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Pre-Columbian land use and human impact in the Bolivian Amazon

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Doctor of Philosophy

University of Edinburgh, 2014.
Declaration

I confirm that I am the author of this thesis. The work included is my own, except where otherwise stated. This work has not been submitted for any other degree or professional qualification.

Signed

John Carson
Thesis Abstract

There is a polarised debate amongst Neotropical archaeologists and ecologists over the extent of Pre-Columbian (pre-AD 1492) anthropogenic environmental impacts in Amazonia. While some maintain the old paradigm of pre-Columbian Amazonia as a “pristine wilderness”, which was sparsely populated by humans, others point to the discovery of an increasing number of archaeological sites across the Amazon basin as evidence for large, complex societies, supported by intensive agriculture and management of forest and aquatic resources. Under this model, pre-Columbian people had profound impacts on biogeochemical cycles through large-scale deforestation and biomass burning, and left an indelible legacy on Amazonian ecosystems by altering edaphics, geomorphology, hydrology, and the distribution of plant taxa. However, so far this debate has continued amongst a paucity of palaeoecological data, meaning that we have little evidence to indicate the scale of impact, and often no palaeoenvironmental context in which to place these societies. Other aspects of pre-Columbian cultures, such as their chronology, land use practices and subsistence strategies, are also poorly understood.

In this thesis, palaeoecological methods are applied to improve our understanding of the scale, nature, and legacy of land use associated with pre-Columbian geometric earthwork cultures in north-east Bolivia. The methodology employs analysis of fossil pollen and macroscopic charcoal from lake core sediments. In total 110 pollen and 628 charcoal samples were analysed from four lakes: Laguna Isireri (14°49’18”S, 65°40’57”W), Laguna Orícore (13°20’44”S, 63°31’31”W), Laguna Granja (13°15’44”S, 63°42’37”W) and Laguna La Luna (13°21’20”S, 63°35’2”W). Chronologies were built for these cores from 12 AMS $^{14}$C dates. Results are presented in four chapters (thesis chapters 2-5), in the form of academic papers.

Chapter 2 describes a new laboratory technique co-developed with Whitney et al., which improves the recovery of cultigen pollen grains from sediments from large, Neotropical lake basins. This technique was applied in the laboratory preparations of sediments analysed for Chapters 3-5.

Chapter 3 employs palaeoecological reconstructions from differently sized lakes (L. Granja and L. Orícore) in the same earthwork region, to obtain both a
regional- and a local-scale history of environmental change/human impact around the archaeological site. Key findings of this paper are: 1) earthworks were built in an originally open savannah landscape, which existed under drier-than-present climatic conditions in the mid-to-late-Holocene; 2) forest expanded into this region from ~2000 (cal yrs) BP and was supressed locally around the settlement to maintain an open landscape; therefore, 3) earthwork construction across southern Amazonia, may not have required extensive deforestation, and pre-Columbian impacts on biogeochemical cycling may have been much less than some authors have suggested.

Chapter 4 looks more closely at the local scale record provided by L. Granja. These data are integrated with phytolith data analysed by co-author J. Watling and existing archaeological data, to discuss the chronology of settlement on the site, the agricultural/land use strategies employed by its inhabitants, and the spatial scale and distribution of impacts locally around the site. Key findings are that: 1) first occupation of the site is much earlier than previously dated from archaeological contexts, beginning ~2500 BP; 2) maize was the staple crop grown on site; 3) land use involved more extensive and intensive burning of the landscape than compared to modern slash-and-burn agriculture; 4) site decline occurred ~ 500 BP, and may have been related to the Columbian Encounter of AD1492; 5) the close integration of local scale palaeoecological records with archaeology, is highly useful in discerning aspects of chronology and spatial variability of land use.

Chapter 5 presents a 6000 year record of palaeoenvironmental change and land use on a pre-Columbian forest island site. Key findings are that: 1) As in Chapter 3, inhabitants exploited an originally open landscape and practiced forest suppression to maintain that open landscape; 2) the earliest recorded evidence for maize agriculture in the region is found at 2100 BP; 3) the economically useful species *Theobroma cacao*, which is abundant on the site today, is not detectable in the pollen record; 4) clear-cutting was not practiced on the site and previous population estimates, based on labour for deforestation, must be reconsidered.

The work in this thesis reveals a new model of human-environment interactions, demonstrating that pre-Columbian earthwork cultures in southern Amazonia occupied and adapted to a region of dynamic, climatically controlled forest-savannah transition during the mid-to-late-Holocene. Obtaining a
palaeoenvironmental context for archaeological landscapes, is shown to be a vital pre-requisite to inferring past environmental impacts. Furthermore, we demonstrate the valuable contribution that palaeoecology can make to a better understanding of the chronology and land use practices of pre-Columbian cultures.
Preface

The four results chapters of this thesis are presented in academic journal style. As such, some of the data included in these chapters was contributed by collaborating authors. Each paper is therefore prefaced with a clear explanation of the contribution of each co-author. Key background and introductory points discussed in the thesis introduction are reinforced in the each relevant chapter, in order to make coherent, stand-alone papers. When there is cross-referral between the three unpublished results chapters, in-line citation style is used, along with the thesis chapter number, i.e. (Carson et al. Chapter 3), (Carson et al. Chapter 4) and (Carson et al. Chapter 5). When reference is made to the technique described in Chapter 2, the published paper is cited.

Chapter 2 is based upon an already published article in the journal *The Holocene*. The writing of the original article was by Dr. Bronwen Whitney (BW), with contributions to the interpretation from Elizabeth Rushton (ER) and John Carson (JC). Data for the published article was contributed by BW, ER, and JC. The paper’s presentation here, as a thesis chapter, incorporates pollen data analysed by BW.

Chapters 3 and 4 include details of unpublished archaeological data provided by Dr. Heiko Prümers (HP) of the Deutsches Archäologisches Institut, in Bonn, Germany. It also incorporates data from a modern botanical survey carried-out by J. Daniel Soto (JDS) of the Museo de Historia Natural ‘Noel Kempff Mercado’, Santa Cruz, Bolivia. Chapter 4 also includes phytolith data analysed by Jennifer Watling (JW) of the University of Exeter.

Chapter 5 incorporates data from a modern botanical survey conducted by DS.

The full pollen and charcoal datasets from Chapters 3-5 will be made publically available on the Neotoma Palaeoecology Database (www.neotomadb.org) and the Global Palaeofire Working Group’s Global Charcoal Database (www.gpwg.org).

All of the radiocarbon dates mentioned in the text are in calibrated years BP, unless otherwise stated.
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A huge part of these acknowledgments must go to my second supervisor Bronwen Whitney. I have no doubt that the work produced during this PhD would not have turned out so well, and the process not been so much fun, without Bronwen’s consistent guidance, reassurances and patience in general. From Bronwen I have gained what I hope will be the start of a long and exciting research collaboration, and I am sure will be a lifelong friendship.

I am thankful for the collaboration and input of José Iriarte, Jennifer Watling, Heiko Prumers and J. Daniel Soto, all of whom contributed to the research presented in this thesis. I also thank Elaine McDougal for her assistance and creative thinking in the labs at the University of Edinburgh. The fieldwork portion of this research would not have been possible without the logistical support of the ‘Noel Kempff Mercado’ Museo de Historia Natural, in Santa Cruz, Bolivia; Douglas Bruckner of the ‘Programa de Conservación de la Paraba Barba Azul’, Trinidad, Beni Department, Bolivia; and the rangers from the ‘Reserva Iténez’ WWF station in the town of Bella Vista, Beni Department, Bolivia.

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I have shared the trials and tribulations of the past 3+ years with an incomparable group of friends from amongst the Geosciences department at Edinburgh. I have these excellent people to thank for maintaining my sanity, by
taking me off on numerous adventures, whether that be trips up Munros in the beautiful highlands of Scotland or down to a comfortable seat in the local pub.

Finally, the ultimate thanks must go to my family who, despite often not comprehending what it is I do or why I do it, have always taken pride in and supported me, in all possible ways.
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List of abbreviations

BP – years before present. All dates are calibrated unless otherwise stated
CE – ‘Columbian Encounter’
GDP – Granja del Padre
ITCZ – Intertropical Convergence Zone
LB – La Luna Bog
LBV – Laguna Bella Vista
LCH – Laguna Chaplin
LEC – Laguna El Cerrito
LF – Laguna Frontera
LG – Laguna Granja
LI – La Luna Island
LIS – Laguna Isireri
LL – Laguna La Luna
LO – Laguna Oricore
LSJ – Laguna San José
METF – moist evergreen tropical forest
PCS – Pre-Cambrian Shield
SDTF – Semi-deciduous tropical forest

List of terms used

*Cerradão* – *terra firme* savannah with dense tree cover, but little understory
*Cerrado* – *terra firme* savannah
*Llanos* – wetland/seasonally inundated grassland
*Terra firme* – non-flooded upland
*Varzea* – floodplain/seasonally inundated land bordering a river
Chapter 1. General Introduction

1.1. Amazonia: pristine wilderness or cultural parkland?

The Amazon rainforest is often considered one of the world’s last great wildernesses. Neotropical lowland rainforest constitute ~50% of the total remaining tropical rainforest area on the planet (Achard et al. 2002), and are recognized as ecosystems of exceptional biodiversity (Dirzo and Raven 2003) and a globally important terrestrial carbon store (Baker et al. 2004). As such, their preservation is considered an international conservation priority. In many ways, the ecosystems of the Amazon basin have become a symbol of the global conservation movement, pitting primordial nature against expanding human exploitation. Modern human incursions into these purportedly highly sensitive and pristine environments are considered detrimental and disruptive to the fragile balance of natural processes.

This view incorporates the traditional anthropological narrative of Amazonian native cultures, before the arrival of Europeans in the Americas in AD 1492 (Steward and Faron 1959; Meggers 1971, 1992b). Under this old paradigm, the South American continent first encountered by Europeans was one of predominantly sparsely populated and untouched natural landscapes. With the exception of the imperial cultures in the high Andes, human population densities were low and their environmental impacts limited. Under this model, pre-Columbian native Amazonians are viewed in the guise of the “noble savage”, existing in small, dispersed groups and utilising benign subsistence practices, which had very little impact upon the natural environment (Meggers 1995, 2003). Any exceptional examples of sedentism and greater cultural complexity in the lowlands (Meggers and Evans 1957), were attributed to short-lived invasions or diffusion from more advanced Andean societies. Significant human impacts began in lowland Amazonia only after colonisation of the New World by Europeans and the introduction of destructive land management practices. This old paradigm denies Native Amazonians any role in the development of the ecosystems they inhabited, and supports an exclusionary, national park
approach to modern conservation, which establishes ecological reserves to keep out anthropogenic influences.

In recent decades, the cataloguing of numerous monumental earthworks and extensive settlements at sites across the Amazon basin, has led to a re-evaluation of the traditional view of pre-Columbian lowland Amazonia as a wild landscape, with a low population density. A new pan-American hypothesis has emerged of a continent inhabited by sizeable, pre-contact, Amerindian populations, which underwent demographic collapse after AD 1492 (Denevan 1992b). This collapse was driven initially by the spread of Old World diseases, such as smallpox, to which native inhabitants had little resistance. Native societies became further dispersed during and post-conquest, through warfare, exploitation for slavery, enforced urbanization, and the introduction of new technologies and new forms of agriculture (Whitmore 1991; Walker 2000). Under this new paradigm, large, sedentary, and socially complex pre-Columbian societies, supported themselves in Amazonia through systems of intensive agriculture and resource management. They had widespread environmental impacts and profoundly altered the environment around them. The cessation of native land use across much of Amazonia following this demographic collapse, allowed forest ecosystems to recover and reforestation to occur on previously occupied sites. These forests, however, bore the legacy of pre-Columbian land use, which had profoundly altered most aspects of the natural environment, including floristics, edaphics, hydrology, and topography (Erickson 2008; Balée 2010). Amazonia, therefore, is not a “pristine wilderness”, but a highly anthropogenic “cultural parkland” (Heckenberger 2003) and, it is argued, our attitudes toward its conservation and the role of modern native groups in its management should reflect this (Heckenberger et al. 2007; Chave et al. 2008). This new paradigm of a heavily settled pre-European Amazon suggests that its ecosystems can sustain large populations, and that far from being fragile, they have shown resilience to and synergy with a long history of pervasive anthropogenic impacts.

These are the two extremes of the argument occurring amongst anthropologists, ecologists, and conservationists working in Amazonia. Yet, although there is now abundant archaeological evidence for settled, complex societies in the Amazon, many important aspects of these societies, such as their land use practices,
chronology, and the palaeoenvironment which they inhabited, are unknown. Fundamentally, there is a lack of palaeoecological data associated with pre-Columbian archaeological sites, without which, it is difficult to confidently discuss the scale and impact of historical land use. The aim of this thesis is to improve our understanding of pre-Columbian land use and environmental impacts, using palaeoecological records from the rich archaeological sub-region of the Bolivian Amazon. An overview of the wider debate and details of the study region are given below to provide context for the results chapters that follow.
1.2. A brief overview of the “pristine wilderness” debate in Amazonia.

1.2.1. Amazonia as pristine wilderness

The accounts of the early European explorers in Amazonia describe encountering numerous, large, riverside chiefdoms, on their journeys down the Amazon’s waterways (Medina 1934; Markham 2010). These accounts were mingled however, with more mystical stories of Amazon warrior women and lost cities of gold. As a result, in the 19th and 20th centuries, many anthropologists dismissed these early accounts as fanciful and unreliable (Mann 2006). The pristine wilderness paradigm of Amazonia emerged largely from studies of extant Amazonian tribes (and other historical and political factors, which are beyond the scope of this brief discussion). From these pioneering ethnographic studies, a picture of native Amazonian life was built in which small, semi-nomadic tribes, supported themselves using swidden agriculture, hunting and resource gathering. The shifting nature of their occupations meant that these groups exerted little detrimental impact on their environment, and were limited in their level of social and technological complexity (Steward and Faron 1959). This view was formalised scientifically in the 1950s, with the hypothesis that the low agricultural potential of nutrient poor tropical soils and a lack of readily available animal protein, were limiting factors to high population in the Amazon (Steward 1949; Meggers 1954; Meggers 1971). The poor nutrient status of the ecosystem and the physical barrier of dense, closed canopy rainforest (Roosevelt 2000), precluded sedentism and sustainable high populations.

1.2.2. Amazonia as cultural parkland

The old pristine wilderness paradigm came under criticism for its environmentally deterministic assessment of population potential. The emergence of archaeological evidence for large, sedentary cultures, in the Bolivian Amazon (Denevan 1966; Denevan 1976) and at Marajo Island at the mouth of the Amazon River (Roosevelt 1991; Schaan et al. 2009; Schaan 2010) (see fig 1.1), suggested that pre-contact populations in some regions were sizeable, and that adaptive subsistence strategies had been developed to sustain permanent settlements. There was also the discovery of widespread black earth or terra preta anthrosols along the banks of the
Amazon River (Glaser and Woods 2004), which are suggestive of sedentism, although there has been disagreement over whether these deposits represent large settlements (Roosevelt 1991) or are the detritus from successive phases of small, shifting occupations (Meggers 2001).
Figure 1.1 Map showing major biomes and rivers of the Amazon basin, major archaeological sites and palaeo sites. Major archaeological find sites are shown: 1) Marajo, 2) Santarem, 3) Manaus, 4) Upper Xingu, 5) Llanos de Mojos and 6) Acre. Southern Amazonian lakes with Holocene palaeo records: BV – Bella Vista, CH – Chaplin, Y – Yaguarú, C-SR – Chalalán & Santa Rosa, PM – Puerta Maranon lakes, C – Carajas and LT – Lago Titicaca. Dashed white line indicate position of transect from Pessenda et al. (1998a), de Freitas et al. (2001), and Gouveia et al. (2002). Black crosses and dashed line indicate soil core sampling points/transects from McMichael et al. (2012b). METF=Moist evergreen tropical forest. SDTF= Seasonally dry tropical forest
The notion that large pre-Columbian New World populations experienced a post AD1492 collapse as a result of a catastrophic epidemic, was first introduced by Dobyns (1963, 1966), who used historical records to demonstrate a collapse in the Peruvian native population, attributed to smallpox, during the early colonial period. Dobyns’ population estimates for the whole American continent pre-1492 was 90 million. In a later paper, and one which remains highly cited in the academic literature, Denevan (1992) estimated an American continental population in the 16th century of 43-65 million, with 8.6 million in lowland South America. However, since then, Denevan has himself criticised the methods employed to make these estimates, and questioned the accuracy with which any estimate of native population pre-1492 can be made (Denevan 2003). Nevertheless, based on estimates of potential land carrying capacity, Denevan maintains that the population was “…at least 5-6 million in Greater Amazonia and at least 3-4 million for the wider Amazon basin” (Denevan 2003, pp.184). Estimates of depopulation have ranged as high as a 95% decrease in some areas of the New World, with a population nadir in AD 1750 (Denevan 1976).

In this scenario, the absence of human management of ecosystems following population collapse allowed the reforestation of habitation areas and agricultural land, and the establishment of what appeared to be a pristine landscape by AD 1750 (Denevan 1992b). Furthermore, it was suggested that the semi-nomadic lifestyle practiced by extant tribes in Amazonia, far from being representative of their ancestors’ way of life, is a product of population collapse and colonization. The historical events surrounding the European colonization fragmented groups, by forcing them deeper into less productive regions, and introducing the steel axe, which made shifting slash-and-burn agriculture a viable subsistence strategy (Denevan 1992a; Denevan 2001).

The continued rediscovery of sizeable artificial earthworks and occupation sites in new regions, including in the Upper Xingu (Heckenberger et al. 1999), Amapa (Mayle and Iriarte 2012), eastern Acre (Parssinen et al. 2009) and the central Amazon (Rebellato et al. 2009; Heckenberger and Neves 2009) in Brazil, and in coastal French Guiana (Renard et al. 2012; Iriarte et al. 2012a), has bolstered the archaeological evidence for large pre-contact populations. The scale and extent of
these earthworks has led Erickson (2008) and Heckenberger (2003) to suggest that the entire Amazon in AD 1492 was an anthropogenic landscape or “cultural parkland”, which had been influenced by extensive anthropogenic management, since perhaps the early-Holocene. Unlike Eurasian prehistoric people, who domesticated numerous animal species, it is argued that Amerindian societies secured their subsistence and achieved greater cultural complexity through the domestication of entire landscapes, allowing them to control wild plant and animal resources, alongside domesticated plant cultivation (Erickson 2008). The construction of raised agricultural fields (Denevan 1970; Walker 2000) and enrichment of soils (Glaser and Woods 2004; Bozarth et al. 2009; Fraser et al. 2009) allowed Amazonians to practice sustainable agriculture, growing domesticates which included yuca (Manihot esculenta), maize (Zea mays), squash (Curcurbita sp.), and sweet potato (Ipomoea batatas) (Piperno 1990; Heckenberger 1998; Dickau et al. 2012). It has even been suggested that these ancient agricultural practices could form the basis for new sustainable exploitation of Neotropical landscapes in the modern day (Erickson 1988; Glaser 2007; Renard et al. 2012). However, others have questioned whether the use of raised field technology actually increases crop yields (Lombardo et al. 2010), and the notion that ancient land use practices can be recreated in the modern day has been heavily criticised (Baveye 2012). It is also not yet clear whether the formation of terra preta soils, although they are highly fertile and nutrient retentive, was an intentional strategy for improving soil quality, or was the accidental result of long occupations (Glaser and Birk 2012).

Some authors have inferred levels of pre-Columbian environmental impact in Amazonia and the wider Americas, sufficient to have had continental to global scale effects on biogeochemical cycling (Nevle and Bird 2008; Dull et al. 2010; Nevle et al. 2011). It has been suggested that the decline in biomass burning and reforestation induced by population collapse in the 15th century, led to a decline in carbon dioxide emissions/sequestration of atmospheric carbon, on a scale which amplified Little Ice Age (~1550-1750 AD) climate cooling (Dull et al. 2010). Similar scenarios have been suggested that incorporate global population and land use shifts as part of an early-Anthropocene impact on climate (Ruddiman 2003; Kaplan et al. 2010). The current net biomass increase observed across Amazonian rainforest plots (Baker et
al. 2004) has also been attributed to post-Columbian forest recovery from human degradation (Chave et al. 2008), rather than to natural factors, such as increased CO$_2$ fertilisation, as suggested by ecologists (Phillips et al. 2008).

In addition to destructive impacts, such as deforestation and biomass burning, it is claimed that pre-Columbian people shaped Amazonian environments in more subtle ways. Historical ecologists and ethnobotanists working in the Amazon have noted the wide variety of plant species which are used or consumed by modern indigenous groups (Balée 1994), and observed how native people plant and manage these wild, but “economically useful” taxa, in “kitchen gardens” around their settlements. Forest inventories made around pre-Columbian sites, such as the monumental habitation mounds in the Bolivian Beni basin, documented high abundance of palms and other economically useful species (Erickson and Balée 2006). These were interpreted as the legacy of pre-Columbian planting and preferential management. This historical ecological approach has been applied to other regions (Shepard and Ramirez 2011; Levis et al. 2012) of the Amazon, and it is now suggested that forest composition has widely been shaped by a long history of anthropogenic management (Clement and Junqueira 2010; Balée 2010). In a review of inventories from floristic plots and archaeological sites, across numerous locations, Balée (1989) concluded that 12% of the total forest area in Amazonia may have an anthropogenic origin. These historical ecological interpretations have, however, been met with alternate explanations from ecologists, who cite natural factors which could produce the same species associations and monospecific forest stands that historical ecologists claim are the legacy of pre-Columbian management (Barlow et al. 2012).

1.2.3. The intermediate hypothesis of pre-Columbian impact in Amazonia

In response to this highly polarised debate, an intermediate scenario of pre-Columbian population densities and the spatial distribution of environmental impacts has emerged. Amazonian archaeologists have commonly split native societies into groups that inhabited the *varzea* (floodplain), and those that inhabited the *terra firme* (interfluvial uplands) (Meggers 1971; Denevan 1976). Denevan later modified this organization with his “bluff model” of pre-Columbian occupation (Denevan 1996),
proposing that settlements were based, not on the floodplain itself, which would be a precarious location for a settlement, but on high river promontories or bluffs on the valley sides. Positioning on the river bluffs kept settlements safe from flooding and allowed cultivation of the rich floodplain soils during non-flood periods. With this model of land use in mind, Bush & Silman (2007) evaluated the spatial scale of pre-Columbian human impacts across Amazonia, by mapping the major archaeological sites, known *terra preta* deposits, and all published lake palaeoecological records which showed evidence of pre-Columbian occupation. They concluded that the larger settlements and more intensive land use practices were centred in the *varzea* zones around major rivers and large lakes, while the interfluvial *terra firme* regions had low population densities and showed the least evidence of anthropogenic impact. It was argued that the more fertile floodplain soils, easy access to the land from the river, and availability of aquatic resources, made the *varzea* attractive for settlement and productive enough to support larger, sedentary groups. The interfluvial zones on the other hand, were less productive and less accessible, and could therefore support only small, nomadic, slash-and-burn agriculturalists, such as those that occupy the rainforest environment today.

A recent set of palaeoecological studies, investigating the scale of pre-Columbian impacts in rainforest regions of west and central Amazonia (McMichael et al. 2011, 2012a,b), produced evidence in support of Bush and Silman’s intermediate hypothesis. This study analysed phytoliths and macroscopic charcoal from terrestrial soil cores taken in transects, which radiated out from known archaeological sites into the surrounding forest. The data were interpreted as showing some evidence for burning, cultivation, and tree clearance near to the rivers, but little or no evidence of disturbance in the interfluvial zones. This intermediate model returns to the environmental limitation arguments of Meggers, and supports the notion that the majority of the rainforest in the interfluvial zones away from major rivers is a pristine environment, which experienced little anthropogenic impact in pre-Columbian times. This model however, does allow for larger populations and higher impact in the *varzea*.

