supratidal depositional environment. The increased silt content, decrease in sand size, preservation of root traces and deposits of Kaharoa tephra (reworked) are all criteria that indicate accretion of the sediment surface to a permanently supratidal elevation (Fig. 9). The process of accretion would have involved a gradual decline in the role of tidal processes and a gradual increase in fluvial processes, with stream flooding becoming the only agent for sediment transport to the terrace surface. Hence the increase in silt content toward the modern surface.

The pollen evidence from Core 3 indicated that *Pteridium,* not surprisingly, was a major component of the hillside vegetation at this end of the beach also (Horrocks *et al*. 2002b). As with Core 2 from the eastern end, the swamp taxa Cyperaceae and *Typha* appeared later in the sequence and the tephra evidence (lack of biotite) showed reworking. Further, the new (upper) date we give for this core is inverted (Fig. 9). The tephra layer would have been deposited on a surface that was open to inundation from overbank stream flow, and perhaps storm tides. However, we looked for background microscopic occurrences of dispersed Kaharoa glass from samples of sediment below the tephra layer at depths of 30 cm, 35 cm, 40 cm, 45 cm and 85 cm and, as with Core 2, found none. The presence of the tephra layer close to the surface (20–25 cm depth) is probably in part due to sediment shrinkage from artificial drainage of the area. European-introduced *Pinus* pollen, which cannot have been deposited before the early nineteenth century, was found in the sample immediately below this layer, showing mixing of sediments.

A radiocarbon date from a few centimetres above the upper coprolite pair in TP 1 suggested a minimum age of 467 ± 60 ^{14}C yr BP for the Harataonga coprolites (Horrocks *et al*. 2002a). In a prehistoric New Zealand context, this can be narrowed down to Polynesian people or their introduced, domesticated dog. However, there may be difficulty in determining between the two because their diets have considerable potential to overlap, with dogs feeding on scraps. Macrofossils are potentially useful, but the coprolites did not provide any evidence in this respect, such as large bone fragments indicating canine origin. The trisodium phosphate test (Bryant 1974) suggests that the coprolites are neither human nor dog in origin (and also that meat was not part of the diet at the time). Instead, they suggest herbivore origin. However, because of poor preservation of organic material in the coprolites, we consider that the probability of their having lost much of their biochemical constituency is high (see also below for lack of steroids and myoglobin, and endoparasite eggs). We thus treat the sodium triphosphate results cautiously, particularly as the method was developed with North American coprolites preserved in very different conditions (dry caves).

All twelve species of bacteria isolated from the coprolites are widely distributed in the environment, including water and soil, and are not specifically of faecal origin. *Kurthia* spp., *Pseudomonas* spp. and *Acinetobacter baumanii* in particular are all organisms that occur as part of the normal gut flora of humans and many other animals. None of the isolated species are considered to be clinically significant or pathogenic to humans. As all of these organisms occur so commonly in a wide range of environments it is impossible to determine if their origin in these specimens was faecal or part of the surrounding soil.

The apparent absence of endoparasite eggs from the coprolites is due to either lack of parasites in the individuals or poor preservation of organic material in the coprolites. Myoglobin is a protein and therefore species-specific, so the lack of human myoglobin in

the coprolites would be expected if the coprolites were of dog origin. However, the lack of steroids is almost certainly a result of poor preservation because steroids are not species specific.

SUMMARY

The evidence presented here for localised human impact on the Harataonga coastal landscape is representative of two different sub-environments: dunes and alluvial plain. Each responded in different ways to human activity. The former is highlighted by strong evidence for dune instability (TP 1 and 10) at the same time as a major increase in disturbance-related plants (especially ground ferns) coupled with the cultivation of introduced plants, notably *Lagenaria siceraria* and *Ipomoea batatas* in Core 2 and coprolites (Horrocks *et al.* 2002a, 2004b). At the eastern end of the beach, this period of dune instability led to infilling of a former stream channel, ponding of stream flow behind the dune to form a swamp, and further infilling with sand and slopewash sediments. The lack of a coherent age profile through the swamp stratigraphy (Core 2) is strongly indicative of a highly disturbed sedimentary sub-system. The evidence all very strongly suggests that the present form of much of the Harataonga dunes, and of the back swamp at the eastern end of the beach, is directly a product of human impact. The timing for this appears to be no earlier than 737 ± 178 ^{14}C yr BP.