However, the limited available palaeoecological evidence in favour of this hypothesis, is rebutted by the archaeological record, which does show evidence of
large, sedentary societies in *terra firme* regions of southern Amazonia, in the form of
geometric earthworks (Ranzi 2003; Parssinen et al. 2009; Heckenberger 2009;
Saunaluoma 2010; Erickson 2010). These are large (100-300m diameter plus),
geometrically shaped, ditched earthworks, which occur in interfluvial zones in the
Upper Xingu, eastern Acre, and north-east Bolivia. The majority of these earthworks
were discovered in recent decades and beneath what was thought to be pristine,
closed-canopy rainforest. Furthermore, it has been estimated that the hundreds of
earthworks revealed so far by modern deforestation, may represent as little as 10% of
the total that lie undiscovered (Parssinen and Korpisaari 2003), and occurring in a
broad arc across the southern Amazon, beneath forest that may cover an area as great
as 1,000,000 km$^2$ (see fig 1.2). The presence of these earthworks, whose construction
it is argued, must have required a level of social organization and population above
that of ephemeral tribes (Parssinen and Korpisaari 2003; Schaan 2012), does not
match with the model of limited population density and impact in the interfluvial
forests. It is also argued that their position beneath modern closed-canopy rainforest
is evidence of large-scale pre-Columbian deforestation (Erickson 2010). However,
apart from the positioning of the earthworks themselves, there is no direct evidence,
in the form of palaeoecological data, that their construction involved widespread
deforestation.
Figure 1.2 Geometric earthworks across southern Amazonia. Major geometric earthwork find sites are shown: 1) Iténez, 2) Pando, 3) Acre and 4) Upper Xingu. Dashed line is an approximation of the hypothesised potential coverage area of geometric earthworks. Solid black line shows modern southern extent of Amazonian rainforest.
1.3. Existing palaeoecological records and Holocene vegetation in the southern Amazon

In comparison to temperate regions of the globe, the Neotropics, and especially the southern American lowland tropics, are poorly covered by palaeoecological studies. In our study region of southern Amazonia, there are a small number of palaeoecological records which reconstruct vegetation change through the Holocene period, and even fewer records that date back to the last glacial period (see fig 1.1). The lowland Amazon basin also contains few palaeoecological studies which aim to identify human occupation, or assess historical anthropogenic environmental impacts. Here we give a review of the published palaeoecological records in the Bolivian Amazon and relevant parts of wider Amazonia.

1.3.1. Holocene biome change in the Southern Amazon

In the north-east Bolivian Amazon, where the majority of the study sites analysed for this thesis are located, there has been a history of dynamic forest-savannah ecotonal shifts and species compositional changes, related to changing climate over the Holocene (Mayle and Power 2008). Lagunas Bella Vista (13° 37’S, 61° 33’W) and Chaplin (14°, 28’S, 61° 04’W), located in the Noel-Kempff-Mercado National Park, in north-east lowland Bolivia, provide pollen and charcoal records of vegetation and burning over the past ~50,000 years (Mayle et al. 2000; Burbridge et al. 2004). The dominant vegetation type surrounding these lakes today is terra firme moist evergreen tropical rainforest (METF). L. Bella Vista (LBV) is located ~150 km north of the modern, climatically-controlled, evergreen forest-semi-deciduous tropical forest (SDTF) transition, and L. Chaplin (LCH) is ~30 km north of this diffuse ecotone. The pollen and charcoal records from these lakes revealed, however, that during the early-to-mid-Holocene, a more open mosaic of savannah and drought-tolerant SDTF characterised the landscape around them, which also experience much higher burning frequency compared to present. From 6,000 to 3,000 BP at LBV, and ~2,200 BP at LCH, a gradual expansion of forest began in the region, with humid evergreen rainforest becoming established around the lakes at ~1650 BP and ~700 BP respectively. This pattern of a southward, time-transgressive rainforest expansion, coincides with evidence for increasing Amazonian precipitation from
~4500 to 2000 BP, as recorded by rising lake level records at Lake Titicaca (Cross et al. 2000; Baker et al. 2001) and the precipitation record from the Pantanal region, in central South America (Whitney et al. 2011).

Similar ecotonal movements are recorded in other southern Amazonian records. Located ~110 km or ~1 degree of latitude south of LCH, L. Yaguarú (15° 36’S, 63° 13’W) lies on the modern ecotone between METF and the Chiquitano dry forest, in the south-central Bolivian Amazon (Taylor et al. 2010). The pollen record from this site indicates that the environment was a mixture of savannah and open SDTF through the late-to-mid-Holocene, and experienced higher-than-modern burning frequency. After ~1200 BP, there was a transformation to the type of closed-canopy SDTF, which surrounds the site today. This transition also saw an increase in more moisture dependent species, such as Celtis sp., and a decrease in burning.

Stable carbon isotope analyses have been conducted using terrestrial soils from transects (7° 21’S, 63° 2’W to 15° 14’S, 59° 19’W) through the south-west Amazonian states of Rondônia and southern Amazonas, Brazil (Pessenda et al. 1998a; de Freitas et al. 2001). These studies indicate expansion of savannah and contraction of forest in the early-to-mid-Holocene, followed by contraction of savannah in favour of forest from ~3,000 BP. This pattern is not uniform however across the test sites. The sample taken from the Pontes e Lacerda site for example, located ~15°S, shows a forest signal throughout the Holocene (Gouveia et al. 2002). This is possibly related to the discrete spatial scale represented by stable carbon isotope analysis of terrestrial soil samples, which reflect highly localised vegetation (Mayle and Iriarte 2012) related to site specific environmental conditions, rather than vegetation on a biome scale. The nature of the analysis also means that interpretations are limited to reconstruction of tree vs. grass cover, and yield no information on possible forest compositional changes, such as an increase in drought-tolerant taxa. However, the fact that results vary from site to site in these stable carbon isotope studies, may also reflect the patchiness of climatically driven biome changes in lowland Amazonia, which would have occurred along diffuse ecotonal boundaries, rather than solid treelines (Pennington et al. 2006).

In the far south-east Amazon basin, palaeoecological studies from lakes around Carajas (6° 36’S, 49° 30’W), in Para State, Brazil, have also shown regional

Several lake records are available from the far south-west Amazon, located in the modern day METF, on the flanks of the Andes (Bush et al. 2007; Urrego et al. 2013). Unlike the records further east, these sites show no evidence of ecotonal movement during the late-to-mid-Holocene. A collection of 4 lake records from the Puerto Maldonado region of south-east Peru, do show evidence, in the form of lake level lowstands, of a dry period between 6,200-4,200 BP (Bush et al. 2007). However, with the exception of Lake Gentry, which records an increase in Moraceae/Urticaceae type pollen after ~ 2300 BP, the composition of the forest around the sites did not change in response to increasing or decreasing precipitation over the Holocene. The authors also inferred that the forests did not experience any increase in natural fire frequency associated with the mid-Holocene dry period. Lakes Santa Rosa and Chalalán, which are situated on the very western edge of the Beni department, also record a history of forest ecotonal stability throughout the Holocene (Urrego et al. 2013). However, these forests did experience species compositional changes and increased burning frequency, related to drier climate in the late-to-mid-Holocene.

The increase in precipitation and shortening of the dry season during the late-Holocene between ~4000-2000 BP, which as indicated by the records discussed above, occurred across the southern Amazon, has been attributed to orbitally driven increases in austral summer insolation and subsequent southward migration of the Intertropical Convergence Zone (ITCZ) (Mayle and Power 2008). The position of the ITCZ during the Holocene has influenced the intensity of the South American summer monsoon, and inversely, the length of drought period experienced across the seasonal southern Amazon basin (fig 1.3). Vegetation and climate in the southern Amazon have, therefore, not been quiescent over the Holocene, and some parts have seen considerable change. This is a factor which is often not reflected in the perspectives of archaeologists, such as Erickson (2010) and Heckenberger (2003)
who have assumed a stable forest ecosystem over the Holocene in north-east Bolivia and the Upper Xingu, Brazil.

Figure 1.3 Modern ITCZ position over S. America. Map showing the approximate position of the ITCZ in austral summer and winter (adapted from (Burbridge et al. 2004)). Green line approximates modern ecotone of evergreen rainforest.

1.3.2. Palaeoecological evidence of human occupation in the south-west Amazon

Palaeoecological techniques have been widely applied to archaeological questions in the Neotropics of Central America and the Andes (Bush et al. 1989; Jones 1994; Alcala-Herrera et al. 1994; Jacob and Halmark 1996; Islebe et al. 1996; Rosenmeier et al. 2002; Hodell et al. 2005; Niemann and Behling 2010; Williams et
al. 2011; Rushton et al. 2012), contributing valuable information on land use, environmental impact, subsistence, and chronology. In the central and eastern Amazon also, there are palaeoecological lake records (Bush et al. 2007b; Bush and Silman 2007), hundreds of terrestrial charcoal records (Bush and Silman 2007; Bush et al. 2008; McMichael et al. 2012b), and equally numerous analysis of terra preta soil deposits (Glaser and Woods 2004; Bozarth et al. 2009; Fraser et al. 2009), which are mainly concentrated along major Amazonian waterways. However, in the south-west Amazon, examples of such palaeoecological studies are rare and, until very recently, there were no such records in the Bolivian Amazon. Furthermore, many of the existing examples of palaeoecological studies in Amazonia which discuss pre-Columbian cultures are not specifically targeted in their methodological approach toward investigating anthropological questions. Often the evidence/discussion of anthropogenic activities is incidental to the original aims of the research. By making integration of palaeoecological records with archaeological data a central aim of the research, there is the potential to gain more detailed information on aspects of chronology and land use (Mayle and Iriarte 2012).

The suite of lakes located in the Maldonado region in south-east Peru (discussed above) were also analysed for evidence of anthropogenic activity, such as cultigen pollen. Three of the lakes (Gentry, Parker and Vargas) contained evidence of pre-Columbian cultivation and anthropogenic burning, from perhaps as early as 7000 BP (Bush et al. 2007b). Agriculture is evident only at the L. Gentry site, with maize recorded from 3,700-500 BP, and manioc in a single horizon at ~2400 BP. Some degree of anthropogenic burning and forest clearance was inferred at all of the sites, although the authors concluded that this was limited to the area locally around each lake, and did not extend into the wider forest. The lack of maize pollen and reduction in charcoal after 500 BP at L. Gentry was interpreted as a signal of site abandonment/population collapse, related to the European Encounter.

An analysis of macroscopic charcoal and phytoliths from soil core transects taken between the Gentry and Parker lake sites, and around the Los Amigos biological research station, located 40 km south of the lakes, confirmed that pre-Columbian burning, forest disturbance, and agriculture, were limited to the local area around the lakes (McMichael et al. 2011; McMichael, et al. 2012a;2012b). The soil
core study by McMichael et al. (2012b) also analysed a single location on the western border of Acre state. Phytolith and charcoal analyses from this location found no evidence of anthropogenic burning or forest clearance, although the sample site is not located close to any known archaeological features.

In the Bolivian Amazon two recent studies, which integrated palaeoecological data from lake cores with archaeological data from excavated earthwork features, have added to our knowledge of pre-Columbian cultures in the seasonally-inundated savannah region (see Study Region section below for description of Bolivian Amazon and fig 1.4 for lake locations). The first (Whitney et al. 2013) investigated the land use history and environmental impact associated with monumental habitation mounds (Prümers 2009a; Lombardo and Prümers 2010). The sample lake, L. San José (LSJ) (14° 56’S, 64° 29’W), is a large (14 km²), flat-bottomed lake, which is surrounded by pre-Columbian artificial habitation mounds. The charcoal record from the lake revealed greater-than-modern anthropogenic burning between 1600-720 BP, although the stability of the forest pollen signal throughout the record, led the authors to conclude that this burning took place mainly in the savannahs, rather than being used to clear forest. Maize pollen was also found throughout the core, indicating that it was cultivated around the lake from pre-Columbian times and into the colonial period. However, the decline in macroscopic charcoal observed around 720 BP in the lake record, was interpreted as an indication of decline in land use intensity and possibly population decline, which matches well with dating of archaeological layers from the nearby habitation mounds (Prümers 2009a). This decline in the monumental mounds culture preceded the arrival of Europeans in AD 1492, and is therefore interpreted not to have been caused by the introduction of Old World diseases (Whitney et al. 2013), as described by the current model of pre-Columbian population collapse (Denevan 1992b).

The second Bolivian study (Whitney et al. 2014) investigated land use and impact associated with raised agricultural fields. This study looked at two oxbow lake sites, L. Frontera (LF) (13° 13’S, 65° 21’W) and L. El Cerrito (LEC) (13° 14’S, 65° 23’W), located ~5 km apart, and <3 km from an excavated raised agricultural field site. The lake pollen and charcoal records indicate anthropogenic burning and maize agriculture on the raised fields from 1690-710 BP. This was corroborated by
the presence of maize phytoliths in terrestrial soils, excavated from the nearby raised fields. The low arboreal pollen abundance during this period was interpreted as an indication of forest clearance, in both the gallery forest and on the savannah, possibly associated with construction of the earthworks. Intriguingly, in contrast to the monumental mounds region, the raised field builders appear to have extensively cleared forest. Anthropogenic burning declined in the area between ~840 BP and 710 BP, and there is a possible change in land use strategy toward a system using less intensive burning, and cultivation of sweet potato and the tree genus Inga sp., alongside maize. This apparent land use change agrees with evidence of two separate occupations recorded in the raised field region by an earlier archaeological study (Walker 2000). Again, this site shows continuation of agriculture into the colonial period (until ~200 BP), although decline of land use may have started around 550 BP, as indicated by the reestablishment of forest in the area.

These two studies have been important in providing some of the first palaeoecological data from earthwork sites in the Bolivian Amazon. They also provide a more detailed interpretation of land use than previous examples of palaeoecological work from the Andean region, and have demonstrated an interesting heterogeneity of land use practices between different earthwork-building cultures in the savannah environment of the central Bolivian Amazon. However, the archaeological sites investigated in the studies by Whitney et al. occur in seasonally-flooded savannahs, outside of the Pre-Cambrian (Brazilian) Shield (PCS) terra firme rainforest. They do not tell us therefore, what the nature of land use was in the terra firme rainforest regions, where the geometric earthworks are found. In order to address questions over the scale of pre-Columbian deforestation in the vast, interfluvial Amazonian rainforest, and its potential impact on biogeochemical cycling, we need archives of land use and environmental change associated with these geometric earthwork cultures.
1.4. Study region

1.4.1. The Llanos de Moxos

The *Llanos de Moxos* is situated in the Beni department, in the northern lowlands of modern Bolivia (fig 1.4). The *Moxos* is a sedimentary basin, which has been infilled up to 1 km depth by Quaternary sediments derived from the Andes (Clapperton 1993). The low elevation, lack of topography and impermeable clay substrate in the basin, means that, during the annual wet season from December to March, the majority of the landscape becomes flooded, with the exception of a few higher areas formed by termite mounds, abandoned river levees, resistant rock outcrops and human construction. This flooding suppresses tree growth, except on those higher areas described, forming a vast (130,000 km$^2$) hydrological savannah-forest mosaic, made up of grassland, gallery forest and forest islands (Orellana et al. 2004). Average precipitation across the region varies on a roughly north-west to south-east gradient, ranging from 2000 mm in the north to 1200 mm in the south. The average temperature at the departmental capital, Trinidad, which is located in the south-central part of the department, is 26°C (Hanagarth 1993).

1.4.1.1. Geoarchaeological sub-regions of the Llanos de Moxos

The *Llanos de Moxos* contains some of the most impressive and varied examples of pre-Columbian artificial earthworks. These include various forms of raised agricultural fields, monumental habitation mounds, ring ditches, canals and causeways (Denevan 1966; Walker 2000; Walker 2009; Lombardo and Prümers 2010; Prümers 2012a) and zig-zagged fish weir structures (Erickson 2000). These different built landscapes can be divided into 4 separate geoarchaeological sub-regions (Lombardo, Denier, et al. 2013), defined by their unique geomorphological and archaeological characteristics (fig 1.4). In the north-west *Moxos*, the dominant earthwork type is platform raised fields. In the south-west *Moxos*, earthworks are ridged agricultural fields and canals and causeways. In the south-central and south-east *Moxos* is the monumental mounds region, which also contains canals and causeways. In the north-east *Moxos*, Iténez province is characterised by ring ditches built on natural forest islands, along with canals and causeways built in the savannah.
In the north of Iténez there are also ditched fields, and in the south of the province, south-east of the provincial capital Baures, there are zig-zagged or fish weir earthworks. The research in this thesis focuses mainly on the ring ditch region of northern Iténez, but also encompasses a wider view of the Bolivian Amazon and geometric earthwork building cultures across the southern Amazon.

*Figure 1.4* Distribution of earthwork types in the south west Amazon: Moxos and Pando, Bolivia, and partial eastern Acre. Adapted from Mann 2008; Lombardo et al. 2013a. Existing palaeoecological lake records in the Moxos, discussed in text, are shown: SJ – San José (Whitney et al. 2013), and F-EC – Frontera/El Cerrito (Whitney et al. 2014). Location of lakes analysed for this thesis are also shown: IT is approximate location of the northern Iténez lakes; IS – Istreri.
1.4.1.2. Environment and archaeology of Iténez province

The modern province of Iténez, in the north-east Beni department, lies on the edge of the basin and, as such, is a transitional zone, both geologically and ecologically. The River San Martín, which flows through the north and east of the province, marks the geological divide between the lowlands of the *Llanos de Moxos* sedimentary basin and the uplands of the PCS (Navarro and Maldonado 2005). To the north and east of this divide, the PCS supports *terra firme* (non-flooded), closed-canopy, evergreen rainforest, which is floristically linked to the Madeira-Tapajós ecoregion (Navarro and Maldonado 2005). This ecoregion extends throughout the central and southern Amazon basin, from south of the Amazon river, down to the Bolivian Amazon (Olson et al. 2010). To the south and west of the San Martín is the seasonally-inundated forest-savannah mosaic of the *Moxos*. This north-east sub-region, however, is different from the rest of the *Moxos*, in that the topographically higher “forest islands” are formed by natural, isolated outcrops of the PCS, which punctuate the savannah. The forest covering these outcrops are also part of the same Madeira-Tapajós ecoregion.

The Iténez province is also culturally distinct from the western *Moxos*. An alternative name for the province is Baures, which is both the name of the provincial capital in the south, and derived from the name and language of the native Baure people, who occupy the region (Erickson 2010). The *Moxos* contains several distinct linguistic groups (fig 1.5) of which, Baure and Mojo are derived from the Arawak diaspora, which dominates the south-west, central and eastern Amazon (Heckenberger 2008). Native cultures of the Amazon and wider South America are divided into two broad linguistic and cultural prehistoric diaspora: the Arawak in the north and west, and the Tupi-Guarani in the south and east. It is hypothesised that these diaspora spread from ~ 3000 to 2000 BP, and that the Arawak cultures were “…characterised by hierarchy, regional integration and settled life” (Heckenberger 2008, pp.947). Heckenberger (2013) has also suggested that circular planned settlements, ring ditch earthworks, and hierarchical settlement patterns, are a feature of Arawak culture, and links geometric earthwork sites across the southern Amazon in this way.
The archaeological landscape of Iténez was first described by Erland Nordenskiöld in the 1910s, who described encountering a large moated settlement while travelling down the Guaporé River, and theorised, based on the earthwork’s impressive scale, that pre-Columbian populations may have been large in the region (Nordenskiöld 1910). Since then, archaeological investigations in the region have been sporadic (Becker-Donner 1956a; Becker-Donner 1956b; Hanke 1957; Dougherty and Calandra 1985), documenting a shared ceramic tradition throughout the province and some evidence for terra preta (Hastik et al. 2013). In the north, ditched fields have also been identified in the savannahs (Lombardo et al. 2013a). In the south of the province, complex zigzag shaped earthworks found in the savannah, which are connected to small artificial ponds, have been interpreted as a system of fish weirs for harvesting river fish during flood periods (Erickson 2000). The most commonly encountered earthworks consist of ring ditches in the terra firme areas and causeways/canals in the savannah, which run between the terra firme outcrops, linking them up. Surveys carried out by Clarke Erickson gave an idea of the extent of earthworks on the PCS forest islands. Erickson reported that ring ditches were encountered on almost all forest islands >2 km² in area. The total, non-continuous
area covered by earthworks in both savannah and forest, was estimated at 12,000 km² (Erickson 2010). Artefactual remains excavated from the ring ditch sites tend to be sparse (Heckenberger and Neves 2009), however, Erickson (2008) has argued that evidence of high pre-Columbian population and social complexity in this region comes from domestication of the landscape, rather than from density of material culture.

The most detailed archaeological excavations in Iténez to date were carried out over several seasons by Prümers’ team, around the modern town of Bella Vista, in the north of Iténez (Prumers et al. 2006; Prümers 2012b). Excavations in the centre of the town and of two ring ditches located on the outskirts, uncovered cremation burials inside of ceramic urns, and lithic tools, within a thin occupation layer. This layer was dated in the two excavated ring ditches to between ~750-550 BP. Dickau et al. (2012) (full dates reported in Chapter 4, Table 4.1) performed archaeobotanical analysis on a range of lithic and ceramic tools excavated from the two ring ditch sites, looking for evidence of plant domesticates and other edible species. Their analyses recovered starch grains and phytoliths almost exclusively diagnostic of *Zea mays* L. (maize). Recent LiDAR scanning over a forested area of 200 km² around the town has mapped numerous further networks of ditched earthworks, located along the river and penetrating further into the PCS forest, some of which enclose areas of up to 200 ha, (Prümers 2012b). The modern town of Bella Vista, therefore, represents a substantial site and was potentially heavily impacted by pre-Columbian deforestation. These factors were taken into account when selecting a study location to meet the aims of this thesis, as is outlined below.
1.4.1.3. Site selection and rationale

Three lakes were selected for coring in the northern Iténez region: Laguna Orícore (LO), L. Granja, (LG) and L. La Luna (LL) (see fig 1.6). As discussed above, the Iténez region is an example of a geometric earthwork landscape, with evidence for extensive pre-Columbian ring ditches underlying apparently pristine, *terra firme* rainforest. Unlike other geometric earthwork regions, a key advantage to locating our study in the northern Iténez region was the availability of numerous potential lake sites of different sizes for coring. In other interfluvial regions where geometric earthworks occur, the sites available are often limited to small oxbow lakes, or there are no lakes at all located close to the archaeological features, and terrestrial soil archives must be used. In Iténez we can take advantage of the availability of both large (LO) and small lake basins (LG and LL), which can be combined to yield regional- and local-scale palaeoenvironmental reconstructions (Sugita 1994; Mayle and Iriarte 2012). Using lake sediment records, rather than terrestrial soils, also potentially provides a continuous chronology, and allows us the greater floristic detail from fossil pollen, which cannot be obtained through stable carbon isotope/phytolith analysis. This is therefore an ideal region to investigate the spatial scale of impact associated with geometric earthwork construction.
Figure 1.6 Map of the Llanos de Moxos, northern Iténez and study site locations. A shows the Llanos de Moxos in northern Bolivia, with study region in northern Iténez highlighted. B shows the study region in Iténez and the location of LO, LL, and of LG within Bella Vista Village, which is highlighted as an orange area. C shows LG and the Granja del Padre ring ditch.

LG was selected because of its position within the Bella Vista earthwork site, and the site’s location within terra firme rainforest on the PCS. The history of excavations from the Bella Vista site also meant that this was an ideal location for integrating our local-scale palaeoecological data, with the archaeological record.

LL was selected because of its close proximity to the La Luna forest island site (LI), which is another example of a large geometric earthwork located beneath modern dense-canopy rainforest. The lake was selected to test the hypothesis that the forest island landscape was clear-cut by its pre-Columbian inhabitants in order to build the ring ditches (Erickson 2010). It was also reasoned that LL would provide a local-scale reconstruction of vegetation change on LI, and allow us to investigate the
suggested role of humans in shaping the species composition of the forest (Erickson 2010).

Laguna Isireri (LIS) was originally selected with the intention, in the early stages of the research project, to investigate land use associated with the ridged agricultural fields in the south-west *Moxos*. The sediment core was analysed for pollen and macroscopic charcoal during the first PhD year (results presented in appendix A.1) however, subsequent radiocarbon dating produced several modern ages, suggesting that the sediments had experienced overturning/mixing throughout most of their depth. The core was, however, well situated (being close to known pre-Columbian earthworks) for developing and testing the new laboratory methodology discussed in Chapter 2.
1.5. Thesis aims

This thesis has a number of specific aims, which are encompassed within the broad aim of using palaeoecological techniques to improve our understanding of the nature, scale and legacy of pre-Columbian land use and environmental impacts, in the Bolivian Amazon and wider southern Amazonia. Three lakes located in the ring ditch region of north-east Bolivia were selected for coring, because of their close proximity to known earthwork sites. Fossil pollen and charcoal from the lake sediments were combined with phytolith and archaeological data, to reconstruct palaeovegetation, burning, and anthropogenic land use change over the last 6000 years. The ring ditch structures in north-east Bolivia (Denevan 1966; Erickson 2010; Prümers 2012b) are an example of so-called “monumental” scale pre-Columbian earthwork construction. Their presence beneath apparently pristine rainforest, in a protected reserve area, has been taken together with other geometric earthwork discoveries across the southern Amazon as evidence of large scale pre-Columbian deforestation (Erickson 2010). This feeds into the larger debate over the scale and legacy of pre-Columbian environmental impacts in Amazonia (Meggers 2003; Heckenberger 2003; Bush and Silman 2007) and its implications for sustainable land use in the Amazon (Glaser 2007; Renard et al. 2012), conservation practices (Heckenberger et al. 2007; Clement and Junqueira 2010), and early Anthropocene influences on global biogeochemical cycling (Dull et al. 2010; Neve et al. 2011). The application of palaeoecological techniques to anthropological/archaeological questions has not been widely applied in Amazonia. We also aim therefore, to demonstrate the value of integrating palaeoecology with archaeobotany and archaeology, in order to improve our knowledge around aspects of chronology, land use, subsistence, and impact, and address the paucity of such data in the Bolivian Amazon.

The thesis aims to address the following overarching hypotheses:

- Geometric earthwork construction in the Southern Amazon required large-scale deforestation of a closed-canopy rainforest environment.
- Pre-Columbian land use in the Bolivian Amazon entailed intensive and frequent burning.
• Pre-Columbian geometric earthwork building societies in the Bolivian Amazon fundamentally altered the floristic structure of the forest around them by encouraging economically favourable plant species, and left a lasting legacy of economic species richness in the modern forests which cover these sites.
• Population collapse occurred in the Bolivian Amazon geometric earthwork region around 500 years BP, as a result of European Contact.
• Geometric earthwork building societies in the Bolivian Amazon had a subsistence strategy based upon Manioc (*Manihot esculenta*) agriculture.

The results chapters of the thesis, presented in the style of academic papers, have the following specific research aims.

Chapter 2 aims to:
• Improve the standard pollen laboratory preparation methodology for recovery of large cultigen grains from Neotropical lake sediments.
• Determine the optimal procedure for this, which does not result in loss of small pollen grains from the sample.
• Demonstrate the new methodology’s effectiveness by testing it on sediments from tropical Bolivian lakes.

Chapter 3 aims to:
• Combine local- and regional-scale palaeoecological reconstructions from lake cores, to determine the spatial scale of pre-Columbian impacts associated with geometric earthwork construction in north-east Bolivia.
• Test the current opposing theories of large- vs. small-scale pre-Columbian deforestation in Amazonia.
• Assess the respective roles of pre-Columbian anthropogenic impact and climate in shaping vegetation change and biogeochemical cycling in the southern Amazon over Holocene timescales.

Chapter 4 aims to:
• Examine in more detail the pollen record from LG, and combine with phytolith data, to discern spatial variations in local-scale land use and impact over time on the Bella Vista earthwork site.
• Determine what the pre-Columbian agricultural practices/subsistence strategies were on the earthwork site.
• Assess the impact of pre-Columbian land use relative to the modern day.
• Extend the chronology of occupation on the site beyond the limited number of dates available from archaeological contexts.
• Assess what role the Columbian Encounter of AD 1492 may have had in the site’s abandonment.

Chapter 5 aims to:
• Provide a chronology of pre-Columbian use of the La Luna forest island site and the first dating of any ring ditch site in the forest island landscape of Iténez province.
• Determine the nature of land use on the forest island.
• Test the hypothesis that earthwork construction involved clear-cutting of the forest on the island (Erickson 2010).
• Test whether *Theobroma cacao* is a detectable economic species in the pollen record.
• Test the hypothesis that pre-Columbian people fundamentally altered the plant species composition on the island through selective management (Erickson 2010).
• Assess what role the Columbian Encounter of AD 1492 may have played in the abandonment of site.
Chapter 2. An improved methodology for the recovery of *Zea mays* and other large cultigen pollen grains, and its application in the Bolivian Amazon.

2.1. Preface

The following chapter describes a new laboratory technique developed jointly by BW, LR, and JC. The chapter is based upon a published article¹ which had the following author contributions: BW designed the research concept and led the writing of the paper. The Bolivian lake sites, Laguna San José and Laguna Isireri, were analysed by BW and JC, respectively. The Belizean site, New River Lagoon (not included here), was analysed by ER. JC performed extensive experimentation with lake sediments from Isireri, to determine the optimum protocol for successful fractionation of clay rich sediments. All authors contributed to the writing of the final paper.

While parts of the original paper have been rewritten (introduction, discussion, conclusions) and figures altered to fit the specific aims of this thesis and explain more fully the work undertaken by JC, other sections from the original manuscript (site description, methodology, results) have been inserted largely unedited. These sections were co-authored by JC and are unaltered, because they are factual descriptions of the work carried out, such as outlining of the laboratory protocol. The published manuscript is included for reference, in thesis section C. The full results of the pollen and macroscopic charcoal analyses carried out on Laguna Isireri are also reported in the Appendix (A.1.).

2.2. Abstract

This paper describes a modified lab methodology for fossil pollen analysis, developed to improve the recovery of *Zea mays* L. (maize) and other large cultigen pollen grains, from Neotropical lake sediments. Cultigen pollen grains, especially maize, are important in Neotropical palaeoecology, both as independent indicators of a historical human presence, and for the information they provide on subsistence strategies. However, these large grains are poorly transported and tend to be rare in the fossil pollen record. In this new methodology, large grains were separated through the addition of a sieving stage to the standard palynological lab protocol, which used a 53 μm sieve to split sediment sub-samples into a coarse (>53 μm) and a fine (<53 μm) fraction. We experimented with the new methodology using sediments from two large lakes situated in the pre-Columbian earthwork landscape of the Bolivian Amazon. In order to test the effectiveness of the new methodology, two samples were prepared from each depth horizon; one using the sieving methodology and the other using the standard methodology. The coarse fraction recovered from the sieved sample was scanned for cultigens, and counts of 300 terrestrial grains were compared from the fine fractions of the sieved and the non-sieved sample. While samples prepared using the standard methodology recovered no cultigen grains, the new methodology recovered maize pollen from both of our study lakes. A known concentration of *Lycopodium* spores was added during the preps to calculate confidence intervals, and confirmed that there was no statistically significant difference between the pollen assemblages of the sieved and non-sieved samples. This new technique greatly enhances our ability to detect pre-Columbian agriculture, and was important for the success of the analyses discussed in the following three chapters of this thesis (Chapters 3-5).
2.3. Introduction

Our ability to identify cultigens in the fossil pollen record is vital for investigating pre-Columbian land use and agriculture in Amazonia. As well as informing us about the agricultural strategies of pre-Columbian people, cultigen pollen can be highly useful for 1) disentangling natural from anthropogenic signals of vegetation change, by providing an independent indicator of human activity in the palaeo record, and 2) dating the start and end of agriculture on an archaeological site.

Maize is a particularly important palynological marker because, although its pollen has a similar size range to its ancestor *Balsas teosinte* (*Z. mays* subsp. *parviglumis*), it is easily distinguishable from other wild grasses using pollen morphological criteria (Holst et al. 2007). Maize produces large pollen grains (55-120 μm), which are not well transported from the source plant. This is both advantageous, as the presence of maize pollen in a lake record indicates cultivation locally around a site (Bryant and Hall 1993; Jones 1994; Islebe et al. 1996; Pohl et al. 1996; Bush et al. 2007b; Lane et al. 2010), and disadvantageous, as poor transportation means that maize is typically not well represented in the pollen record (Raynor et al. 1972; Bush et al. 1989; Behling and Hooghiemstra 1998; Jarosz et al. 2003; Lane et al. 2010; Niemann and Behling 2010). Other important cultigen species, such as sweet potato (*Ipomoea batatas* L.), yuca (*Manihot esculenta*), and squash (*Curcurbita* spp.), are also useful indicators of past cultivation. Although, unlike maize, these taxa cannot be identified to the domesticated species level using pollen, their co-occurrence with abundant macroscopic charcoal, maize pollen, and changes in arboreal to grass pollen ratios, can also be indicative of human activity. These taxa are also typically poorly represented in the pollen record, because of the large size of the grains (80-250 μm) (Herrera and Urrego 1996), and because they are relatively unproductive of pollen (Rogers 1965; Hurd et al. 1971; Real 1981; Rival and McKey 2008). The low pollen productivity and poor dispersal of large cultigen grains, means that agricultural production is typically inferred in the palaeoecological record from simple comparisons of presence/absence of cultigen pollen at each horizon.

Given the rarity of these cultigen types in the pollen record and their importance as indicators of past human land use, palynologists will often invest great effort in extending their pollen counts outside of the main terrestrial pollen sum of
300 grains, scanning multiple slides to find them (Bush et al. 1989; Clement and Horn 2001; Horn and Kennedy 2001; Anchukaitis and Horn 2005; Kennedy and Horn 2008). Modern lake studies from sites surrounded by maize fields have demonstrated that, even in these circumstances, where source plant density is high and proximity is close, when using the standard laboratory preparation and scanning outside of the sum maize pollen densities are low at < 80 grains per cm³ (Lane et al. 2010). Maize pollen grains become even rarer as lake basin size and/or distance of cultivation from the lake shore increase (Lane et al. 2010). This means that encountering cultigen pollen in the very large (≥ 10 km²), flat-bottomed lakes, which are typical in the pre-Columbian earthwork region of the Bolivian Amazon, would be extremely unlikely using the standard preparation methods.

The recovery rates of large pollen grains from lake sediments can be improved through the inclusion of a size fractioning stage in the laboratory preparations. This has been applied to European sediments, in a methodology which uses a 30 μm mesh sieve to separate out cereal pollen (Bowler and Hall 1989). This method, however, requires that two sediment sub-samples be processed, and therefore has the disadvantages of being more time consuming than processing a single sample and of being more wasteful of sediment. Sediment from a 5cm diameter core is a limited resource, not only because the study sites are on a different continent, but because many of the lakes are in remote rural areas, only reachable on foot or by horse; which therefore, precludes the possibility of taking and transporting multiple sediment cores. Sediments must be used as efficiently as possible when applying multiple analyses. A lab preparation technique which necessitates two destructive sediment sub-samplings is, therefore, not ideal.

2.3.1. Aims

Here, we present a new methodology, which introduces a sieving stage using a 53 μm sieve to the standard pollen preparation protocol, with the aim of improving the recovery of large cereal grains (Faegri and Iversen 1989). The technique was tested on sediments from two large, flat bottomed lakes, in the Bolivian Amazon, to demonstrate its effectiveness in recovering cultigen pollen where previously none had been found using the standard protocol. We also experimented with different variables in the pollen preparation protocol, such as re-ordering of the chemical
digestion stages, use of different sizes of centrifuge tubes, and increasing spin speeds, to determine the optimal procedure for effective fractionation of the sample.
2.4. Study region

2.4.1. Llanos de Moxos, Bolivia

The *Llanos de Moxos* is a large hydrological basin (130,000 km$^2$) in SW Amazonia, situated in the Beni department, lowland Bolivia (fig. 2.1). The *Moxos* is characterized by a mosaic landscape of seasonally-inundated savannahs, interspersed with forested mounds and river levees (Orellana et al. 2004). Large-scale pre-Columbian earthworks, including habitation mounds, raised fields, ring ditches, canals, causeways and fish weirs (Denevan 1966; Erickson 2000; Mann 2008; Walker 2009; Lombardo and Prümers 2010; Lombardo et al. 2011), have been identified throughout the region.

2.4.2. Study sites

The methodology was tested using sediments from two large lakes in two different archaeological sub-regions of the Bolivian Amazonia (fig. 2.1). Approximately 50 km east of Trinidad, the capital of the Beni department, the pre-Columbian earthworks consist of extensive large habitation mounds (> 100), canals and causeways (Lombardo and Prümers 2010). Situated amongst these archaeological features, Laguna San José (LSJ) (14°56′58″S, 64°29′42″W), is one of the many large (14.4 km$^2$), flat-bottomed, shallow (1 m water depth), rectilinear lakes, which are dispersed across the *Moxos* (Clapperton 1993). Two large habitation mounds, Loma Salvatierra and Loma Mendoza, lie within 2 km of LSJ, and have been extensively excavated by archaeologists (Prümers 2008; Prümers 2009a; Prümers 2009b; Lombardo and Prümers 2010; Dickau et al. 2012). Several additional pre-Columbian earthworks surround the lake, including a canal located within 100 m of its north-western shore (Lombardo and Prümers 2010). Dominant vegetation around LSJ includes seasonally-inundated savannahs, some of which are used now for cattle pasture. Forest patches are located near the lake shore.

Situated approximately 80 km west of Trinidad, Laguna Isireri (LIS) (14°59′16″S, 65°41′04″W) is another large (19 km$^2$), shallow (1.5 m water depth), rectilinear lake, located adjacent to the town of San Ignacio de Moxos. Typical of the northern and western region, evidence of pre-Columbian land-use in this area consists of raised agricultural fields (Mann 2008). Several raised fields have been
identified adjacent to LIS (Saavedra 2009), within a few hundred metres of its southern shore (per comms. Machicado-Murillo, E.P.). Although the local vegetation has been heavily modified in recent centuries, the dominant vegetation in the region is seasonally-inundated savannah, with forest fringing the lake shore.

Figure 2.1 Location of Lagunas San José and Isireri, situated near to habitation mounds and raised fields, respectively, in the Beni department, lowland Bolivia.
2.5. Methods

2.5.1. Sediment collection

Surface-sediment coring of the two lakes was performed in June and July 2010, using a 5 cm diameter Perspex® tube and piston from a floating platform. Surface cores of 31 cm (LSJ) and 51 cm (Isireri) were extruded in consecutive 0.5 cm increments in the field, into sealed plastic bags or screw-lid bottles, and shipped to the University of Edinburgh, where they were stored at 4°C. Sediments from both cores are clay-rich and contain very little organic matter (< 5%), as estimated through loss-on-ignition at 550°C.

2.5.2. Summary of Protocol

For each of the two study lakes, five pairs of 1 cm³ sediment samples were prepared. For each pair, both samples received identical chemical treatments, but one sample was treated with an extra sieving stage (53 µm), while the other control sample, taken from the same stratigraphic horizon, was not sieved.

Standard chemical digestion protocol was used for the preparation of all fossil pollen samples (Faegri and Iversen 1989; Bennett and Willis 2001), including hot 10% NaOH, 40% HF, and acetolysis treatments. Lake sediments in the Beni commonly have very high clay contents and require intensive HF treatments in order to digest the clays. Removal of clays was enhanced by beginning the sample preparation with a hot 5% sodium pyrophosphate treatment to disaggregate clays (Bates et al. 1978), followed by repeated rinses with water until the supernatant became clear. The hydrofluoric acid and acetolysis treatments followed, after which, the samples were passed through a 53 µm sieve to isolate the large cultigen pollen grains. The modified protocol is outlined diagrammatically in Fig. 2.2.

2.5.3. Isolation of large pollen grains

We used a 53 µm aperture sieve to isolate maize and other large cultigen pollen, based on the minimum size of maize pollen in the Americas determined by Holst et al. (2007). The pollen of Curcubita spp., Ipomoea batatas, and Manihot esculenta all have diameters > 80 µm (Herrera and Urrego 1996). The majority of other lowland Neotropical pollen types are <53 µm and should therefore not be
retained by the sieve. After going through all of the chemical digestion stages, the sieving stage of the preparations was begun by suspending the pellet in 10 ml of 10% NaOH, and heating in a boiling water bath for 5 min, stirring occasionally to disaggregate the sample. Samples were transferred to a small beaker, using deionized water to rinse the contents of the tube into the beaker. Approximately 30 ml of water was used to transfer the sample.

The sample was passed through a 53 µm brass sieve with a few short blasts of deionized water from a wash bottle, and in doing so, we tried to keep the total volume of filtrate <100 ml. The filtrate was saved for the standard terrestrial pollen count, and the residue was easily washed off the sieve into a 15 ml centrifuge tube, reserved for large (>53 µm) pollen grains. The filtrate was concentrated by centrifuging in 15 ml tubes at 3500 rpm, decanting, and topping up with additional filtrate until all of the fine fraction was contained in the pellet. All 15 ml tubes, now double the original number, were centrifuged and decanted.

At this stage, we checked to ensure whether the sieving had been successful (Fig. 2.2). Selected tubes containing the coarse fraction (>53 µm) of a sample were whirly-mixed, and, using a clean pipette, a small aliquot of the sample was transferred onto a slide and scanned at 100x magnification to check if the sample contained small (<53 µm) pollen and/or ‘clumped’ organic material, which binds small pollen and prevents it from passing through the sieve. Although this was rarely the case, if the sieving stage was found to be ineffective, the coarse fraction (residue) can be re-suspended in 10% NaOH and re-sieved. The resulting filtrate can be concentrated as above, and combined with the fine fraction from the first sieving attempt.

After the sieving stage, an equal number of Lycopodium tablets (Stockmarr 1971) were added to each tube containing the residue and filtrate. The separated coarse and fine fractions were then dehydrated in tertiary-butyl alcohol and mounted in silicone oil for analysis.
Figure 2.2. Flowchart of the pollen preparation method used and incorporating the sieving stage
2.5.4. Pollen Counting and statistics

The separated coarse and fine fractions were both mounted on slides for analysis. Identifications of all pollen types were made according to published tropical pollen floras (Roubik and Moreno 1991; Herrera and Urrego 1996; Colinvaux et al. 1999), a digital tropical pollen database (Bush and Weng 2007), and a Neotropical pollen reference collection consisting of >1000 specimens (collected from herbaria at the ‘Noel Kempff Mercado’ Natural History Museum in Santa Cruz, Bolivia, and the Royal Botanic Garden Edinburgh (RBGE)), which is held at the University of Edinburgh and University of Reading. The identification of *Z. mays* was confirmed using the criteria outlined in Holst et al. (2007), including analysis of the distribution of exine intertectile columnellae using phase contrast at 1000x magnification, to distinguish it from the large grains of the genus *Tripsacum* (Poaceae).

Fine fractions (<53 µm) were counted at 400x magnification for the standard terrestrial pollen count of 300 grains, and coarse fractions (>53 µm) were counted at 100x magnification for large cultigen pollen grains, in particular, *Z. mays* (maize), *Curcurbita* (squash), *Manihot* (yuca), and *Ipomoea batatas*-type (sweet potato). Results of cultigen pollen counts from each sample are presented in Table 2.1. Counting the coarse fraction also allowed us to determine the number and type of small (<53 µm) pollen grains caught on the sieve that should have been washed through the sieve into the filtrate. This was calculated using the *Lycopodium* counts of each fraction to relate the standard terrestrial pollen count (fine fraction) to the coarse fraction count. However, we found that only a negligible number of small pollen grains were inadvertently caught in the coarse fraction (Table 2.1).

Large, non-cultigen, pollen grains, that palaeoecologists would usually include as components of the standard terrestrial pollen count, for example *Inga* spp. and *Annona* spp., are also concentrated in the coarse fraction. These grains usually comprise a very low proportion of the pollen sum, but their presence can be of high ecological value for the interpretation of the pollen signal.

2.5.5. Statistics

Confidence intervals (95%) were calculated using the modification of Maher’s lognormal distributions method in Psimpoll (Maher 1972; Bennett 2007).
To relate the volume of sample analysed from the coarse fraction to that of the standard terrestrial pollen count, or the ‘count equivalent’ (Table 2.1), the number of *Lycopodium* spores found on the coarse fraction slide was expressed as a proportion of the *Lycopodium* count of the main pollen sum. For example, if 100 *Lycopodium* spores were encountered in the standard terrestrial pollen count and 1000 spores were counted on the coarse fraction slide, then the volume of material on the coarse fraction slide represents 10x the amount of sample analysed for the standard terrestrial pollen count.

To determine what proportion of small pollen grains (<53 µm) were unintentionally retained in the coarse fraction (sieve residue) during preparation, the number of small grains that were encountered in the coarse fraction slide were expressed as a percentage of the main pollen sum using the aforementioned ‘count equivalent’. Expanding on the above example, and assuming a standard count of 300 grains, the amount of residue scanned in the coarse fraction is equivalent to a count of 3000 pollen grains. Thus, if 30 small pollen grains were found on the coarse fraction slide, we can estimate that this equates to approximately 1% of the 300-grain standard pollen sum (Table 2.1).
2.6. Results and Analysis

Our results demonstrate that sieving at 53 µm successfully isolates large cultigen pollen grains such as Z. mays (Table 2.1). Although this was the only cultigen pollen type found in the test samples, subsequent use of this technique has yielded other large cultigens, including Curcurbita (squash), Manihot (yuca), and Ipomoea batatas-type (sweet potato), from additional sites in the Beni (Whitney et al. 2014), French Guiana (Iriarte et al. 2012a) and Belize (Rushton et al. 2012). The sieving methodology has also been used to recover maize pollen from the Bolivian lake sites discussed in chapters 3, 4 and 5 of this thesis.

Table 2.1 Results of the coarse (>53 µm) fraction for each sample analysed, including number of slides scanned for cultigen pollen, the proportion of small (<53 µm) pollen grains inadvertently retained within the coarse fractions, and number and type of cultigen pollen recovered. The equivalent terrestrial pollen count refers to the number of pollen grains that the volume of residue scanned would have contained, were it not sieved.