The sedimentary record from the alluvial plain of the main watercourse, at the western end of the beach, appears less sensitive to human impact. That is, in the context of a former tidal inlet that would have been open to storm surge and stream floods it is difficult to resolve sedimentary inputs that directly relate to human presence. The sequences reported from the Stream Section at the mouth of Harataonga Stream and Core 3 further landward are largely made up of deposits that typically occur in such environments. The only exception would be the surficial slopewash materials forming the terrace surface. These sediments bear pollen that is consistent with vegetation disturbance. Our conclusion would therefore be that the landforms are natural but the *rate* at which the tidal inlet was infilled to form a terrace was probably accelerated by human presence.

The nature and timing of the localised human impacts at Harataonga are consistent with those observed elsewhere on Great Barrier Island and mainland New Zealand. Some of our techniques (e.g., bacteria, steroids) are newly applied to coprolites in New Zealand but none provided any useful information because of poor preservation.

ACKNOWLEDGMENTS

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APPENDIX 1

Measurement of estradiol and testosterone concentrations and human myoglogin in the Harataonga coprolites

METHODS

Dried faecal material was separated from sand and each faecal sample then crushed to a fine powder with a spatula. Samples of fresh human faecal material were also obtained for comparison with the coprolites. The fresh samples (one male and one female) were freeze-dried and crushed to a fine powder.

Steroid hormones present in the faecal samples were extracted into ethanol. 0.05 g of dried powdered sample was transferred to a glass screw top tube, 2.5 ml of 90% ethanol added, and tubes were vortexed for 60 seconds. The tubes were shaken horizontally for one hour on an orbital shaker and then vortexed for another 60 seconds. The tubes were capped and samples were centrifuged for 20 minutes at 1900 g (Beckman TJ-6 centrifuge). The supernatant was pipetted into a second glass tube. A further 1.25 ml of 90% ethanol was added to the pellet, which was vortexed for 60 seconds, and centrifuged for 20 minutes at 1900 g. The supernatant was added to that from the previous spin, and the pellet was discarded. The ethanol extracts were dried under a stream of air in a heating block at 37°C. The sides of the tubes were rinsed with 90% ethanol twice during this process to ensure that the final level of extract residue was below the level of 1000 µl, then allowed to cool before being reconstituted in 1 ml of phosphate-buffered saline with gelatine (PBSG; 0.1M, pH 7.0) and left overnight at 4°C. The next day, samples were vortexed for 30 seconds, shaken for one hour at room temperature, revortexed, and left overnight again at 4°C. After the second night of refrigeration, samples were vortexed, and the extract transferred into a 1.5 ml polypropylene eppendorf tube and centrifuged for 10 minutes at 14,000 g (IEC Micromax ventilated microcentrifuge OM3590). The supernatant was transferred into a 1 ml polypropylene titre tube, and frozen at −20°C.

The recovery of steroid during the extraction process was measured by adding 100 µl of tritiated steroid (Amersham; 5000 cpm) to each sample before shaking. Aliquots of tritiated steroid were also placed in scintillation vials for measurement of the total counts. A 100 µl aliquot of the reconstituted extract was removed after the extraction was completed and used to determine the percentage recovery of steroid.

A modification of the extraction procedure was used for one of the coprolite samples after the initial results showed that there was no detectable steroid in the coprolites. Our standard method for faecal steroid extraction was modified to increase the sensitivity of the method by a factor of 10. 1.00 g of dried sample was used instead of 0.05 g, 5 ml instead of 2.5 ml of 90% ethanol was added, and the final dried ethanol extract was reconstituted in 2100 µl PBSG instead of 1000 µl.

Testosterone concentrations in faecal extracts were measured by radioimmunoassay using a modification of the method described by Wingfield *et al*. (1997). Samples were assayed in duplicate. Reconstituted extracts were incubated with 100 µl of antibody (Endocrine Sciences, California; testosterone antiserum T3-125) and 100 µl of tritiated testosterone (approximately 5000 cpm; ^{3}H-testosterone TRK.406 Amersham, UK) at 4°C overnight. The cross-reactions of the antibody with other steroids tested by Endocrine Sciences were dihydrotestosterone (20%), corticosterone (<0.01%), oestradiol (0.14%), Δ-1-testosterone (52%), 4-androsten-3β-17β-diol (3%), 5α-androsten-3β-17β-diol (1.8%), Δ-4-androstenedione (0.5%), and other steroids (<0.5%). Bound and free steroids were separated

by the addition of 500 µl dextran-coated charcoal (2.5 g/l charcoal, 0.25 g/l dextran (Dextran T70, Amersham Pharmacia) in PBSG) for 15 min at 4°C. Tubes were then centrifuged at 4500 g for 15 minutes at 4°C and the supernatant poured off. 3 ml of scintillant (5 g/l PPO (2,5-diphenyl-oxazole, Sigma), 0.3 g/l dimethyl POPOP (1,4-bis-[4-methyl-5-phenyl-2-oxazoly]-benzene, Sigma) in toluene) was added, the samples were shaken for one hour, left for one hour at room temperature, and then each sample was counted for five minutes in a Wallace 1409-411 liquid scintillation counter.