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of slides scanned for cultigen pollen</th>
<th>Equivalent terrestrial pollen count</th>
<th>No. of small (&lt;53 µm) pollen counted on slides from coarse fraction</th>
<th>% small pollen on coarse fraction slides, relative to standard count</th>
<th>No. of cultigen pollen grains recovered</th>
</tr>
</thead>
<tbody>
<tr>
<td>L. San José</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>3</td>
<td>2876</td>
<td>2</td>
<td>0.07 %</td>
<td>nil</td>
</tr>
<tr>
<td>7</td>
<td>3</td>
<td>16168</td>
<td>10</td>
<td>0.06 %</td>
<td>2 x Z. mays</td>
</tr>
<tr>
<td>8</td>
<td>2</td>
<td>14202</td>
<td>29</td>
<td>0.20 %</td>
<td>3 x Z. mays</td>
</tr>
<tr>
<td>9</td>
<td>3</td>
<td>23519</td>
<td>8</td>
<td>0.03 %</td>
<td>2 x Z. mays</td>
</tr>
<tr>
<td>10</td>
<td>3</td>
<td>33672</td>
<td>31</td>
<td>0.09 %</td>
<td>1 x Z. mays</td>
</tr>
<tr>
<td>L. Isireri</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>3</td>
<td>788</td>
<td>4</td>
<td>0.5 %</td>
<td>nil</td>
</tr>
<tr>
<td>12</td>
<td>6</td>
<td>6382</td>
<td>2</td>
<td>0.03 %</td>
<td>nil</td>
</tr>
<tr>
<td>13</td>
<td>3</td>
<td>4943</td>
<td>7</td>
<td>0.1 %</td>
<td>nil</td>
</tr>
<tr>
<td>14</td>
<td>3</td>
<td>2986</td>
<td>4</td>
<td>0.1 %</td>
<td>nil</td>
</tr>
<tr>
<td>15</td>
<td>7</td>
<td>1891</td>
<td>4</td>
<td>0.2 %</td>
<td>1 x Z. mays</td>
</tr>
</tbody>
</table>

Both study sites are substantial water bodies and the sediment cores were taken far from the lake shore (1000 m and 500 m at LIS and LSJ respectively) to ensure the recovery of a continuous sediment sequence in case of lower lake levels due to past drought. Despite the considerable distance of the core location from shore, which is shown to reduce maize pollen concentrations in the sediment (Lane et al. 2010), maize pollen was recovered from both lakes. The concentration of large pollen (>53 µm) by sieving was particularly successful for LSJ, where in some
horizons, the proportion of 1 cm³ sediment sample examined for the coarse fraction equated to over 100x the proportion of the sample examined for the standard terrestrial pollen sum, and maize pollen grains were found in four of the five horizons analysed (Table 2.1). It follows then, that even if the samples were scanned outside the standard terrestrial pollen sum, it is highly improbable that any *Z. mays* grains would have been found at LSJ and LIS, if this additional sieving stage had not been employed.

Confidence intervals calculated for the paired samples (Bennett 2007) demonstrate that the added sieving stage does not concentrate small pollen grains, such as *Cecropia*, or reduce the relative abundance of larger grains, such as Poaceae (fig 2.3). Furthermore, whatever small (<53 µm) grains are caught in the coarse fraction, they are not disproportionately-represented by any particular pollen type. In total, the number of small grains trapped in the coarse fraction is negligible (<1%), if care is taken with the digestion and sediment re-concentration stages, as explained in the discussion below.
Figure 2.3 Bar plots with 95% confidence intervals comparing sieved and non-sieved percent abundances of key taxa found in each study site.
A further key advantage to including our coarse-sieving stage is that the total pollen assemblages in the fine fraction are more concentrated, and comprise ‘cleaner’ preparations, than those for non-sieved samples, making the process of pollen counting significantly faster and easier. To reach a count of 300 terrestrial grains can be relatively time-consuming in the analysis of largely inorganic sediments from large lakes in the Bolivian lowlands, where pollen concentrations are often relatively low. In the case of LIS, the number of fine fraction slides processed in order to reach a terrestrial count of 300 grains was reduced from two or more slides to, at some horizons, one partial slide.
2.7. Discussion

2.7.1. Development of sieving protocol best practice

The development of this protocol involved several weeks of experimentation with lake sediments from LIS by JC. For this new sieving methodology to be viable, it was essential that the resulting pollen assemblages were not significantly different from those produced using a standard laboratory protocol i.e. that pollen <53 µm in size would not be retained in the course fraction after sieving or otherwise lost during the process. The first set of pollen samples prepared from LIS using the sieving methodology did produce significantly different pollen assemblages when counted. Through a series of trial and error experiments, three key elements were identified which could affect the sieving process: 1) blocking of the sieve by undigested clays, 2) sediment clumping and the formation of a sticky pellet, and 3) loss of very small (<5 µm) pollen grains during the re-concentrating of the pellet after sieving.

2.7.1.1. Removal of clays and preparation of sample for sieving

Ensuring that as of the much of the clays as possible were removed from our samples was an essential step before sieving, as cohesive clay minerals would block the sieve and prevent complete fractionation of the sample. The abundance of very small pollen (e.g. Cecropia and Mimosa, <5 µm diameter) in tropical ecosystems, precludes the possibility of fine sieving (Cwynar et al. 1979) for the removal of clays and fine silts, as is sometimes done for European pollen. To deal with these very clay rich sediments, we found that an initial treatment for 10-15 minutes with hot 5% sodium pyrophosphate (Na₄P₂O₇), which is an anti-flocculent, was an effective first step for treating clays. Treatment with sodium pyrophosphate caused clay minerals to float in the water column after spinning in the centrifuge, which allowed the excess clays to be poured-off, without losing sediment and pollen grains from the main pellet. Up to 10 repeated water washes were required before the supernatant became clear, after which a significant amount of clays had been removed from the pellet. The very high clay content of the sediments recovered from the Beni lakes requires additional long treatments with hydrofluoric acid (up to two 30 minutes treatments with 40% HF). It was found during preparations that this intensive chemical process
often resulted in a “sticky” pellet, which did not pass easily through the sieve, and resulted in smaller pollen grains being retained in the coarse fraction. The recommended method for dispersal of fluorosilicates from Bennett & Willis (2001) is treatment with hot 10% HCl. However, we did not find this treatment effective with these lake sediments. A sticky pellet can result from a build-up of chemical residues in the sample from each successive acid treatment (Bennett & Willis 2001). The addition of 3-4 wash stages with deionised water after HF treatment, and repeated careful washing with deionised water after every other chemical treatment, removed these chemical residues and produced a well dispersed pellet for sieving. We also found that by reordering the laboratory protocol so that the stage using hot sodium hydroxide (NaOH), which also acts as a deflocculent, was placed immediately before the sieving, dispersed the pellet and allowed for easier sieving.

2.7.1.2. Retention of small pollen grains

It was noted during development of the new methodology, that very small pollen grains (<5 µm diameter), such as Cecropia and Mimosa, were less well represented in the fine fractions of the sieved compared to the non-sieved samples. Scanning of the coarse fractions of the sieved samples however, showed that these missing small pollen were not being retained by the sieve, suggesting that they were being lost during another stage(s) of the sample processing. These rounds of samples were prepared using 50 ml sample tubes, which when spun-down in the centrifuge, did not produce a discreet pellet, and could result in loss of finer sediment and smaller pollen grains when decanting. It was also hypothesised that the very well digested, well dispersed nature of the sediments, meant that there was a lack of minerogenic material for very small pollen grains to colloid to and, as a result, small grains did not settle-out and remained in the water column after centrifuging. These issues where addressed by 1) judicious use of deionised water when sieving (≤100 ml), to avoid over-diluting the sample, 2) using 15ml centrifuge tubes, which formed a discreet pellet after centrifuging, and 3) increasing the centrifuge speed from 3000 to 3500 rpm, all of which ensured better retention of small pollen grains in the filtrate.
2.7.2. Implications for palaeoecology and archaeology in Amazonia

Palaeoecological studies of past human-environment interactions are faced with the complication of disentangling natural, climatic drivers of vegetation change and burning, from anthropogenic ones. In our study region of southern Amazonia, there is ample evidence of pre-Columbian settlement (Erickson 2000; Prumers et al. 2006; Lombardo and Prümers 2010; Saunaluoma 2012; Lombardo, Szabo, et al. 2013), which could potentially have had great environmental impacts, but we are also aware that this region was subject to naturally shifting forest-savannah biomes, driven by increasing precipitation in the late-Holocene (Absy et al. 1991; Mayle et al. 2000; Baker et al. 2001; Burbridge et al. 2004). It could be difficult, therefore, to distinguish climatically driven ecosystem changes from human impacts, such as deforestation. The pollen of cultivated plant taxa can provide unequivocal evidence for human land use in a palaeoecological record, and help to address our questions over the importance of pre-Columbian environmental impacts in the development of Amazonian ecosystems. Understanding the subsistence strategies and agricultural practices of pre-Columbian people, also requires more data on the types of crops grown and the environment of cultivation. However, the rarity of cultigen pollen means that it would be difficult to recover from lake core sediments using the standard laboratory protocol, without wasting valuable sediment (Bowler and Hall 1989). The technique presented here, which maximises our chance of recovering evidence of cultivation from sediment archives, is therefore a timely additional tool for investigating pre-Columbian land use.

Additionally, the use of this technique will reduce the likelihood of encountering false negatives when searching for cultigen pollen, especially in sediments from large lakes. As cultigen pollen concentrations in lake sediments are inversely proportional to distance from the plant source (Lane et al. 2010), the probability of recovering cultigen pollen from very large lake basins is greatly reduced. By selecting two large study lakes to test our new methodology, we have demonstrated that it greatly improves the likelihood of recovering cultigen pollen, even from large lake basins. Using this technique, we can therefore have greater confidence that patterns of presence vs. absence in records where cultigens are
encountered genuinely reflect patterns of change in agricultural production or the spatial distribution of agriculture around a lake, rather than being the result of dilution of the pollen signal.
2.8. Conclusions

The most effective approach to investigating pre-Columbian land use practices and past human-environment interactions, is through the combination of archaeological, archaeobotanical, and palaeoecological data (Pearsall et al. 2003; Iriarte et al. 2004; Piperno et al. 2007; Piperno et al. 2009; Duncan et al. 2009; Dickau et al. 2012). However, where this ideal line of multiple datasets is not available, the identification of cultigen pollen grains from lake sediments, combined with a palynological signal of disturbance and charcoal evidence of anthropogenic burning, can provide compelling evidence for human activity. Such data will be particularly important in regions, such as the Bolivian Amazon, where archaeological research is still developing, and little data of any kind exists. This paper has described the development of a new methodology, which maximises the potential to recover maize and other large cultigen pollen grains from lake sediments, and therefore provides a valuable tool to archaeologists and palaeoecologists for the detection of early agriculture and land use in the Neotropics.

2.9. Acknowledgements

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Chapter 3. Environmental impact of geometric earthwork construction in pre-Columbian Amazonia


3.1. Preface

The following chapter is adapted from a paper submitted for publication at the journal Proceedings of the National Academy of Sciences. As such, the chapter is written in the style of a short journal article, conforming to the strict length regulations and the publication aims of its intended journal. The author contributions to this paper were as follows: JC led the writing of the paper. JC, BW, and FM designed the research project. Pollen and charcoal analysis of Laguna Granja and Orícore was carried out by JC, although BW analysed the surface sample from L. Orícore for an earlier study. JC, BW, FM, JI, HP, and JW interpreted the data. HP contributed data on the archaeology around L. Granja. JDS conducted the botanical survey around both lake sites. All authors contributed to the writing of the final manuscript.


3.2. Abstract

There is considerable controversy over whether pre-Columbian (pre-AD 1492) Amazonia was largely ‘pristine’ and sparsely populated by slash-and-burn agriculturists, or instead a densely populated, domesticated landscape, heavily altered by extensive deforestation and anthropogenic burning. The discovery of hundreds of large geometric earthworks beneath intact rainforest across southern Amazonia challenges its status as a ‘pristine’ landscape, and has been assumed to indicate extensive pre-Columbian deforestation by large populations. We tested these assumptions, using coupled local and regional scale palaeoeological records to reconstruct land use on an earthwork site in north-east Bolivia, within the context of regional, climate-driven biome changes. This revealed evidence for a novel third scenario of Amazonian land use, which did not necessitate labour-intensive rainforest clearance for earthwork construction. Instead, we show that the inhabitants exploited a naturally open savannah landscape that they maintained around their settlement, despite the climatically-driven rainforest expansion that began ~2000 years ago across the region. Earthwork construction and agriculture in terra firme regions of what are now the seasonal rainforests of southern Amazonia may, therefore, not have necessitated large-scale deforestation using stone tools. This implies far less labour, and hence lower population density, than previously supposed. Our findings demonstrate that current debates over the magnitude and nature of pre-Columbian Amazonian land use and its impacts on global biogeochemical cycling, are potentially flawed, because they do not consider this land use in the context of climate-driven ecosystem dynamics (e.g. forest-savannah biome shifts) through the mid-to-late-Holocene.
3.3. Introduction

Evidence for the existence of large and socially complex societies in Amazonia before the arrival of Europeans (pre-AD1492) is emerging from a growing number of archaeological sites across the Amazon basin (Denevan 1966; Roosevelt 1991; Heckenberger et al. 2008; Parssinen et al. 2009; Walker 2009; Heckenberger and Neves 2009; Saunaluoma 2010; Schaan 2012; Iriarte et al. 2012a). The scale of these societies’ environmental impact and its potential legacy in modern Amazonian forest ecosystems are hotly debated. While some argue for a relatively limited and localized human influence (Meggers 1992b; Bush and Silman 2007; Peres et al. 2010; Barlow et al. 2012), others have described pre-Columbian Amazonia as a “cultural parkland”, which was widely impacted by human disturbance (Heckenberger 2003; Erickson and Balée 2006; Clement and Junqueira 2010). It has been proposed that deforestation and biomass burning before the collapse of native Amazonian populations, following European Contact in AD1492, occurred on a scale large enough to contribute to an early anthropogenic influence on the global carbon cycle (Nevle et al. 2011), and was a significant forcing of Holocene climate perturbations (Chave et al. 2008; Dull et al. 2010). It has also been suggested that this pervasive historical human influence on Amazonian forest ecosystems should change our views on their resilience to human impacts, and influence our approach to their conservation (Heckenberger et al. 2007). However, in many regions, a lack of appropriately scaled palaeoecological data means that we have no palaeoenvironmental context in which to place these societies and assess their environmental impacts.

Palaeoecological studies (Bush et al. 2007; McMichael et al. 2011; McMichael et al. 2012b; Urrego et al. 2013) conducted in aseasonal western and central Amazonia, have shown stability of the rainforest biomes throughout the Holocene, and found little evidence for significant deforestation or biomass burning by their pre-Columbian inhabitants. However, the presence of regionally extensive pre-Columbian geometric earthworks underlying the seasonal southern Amazonian rainforests (SSAR: fig 3.1A) (Heckenberger 2003; Mann 2008; Saunaluoma 2010; Erickson 2010; Schaan 2012) is suggestive of large-scale historical deforestation, by substantial populations. These earthworks, uncovered by modern deforestation, are
thought to represent only a fraction of the total which lie undiscovered beneath the intact SSAR (Parssinen et al. 2009), a sub-region of ~1,000,000 km² (see Study Area), which constitutes one fifth of the Amazon basin.

3.3.1. Aims

Here, we investigated the environmental impact of geometric earthwork building, by employing palaeoecological techniques to reconstruct vegetation change and human land use on both a local and a regional scale around a pre-Columbian earthwork site in north-east Bolivia. To gain a temporal understanding of the nature and scale of human impact on vegetation in the ring ditch region of north-east Bolivia, we analysed fossil pollen and macroscopic charcoal from radiocarbon-dated lake sediment cores, to reconstruct palaeovegetation and fire history over the past ~6000 years.
Figure 3.1 Map of site locations. *Full caption over page*. 
(A) Present day precipitation across Amazon basin during three driest months of the year (Hijmans et al. 2005); solid black line delimits modern extent of Amazonian forest; approximate extent of SSAR/geometric earthwork region is within the <125mm isohyet. Over 400 geometric earthworks have been discovered in eastern Acre, Brazil (3), and many more across Iténez (1) and Riberalta (2), Bolivia, and the Upper Xingu (4), Brazil. Previously-published lake records showing evidence for late-Holocene biome shifts are represented by white circles: a. Bella Vista, b. Chaplin (Mayle et al. 2000), c. Yaguara (Taylor et al. 2010), d. Carajas (Absy et al. 1991). Previously published lake sediment pollen records in central and western Amazonia, which show stability of the forest biome and limited or no human impact over mid-to-late-Holocene, are represented by grey circles: e. Siberia (Mourguiart and Ledru 2003), f. Chalalán and Santa Rosa (Urrego et al. 2013), g. Gentry, Parker, Vargas, Werth (Bush et al. 2007a). Black crosses represent soil-pit sampling locations/transects discussed in the main text, which show little evidence of pre-Columbian forest disturbance (McMichael et al. 2011; McMichael et al. 2012b). (B) Study area showing Laguna Orícore, main rivers and biome distribution. (C) Laguna Granja and Granja del Padre ring ditch.
3.4. Study area

Our study was conducted in Iténez province, in the Beni department of north-east Bolivia. This province lies at the geologically defined boundary between the *terra firme* (non-flooded) humid evergreen rainforest on the uplands of the pre-Cambrian Shield (PCS), and the seasonally-flooded savannahs of the adjacent sedimentary basin (Navarro and Maldonado 2005). The two study lakes are situated on either side of the confluence between the Blanco River and the San Martín River which, in northern Iténez, marks the geological divide between the uplands and the lowlands (fig 3.1B). The large lobe of the PCS to the north of the San Martin covers an estimated 5500 km$^2$ in area. The dominant vegetation type of the PCS lobe is mostly undisturbed, dense, humid, evergreen rainforest, which is floristically linked to the Madeira-Tapajós ecoregion (Navarro and Maldonado 2005) and part of the Iténez Protected Area. The *Llanos de Moxos*, a vast (135,000 km$^2$), seasonally-inundated, forest-savannah mosaic, is located to the south of the San Martín. The low elevation and thick impermeable clay soils of the basin (Clapperton 1993) means that it becomes flooded during the months of November to March, due to the passage of the South American (austral) summer monsoon. This seasonal inundation restricts forest growth to local non-flooded areas of higher elevation, such as natural outcrops of the PCS that occur in the region, but also artificial causeways that were created by pre-Columbian earth-moving cultures (Erickson 2010).

Although the modern forest-savannah boundary of our study area is locally controlled by geological, edaphic, and hydrological conditions, it is situated well within the SSAR, the southern ecotone of which is controlled by precipitation at a broad regional scale (Fig. 3.1A). These forests experience marked seasonality of precipitation in comparison to the west and central Amazon basin. This is the potential area of forest ecotonal movement over the mid-to-late-Holocene and, as estimated from the distribution of earthworks discovered so far, the potential area of geometric earthworks, which may lie undiscovered beneath the modern rainforest.

3.4.1. Geometric earthworks

The practice of geometric earthwork building appears to have developed and diverged across southern Amazonia over a long time period from prehistory (~2,500
BP) to European contact (Schaan 2012). These archaeological features show variations between the four regions of Amazonia where they have been discovered – Iténez province and Pando department in north-east Bolivia, and Acre and the Upper Xingu in Brazil. In Pando, Iténez, and the Upper Xingu region, the earthworks tend to be simple circular ditched enclosures and are typically referred to as “ring ditches” (Heckenberger et al. 2008; Saunaluoma 2010; Erickson 2010). In eastern Acre state, Brazil, the earthworks often have more complex geometric shapes and are referred to as “geoglyphs” (Parssinen et al. 2009). For convenience, we refer here to all of those features which occur along the SSAR (Mann 2008), collectively as “geometric earthworks”.

In Iténez province, the *terra firme* land of the PCS is covered by dense clusters of ring ditches, ranging from circular forms up to several hundred meters in diameter, to kilometre-long curvilinear ditches up to 3 m deep and 4 m wide. These ring ditches are part of a wider artificial earthwork landscape, covering an estimated area of 12,000 km² (Erickson 2010). The seasonally-flooded savannah in this region is crisscrossed by causeway features, which link-up small *terra firme* outcrops of the PCS or “forest islands”. In the south of the Iténez province, around the town of Baures, Erickson (2000) has identified networks of zigzagged earthworks and small ponds, which functioned as fish weirs. In northern Iténez, ridged agricultural fields have also been discovered in the savannahs (Lombardo et al. 2013a).

### 3.4.2. Study lakes

The pollen source area represented by fossil pollen in lake sediments is determined by lake surface area (Sugita 1994). We therefore selected a pair of lakes of contrasting size, to capture vegetation on two distinct spatial scales (fig. 3.1B). The smaller of the two lakes, Laguna Granja (LG), reflects local-scale vegetation at an adjacent ring ditch site (fig. 3.1C), and is nested within the regional-scale (ca. 100 x 100 km) pollen catchment of the larger lake, Laguna Orícore (LO), which reveals regional, biome-level vegetation changes on the PCS.

#### 3.4.2.1. Laguna Granja (LG)

LG (13°15’44” S, 63°42’37” W) is an oxbow lake located approximately 1 km north of the boundary of the modern village of Bella Vista, and 300 m from the
present course of the San Martín River, at its closest margin. It lies on the north, PCS side of the San Martín River. The surface area of the lake is approximately 0.2 km², with a maximum water depth of 2 m. The site was selected for its close proximity to archaeological remains, in particular, a ring ditch site named “Granja del Padre” (GDP) that lies 100 m from the eastern shore of the lake (figs. 3.2A and B). LG therefore provides a record of local-scale human impact at a ring ditch site on the PCS.

The GDP ditch is 150 m in diameter and 2 m deep, and is connected to a second ring ditch by a linear ditch that bisects an area of 150 ha. Excavations within the GDP ring ditch have uncovered 15 separate burials and associated ceramics. Surveys over an area of 200 km² around LG have documented numerous further earthworks, some enclosing areas up to 200 ha.

The vegetation immediately around LG is riverine forest, and this forest shows evidence of recent burning and clearance for shifting cultivation. On the east side of the lake, forest has been cleared to create a 0.3 km² area of grassland for cattle ranching. The most abundant terrestrial tree species in the riverine forest are Vochysia mapirensis (Vochysiaceae) and Buchenavia oxycarpa (Combretaceae). The littoral margins of the lake, especially on its north side, are dominated by floating mats of aquatic vegetation, including the water fern Marsilea polycarpa (Marsileaceae) and the water hyacinth Eichhornia azurea (Pontederiaceae). The wider area of the PCS surrounding the lake is covered with closed-canopy terra firme rainforest, which is degraded by modern anthropogenic disturbance to a distance of ~5 km from the lake.
3.4.2.2. Laguna Orícore (LO)

LO (13°20'44.02"S, 63°31'31.86"W) is one of the large (11.2 km$^2$), shallow, flat-bottomed lakes, common in the Beni department. Much of the lake margin is fringed by a 10 m strip of seasonally-inundated gallery forest. A 1.5 km wide outcrop of the PCS, currently supporting terra firme forest and containing some drought tolerant taxa, is situated at the northeast margin of the lake. Seasonally-flooded savanna dominates the wider landscape in which LO is situated, and the margin of the PCS lobe is situated 3 km north of the lake. Despite its location within open savannah, the surface sediment pollen assemblage from LO reflects humid evergreen rainforest (Gosling et al. 2005; Gosling et al. 2009; Burn et al. 2010; Jones et al. 2011), demonstrating that most of its regional pollen signal originates from the closed-canopy terra firme rainforest atop the PCS, north of the San Martín river.
3.5. Methods

3.5.1. Sediment acquisition

Fieldwork was carried out in June-July 2011. Samples were taken from a stable floating platform in the central, deepest part of both lakes, using a drop-hammer Colinvaux-Vohnout modified Livingston piston corer (Wright 1967; Colinvaux et al. 1999). Surface sediments were taken using a 5-cm diameter Perspex® tube to capture the uppermost unconsolidated sediments. Softer sediments from the surface core were divided in the field into 0.5-cm increments and stored in watertight plastic tubes. Firmer surface core sediments were extruded as intact cores in the field and shipped back to the UK in robust, watertight packaging. Livingstone core sections were transported in their aluminium core tubes and extruded in the lab in the UK. In the lab, the sediment cores were split lengthways into equal core halves, one of which was used for destructive sampling, while the other was retained as an archive core. All samples were kept in cold storage at 4°C.

3.5.2. Lab protocol: pollen analysis

The LG core was sub-sampled for pollen analysis at 5-cm intervals between 0-110 cm depth (0 to ~3,300 BP), and at 10-cm intervals between 110-150 cm depth (~3,300 to ~6100 BP). The LO core was sub-sampled at 1-cm intervals between 0-22 cm depth (0 to ~2550 BP), and at 2- to 3-cm intervals between 22-40 cm depth (~2550 to ~6700 BP). A 1-cm³ sub-sample of sediment was prepped from each horizon using a modified sieving protocol designed for optimal recovery of large cultigen pollen (Whitney et al. 2012). All other stages follow the standard pollen preparation protocol (Faegri and Iversen 1989). Samples were spiked with a known concentration of Lycopodium marker spores for calculation of pollen concentration. Pollen concentration values were calculated from pollen samples from sediment cores in both lakes, to confirm that observed changes in pollen percentage abundance in the palaeo record were not the result of changes within a closed sum (figs. 3.6 and 3.7). Pollen influx rates were also calculated for each late (figs. 3.9 and 3.10). The pollen in the fine fractions (material <53 μm) was counted to the standard 300 terrestrial grains. The coarse fractions were scanned for pollen up to a standardized equivalent count of 2000 Lycopodium grains, representing ~0.4 cm³ of sediment
scanned. Fossil pollen was identified with reference to the collection of over 1000 Neotropical pollen specimens housed at the University of Edinburgh and University of Reading. Maize pollen grains were distinguished from those of other wild grasses according to the morphological criteria described in Holst et al. (2007). Where possible, members of the Moraceae family were identified to genus using pollen reference material and morphological descriptions from Burn et al. (Burn and Mayle 2008). Where genus level identification was not possible, grains were assigned to the Moraceae/Urticaceae undifferentiated category.

In a botanical survey of the modern vegetation surrounding LG conducted by DS, Cyperaceae, which can be both a terrestrial and an aquatic herb, was identified exclusively as an aquatic/semiaquatic type on the lake margins. Cyperaceae pollen in the LG assemblages was, therefore, classified as an ‘aquatic’ type and excluded from the terrestrial pollen sum of 300 grains. (Note: The inclusion/exclusion of Cyperaceae pollen from the terrestrial pollen sum was found to have a negligible impact upon the pattern of the arboreal pollen rise seen in the LG record at 500 BP, when plotted in a percentage diagram as part of the terrestrial sum). Cyperaceae, however, is known to be an important taxon in the seasonally-flooded Moxos savannah (Jones et al. 2011) which surrounds LO, therefore, its pollen was included in the terrestrial count of 300 grains from LO.

3.5.3. Lab protocol: charcoal analysis

Samples for charcoal analysis were initially taken at 10-cm intervals, and after preliminary analysis, sampling resolution was increased where significant vegetation changes and/or burning were recognized. The final sampling strategy for LG was at every 0.5-cm interval from 0-110 cm depth, and in the remainder of the core where charcoal abundance was consistently low, sampling resolution was lowered to 5-cm intervals. LO was sub-sampled at 0.5-cm intervals from 0-40 cm depth.

Charcoal analysis was performed on sub-samples of 1 cm³ that were first treated with hot 10% sodium pyrophosphate for 10 minutes, to disaggregate the clayey sediments. The samples were then sieved using 250 μm and 125 μm nested sieves, and charcoal particles from each recovered fraction were counted in water under x40 magnification.
3.6. Results and Interpretation

3.6.1. Coring and pollen preservation

A 240-cm core was extracted from LG, including a 58-cm surface core. Sediments consisted of uniform light-brown clay throughout. Particle size analysis found that the core sediments were fine clays and silts throughout (see fig. 3.10). Pollen preservation was good throughout the core, except in horizons from 170-150 cm. Pollen/charcoal analyses and radiocarbon dating of sediments were therefore focused above 150 cm, where we could be confident of continuous sedimentation. Overlapping core sections were correlated by matching curves from high-resolution (0.5 cm) charcoal analysis.

The LO sediment core measures 120 cm, including a 54-cm surface core. Sediments consisted of uniform, stiff, light-grey clay throughout. Pollen analysis was performed on sediments between 0 cm and 40 cm, where concentrations were sufficient for analysis.

3.6.2. Radiocarbon dating

A total of five AMS $^{14}$C dates were obtained to construct a chronology for LG and 3 dates for LO. All the dates were obtained from the organic silt fraction of non-calcareous bulk sediments and the results are presented in Table 3.1 below. To construct the age models for both sets of lake cores, single age estimates for each date were derived by calculating the weighted mean of the probability distribution of the calibrated age ranges (Telford et al. 2004b), and an age-depth curve was drawn by linear interpolation between these calibrated ages (Figs 3.3 and 3.4). This simple linear interpolation is the most appropriate method, given the small number of dates (Telford et al. 2004a).
Table 3.1 AMS $^{14}$C dates for LG and LO. The dates were calibrated using the IntCal09 calibration curve in OxCal version 4.1 (Bronk Ramsey 2009). The 2σ (95%) calibrated age ranges of each date are presented. Single age estimates for each date were calculated from the weighted means of the probability distribution of the calibrated age ranges (Telford et al. 2004b).

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<th>Publication code</th>
<th>Depth below sediment-water interface (cm)</th>
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<th>Calibrated age range (cal yr BP) ± 2 σ</th>
<th>Area under probability curve</th>
<th>Weighted mean calibration (cal yr BP)</th>
<th>$\delta^{13}$C_{VPDB}(‰)</th>
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<td>0.020 0.980</td>
<td>5683</td>
<td>-19.8</td>
</tr>
</tbody>
</table>
Figure 3.3 Age-depth model for LO core, based on linear interpolation between three calibrated AMS radiocarbon dates (see Table 3.1). Error bars represent 2σ (95%) calibrated age ranges. Surface sediment is assumed to be modern or 0 years BP.

Figure 3.4 Age-depth model for LG core, based on linear interpolation between five calibrated AMS radiocarbon dates (See Table 3.1). Error bars represent 2σ (95%) calibrated age ranges. Surface sediment is assumed to be modern or 0 years BP.
3.6.3. Laguna Orícore pollen and charcoal

In zone LO-1 (5700-2000 BP) (fig. 3.5), LO shows low arboreal pollen levels (≤30%) and high Poaceae (grass) abundance (up to 50%), which, together with peak abundance of drought-tolerant taxa such as *Anadenanthera* and maximum charcoal concentrations, indicates a drier and regionally more open environment compared with present (Gosling et al. 2005; Gosling et al. 2009; Burn et al. 2010; Jones et al. 2011). At the zone LO-1/LO-2 boundary (~2000 BP) total arboreal pollen percentages begin to increase, with especially marked increases in abundance of evergreen taxa such as *Brosimum* and *Alchornea*, while Poaceae, *Anadenanthera* and charcoal levels decline, signalling an expansion of closed-canopy humid evergreen rainforest (Gosling et al. 2005; Gosling et al. 2009; Burn et al. 2010). By ~1700 BP, arboreal pollen percentages double, indicating that the regional *terra firme* PCS landscape reached a level of forest cover comparable to the modern closed-canopy rainforest.
Figure 3.5 L. Orícore stratigraphy, pollen and charcoal record. Pollen and charcoal presented as percent abundance and particles/cm³, respectively. “Total Arboreal” is the sum of all tree taxa. Dominant tree taxa (> 2% abundance) are shown. Plotted against calibrated radiocarbon years BP and individual radiocarbon dates shown on right (See table 3.1 and figure 3.3).
Figure 3.6 Pollen concentration values for L. Orícore. Dominant taxa, total arboreal and total overall pollen concentrations are shown as grains per cm$^3$. 
Figure 3.7 Pollen influx values from L. Oriocore. Key taxa, total arboreal and total pollen influx values are shown in grains per cm² per year.
3.6.4. L. Granja pollen and charcoal

In zone LG-1 (6100 – 2500 BP) (fig. 3.8) the high ratio of Poaceae to arboreal pollen shows that, prior to occupation, the environment around LG was an open savannah, in contrast to the dense evergreen rainforest that exists there today (notwithstanding small-scale clearings made for cattle pasture) (Gosling et al. 2005; Gosling et al. 2009; Burn et al. 2010; Jones et al. 2011). The LG-1/LG-2 zone boundary marks the onset of agriculture and anthropogenic burning at LG, at 2500 BP, as indicated by a sharp charcoal increase and the presence of maize pollen (Zea mays L.). Although the charcoal record shows variations, with two distinct periods of more intensive anthropogenic burning at ~2500-1600 BP and ~700-500 BP, the almost continual presence of maize pollen shows that the site was occupied throughout zones LG-2 and LG-3 (2500-500 BP and 500 BP- present, respectively). The second charcoal peak (700-500 BP) coincides with peak Poaceae pollen percentages and ring ditch construction/occupation (750-550 BP) (Dickau et al. 2012), which indicates either a more intense period of burning associated with the ring ditch culture, or burning closer to the lake itself. Open savannah/grassland vegetation persists throughout zone LG-2, and expansion of dense-canopy rainforest around LG does not occur until ~500 BP (zone LG-2/3 boundary). This follows a probable decline of human activity on the site, possibly linked to the introduction of Old World diseases associated with the ‘Columbian Encounter’ (CE) in AD1492 (Denevan 1992b). Our age-depth model for LG (fig. 3.4) shows an increase in sedimentation rate from ~2000 BP. As expected, this translates to an increase in the pollen influx rate (fig. 3.10), but does not appear to impact pollen concentration values (fig. 3.9) or pollen percentages/charcoal abundance (fig. 3.8).
Figure 3.8 L. Granja stratigraphy, pollen and charcoal records. Pollen and charcoal presented as percent abundance and particles/cm$^3$, respectively. Zea mays plotted as no. of grains per ~0.4 cm$^3$. “Total Arboreal” is the sum of all tree taxa. Dominant tree pollen types (>2% abundance) are shown. Dashed line indicates timing of regional forest expansion shown in LO. Plotted against calibrated years BP, with individual radiocarbon dates shown on the right (see table 3.1 and figure 3.4).
Figure 3.9 Pollen concentration values from L. Granja. Key taxa, total arboreal and total overall pollen concentrations are shown as grains per cm$^2$. 
Figure 3.10 Pollen influx values and sediment particle size from L. Granja. Dominant taxa and overall total pollen influx values are shown as grains per cm$^2$ per year. Particle size data is split into three size categories: clays (<2μm, <5μm) and silts (<30μm).
The timing of this local forest expansion at LG (~500 BP), contrasts with the much earlier regional-scale pattern of forest expansion seen at LO (~2000 BP). The most parsimonious explanation for the persistence of this open landscape at LG is that early settlers maintained open ground on a local scale, by suppressing forest expansion around the settlement. The maintenance of this opening is not seen in the LO record, because the much larger pollen catchment area of LO means that it is insensitive to detecting openings on the scale of the ring ditch site (Sugita 1994). The pollen record, therefore, demonstrates that construction of the earthworks and agricultural activity around LG occurred in an open environment, maintained since the first occupation of the site. Rather than having to rely upon stone axes and burning to clear dense-canopy rainforest, a feat that would have been hugely labour intensive and highly impractical before the introduction of steel tools (Denevan 2001), our data show that the inhabitants took advantage of an existing open savannah landscape. The maintenance of a locally open landscape around LG, by the suppression of tree growth associated with the climate-driven rainforest expansion, would have required far less human effort (i.e. lower population density) than that needed for the removal of dense-canopy rainforest.
3.7. Implications

It is intriguing that most geometric earthworks found beneath *terra firme* tropical forest have been concentrated in seasonal southern Amazonia (i.e. north-east Bolivia, eastern Acre state and the upper Xingu region of Brazil), rather than wetter, less seasonal parts of the basin (fig. 3.1A). Late Holocene climate-driven rainforest expansion has been documented in other parts of southern Amazonia (Absy et al. 1991; Mayle et al. 2000; Taylor et al. 2010), coinciding with rising lake levels in the tropical Andes (Cross et al. 2000; Baker et al. 2001), which demonstrates that the forest expansion at LO reflects a broad-scale vegetation response to increasing precipitation across the southern Amazon. However, the geographic scale of this biome shift in the context of large geometric earthwork construction, has hitherto not been considered. Our discovery raises the strong possibility that other geometric earthworks across southern Amazonia were also built in an open savanna/woodland that subsequently became covered by closed-canopy forest.

Gaining a sound understanding of the historical role of humans in shaping Amazonian landscapes, and the extent to which Amazonian forests were resilient to historical disturbance, is critical to informing policy decisions on sustainable Amazonian futures (Heckenberger et al. 2007; Clement and Junqueira 2010; Barlow et al. 2012; Renard et al. 2012; Iriarte et al. 2012a). However, the debate so far has considered only the extent of past human impact and land use in a forested landscape, assumed to be analogous to the modern environment. Our study demonstrates that current debates over the magnitude and nature of pre-Columbian Amazonian land use are potentially flawed, as they do not consider this land use in the context of climate-driven ecosystem dynamics through the mid-to-late-Holocene (Absy et al. 1991; Mayle et al. 2000). Our findings show that, in order to determine the scale of environmental impact associated with pre-Columbian land use, the type of vegetation cover (e.g. closed-canopy forest versus open woodland or savannas) antecedent to settlement, must first be known. This is a vital pre-requisite to drawing inferences regarding population size and subsistence strategies.

Our findings have implications too for biogeochemical cycling. It has been speculated that post-Columbian secondary forest re-growth and the reduction in biomass burning across the Neotropics following indigenous population collapse,
caused sufficient carbon sequestration to reduce atmospheric CO$_2$ concentrations and amplify Little Ice Age cooling (Dull et al. 2010; Nevle et al. 2011). It has also been suggested that observed biomass gains amongst old-growth, lowland Neotropical forests, over recent decades, may be due to continued recovery from pre-Columbian anthropogenic disturbance (Chave et al. 2008). Our data support an alternative scenario, whereby pre-Columbian terra firme agriculture and construction of vast and numerous earthworks, did not necessitate large-scale deforestation, and the forest in these regions expanded in response to increasing precipitation over the past 2000 years, rather than solely as part of a post-disturbance forest recovery over the last 500 years, following indigenous population collapse associated with the CE.
3.8. Acknowledgments:

This research was supported by a Leverhulme Trust research grant (F/00158/Ch) awarded to FM and JI and a NERC Doctoral Training Scheme grant (NE/152830X/1) and funds from the University of Edinburgh’s Principal’s Career Development Scholarship, awarded to JC. A NERC radiocarbon facility date was granted to FM (1623.0312). Fieldwork support was provided by the Noel Kempff Mercado Natural History Museum, Santa Cruz, Bolivia. We thank Douglas Bruckner of the ‘Programa de Conservación de la Paraba Barba Azul’, Trinidad, Beni Department, Bolivia and the rangers from the ‘Reserva Iténez’ WWF station in the town of Bella Vista, Beni Department, Bolivia, for logistical support in the field. We also thank José Manuel Barrios Fernández for allowing us access to core the L. Granja site.
Chapter 4. A palaeoecological record of land use and habitation at a pre-Columbian ring ditch site in NE Bolivia

Authors: Carson, J.F., Watling, J., Mayle, F.E., Whitney, B.S., Soto, J.D. and Iriarte, J.

4.1. Preface

The following chapter has been written in academic publication style for submission to the journal Quaternary Research and conforms to the length requirements of that publication. The author contributions to this paper were as follows: JC led the writing of the paper. JC, BW, and FM designed the research project. Pollen and charcoal analysis of Laguna Granja was carried out by JC. Phytolith analysis from L. Granja was carried out by JW. Interpretation and integration of pollen and phytolith data was done by JC, with assistance from JW. JC, BW, FM, JI, HP, and JW interpreted the data. HP contributed data on the archaeology around L. Granja. JDS conducted the botanical survey around L. Granja. All authors contributed to the writing of the final manuscript.
4.2. Abstract

The Bolivian Amazon has yielded some of the most impressive evidence for large and complex pre-Columbian societies in the Amazon basin. However, there remains relatively little data concerning the chronology, land use practices and environmental impact of these societies. Palaeoecology, when closely integrated with archaeological data, has the potential to help fill these gaps in our knowledge. We present a 6,000 cal year BP record of burning and vegetation change inferred from pollen, charcoal, and phytolith remains, from an oxbow lake located adjacent to a pre-Columbian ring ditch site in north-east Bolivia (13°15’44” S, 63°42’37” W). Human occupation is inferred from pollen and phytoliths of *Zea mays* and macroscopic charcoal evidence of anthropogenic burning. The persistence of maize in the record suggests that it was the staple crop grown in this region in pre-Columbian times, and abundant macroscopic charcoal suggests that pre-Columbian land management employed more extensive burning of the landscape than the slash-and-burn agriculture practiced around the site today. Radiocarbon dates from this lake have extended the beginning of human occupation of the site to 2500 years BP, with the first evidence for maize cultivation at 1850 BP. Maize was grown continuously on the site through to near-modern times, however there is evidence for a post-500 BP decline in agricultural intensity or change in land use strategy, and possible population decline, post European contact. We demonstrate the value of closely integrating archaeological data from pre-Columbian Amazonian sites with palaeoecological records from nearby small lakes for 1) establishing a continuous chronology of occupation, 2) gaining a better understanding of subsistence strategies and the spatial variations in land use, and 3) establishing the scale of pre-Columbian sites relative to modern disturbance.
4.3. Introduction

In recent decades we have seen a paradigm shift in our ideas over the size and complexity of pre-Columbian (pre-AD 1492) Amazonian societies. Rather than being limited to small, semi-nomadic, hunter-gatherer groups (Meggers 1992b), there is abundant archaeological evidence, in the form of settlement remains, artificial earthworks and black earth (terra preta) soils, for sedentary groups with relatively large populations in many different parts of the Amazon basin (Roosevelt 1991; Erickson 2000; Walker 2000; Heckenberger 2003; Parssinen and Korpisaari 2003; Glaser 2007; Walker 2009; Saunaluoma and Schaan 2012; Iriarte et al. 2012b). However, there is still considerable debate over the nature of land use, scale of environmental impact and chronology of these societies.

The Bolivian department of the Beni (fig. 4.1A) in south-west Amazonia has some of the most diverse and extensive examples of pre-Columbian earthworks. These include raised agricultural fields, monumental mounds, and in the north-east province of Iténez, ring ditches, causeways and fish weirs, covering an estimated area of 12,000 km² (Denevan 1966; Erickson 2010; Lombardo et al. 2011). Remote sensing and ground based surveys in Iténez have begun to map the spatial extent of these earthworks (Erickson 2010; Prümers 2012a,b), while archaeological excavations are documenting the material culture of the societies that built them, and have provided dating from occupation layers within excavated ring ditch features (Prumers et al. 2006). Archaeobotanical analyses have been employed to uncover aspects of palaeo diet (Dickau et al. 2012), and historical ecological studies of the vegetation surrounding earthwork sites, which are informed by modern ethnographic data, have attempted to reconstruct the legacy of pre-Columbian land management on extant forest (Erickson and Balée 2006). However, these studies often lack the temporal depth/continuity to be able to discern changes in land use, agriculture, and legacy of environmental impact in the ring ditch region, over Holocene timescales.

There is a paucity of dating evidence from archaeological sites in Iténez, meaning that the timing of occupation and earthwork construction in this region are not well constrained. Furthermore, the limited number of dates that have been published are from archaeological layers, meaning that they tell us the timing of construction and occupation of individual ring ditches, but may not date the first
occupation/abandonment of a site, or any occupation which did not leave behind visible evidence in the form of earthworks and artefacts. A key question is whether epidemic crises, warfare and cultural changes following European contact, precipitated a collapse of indigenous societies after AD 1492, or whether site abandonments may have pre-dated European contact, and were instead caused by internal social, political, or environmental factors. Gaining a sound chronology of occupation is therefore vital to informing such debates over the antiquity and decline of indigenous Amazonian societies.

Similarly, archaeobotanical studies using phytoliths and starch grains recovered from stone tools identify the cultigens processed at a particular point in the archaeological record, but do not tell us whether the same staples have always been cultivated or whether this changed over time. Such studies also yield no information on the type or intensity of land use associated with subsistence at these sites. Mapping surveys delineate the spatial extent of earthworks, however we cannot directly infer from these the extent of environmental impacts such as burning or forest clearance associated with their construction, or indeed the extent of any subsequent reforestation. Understanding this will inform debates over the resilience of Amazonian ecosystems to pre-Columbian environmental impacts and the pristine nature of Amazonian forests (Meggers 2003; Heckenberger 2003). Defining the scale and nature of impact is also important for discussions of the proposed role of pre-Columbian peoples in the early Anthropocene, and whether they had a significant impact on Holocene biomass levels and carbon emissions, through deforestation and burning (Dull et al. 2013; Nevle et al. 2011; Power et al. 2012).

To address this paucity of chronological and spatial detail, Mayle & Iriarte (2012) proposed an integrative approach, combining local-scale palaeoecological records from small lakes with archaeological/archaeobotanical data from nearby earthwork sites. The regular and continuous nature of sediment accumulation in lakes, and our ability to isolate and identify cultigens through their pollen and phytoliths, offers the chance of establishing a continuous chronology of human settlement/land use. Pollen and phytoliths have been shown to be complimentary proxies when reconstructing tropical environments from lake sediments (Whitney et al. 2013), with pollen providing higher taxonomic resolution within arboreal types,
and phytoliths better able to distinguish between grass and herb taxa. By combining palaeoecological and archaeological data, we gain insights into the land use strategies of pre-Columbian peoples, the spatial extent of past impacts, and the palaeoenvironmental context of those human-environment interactions.
4.3.1. Aims

Here, we apply this integrative approach to an earthwork site in the modern town of Bella Vista, in north east Bolivia, by analysing pollen, phytoliths, and macroscopic charcoal from Laguna Granja (LG), an oxbow lake within the earthwork site. The pollen and charcoal records from LG were previously discussed in Carson, et al. (Chapter 3) in comparison with a regional-scale lake record, to determine the palaeoenvironment of pre-Columbian geometric earthwork construction in the north-east Bolivian Amazonia. Here, these are combined with new phytolith data from LG, and archaeological data from previous excavations (Prumers et al. 2006; Dickau et al. 2012), to determine the occupation history of the Bella Vista earthwork site, and discuss the land use practices and potential environmental impacts of its pre-Columbian inhabitants. Specifically we address the following questions:

1. What was the period of occupation on the site and did abandonment coincide with the arrival of Europeans ~500 years BP?

2. Was maize the staple crop as suggested by Dickau et al. (2012) or did subsistence involve inputs from other cultigen types? Did this change over time?

3. What was the nature of pre-Columbian land management (i.e. did it involve extensive burning, clearance or manipulation of economically useful forest resources) and what was its spatial extent?
4.4. Study site and archaeological background

The modern village of Bella Vista is located in Iténez, the easternmost province of the Beni department, Bolivia, on the north side of the San Martín River (fig. 4.1). The river marks the geo-ecological divide between two Amazonian ecosystems. To the north and west, the highlands of the Pre-Cambrian Shield (PCS) support dense canopy, evergreen rainforest, which is taxonomically linked to the Madeira-Tapajós ecoregion. To the south and east is the Llanos de Moxos, a vast, low lying sedimentary basin which, due to the impermeability of its clay sediments (Clapperton 1993), becomes largely flooded during the annual wet season from November-March. As a result of this annual flooding, the landscape is maintained as a wetland savannah, interspersed by small outcrops of the PCS (Clapperton 1993), which support terra firme rainforest, and are known as “forest islands”. Earthworks in this region consist of ring ditches on the terra firme, ranging from hundreds of metres to kilometres in length, and in the savannah, linear causeway structures which link up the terra firme sites (Lombardo et al. 2011).

![Map of the Llanos de Moxos, northern Iténez and study site location. A shows the Llanos de Moxos in Bolivia with study area highlighted. B shows position of LG on the Shield and relative to the village of Bella Vista. Approximate land area of Bella Vista Village is represented by orange shaded area. C shows vegetation cover around LG and position of the Granja del Padre ring ditch.](image-url)
Surveys around Bella Vista village have documented numerous ditched earthworks across the site, enclosing areas of up to 200 ha (Prümers 2012a,b). Two of the circular ditches, Granja del Padre (GDP) and BV-3, were excavated by Prumers et al. (2006). The two ring ditches are located 1 km apart and are connected by a long, semi-circular ditch, which surrounds an area of 150 ha. The GDP ring ditch has a 2 m deep trench and measures 150 m in diameter. A total area of 600 m² of GDP was excavated, uncovering 15 urn burials and a single thin cultural layer, which was radiocarbon dated from soot on ceramic sherds to between ~650-750 years BP. BV-3 also had a single, thin occupation layer, which was radiocarbon dated from ceramic sherds to ~550-750 years BP (full data on radiocarbon dates are given in Table 4.1 below).