Faecal hormone concentrations were measured in diluted PBSG buffer extracts. Buffer had been added to the droppings in proportion to their dry weight, so the hormone concentrations in buffer were directly proportional to the amount of steroid per unit dry weight of the original dropping. The raw assay results were therefore converted to give final values as ng steroid/g dry weight dropping. The conversion included multiplication factors for the dilution of the buffer and a correction factor for the recovery of labelled steroid in the sample preparation procedure.

The limit of sensitivity of the radioimmunoassay, defined as the smallest amount of steroid on the standard curve distinguishable from the zero hormone tube and expressed as testosterone concentration in dried faeces, was 0.41 ng/g for the standard extracts and 0.06 ng/g for the 1.00 g extract.

Solutions of testosterone in PBSG were prepared at concentrations that gave approximately 20, 50, and 80% binding on the standard curve. These solutions were used as high, medium and low quality control samples in every assay. The intra-assay coefficients of variation for these three samples were 19.3, 7.8, and 5.5% (n=20). The inter-assay coefficients of variation for these three samples in 10 assays were 8.1, 13.4, and 13.5% (n=10).

Estradiol concentrations in extracted quail plasma were measured by radioimmunoassay using the same method as that described for testosterone. Samples were assayed in duplicate. Reconstituted extracts were incubated with 100 µl of antibody (Dr R.J. Etches, University of Guelph, Canada; estradiol antiserum 41–12 raised in sheep) and 100 µl of tritiated oestradiol (approximately 5000 cpm; ^{3}H-estradiol TRK.322 Amersham, UK) at 4°C overnight. Cross reactions of the antibody with other steroids were estradiol-17β (<1.0%), estriol (<1.0%), and estrone (4.3%; Etches *et al*. 1981).

Faecal hormone concentrations were measured following the methods used for testosterone. The limit of sensitivity of the radioimmunoassay, defined as the smallest amount of steroid on the standard curve distinguishable from the zero hormone tube and expressed as testosterone concentration in dried faeces, was 1.07 ng/g for the standard extracts and 0.07 ng/g for the 1.00 g extract.

Solutions of estradiol in PBSG were prepared at concentrations that gave approximately 20, 50, and 80% binding on the standard curve. These solutions were used as high, medium and low quality control samples in every assay. The intra-assay coefficients of variation for these three samples were 14.5, 8.5, and 11.8% (n=20). The inter-assay coefficients of variation for these three samples in 12 assays were 8.7, 12.0 and 17.9%.

Coprolite samples were prepared for human myoglobin analysis by adding 600 µl PBSG to 0.07 g of dried sample. Samples were shaken for one hour, and then spun at 13,000 rpm for 10 minutes. The supernatant was removed and a 100 µl aliquot frozen for myoglobin measurement. Human myoglobin was measured by electrochemiluminescence immunoassay.

RESULTS

Testosterone

The extraction efficiencies for testosterone in the faecal samples were high and consistent between samples (Table 2). The measurement of hormone concentrations by radioimmunoassay (RIA) is based on a competitive binding reaction between an antibody and the hormone of interest. The reaction tube contains an antibody that binds to the hormone, some radioactive hormone (labelled hormone) and an unknown amount of hormone from the biological sample. The hormone and labelled hormone compete for binding to the antibody, so that the more sample hormone in the tube the less radioactive hormone will be bound to the antibody. The bound and free labelled hormone are separated at the end of the reaction period and the amount of bound labelled hormone is counted in a scintillation counter that detects the radioactivity emitted by the labelled hormone.

TABLE 2
Extraction efficiencies for labelled steroid added to Harataonga coprolites

Hormone	**Extraction efficiency (%)**					
	Sample F1	**Sample F6**	**Sample 61**	**Sample 62**	**Mean**	**S.E.**
Testosterone	82.12	83.95	91.26	78.02	83.84	2.40
Estradiol	76.08	73.49	78.55	66.01	73.53	2.35

The antibody against the hormone may also bind to other hormones, so it is important to choose an antibody that has a low level of binding (cross-reaction) with other hormones. The antibody can also bind to other substances that are not hormones, so that an apparent measurement of a hormone concentration could be a measurement of some other substance. The first step in establishing a new RIA or using an existing RIA for a new sample material is to check that the antibody binds to the hormone to be measured without significant cross-reactions with other hormones or substances. The process of validation of the RIA starts with the preparation of serial dilutions of the sample material. A serial dilution using halving dilutions should give measured hormone concentrations that also halve with each dilution.