The dates are reported as median ages of the calibrated age range, which is how they were reported in the original publication. The calibrated age ranges show unimodal distributions, therefore this method of reporting a single age estimate was deemed acceptable. As is common with radiocarbon dates from material recovered from archaeological layers, the dating sequence does not show a perfect age-depth relationship. The dates from the GDP site show age reversals at 92-93 cm depth and at 120-130 cm depth. The date of 754 cal BP at 93 cm depth is not incongruous with the date of 649 cal BP above at 90-100 cm, when we consider that this compares a bulk sediment date taken across a 10 cm depth range, with a macroscopic charcoal date taken from a 2 cm horizon within that depth range. The lowermost date of 752 cal BP at 120-130 cm however is not congruous with the dating above it and must be the result of younger material being transported further down the soil profile. Dating at 70-80 cm depth produced an age of 767 cal BP, which is older than the date below. This was attributed to older material being reworked into a higher layer in the sediment profile (Prümers et al 2006). Prümers et al. (2006) noted that the oldest dates from both sites (4517 cal BP; 5231 cal BP from GDP, and 7964 cal BP from BV-3) were recovered from a “sterile”, pre-cultural layer. This led them to conclude that there had been no older occupation at either of the ring ditch sites before ~750 BP. These archaeological data highlight the difficulty of obtaining a continuous and linear age-depth relationship from terrestrial archaeological contexts, and the potential value of lake sediment records in providing a continuous chronology.
Table 4.1 Radiocarbon dates from archaeological contexts in GDP and BV-3 ring ditch sites, Bella Vista. Dates where calibrated to 95% (2σ) confidence interval using the IntCal09 calibration curve in the OxCal program version 4.1 (Bronk Ramsey 2009)

<table>
<thead>
<tr>
<th>Laboratory no.</th>
<th>Site</th>
<th>Depth (cm)</th>
<th>Radiocarbon age ($^{14}$C yrs BP)</th>
<th>Calibrated age range (yrs BP)</th>
<th>Median age (yrs BP)</th>
</tr>
</thead>
<tbody>
<tr>
<td>KIA 38831</td>
<td>GDP</td>
<td>70-80</td>
<td>782 ±27</td>
<td>786-721</td>
<td>767</td>
</tr>
<tr>
<td>KIA 48488</td>
<td>GDP</td>
<td>90-100</td>
<td>607 ±28</td>
<td>705-595</td>
<td>649</td>
</tr>
<tr>
<td>KIA 38833</td>
<td>GDP</td>
<td>92-93</td>
<td>783 ±25</td>
<td>784-723</td>
<td>754</td>
</tr>
<tr>
<td>KIA 38830</td>
<td>GDP</td>
<td>110-120</td>
<td>3978 ±27</td>
<td>4573-4461</td>
<td>4517</td>
</tr>
<tr>
<td>KIA 38829</td>
<td>GDP</td>
<td>113.5</td>
<td>4526 ±26</td>
<td>5359-5104</td>
<td>5231</td>
</tr>
<tr>
<td>KIA 48489</td>
<td>GDP</td>
<td>120-130</td>
<td>775 ±25</td>
<td>780-722</td>
<td>752</td>
</tr>
<tr>
<td>KIA 40610</td>
<td>BV-3</td>
<td>10-20</td>
<td>432 ±18</td>
<td>658-528</td>
<td>550</td>
</tr>
<tr>
<td>KIA 40611</td>
<td>BV-3</td>
<td>80-90</td>
<td>470 ±27</td>
<td>758-709</td>
<td>748</td>
</tr>
<tr>
<td>KIA 40612</td>
<td>BV-3</td>
<td>140</td>
<td>6936 ±30</td>
<td>7589-7740</td>
<td>7964</td>
</tr>
</tbody>
</table>

An analysis by Dickau et al. (2012) of the phytoliths and starch grain residues left on stone tools excavated from these ditches, found maize to be the main identifiable cultigen, and led the author to tentatively conclude that maize was the staple crop grown on site. Numerous theories exist as to the function of the ring ditches, ranging from a method of hydrological engineering (Walker 2009) to ceremonial purposes, however the architecture of the ring ditches around Bella Vista led Prumers et al. (2006) to conclude that they were probably defensive features.

The lake cored for this study, LG (13°15’44” S, 63°42’37” W), is a small (0.2 km$^2$), oxbow lake, located 100 m from the GDP ring ditch (fig. 4.1C) and 1 km north of the modern town. Maximum water depth during the dry season is 2 m. The majority of the lake margins are dominated by gallery forest which, according to local inhabitants, becomes flooded up to a height of 2 m every year during the annual wet season. On the east side of the lake, a small area of land (~0.3 km$^2$) has been cleared for cattle grazing.
4.5. Materials and methods

4.5.1. Sample acquisition

Fieldwork was carried out in June-July 2011. Samples were taken from a stable floating platform in the central, deepest part of the lake, using a modified Livingston piston corer (Colinvaux et al. 1999). Surface sediments were taken using a 5-cm diameter Perspex® tube to capture the uppermost unconsolidated sediments. Softer sediments from the surface core were sub-sampled in the field at 0.5-cm intervals and stored in watertight plastic tubes. Firmer sediments were extruded in the field as intact cores and shipped back to the UK in robust, watertight packaging. Livingstone core sections were transported in their aluminium core casings and extruded in the lab in the UK. In the lab, the sediment cores were split lengthways into equal core halves, one of which was used for destructive sampling while the other was retained as an archive core. All samples were kept in cold storage at 4°C.

4.5.2. Modern vegetation survey

During fieldwork a rapid assessment botanical survey was made of the vegetation around LG to aid with pollen and phytolith identification, and in determining the spatial representation of the microfossil record. Species encountered on a 100 m transect were classified and voucher specimens were collected for the herbarium of the Museum of Natural History ‘Noel Kempff Mercado’, in Santa Cruz, Bolivia (table 4.3).

4.5.3. Chronology

The core was dated using the AMS $^{14}$C method. All the dates were from the organic silt fraction of non-calcareous bulk sediments, because the core lacked plant macrofossils and macroscopic charcoal particles large enough for radiocarbon dating. Dates were calibrated to 95% (2σ) confidence interval using the IntCal09 calibration curve in the OxCal program version 4.1 (Bronk Ramsey 2009). Given the small total number of dates, the best representation in an age model was achieved using simple linear interpolations between data points (Bennett 1994; Telford et al. 2004a) (fig.4.2). Single age estimates for each date were calculated using the weighted means of the probability distribution of the calibrated ages (Telford et al. 2004).
4.5.4. Pollen analysis

After an initial coarse resolution pollen analysis was made of the core, sample resolution was increased, focusing on depths where significant changes in vegetation were observed. From 0-110 cm depth, sampling resolution was increased to 5-cm intervals, and from 110 cm to the base at 150 cm, resolution was increased to 10-cm intervals.

A 1 cm$^3$ sub-sample of sediment was prepped from each horizon using a modified sieving protocol designed for optimal recovery of large cultigen pollen (Whitney et al. 2012). All other stages followed the standard pollen preparation protocol (Faegri and Iversen 1989). Samples were spiked with a known concentration of *Lycopodium* marker spores for calculation of pollen concentration values, and to confirm that observed changes in pollen percentage abundance were not the result of changes within a closed sum (fig 4.5). Pollen influx values were also calculated (fig 4.6). The pollen in the fine fractions (material <53 μm) was counted to the standard 300 grains. The coarse fractions were scanned for large cultigen pollen grains up to a standardised equivalent count of 2000 *Lycopodium* spores, representing ~0.4 cm$^3$ of the original 1 cm$^3$ of sediment processed. Fossil pollen was identified with reference to the collection of over 1000 tropical pollen specimens housed at the University of Edinburgh and University of Reading, and from atlases of Neotropical pollen (Colinvaux et al. 1999; Bush and Weng 2007). Maize grains were distinguished from other wild grasses according to the morphological criteria described in Holst (et al. 2007). Where possible, members of the Moraceae family were identified to genus using morphological descriptions from Burn & Mayle (2008). Where genus level identification was not possible, grains were assigned to the Moraceae/Urticaceae undifferentiated category. Cyperaceae and *Alternanthera* were identified in the modern botanical survey as common aquatic/semi-aquatic types around the modern lake. They were therefore counted but excluded from the terrestrial count of 300 grains. They are presented as part of the aquatic flora.
4.5.5. Phytolith Analysis

Sediments were sub-sampled for phytolith analysis at 10-cm resolution throughout the core, with an additional three samples at 22, 35, and 45 cm depth. Phytolith extraction followed standard procedures established by Piperno (2006). Samples were pre-treated to remove clays through deflocculation and gravity sedimentation using a centrifuge, carbonates were removed using 36% HCl, and organics were removed by heating the sample in a solution of 70% HNO₃. Phytoliths were extracted by heavy liquid flotation in ZnBr₂ (specific gravity 2.3g/cm³) and mounted with Entellan® mounting agent to allow 3D rotation under the microscope. Due to the small volumes of sediment we were working with, phytoliths were not separated into size fractions (Piperno 2006). Despite the small sediment volumes, all samples yielded abundant quantities of phytoliths. A minimum of 200 phytoliths were counted per slide and the whole slide was scanned for diagnostic crop phytoliths. Phytoliths were identified by comparison with a phytolith reference collection of over 750 neotropical plant taxa held at University of Exeter (Watling and Iriarte 2012). Identification of Poaceae short cell phytoliths followed a system first proposed by Twiss et al. (1969) and later expanded to include other aspects of 3-dimensional morphology (Brown 1984; Piperno and Pearsall 1998; Pearsall 2000). As with the pollen data, Cyperaceae phytoliths are presented separately from the terrestrial sum.

4.5.6. Charcoal Analysis

Macroscopic charcoal analysis was carried out on the core from 0-150 cm. Samples were initially taken at 10-cm intervals. Where this process identified significant vegetation changes and/or burning, the sampling resolution for charcoal analysis was increased. From 0-110 cm sampling resolution was increased to 0.5-cm intervals, while at the base of the core, between 230-190 cm, sampling resolution was increased to 5-cm intervals. Sub-samples of 1 cm³ were taken from each horizon and heated in 10% sodium pyrophosphate to disaggregate clay sediments. The samples were then sieved at 250 μm and 125 μm, and charcoal particles counted in water under x40 magnification. All graphs were drawn using the program C2 (Juggins 2007).
4.6. Results and Interpretation

4.6.1. Core stratigraphy and chronology

A 240 cm core was recovered from LG, including a 58-cm surface core. The sediment throughout was a light to medium brown clay, with some fine sands. Initially, this suggested that sedimentation rates were continuous throughout the core, however, pollen preservation between 150-170 cm was found to be poor, and therefore, palaeoecological analysis and radiocarbon dating of sediments were focused above 150 cm depth. Particle size analysis of the core revealed that the sediment consists of fine clays and silts throughout (fig. 4.6).

A total of five AMS $^{14}$C dates were obtained to build a chronology for the LG record (table 4.2). No reversals were observed in the chronology (fig. 4.2).
<table>
<thead>
<tr>
<th>Site and sample identifier</th>
<th>Publication code</th>
<th>Depth below sediment-water interface (cm)</th>
<th>Conventional C^{14} age (yr BP±1σ)</th>
<th>Calibrated age range (cal yr BP) ± 2 σ</th>
<th>Area under probability curve</th>
<th>Weighted mean calibration (cal yr BP)</th>
<th>δ^{13}C_{VPDB}‰</th>
</tr>
</thead>
<tbody>
<tr>
<td>Granja Gr 21.5</td>
<td>Beta - 339227</td>
<td>21.5-22.5</td>
<td>240 ±30</td>
<td>430-357 332-280 170-150 11-0</td>
<td>0.299 0.547 0.131 0.023</td>
<td>306</td>
<td>-26.4</td>
</tr>
<tr>
<td>Granja Gr 45</td>
<td>Beta - 339228</td>
<td>45-45</td>
<td>750 ±30</td>
<td>730-660</td>
<td>1</td>
<td>696</td>
<td>-23.1</td>
</tr>
<tr>
<td>Granja Gr 91</td>
<td>SUERC-43148</td>
<td>91-92</td>
<td>1782 ±38</td>
<td>1820-1610</td>
<td>1</td>
<td>1716</td>
<td>-22.2</td>
</tr>
<tr>
<td>Granja Gr 123</td>
<td>Beta-347192</td>
<td>123-124</td>
<td>4070 ±30</td>
<td>4800-4770 4700-4670 4650-4500 4490-4440</td>
<td>0.128 0.021 0.691 0.160</td>
<td>4585</td>
<td>-23.3</td>
</tr>
<tr>
<td>Granja GR 146</td>
<td>Beta - 339229</td>
<td>146-147</td>
<td>5200 ±30</td>
<td>6000-5910</td>
<td>1</td>
<td>5954</td>
<td>-24.2</td>
</tr>
</tbody>
</table>
Figure 4.2 Age depth model for LG from 2 σ calibrated radiocarbon dates.

4.6.2. Modern vegetation and surface microfossils

The results of the modern vegetation survey are presented in table 4.3. Before discussing the palaeoecological record, we compare this survey data to the pollen and phytolith assemblages from the surface lake sediments, to determine the taxonomic and spatial representativeness of the fossil record. The vegetation survey identified 62 species, mostly representing the inundated/gallery forest zone around the lake. The most common terrestrial tree types were *Vochysia mapirensis* (Vochysiaceae) and *Buchenavia oxycarpa* (Combretaceae). The dominant aquatic species were the water fern *Marsilea polycarpa* (Marsileaceae) and the water hyacinth *Eichhornia azurea* (Pontederiaceae).
Table 4.3 Results of modern vegetation survey around LG. An * indicates that the Family or Genus was identified in the surface pollen assemblage. This symbol ǂ indicates that the Family or Genus was identified in the surface phytolith assemblage.

<table>
<thead>
<tr>
<th>Family</th>
<th>Genus (sp.)</th>
<th>Occurrence in modern environment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acanthaceae</td>
<td>Ruellia cf. nitida (Nees) Wash. &amp; J.R.I. Wood</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Amaranthaceae*</td>
<td>Alternanthera* paronychoides</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td>Anacardiaceae*</td>
<td>Tapirira sp.*</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Apocynaceae</td>
<td>Tabernaemontana cf. linkii</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Tabernaemontana sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Asteraceae*</td>
<td>Eupatorium sp.</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Elephantopus mollis Kunth</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td>Chrysobalanaceae</td>
<td>Licania cf. canescens Benoist</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Licania kunthiana Hookf.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Licania sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Clusiaceae</td>
<td>Rheedia brasiliensis (Mart.) Planch &amp; Triana</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Vismia cf. latifolia (Aubl.) Choisy</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Combretaceae*</td>
<td>Combretum lanceolatum Pohl ex Eichler</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Buchenavia cf. oxycarpa</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Convolvulaceae</td>
<td>Merremia macrocalyx (Ruiz &amp; Pav.) O'Donell</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Costaceae</td>
<td>Costus scaber</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Cyperaceae**</td>
<td>Scleria cf. melaleuca Reichen. Ex S. &amp; C.</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Cyperus* luzulae</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Cyperus sp. *</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td>Erythroxylaceae</td>
<td>Erythroxylon anguijugum Mart.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Euphorbiaceae*</td>
<td>Mabea fistulifera Mart.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Nealchornia sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Sapium glandulosum (L.) Morong</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Alchornea sp.*</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td>Family</td>
<td>Species</td>
<td>Habitat</td>
</tr>
<tr>
<td>----------------------------</td>
<td>-------------------------------------------------------------------------</td>
<td>-----------------------</td>
</tr>
<tr>
<td>Fabaceae (Caesalpinioideae)</td>
<td>Dalechampia sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Fabaceae (Caesalpinioideae)</td>
<td>Macrolobium acaciifolium (Benth.) Benth.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Fabaceae (Caesalpinioideae)</td>
<td>Senna obtusifolia (L.) H.S. Irwin &amp; Barneby</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td>Fabaceae (Mimosoideae)*</td>
<td>Zygia cauliflora (Willd.) Killip</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Mimosa* pigra L.</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td></td>
<td>Albizia subdimiata (Splitg.) Barneby &amp; J.W. Grimes</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Fabaceae (Papilionoideae)</td>
<td>Indigofera fruticosa J.N. Rose</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Loganiaceae</td>
<td>Strychnos cf. darianensis Seem.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Malpighiaceae</td>
<td>Byrsonima riparia W.R. Anderson</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Stigmaphyllon florosum C.E. Anderson</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Marsileaceae</td>
<td>Marsilea polycarpa Hook. &amp; Grev.</td>
<td>Aquatic</td>
</tr>
<tr>
<td>Melastomataceae*</td>
<td>Toccoca guianensis Aubl.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Mouriri sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Monimiaceae</td>
<td>Siparuna guianensis Aubl.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Myrtaceae*</td>
<td>Eugenia ochrophyloea Diels</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Psidium sp.</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td></td>
<td>Eugenia florida DC.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Onagraceae*</td>
<td>Ludwigia helminthorrhiza (Mart.) H. Hara</td>
<td>Semi-inundated</td>
</tr>
<tr>
<td>Piperaceae</td>
<td>Piper sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Poaceae**</td>
<td>Sporobolus sp.</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Panicum laxum Sw.*</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Guadua paniculata Munro</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Polygonaceae</td>
<td>Polygonum hispidum (Kunth)</td>
<td>Aquatic</td>
</tr>
<tr>
<td>Pontederiaceae*</td>
<td>Eichhornia* azurea (Sw.) Kunth</td>
<td>Aquatic</td>
</tr>
<tr>
<td>Pteridophyta</td>
<td>Adianthum sp.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td>Rubiaceae*</td>
<td>Duroia micrantha (Ladbrook) Zarucchi &amp; J.H. Kirkbr.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Genipa spruceana Steyerm.</td>
<td>Gallery forest</td>
</tr>
<tr>
<td></td>
<td>Uncaria* guianensis (Aubl.) J.F. Gmel</td>
<td>Gallery forest</td>
</tr>
</tbody>
</table>
The surface pollen assemblage from LG has an almost even contribution from arboreal (48% abundance) and herb types (43%). The most abundant arboreal species are *Cecropia* (13%), *Brosimum* (8%), Moraceae/Urticaceae (8%), *Alchornea* (4%), Arecaceae (palms) (4%), *Trema* (4%) and *Ampelocera* (2%). Poaceae (grasses) accounts for 35% of the surface pollen, while other common herb types include the weedy taxa Asteraceae (4%) and *Borreria* sp. (4%). Aquatic grasses were not found to be abundant around the modern lake shore; therefore it is assumed that the majority of grass pollen in the lake record derives from a terrestrial source. Of the aquatic types documented in the botanical survey, *Eichhornia* (<1%) is underrepresented relative to its abundance on the land/water surface, while Cyperaceae (23%) is overrepresented.

The surface pollen assemblage contrasts markedly with the phytolith assemblage, which is dominated by herb taxa (75%) over arboreal types (26%). Grasses (62%) are the most abundant herb, comprising mostly Panacoideae types (bilobates (29%), crosses (22%)), followed by rondels (10%), which are a general Poaceae indicator with a cosmopolitan distribution (Pearsall, 2010), and a very small proportion of Chloridoideae, represented by saddle-shaped phytoliths (1%). *Heliconia* sp. (5%) and bamboos (4%) are present in small abundance. Arboreal
types (26%) are represented primarily by globular granulates (20%) and palms (6%). The main aquatic is Cyperaceae (3%).

This discrepancy between the pollen and phytolith assemblages is attributable to differences in the transport of these two microfossils and subsequently their spatial representation of the landscape. Phytoliths, which are silica bodies derived from plant materials, are released by in-situ decomposition of plant organic matter (Piperno 1988; Piperno 2006). Although long distance transport of phytoliths is possible via rivers or advection from large fires, their transport is often through remobilisation by water across land surfaces (Birks 2001) and so, in most instances, they can be said to represent vegetation on a local scale around the site of deposition. In contrast, pollen has potential for longer distance transport, especially from anemophilous species such as members of the Moraceae family (Bush 1995; Bush and Rivera 1998; Gosling et al. 2005). The pollen record at LG therefore, represents extra local vegetation around the lake, while the phytoliths represent a more localized catchment area, including the shoreline vegetation.

We can confirm this by comparing the surface pollen and phytolith assemblages with the modern plant inventory (table 4.2). Species which occur in both the plant inventory and the surface pollen assemblage include members of the genera Cecropia, Alchornea, Pourouma and Uncaria. In pollen trap and lake core studies from other terra firme sites in eastern lowland Bolivia, Cecropia and Alchornea were shown to be common members of humid evergreen riparian forest (Burn et al. 2010). Brosimum is found in the surface pollen assemblage from LG, but was not identified in the plant inventory. We therefore determine that it and other anemophilous, terra firme species found in the pollen record, such as Pseudolmedia and Trema (Burn et al. 2010), represent an extra-local signal, which derives from the terra firme evergreen rainforest, outside of the riparian zone around the lake. The modern vegetation around LG includes a cleared area of farm land (~0.3 km²) on its eastern shore. This and other cleared patches visible immediately around the lake, are likely the source for the abundance of grass phytoliths found in the lake surface sediment, but also contribute to the grass pollen signal.
4.6.3. Zone 1 6000-2500BP

From 0-150 cm in the sediment core, pollen and phytolith preservation were good. Pollen and charcoal results are presented in figures 4.3 and 4.4 and phytolith results in figure 4.7. In Zone LG-1 (comprising 11 pollen and 5 phytolith samples), the pollen assemblage is dominated by Poaceae (40-77% abundance), indicating that the wider landscape around the lake was savannah during this period. Pollen assemblages with Poaceae proportions of >40% typically represent savannah (Gosling et al. 2009). The very high proportion of Poaceae pollen (≥50%) compared to arboreal types (18-38%) in almost the entirety of this zone, suggest that the palaeo savannah was one of the more open sub-types with low tree density, rather than a more densely forested form such as cerradão or campo carrado (Gosling et al. 2009; Jones et al. 2011). Of the arboreal taxa present, common types are Cecropia sp. (5-17%), Moraceae/Urticaceae (3-10%), Alchornea (0-6%) and Pseudolmedia sp. (0-3%). The herb taxon Asteraceae (2-10%) is present throughout the zone.

The phytolith record in this zone is dominated by arboreal globular granulate type phytoliths (32-75%), with low Poaceae levels (7-16%), low level of palms (4-21%) and abundant Asteraceae (5-41%), suggesting seasonally inundated semi-deciduous dry forest (Dickau et al. 2012). The small contribution of Poaceae phytoliths indicates that the high Poaceae pollen levels in this zone derive from the terra firme landscape, beyond the lake margins. In the pollen record the presence of Anadenanthera (≤2%), which has both semi-deciduous dry forest inhabiting species and seasonally flooded forest inhabiting species, appears to support this interpretation of seasonally-flooded forest around the lake margins.

The appearance and high abundance in the pollen record of the semi-aquatic genus Alternanthera sp. (1-8%) in this zone (presented outside the terrestrial sum) is also indicative of swampy conditions around the lake, possibly related to a low stand in the lake’s history, which exposed more of the shoreline for colonization by Alternanthera. Charcoal levels throughout this zone are low, suggesting that locally around the lake at least, burning frequency was low at this time.
4.6.4. Zone 2 2500-500BP

In zone LG-2 (14 pollen and 10 phytolith samples), there is a sharp increase in charcoal abundance at ~2500 BP, likely indicating an increase in both the frequency and intensity of burning. The diversion of this charcoal peak from low level burning seen in zone 1 and its contrast with patterns of decreasing natural burning regionally at this time, as seen in the regional-scale charcoal records at Lagunas Orícore (LO) (Carson et al. Chapter 3), Bella Vista (LBV) and Chaplin (LCH) (Mayle et al. 2000; Burbridge et al. 2004), suggest that it is anthropogenic in origin. The first maize pollen of the record is found at ~1850 BP, and maize is found throughout the rest of the zone. The high Poaceae levels (50-77%) and low total arboreal pollen (15-30%) indicate that the landscape around LG continued to be open savannah throughout this zone. However, there is evidence for floristic changes to include more evergreen rainforest species such as Brosimum (1-6%) and Pseudolmedia (1-4%) (Gosling et al. 2009; Burn et al. 2010). Between ~1600 and 800 BP, charcoal levels decline and in the pollen record there is a moderate reduction in Poaceae (50%) in favour of the weedy taxa Asteraceae (2-10%), Chenopodiaceae/Amaranthus (1-6%) and Borroria (0-2%).

At ~800 BP there is another sudden spike in charcoal, indicating a second intensive burning period between 800-500 BP, with peak Poaceae levels (78%) occurring at 680 BP. The disappearance of Alchornea and Celtis and reduction of Cecropia (1-3%) from ~ 800-600 BP, suggest opening of the gallery forest.

The phytolith record in zone LG-2 shows an expansion of grasses (35-75%) and decrease in woody dicots (18-64%), signalling the opening of the landscape around the lake margins. Bamboos (1-8%) appear in the phytolith record in this zone, and may reflect floristic transformation toward evergreen forest (Dickau et al. 2013), but may also be the result of human disturbance. Bamboos are both common understory components in disturbed forest and are also constituent flora of evergreen terra firme rainforest (Dickau et al. 2013). This zone also sees the first appearance of maize phytoliths alongside maize pollen in the record at ~1000 BP and throughout the rest of zone LG-2.
4.6.5. Zone 3 500 BP-present

In zone LG-3 (7 pollen and 5 phytolith samples) between 600-500 BP, the decline in charcoal, decrease in Poaceae pollen (34-44%) and increase in arboreal pollen types (35-58%), most notably *terra firme* taxa such as *Brosimum* (7-10%) and *Pseudolmedia* (0-6%), signal reduced frequency/intensity of burning and an expansion of evergreen forest into the *terra firme* areas of the site (Burn et al. 2010). Both the pollen and phytolith records also appear to show expansion of the gallery forest, with woody phytoliths (25-45%) increasing, and the reappearance of *Cecropia* (3-12%), *Celtis* (1-6%) and *Alchornea* (2-9%) in the pollen record. However, grasses remain the dominant component of the phytolith assemblage (48-62%), suggesting that there is still open ground around the lake margins. Maize is found in the pollen and phytolith records until close to modern times.
Figure 4.3 Pollen and charcoal from Laguna Granja plotted against calibrated years BP. Pollen of all taxa with >2% abundance are shown. Pollen is presented as percentage abundance of the terrestrial count of 300 grains, with the exception of Z. mays, which is presented as no. of grains. Charcoal is presented as particles per cm$^3$. Calibrated radiocarbon ages are displayed on the right (see table 4.1 and fig. 4.2)
Figure 4.4 Rare pollen types (1-2% abundance) from Laguna Granja plotted against calibrated years BP
Figure 4.5 Pollen concentration values from L. Granja. Dominant taxa and total overall pollen concentration are shown as grains per cm$^3$. 
Figure 4.6 Pollen influx values and sediment particle size from L. Granja. Dominant taxa and overall total pollen influx values are shown as grains per cm$^2$ per year. Particle size data is split into three size categories: clays (<2μm, <5μm) and silts (<30μm).
Figure 4.7 Phytoliths from Laguna Granja plotted against calibrated years before present. Expressed as percentage abundance of 200 phytolith count.
4.7. Discussion

4.7.1. Timing of occupation at LG

Archaeological excavations of the two ring ditches, GDP and BV-3, identified a single, thin occupation zone, which was dated between 750-550 BP. The construction/occupation of the ring ditch is clearly seen in the palaeo record as a period of intense burning and degradation of the gallery forest, confirming the tight dating constraints on the construction and occupation of the GDP and BV-3 earthworks. Our palaeoecological analyses however, have revealed a much earlier start to human occupation on this site, with anthropogenic burning evident from ~2500 BP. The palaeoecological record also shows that the dating of this cultural layer does not mark the end of occupation on the site. There is a probable decline in population and activity following ~500 BP, when burning declines and afforestation begins. Maize agriculture is evidently still practiced after this date, until close to present, however, this could have been the result of cultivation on the lake shore by a relatively small population.

The timing of this settlement decline or change in land use strategy at LG is intriguing, as it approximately coincides with the arrival of Europeans in the Americas in AD 1492. Historical records tell us that the Spanish did not begin to formally colonize the Iténez region until the 18th century (Altamirano 1891; Block 1994). However, the hypothesis of post-Contact Native American demographic collapse, proposes that Old World diseases could have been spread rapidly via extensive native trade routes (Dobyns 1963; Denevan 1976; Denevan 1992b), without the need for direct contact with Europeans. Taken on its own, the timing of settlement decline at LG is compelling evidence in support of a possible rapid propagation of Old World diseases through southern Amazonia following the Columbian Encounter (CE). However, other recently published records of pre-Columbian occupation at earthwork sites contemporary to LG, located in the central Llanos de Moxos (Whitney et al. 2013; Whitney et al. 2014), have dated site decline/land use change to shortly before the CE. Decline on these sites was evidently the result of internal factors, rather than a post-CE epidemic. Furthermore, occupation at other ring ditch sites in Pando State, Bolivia, and Acre and the Upper Xingu, Brazil have been dated to well past the contact period (Heckenberger 2003;
Saunaluoma 2010; Saunaluoma 2012), demonstrating that southern Amazonian societies in some places continued beyond this historical turning point.

Although a definite causal link between the CE of 1492 AD and site decline observed at LG cannot be established from our data, exchanges between Europeans and native Amerindians nevertheless likely contributed to changes in land use strategy on sites like Bella Vista in the centuries following contact through, for example, the introduction of metal tools (Denevan 2001). These new tools made slash-and-burn agriculture a possibility, by reducing the labour and time required to fell a tree, and as such may have fundamentally changed the way native Amazonians impacted the forest landscape.

4.7.2. Agricultural and land use strategy

In our pollen analyses, coarse fractions were scanned for common cultigen types, including squash (Curcurbita sp.), sweet potato (Ipomoea batatas), yuca (Manihot esculenta), and maize (Zea mays L.), all of which have large pollen grains and are readily isolated by sieving (Whitney et al. 2012). These species also have diagnostic phytoliths, as do arrowroot (Maranta arundinacea) and Léren (Calathea allouia). In both the pollen and phytolith records, the only cultigen identified was Z. mays. An analysis by Dickau et al. (2012) of phytoliths and starch grains from stone tool and pottery remains recovered from the GDP ring ditch, also found maize to be the dominant cultigen, and led the author to tentatively conclude that this was the primary crop grown on the site. Our data confirm that maize was the staple crop of the inhabitants around LG, not only during the GDP occupation, but from as early as 1850 BP. The high charcoal levels associated with the pre-Columbian occupation suggest that this agricultural strategy may have involved burning of the terra firme savannah around the site; however, other sources of charcoal are also likely, such as inputs from cremations and from everyday fire use from hearths. Macroscopic charcoal is deposited in a lake basin by various processes, meaning that peaks and troughs in the charcoal record may represent both changes in intensity of burning around a lake and in the proximity of the burning to the lake (Whitlock and Millspaugh 1996).

Maize phytoliths appear for the first time in the record alongside maize pollen at ~1000 BP. Again this discrepancy between the two proxies likely represents a
difference in catchment area between phytoliths and pollen, and indicates closer proximity of maize growth to the lake after 1000 BP. This conclusion is supported by the apparent clearance of gallery forest during the latter occupation phase of zone 2, suggesting that cleared land around the lake was used for agriculture, possibly associated with the construction and use of the nearby GDP ring ditch.

It is interesting that maize is not found before 1850 BP, despite evidence for first settlement around LG from 2500 BP. Two possible explanations for the absence of maize before 1850 BP are that 1) subsistence before this point did not include maize agriculture or 2) maize was grown on the site, but not in great abundance or close enough proximity to the lake to be detected in the fossil record. Although maize pollen may be transported further than phytoliths, maize pollen grains are nevertheless large and relatively poorly transported, and therefore reflect cultivation locally around the site of pollen deposition (Lane et al. 2010). Another lake core study by Carson, et al. (Chapter 5) from a forest island located <15km away from LG found maize pollen dated from 2100 BP, which is the earliest evidence for maize cultivation in this region. Subsistence at LG during first occupation at 2500 BP may therefore, have been based on a different, unidentified staple, or relied more heavily on none-agrarian resource gathering.

The fact that from 1850 BP the pre-Columbian inhabitants around LG grew maize as the staple crop is interesting. Today the staple crop grown as part of subsistence agriculture in this region is yuca, which is often identified as a hardier crop, more suitable for nutrient poor tropical soils (Edwards et al. 1976). From our palaeoecological record, it is evident that maize was being grown on the PCS around LG, and that the soils of the PCS were fertile enough to support maize agriculture. Evidence of maize agriculture has also been found at sites in the central and southern Beni Basin (Whitney et al. 2013; Whitney et al. 2014), which together with the data presented here, highlight the importance of maize as a cultigen across the Bolivian Amazon region during pre-Columbian times.

Comparing the lake fossil record from the surface sediments of LG with the record of vegetation cover over the last ~2500 years, gives an interesting insight into the scale and intensity of pre-Columbian land management on this site, relative to modern land use. Despite plentiful field evidence for modern disturbance of the
gallery forest around the lake and the presence of numerous cleared patches visible on Google Earth® satellite imagery (fig. 4.6), macroscopic charcoal levels in the surface sediments of LG are very low when compared to the much higher charcoal levels recorded during pre-Columbian times. From this comparison, we must conclude that Amerindian subsistence involved more intensive and extensive burning of the landscape than the slash-and-burn agriculture, which is practiced around the lake today. Burning was likely an important tool for maintaining an open landscape and would have been a self-reinforcing strategy, as a maintained grassland would have been more easily combustible than dense evergreen rainforest. The modern town of Bella Vista, which is less than one kilometre south of LG, covers a non-contiguously cleared area of ~1.5 km². Again, comparing surface pollen records to the palaeo record, reveals that the open grassland area maintained by pre-Columbian people was likely significantly more extensive than the area of the modern town and/or had a much lower density of trees compared to the patchily degraded landscape that exists today.

![Google Earth satellite image of Laguna Granja and surrounded landscape](image)

*Figure 4.8 Google Earth® satellite image of Laguna Granja and surrounded landscape, showing modern town and patchy degradation of forest.*

The fact that pre-Columbian people maintained an open landscape following regional rainforest expansion from ~2000 BP, suggests a land use strategy which
necessitated large areas of open land on the PCS for agriculture. Open ground may also have been maintained for the construction of ring ditches, which appear often to have existed as collections of interconnected features (Prümers 2012b). Inter-visibility between ring ditch sites may also have been desirable, and therefore required the maintenance of a cleared landscape.

4.7.3. Legacy of pre-Columbian land use

The implications of pre-Columbian land use for questions over the resilience of Amazonian rainforest to human impacts and the pristineness of Amazonian ecosystems, is a key controversy (Heckenberger et al. 2007; Peres et al. 2010; Barlow et al. 2012). The LG record gives some insights into the nature, scale and longevity of such impacts around an earthwork site. We found little evidence in either the pollen or phytolith records that the pre-Columbian inhabitants at LG altered the floristic composition of forest around the site to favour economically useful species. Palms, for example, provide many useful resources, including the edible heart of palm and building materials (Posey and Balée 1989). However, while palm pollen abundance does increase slightly in zones LG-2 and LG-3, this may have been the result of natural factors, such as a response to greater fire disturbance, rather than cultivation. This of course does not rule out the possibility that forest resources were managed at Bella Vista. Increases in economic species may not be seen in the palaeo record, because of insufficient taxonomic resolution in our identifications. Alternatively, the pollination syndrome of some economic taxa may mean that they are underrepresented in the pollen record (Bush and Rivera 2001) and cannot be captured in a standard 300 pollen grain count.

The wider site around LG was maintained as an open anthropogenic landscape until 500 BP, after which afforestation took place. We can see, therefore, that much of the forest now covering the modern village was established in the last 500 years. On a regional scale also, beyond the extent of the ring ditch site, the evergreen forest was established relatively recently, expanding from ~2000 BP in response to changing climatic conditions (Carson et al. Chapter 3). The pollen record suggests that there was regeneration of the gallery forest after 500 BP. However, the phytolith record suggests that this area continued to be exploited following afforestation of the terra firme after 500 BP, and into the modern era. The forest
now covering these sites is, therefore, neither ancient nor pristine. However, the model of humans maintaining an open landscape around their settlements rather than deforesting land, as suggested by the palaeoecological data from LG, does not support suggestions that pre-Columbian earthwork builders in Amazonia contributed strongly to biomass losses and atmospheric carbon increases during the late-Holocene (Chave et al. 2008; Dull et al. 2010; Nevle et al. 2011).
4.8. Conclusion

Our approach of integrating pollen and phytolith analysis of lake sediment cores with terrestrial analyses has revealed important aspects of the chronology and nature of pre-Columbian occupation on the Bella Vista site. The site is shown to have been occupied since ~2500 BP, long before the construction of the two previously dated ring ditches. This presents the possibility that other ring ditches that have been mapped around the site, and in the wider Iténez province, were constructed before the GDP and BV-3 ditches. However, only further excavation and dating of archaeological contexts can confirm this. We demonstrate that even in a small lake such as LG, pollen and phytoliths represent palaeovegetation over different, local and extra local, spatial scales. This is highly useful in discerning spatial patterns of land use on an archaeological site. Anthropogenic burning and suppression of trees maintained an open area on the PCS around LG, greater than that exposed by modern clearance, which was used for maize agriculture and earthwork construction. We confirmed that maize was the staple crop grown on site, although the spatial pattern and intensity of agriculture changed over time, with greater exploitation of the gallery forest occurring from ~1000 BP. Rather than experiencing site abandonment after European contact in AD 1492, the Bella Vista site continued to be occupied through to near the present day. There does, however, appear to have been a decline in human activity after ~500 BP, possibly related to the CE, which allowed expansion of evergreen forest into the terra firme areas, and the establishment of the forest that exists around the site today.
4.9. Acknowledgments:

This research was supported by a Leverhulme Trust research grant (F/00158/Ch) awarded to FM and JI, and a NERC Doctoral Training Scheme grant (NE/152830X/1) and funds from the University of Edinburgh’s Principal’s Career Development Scholarship, awarded to JC. A NERC radiocarbon facility date was granted to FM (1623.0312). Fieldwork support was provided by the ‘Noel Kempff Mercado’ Natural History Museum, Santa Cruz, Bolivia. We thank Douglas Bruckner of the ‘Programa de Conservación de la Parába Barba Azul’, Trinidad, Beni Department, Bolivia and the rangers from the ‘Reserva Iténez’ WWF station in the town of Bella Vista, Beni Department, Bolivia, for logistical support in the field. We also thank José Manuel Barrios Fernández for allowing us access to core the L. Granja site.
Chapter 5. Palaeoecological evidence for pre-Columbian land use on a “chocolate island” in NE Bolivia.

Authors: Carson, J.F., Mayle, F.E., Whitney, B.S. and Soto, D.S.

5.1. Preface

The following chapter has been written in academic publication style for submission to the Journal of Archaeological Science and conforms to the length requirements of that publication. The author contributions to this paper were as follows: JC led the writing of the paper. Pollen and charcoal analysis of Laguna La Luna and La Luna Bog was carried out by JC, although BW analysed the surface sample of L. La Luna for an earlier study. The research design was devised by JC, FM, and BW. Analysis and interpretation of the data was done by JC, with input from FM and BW. JDS conducted the botanical survey. All authors contributed to the final writing of the manuscript.
5.2. Abstract

We present a palaeoecological record of pre-Columbian occupation on La Luna Island, a ~7 km² “forest island” ring ditch site, within the seasonally-inundated forest-savannah mosaic of the Llanos de Moxos, in the Beni department, Bolivian Amazonia. A <5700 BP sediment core record from a lake located adjacent to the forest island, was analysed for fossil pollen and macroscopic charcoal, to reconstruct the history of vegetation, burning and land use on the site. Evidence for anthropogenic burning and the pollen of Zea mays L. indicate that human occupation began ~2090 BP, at a time when the island was not forested due to drier-than-present climatic conditions. Despite a regional, climatically driven expansion of terra firme forest ~2000 BP, the site was maintained as an open grassland, through anthropogenic burning and suppression of tree growth. This continued until ~1240 BP, when land use intensity declined on the site and afforestation of the island took place. The construction of a large pre-Columbian ring ditch did not involve deforestation of the island. We infer from this, that earthwork construction at hundreds of sites across the forest island landscape, did not involve large-scale deforestation, and may have been carried out by smaller populations than previously estimated, and with significantly lower impacts on biogeochemical cycling than previously assumed. Extended pollen counts from the lake sediment surface and from a terrestrial bog located in the centre of the island revealed that the chocolate tree Theobroma cacao, which is abundant on the forest islands today, is silent/rare in the pollen record, and not useful as a marker of pre-Columbian land use. Other economic plant taxa which are common on the island today, including palms and Inga, were not found to increase in abundance in association with pre-Columbian land use. We go on to suggest, however, that the presence of humans on the site since before and throughout the expansion of forest, implies an anthropogenic influence on the forest’s development, and should be taken into account when considering the conservation of forest in this protected region.
5.3. Introduction

The discovery of spatially extensive and structurally diverse earthworks across the Bolivian Amazon, has provided some of the strongest evidence for large and socially complex pre-Columbian (pre-AD1492) societies in Amazonia (Denevan 1966; Denevan 1976; Walker 2009; Lombardo and Prümers 2010; Saunaluoma 2010). These earthworks have stimulated debate, not only over the size of pre-Columbian populations in the Amazon basin, but also the scale of their influence on the natural environment, and its legacy in the modern ecosystem. It has been suggested that pre-Columbian deforestation and biomass burning occurred on a scale large enough to influence Holocene climate (Dull et al. 2010; Nevle et al. 2011). It has also been posited by historical ecologists that pre-Columbian management of plant resources fundamentally altered the structure and species composition of Amazonian forests in previously inhabited areas (Posey and Balée 1989; Balée 1994; Erickson and Balée 2006), and that the legacy of this pervasive human impact can be seen in the abundance of economically useful plant taxa found on pre-Columbian sites today (Chave et al. 2008). However, a paucity of palaeoecological data means that we often have no palaeoenvironmental context in which to place these sites, and drawing inferences regarding past environmental impact becomes difficult. Recent studies have begun to redress this problem, and have demonstrated the value of palaeoecological data in this debate (Whitney et al. 2013; Whitney et al. 2014), but significant gaps still remain.

The forest island landscape of the Iténez Province in north-east Bolivia, constitutes a unique geoarchaeological sub-region of the Bolivian Amazon (Lombardo et al. 2011). The region is characterised by hundreds of natural terra firme “forest islands”, ranging from 100 m in diameter to tens of square kilometres in area, which punctuate the flat, low-lying, seasonally inundated savannahs of the Beni Basin (fig. 5.2). Although some have been recently partially cleared, the majority of these islands are covered by intact closed canopy rainforest, which are taxonomically linked to the vast Medeira-Tapajós rainforest ecoregion (Olson et al. 2010), on the uplands of the main Pre-Cambrian Shield (PCS), and managed within the ‘Iténez Protected Area’. Archaeological surveys have revealed however, that this is also a complex pre-Columbian built landscape, with artificial earthworks found
across both the forest islands and the savannahs (Erickson 2000). This has raised questions over the assumed “pristine” nature of forest in this region, and the impact of past human management on the development of vegetation communities. The presence of ring ditch earthworks beneath what today is closed canopy rainforest, has been taken as evidence for extensive pre-Columbian deforestation, by substantial populations, across the forest island landscape (Erickson 2010). However, a palaeoecological study by Carson et al. (Chapter 4) at the Bella Vista earthwork site in northern Iténez, demonstrated that its inhabitants did not deforest, but built in an originally open savannah landscape, which existed during drier-than-present climatic conditions. After ~2000 BP, when increasing precipitation caused rainforest to expand south into the Iténez region, the inhabitants at Bella Vista suppressed forest expansion around the site and maintained locally an open environment. Without the need for labour intensive deforestation using stone tools, it was argued that ring ditch construction at Bella Vista could have been achieve with less labour and a smaller population than would be necessary in a forested environment. Whether this scenario applies to other ring ditch earthwork sites in Iténez remains to be determined.

The forest islands in Iténez also are held as a prime example of extant pre-Columbian anthropogenic forest, with notably high abundances of economically useful taxa, such as palms and the chocolate tree (*Theobroma cacao*). Such is the concentration of *Theobroma* on these islands that they are commonly referred to as “chocolate islands”. The high density of *Theobroma* has been attributed either to plantations established by Jesuit priests in the 18th century or to earlier planting by pre-Columbian people (Erickson 2010). However, whether high abundances of economic taxa constitute a genuine signal of anthropogenic management is disputed (Barlow et al. 2012).

While some in depth archaeological survey and excavation work has been completed at the Bella Vista site in northern Iténez (Prumers et al. 2006; Prümers 2012a; Dickau et al. 2012), and surveys conducted around the town of Baures in southern Iténez (Erickson 2010), there is no published excavation work from the forest islands themselves. A great deal remains unknown about the subsistence strategies/resource management that these groups employed, the chronology of settlement in the region, and the potential legacy of human impact, which could have
implications for approaches to conservation in the region (Clement and Junqueira 2010).

5.3.1. Aims

Here, we investigate the land use history of the forest island region, by reconstructing the palaeoenvironment of a ring ditch site, located on a forest island in northern Iténez. As in other studies (Carson et al. Chapter 4; Whitney et al. 2013), by selecting to core a small lake in close proximity to archaeological remains, we are able to reconstruct land use history and anthropogenic impacts on a local scale (Mayle and Iriarte 2012). The aim of the study was to test whether the scenario of human-environment interaction recorded at Bella Vista by Carson et al. (Chapter 3; Chapter 4), applies to the forest island site. We also employ extended pollen counts from both lake core sediments and a terrestrial bog located on top of the forest island, to search for the pollen of Theobroma and other known economic taxa, such as palms, as a potential marker of historical horticultural management. By comparing the occurrence of economically useful taxa throughout the palaeoecological record, we aimed to determine whether there are appreciable increases in the abundance of these species associated with human occupation, and if their wealth in the modern environment is in fact a legacy of pre-Columbian land management.

5.4. Study area

Our study was conducted in the province of Iténez, situated in the north-east of the Department of the Beni, Bolivia (fig. 5.1). The Beni encompasses a large, low lying, sedimentary basin filled with Quaternary sediments. The lack of topography and impermeability of the clay sediments means that, during the annual wet season from November to March, the majority of the landscape is flooded, forming the Llanos de Moxos, a vast, seasonally-inundated savannah, interspersed with occasional forest on elevated areas of land (Navarro and Maldonado 2005). While in the western parts of the basin these high points are formed by abandoned river levees, termite mounds and, in some cases, human construction, in Iténez, exposed outcrops of the PCS have created terra firme ‘forest islands’ within the savannah.
Figure 5.1 Map of study region and sample sites. (A) Position of study site within Bolivia and the Llanos de Moxos. (B) Study region highlighting position of the La Luna sites, Bella Vista and Laguna Oricore. (C) Google Earth® satellite image of La Luna forest island, La Luna Bog and La Luna Lake. The approximate position of the boundary ring ditch around the island is shown by red dashed line.