Hormone concentrations in samples are determined using standard curves in which the percentage of the radioactive hormone that has bound to the antibody is plotted against known concentrations of hormone in standards. The percentage bound values increase as the concentration of hormone in the assay tubes decreases. A plot of a serial dilution binding curve for a sample should be parallel to the standard curve. If the plot of the serial dilution is not parallel, then this indicates that the antibody is cross-reacting with substances other than the hormone to be measured, and hence the assay is not valid for that sample and cannot be used.

Serial dilutions were prepared for the four coprolite samples. The percentage bound values for all these samples were very high for undiluted samples, so that if any hormone was present it was at concentrations below the least detectable dose for the assay. It was therefore not possible to determine whether serial dilutions were parallel to the standard curve since the starting percentage bound values were so high (Fig. 10). One sample was prepared at a ten-fold higher concentration than previously, but there was still no detectable testosterone (Fig. 10).

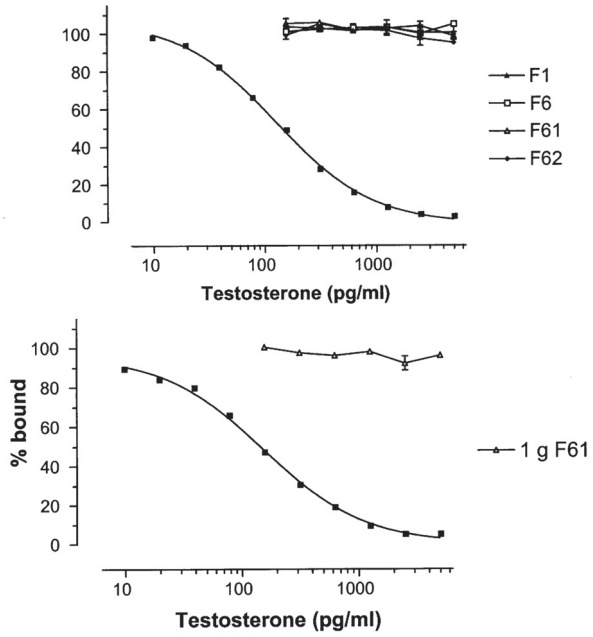

Figure 10: Parallelism test for binding of antibody against testosterone to tritiated testosterone and to Harataonga coprolite extracts. The upper panel gives results for four coprolite samples and the lower panel for a more concentrated extract of Sample F61.

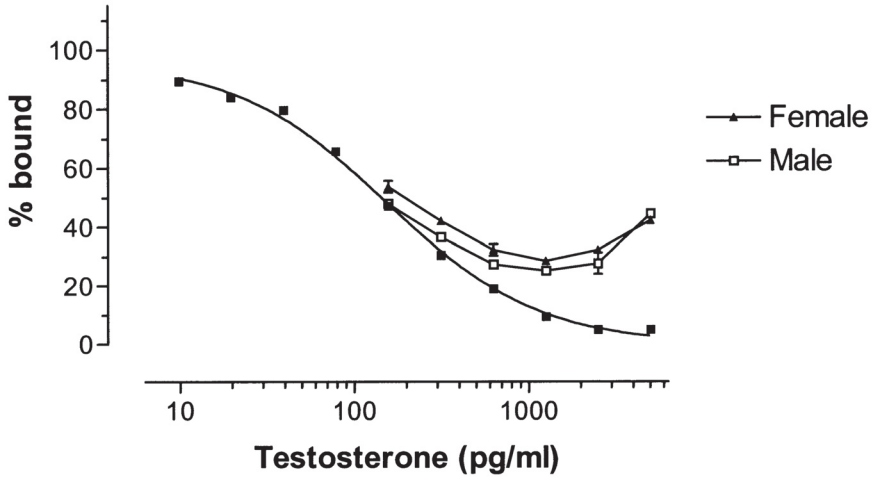

Figure 11: Parallelism between binding of antibody against testosterone to tritiated testosterone and to extracts of fresh human faeces.

Samples of fresh human faecal material were prepared and assayed by the same methods used for the coprolites. Samples from both a male and female had detectable concentrations of testosterone and serial dilutions of both samples were parallel to the standard curve after the first two dilutions (Fig. 11).

Testosterone concentrations for the coprolites and the fresh human samples are given in Table 3. Testosterone concentrations in the coprolites were below the least detectable dose of the assay. Faecal testosterone concentrations were similar in the two human faecal samples.

TABLE 3
Concentrations of testosterone and estradiol
in Harataonga coprolites and fresh human faecal samples

Sample	**Testosterone**	**Estradiol**
(ng/g)	**(ng/g)**	
Coprolite F1	<0.41	2.2
Coprolite F6	<0.41	1.46
Coprolite 61	<0.41	1.51
Coprolite 62	<0.41	1.73
Coprolite 61 (1g extract)	<0.06	<0.07
Fresh male	14.24	65.86
Fresh female	12.78	111.35

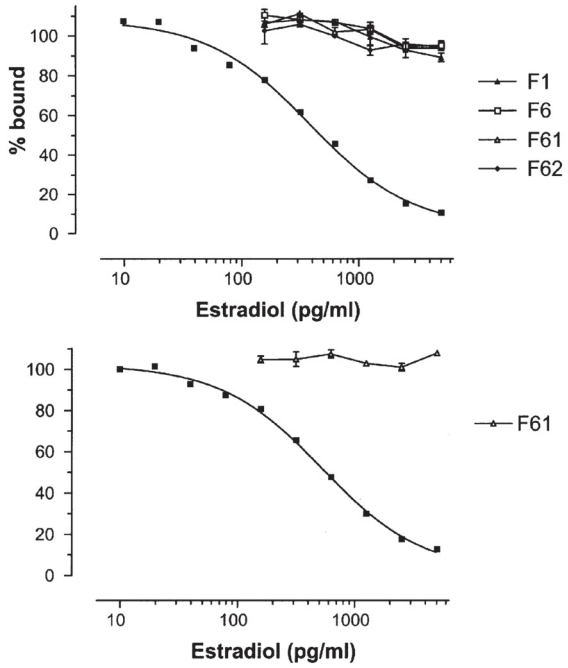

Figure 12: Parallelism test for binding of antibody against estradiol to tritiated estradiol and to Harataonga coprolite extracts. The upper panel gives results for four coprolite samples and the lower panel for a more concentrated extract of Sample F61.

Estradiol

The extraction efficiencies for estradiol in the faecal samples were high and consistent between samples (see Table 2).

Serial dilutions were prepared for the four coprolite samples. The percentage bound values for all these samples were high for undiluted samples, so that if any hormone was present it was at low concentrations. The high initial percentage bound values meant that it was not possible to determine whether serial dilutions were parallel to the standard curve (Fig. 12). A similar result was found for a sample that was prepared at a ten-fold higher concentration (Fig. 12).

Samples of fresh human faecal material were prepared and assayed by the same methods used for the coprolites. Samples from both a male and female had detectable concentrations of estradiol and serial dilutions of both samples were parallel to the standard curve after the first dilution (Fig. 13).

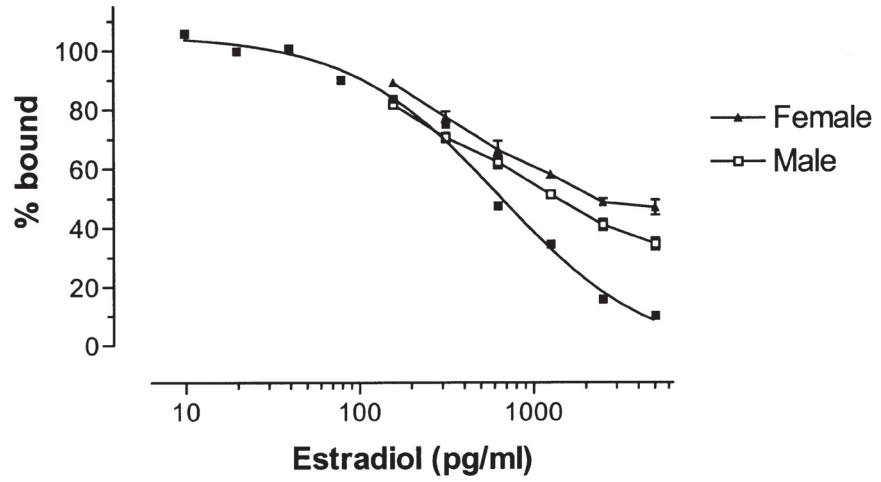

Figure 13: Parallelism between binding of antibody against estradiol to tritiated estradiol and to extracts of fresh human faeces.

Estradiol concentrations in the coprolites at the standard dilution were low and in the more concentrated sample were below the least detectable dose of the assay (Table 3). These results indicate that estradiol could not reliably be detected in the coprolites. Faecal estradiol concentrations were twice as high in the fresh human female sample than the fresh male samples.

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