The pre-Columbian earthworks in northern Iténez consist of ring ditches on the terra firme forest islands, and linear causeways in the wetland savannah. Ridged fields have also been discovered in the savannas in the north of the province, and in the south there are zig-zagged earthworks, which may have functioned as fish weirs (Erickson 2000). The ring ditch earthworks found on the forest islands range from true ring ditches, which are circular and hundreds of metres in diameter, to larger, more irregular shaped boundary ditches, which can be kilometres in length. These structures often have deep trenches, typically ranging from 2-4 m deep and 2-5 m wide, but can be up to 10 m deep (Erickson 2010). Surveys conducted in the south of Iténez documented ring ditches on almost every island of >2 km² (Erickson 2010) and in the north, earthworks are easily encountered on field survey, although their full extent is unknown (Prumers et al. 2006; Prümers 2012b). In the wetland
savannah, linear causeway earthworks have been constructed to run between the forest islands (fig. 5.2). These causeways may have served to link-up the forest islands during the flood season, or may have been used to control the movement of water in the wetlands (Lombardo and Prümers 2010).

The lake cored for this study was Laguna La Luna (LL), a 0.48 km² lake, located within the savannah (13°21′20″ S, 63°35′2″W) and adjacent to La Luna Island (LI), a forest island of approximately 7.4 km². The island is almost completely covered by closed canopy rainforest, except for a 480 m diameter bog (La Luna Bog (LB)) near the centre (see fig. 5.1C). A brief survey of the island revealed that its entire circumference is bounded by a ditch, approximately 3 m deep and 4 m wide, and which lies 10 m in from the edge of the island (see fig. 5.4). A pair of causeway structures runs between LI and the adjacent Esperanza Island to the west, and another pair of causeways runs between LI and a second island to the north. The La Luna site is also located 15 km south-east of the previously studied Laguna Granja site, at the town of Bella Vista, and 5 km west of Laguna Orícore, which provided a reconstruction of forest-savannah ecotonal shifts over the last 6000 years, in the northern Iténez region (Carson et al. Chapter 3; Chapter4).
Figure 5.2 Google Earth® satellite image showing forest island landscape of Iténez province.

Figure 5.3 Google Earth® satellite image showing causeways running between forest islands in southern Iténez, ~40 km south-east of the town of Baures (13°35'03"S, 63°34'38"W).
Figure 5.4 Photograph of boundary ditch on La Luna Island. Dashed line represents modern land surface at the top of the ditch. Taken July 2011
5.5. Materials and Methods

Fieldwork was carried out in June-July 2011. The lake sediment core was taken from a stable floating platform in the central, deepest part of LL, using a modified Livingston piston corer (Colinvaux et al. 1999). The surface lake sediment core was taken using a 5-cm diameter Perspex® tube to capture the uppermost unconsolidated sediments. A surface sediment sample was also taken from the edge of LB (sample location 13°21’24” S, 63°35’33” W), using a sterile plastic sample tube. Softer sediments from the surface lake core were sub-sampled in the field at 0.5-cm intervals and stored in watertight plastic tubes. Firmer surface core sediments were extruded in the field as intact cores and shipped back to the UK in robust, watertight packaging. Livingstone core sections were transported in their aluminium core casings and extruded in the lab in the UK. In the lab, the sediment cores were split lengthways into equal core halves, one of which was used for destructive sampling, while the other was retained as an archive core. All samples were kept in cold storage at 4°C.

5.5.1. Modern vegetation survey

During fieldwork, a rapid assessment botanical survey was made of the vegetation around the lake and of nearby LI, by JDS. Presence of species encountered around the site was noted and voucher specimens were collected for the herbarium of the Museum of Natural History ‘Noel Kempff Mercado’ in Santa Cruz, Bolivia (table 5.2).

5.5.2. Pollen analysis

After an initial coarse resolution sub-sampling of the entire core to determine pollen preservation quality, the core was sub-sampled for pollen analysis at intervals of 2.5 to 3-cm, from 0 to 58 cm depth, below which point pollen was not preserved. A 1 cm³ sub-sample of sediment from each horizon was prepared using the standard pollen preparation protocol (Faegri and Iversen 1989), with the addition of a sieving stage designed for optimal recovery of large cultigen pollen (Whitney et al. 2012). A known concentration of Lycopodium marker spores was added to each sample for calculation of pollen concentration values, and to confirm that observed changes in
pollen percentage abundance were not the result of changes within a closed sum (fig. 5.7). The pollen in the fine fractions (material <53 μm) of most samples was counted to the standard 300 grains. However, to search for Theobroma and other potentially rare but identifiable pollen types of economically useful species, the surface sample (the topmost 0.5 cm of sediment) from the LL core, and the LB sample, were counted to a sum of 1000 terrestrial pollen grains.

The coarse fractions from throughout the LL core were scanned for large cultigen pollen grains up to a standardised equivalent count of 2000 Lycopodium spores, representing an average of ca. 0.1 cm$^3$ of the original 1 cm$^3$ of sediment processed. Fossil pollen was identified with reference to the collection of over 1000 tropical pollen specimens housed at the University of Edinburgh and University of Reading, and from atlases of Neotropical pollen (Roubik and Moreno 1991; Colinvaux et al. 1999; Bush and Weng 2007). Maize grains were distinguished from other wild grasses according to the morphological criteria described in Holst (et al. 2007). Where possible, members of the Moraceae family were identified to genus using morphological descriptions from Burn & Mayle (2008). Where genus level identification was not possible, grains were assigned to the Moraceae/Urticaceae undifferentiated category.

5.5.3. Charcoal Analysis

Macroscopic charcoal analysis was carried out at 0.5-cm intervals throughout the core, from 0 – 58 cm depth. Sub-samples of 1 cm$^3$ were taken from each horizon and heated in 10% sodium pyrophosphate to disaggregate clay sediments. The samples were then sieved at 250 μm and 125 μm and charcoal particles counted in water, under x40 magnification. All graphs were drawn using the program C2 (Juggins 2007).
5.6. Results and Interpretation

5.6.1. Core stratigraphy and chronology

A 167-cm core was recovered from LL, including a 51-cm surface core. The sediments consisted of fine clay throughout, but changes in colour and the degree of compaction were observed. From the base of the core up to 47 cm depth, sediments were light grey, very stiff clay. At 47 cm there was a sharp transition to dark grey, medium-stiff clays up to ~ 25 cm depth. This sharp transition was used to correlate the surface and Livingstone lithologies. From 0 – 25 cm depth, the sediments were comprised of very soft, medium-grey clay.

A total of 5 AMS \textsuperscript{14}C dates were obtained from the LL core (table 5.1). Two samples, from the softer sediments in the top 25 cm of the surface core, produced near modern radiocarbon ages. These ages are anomalously young given the depth of the samples below the surface, suggesting that the very soft sediments in the top 25cm of the core have been affected by bioturbation or mixing. It was not possible with this chronology to produce an age-depth model for the core, but three congruous dates, not contaminated by modern carbon, were obtained for the base of Zone 1 (5760 BP) and the base and middle of Zone 2 (1240 BP; 2090 BP). The sharp stratigraphic boundary at 47 cm depth suggests that there may be a sedimentary hiatus between these two zones.
Table 5.1 AMS $^{14}$C results from Laguna La Luna. Calibrated ages are presented as weighted mean of the age distributions (Telford et al. 2004b). Samples Lu 14 and LU 23 given as parts modern carbon (pMC) rather than a conventional radiocarbon age.

<table>
<thead>
<tr>
<th>Sample identifier</th>
<th>Publication code</th>
<th>Depth below sediment-water interface (cm)</th>
<th>Conventional $^{14}$C age (yr BP±1σ)</th>
<th>*pMC</th>
<th>Calibrated age range (cal yr BP) ± 2σ</th>
<th>Area under probability curve</th>
<th>Weighted mean calibration (cal yr BP)</th>
<th>$\delta^{13}$CVPDB(‰)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lu 14</td>
<td>Beta-355052</td>
<td>14-15</td>
<td>*118.2 +/- 0.3</td>
<td></td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-27.1</td>
</tr>
<tr>
<td>Lu 23</td>
<td>Beta-347193</td>
<td>23-24</td>
<td>90 +/- 30</td>
<td></td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-26.1</td>
</tr>
<tr>
<td>Lu 33</td>
<td>Beta-364772</td>
<td>33-34</td>
<td>1310 +/- 30</td>
<td></td>
<td>1294-1223 1213-1181</td>
<td>0.713 0.287</td>
<td>1240</td>
<td>-22.7</td>
</tr>
<tr>
<td>Lu 45</td>
<td>Beta-355053</td>
<td>45-45.5</td>
<td>2120 +/- 30</td>
<td></td>
<td>2322-2345 2050-2206</td>
<td>0.044 0.956</td>
<td>2090</td>
<td>-24.0</td>
</tr>
<tr>
<td>Lu 56</td>
<td>Beta-339230</td>
<td>56-57</td>
<td>5010 +/- 30</td>
<td></td>
<td>5857-5942 5706-5818</td>
<td>0.358 0.642</td>
<td>5760</td>
<td>-26.3</td>
</tr>
</tbody>
</table>
5.6.2. Modern vegetation survey and surface pollen

Pollen preservation was found to be good from 0 – 58 cm depth in the lake core. Below this depth pollen were absent. The LB surface sample was also found to contain abundant identifiable pollen. Here, we discuss first the surface pollen assemblages from LL and LB, and compare to the results of the rapid botanical survey, before going on to discuss the palaeoecological record.

Table 5.2 Results of the rapid assessment botanical survey. An * indicates that the Family/Genus was identified in the Laguna La Luna surface pollen assemblage. This symbol ‡ indicates that the Family/Genus was identified in the La Luna Bog pollen assemblage.

<table>
<thead>
<tr>
<th>Family</th>
<th>Genus species</th>
<th>Occurrence in modern environment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alismataceae</td>
<td>*Echinodorus bolivianus (Rusby) Holm-Niels</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td></td>
<td>Limnocharis laforestii Griseb.</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Annonaceae</td>
<td>Guatteria sp.</td>
<td>Forest island</td>
</tr>
<tr>
<td></td>
<td>Unonopsis sp.</td>
<td>Forest island</td>
</tr>
<tr>
<td>Apocynaceae</td>
<td>Tabernaemontana siphilitica Leeuwenb.</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Araliaceae</td>
<td>Hydrocotyle bonariensis Lam.</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Bignoniaceae</td>
<td>Godmania sp.</td>
<td>Forest island</td>
</tr>
<tr>
<td></td>
<td>*Tabebuia elliptica (A. DC.) Sandwith</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Bixaceae</td>
<td>Cochlospermum sp.</td>
<td>Forest island</td>
</tr>
<tr>
<td>Caryophyllaceae</td>
<td>Indeterminate</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>‡Cyperaceae</td>
<td>Cyperus haspan L.</td>
<td>SI-savannah</td>
</tr>
<tr>
<td></td>
<td>Echinodorus tenellus (Mart.) Buchenau</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td></td>
<td>Eleocharis elegans (Kunth) Roem. &amp; Schult.</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Eleocharis minima Kunth</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Eleocharis mutata (L.) Roem. &amp; Schult.</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Eleocharis sp.</td>
<td>Aquatic</td>
</tr>
<tr>
<td></td>
<td>Rhynchospora sp.</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td></td>
<td>Scleria obtusa Core</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Euphorbiaceae</td>
<td>Caperonia cf. castaneifolia (L.) A. St.-Hill</td>
<td>SI-Savannah</td>
</tr>
<tr>
<td></td>
<td>Caperonia sp.</td>
<td>Wetland</td>
</tr>
<tr>
<td></td>
<td>*Hura crepitans L.</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Fabaceae</td>
<td>Aeschynomene cf. denticulata Rudd</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td></td>
<td>Aeschynomene pratensis Small</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td></td>
<td>Aeschynomene sp.</td>
<td>SI-Savannah</td>
</tr>
<tr>
<td></td>
<td>Andira inermis (W. Wright) Kunth ex DC.</td>
<td>Forest island</td>
</tr>
<tr>
<td></td>
<td>*Inga sp.1</td>
<td>Forest island</td>
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<td>Family</td>
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<tr>
<td>Lamiaceae</td>
<td><em>Hyptis lantanifolia</em> Poit.</td>
<td>SI-savannah</td>
</tr>
<tr>
<td>Lythraceae</td>
<td><em>Physocalymma scaberrimum</em> Pohl</td>
<td>Forest island</td>
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<tr>
<td>Malvaceae</td>
<td><em>Apeiba tibourbou</em> Aubl.</td>
<td>Forest island</td>
</tr>
<tr>
<td>Lecythidaceae</td>
<td><em>Helicteres</em> sp.</td>
<td>Semi-aquatic</td>
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<tr>
<td>Luehea grandiflora Mart.</td>
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<td>Forest island</td>
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<tr>
<td>Melastomataceae</td>
<td><em>Melochia hirsuta</em> Cav.</td>
<td>SI-savannah</td>
</tr>
<tr>
<td>Ochnaceae</td>
<td><em>Ochroma pyramidale</em> (Cav. ex Lam.) Urb.</td>
<td>Forest island</td>
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<tr>
<td>Pavonia sp.</td>
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<tr>
<td>Lamiaceae</td>
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<td>Lecythidaceae</td>
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<td>Theobroma cacao</td>
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<td>Forest island</td>
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<td>Thalictroideae</td>
<td><em>Thalictroideae crenulata</em> L.</td>
<td>Si-savannah</td>
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<tr>
<td>Meliaceae</td>
<td><em>Mouriri apiranga</em> Spruce ex Triana</td>
<td>Forest island</td>
</tr>
<tr>
<td>Moraceae</td>
<td><em>Pseudolmedia laevis</em> (Ruiz &amp; Pav.) J.F. Macber.</td>
<td>Forest island</td>
</tr>
<tr>
<td>Myrtaceae</td>
<td><em>Myrciaria</em> cf. <em>delicatula</em> (DC.) O. Berg</td>
<td>Forest island</td>
</tr>
<tr>
<td>Onagraceae</td>
<td><em>Ludwigia leptocarpa</em> (Nutt.) H. Hará</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Polygonaceae</td>
<td><em>Polygonum acuminata</em> Kunth</td>
<td>SI-savannah</td>
</tr>
<tr>
<td>Rhamnaceae</td>
<td><em>Rhamnium elaeocarpum</em> Reissek</td>
<td>Forest island</td>
</tr>
<tr>
<td>Rubiaceae</td>
<td><em>Borreria scabiosoides</em> var. <em>anderssonii</em> (Standl.) Steyerm.</td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Diodia kunzei K. Schum</td>
<td></td>
<td>Semi-aquatic</td>
</tr>
<tr>
<td>Rutaceae</td>
<td><em>Zanthoxylum jagara</em> var. <em>culantrillo</em> Reynel</td>
<td>Forest island</td>
</tr>
<tr>
<td>Salicaceae</td>
<td><em>Casearia</em> cf. <em>javitensis</em> Kunth</td>
<td>Forest island</td>
</tr>
<tr>
<td>Sapindaceae</td>
<td><em>Matauba</em> sp.</td>
<td>Forest island</td>
</tr>
<tr>
<td>Sapotaceae</td>
<td><em>Chrysophyllum gonocarpum</em> (Mart. &amp; Eichler ex Miq.) Engl.</td>
<td>Forest island</td>
</tr>
<tr>
<td></td>
<td><em>Chrysophyllum venezuelanense</em> (Pierre) T.D. Penn.</td>
<td>Forest island</td>
</tr>
</tbody>
</table>

The results of the botanical survey are presented in table 5.2. The survey identified 55 plant species in 26 families, across the four habitat types which occur around the lake. These included *terra firme* forest island, a semi-aquatic zone in the margin between the forest island and the lake, the seasonally-inundated savannah, and aquatic habitats on the lake margins. The most common species encountered was the shrub *Melochia hirsute*, which grew mostly on the small circular mounds found across the savannah landscape. In the transitional, semi-inundated zone around the lake edges, the species *Tabebuia elliptica, Hura crepitans*, and *Tabernaemontana* sp. grew as shrubs or small trees, forming mats of vegetation around the lake edge. The
aquatic grasses identified on the lake margins and semi-aquatic grasses in the savannah were exclusively from the genus Cyperaceae. However, this was a rapid assessment survey covering a limited area, therefore it is very likely that Poaceae occurs in both of these habitats within the landscape, but was not encountered on the survey. On the terra firme forest island *Theobroma cacao* and leguminous tree *Inga spp.* were notably frequent economic species.

The surface pollen assemblage from the LL core (fig. 5.6) is dominated by arboreal types (48%). The most abundant arboreal pollen types included *Pseudolmedia* (9%), Moraceae/Urticaceae undif. (8%), *Brosimum* (6%), *Cecropia* (5%), *Ampelocera* (5%), *Celtis* (5%), *Helicostylis* (3%), *Alchornea* (3%), *Gallesia* (2%), and Arecaceae (2%). *Pseudolmedia, Brosimum, Helicostylis*, and other members of the Moraceae family, along with *Ampelocera* and *Celtis*, are common components of terra firme rainforest (Gosling et al. 2005; Gosling et al. 2009; Burn et al. 2010), which suggests that the forest island constitutes a large part of the pollen source area of the lake. *Gallesia* can grow in the savannah or the riparian zone (Smith et al 2004). The most abundant herb type identified was Poaceae (31%) followed by Cyperaceae (7%). A number of taxa which were identified in the botanical survey, such as *Borroria, Hyptis, Hura, Tabebuia*, and *Zanthoxylum*, were also found in the lake surface pollen assemblage, but only as trace elements (≤1%).

The pollen assemblage from LB surface (fig. 5.5) is also dominated by arboreal types, with well represented species including *Maclura* (11%), *Cecropia* (9%), Moraceae/Urticaceae undif. (9%) *Ficus* (4%), *Alchornea* (8%), Arecaceae (5%), and *Schinopsis* (2%). Malpighiaceae (7%), whose growing habit can be trees, shrubs or herbs, was also an abundant type. This family is generally interpreted as a savanna indicator (Smith et al 2004; Jones et al. 2011) however, its presence in the LB sample, indicates that it is growing on the forest island or possibly on the fringes of the bog. Common herb types included Poaceae (17%) and Cyperaceae (4%). The genus *Roupala*, a member of the Proteaceae, which occurs as trees or woody shrubs (Smith et al. 2004), was found to be very abundant in the assemblage (>20%) when included in the total. The hyper-abundance of this genus suggests that there was an individual growing immediately over the sample collection point. For this reason
Roupala is presented outside of the main pollen sum. Theobroma was not identified in either the lake or bog pollen assemblages.
Figure 5.5 Pollen from La Luna Bog. Presented as percentage abundance. Rare taxa are represented by a + symbol. Roupala is presented outside of the terrestrial sum.
Figure 5.6 Pollen, charcoal and core stratigraphy from Laguna La Luna. Pollen presented as percentage abundance and charcoal particles/cm$^2$. Z. mays is presented as no. of grains/cm$^2$. Rare plant taxa (≤1%) which were also identified in the modern botanical survey, are included and represented by + symbol. Results are plotted against depth (cm), with radiocarbon dates plotted on the right (table 5.1).
Figure 5.7 La Luna pollen concentration values. Red box highlights forest rise.
5.6.3. Zone 1 (47-58 cm, <5,670 BP)

The lowest unit of the lake core record, which comprises 5 pollen samples and has a maximum age at its base of 5760 BP, is dominated by Poaceae pollen (62-75%) with some Cyperaceae (4-10%) and very low abundance of arboreal types (≤20%). The most common arboreal types are Moraceae/Urticaceae undif. (3-6%), Cecropia (0-6%), and Arecaceae (2-5%). Macroscopic charcoal levels in this zone are high in comparison to the modern charcoal levels, indicating that burning was more frequent and/or intense in this period, compared to present.

5.6.4. Zone 2 (25-47 cm, <2090 BP)

The central unit of the lake core record, from which 8 pollen samples were analysed, has a maximum age of 2090 BP. The lower portion of the zone from 47-35 cm depth, dated to before 1240 BP, is dominated by Poaceae (65-70%) with relatively low arboreal abundance (18-23%). However, from 1240 BP, between 35-25 cm, there is a steady decline in the percentage of Poaceae (56% to 46%) and an increase in arboreal abundance (30 to 38%). This arboreal increase is driven mostly by evergreen taxa such as Brosimum (0-6%), Pseudolmedia (1-5%), Cecropia (1-4%) and Alchornea (1-5%), which appears for the first time in the record in this zone. This zone also sees the first appearance of maize (Zea mays L.) in the record at 2090 BP, which is present throughout Zone 2. Charcoal abundance in the lower half of the zone is high, with a notable spike in the >250 μm fraction at 46 cm depth. In the upper half of the zone, from ~37 cm depth, charcoal in the >125 μm fraction begins to decline toward modern levels.

5.6.5. Zone 3 (25-0 cm)

The upper unit of the surface core is made up of 11 pollen samples. Both radiocarbon dates from this unit are modern. Given that this may be the product of sediment overturning, it is not possible to discuss changes within the zone, but we can make comparisons between this zone and Zone 2. In Zone 1, the proportion of arboreal taxa to Poaceae reaches levels comparable to the modern surface sediment assemblage (~30%). The arboreal total (40-50%) is dominated by members of the Moraceae/Urticaceae (27-40%), Ampelocera (2-7%), Celtis (2-6%), Alchornea (2-
and some Arecaceae (0-6%). Maize pollen is also present, with the last grain identified at 15 cm depth. The tree *Gallesia* is found for the first time in notable abundance (≥2%) in the record in this zone. Charcoal levels are consistent with modern abundance throughout the zone however, we cannot say whether this reflects genuine consistency of low frequency burning across the time period represented by Zone 1, or whether there has been an averaging effect across the zone due to mixing of sediments.
5.7. Discussion

5.7.1. Palaeoenvironmental change, settlement history and land-use

The earliest part of the LL record indicates that LI was not forested from 5670 BP, but was covered mostly by grasses, with low density tree cover. This reflects the wider terrestrial savannah environment that existed in northern Iténez during the mid-Holocene, as indicated by a regional-scale palaeoecological record from Laguna Orícore (Carson et al. Chapter 3; Chapter 4), and across the wider southern Amazon. Precipitation during the mid-to-late-Holocene (before ~2000 BP) across southern Amazonia was lower than present (Cross et al. 2000; Baker et al. 2001), meaning that the southern extent of Amazon rainforest was much further north than the current forest-savannah ecotone (Absy et al. 1991; Mayle et al. 2000; Burbidge et al. 2004). Laguna Orícore is located <5 km from La Luna, therefore we can be confident that LI is encompassed within the pollen catchment area of the larger lake (Sugita 1994). A drier climate during the mid-to-late-Holocene would also have meant greater burning potential and higher natural fire frequency, as seen in the charcoal record of both LO and LL during this period. There is no direct evidence, in the form of either archaeological remains or palaeoecological indicators such as cultigens, for human occupation in Iténez during the mid-Holocene. However, shell midden deposits excavated in the central Llanos de Moxos, indicate a hunter-gatherer presence in the south-west Amazon since the early-Holocene (Lombardo, Szabo, et al. 2013). Archaeological finds from elsewhere in the Amazon indicate that human occupation in the basin may have begun as early as the late-Pleistocene (12,000 BP) (Roosevelt et al. 1996). We cannot, therefore, rule out the possibility that humans where active in the Iténez landscape as early as the mid-Holocene, and may have contributed to the high fire frequency seen in the LL and Orícore records, through burning of the savannahs.

The earliest direct evidence for human impacts on LI is the appearance of maize pollen in the LL record at 2090 BP. At this point in the record, savannah is still the dominant vegetation on the island. The appearance of maize in the record is immediately preceded by a spike in the >250 µm charcoal curve, which may indicate
anthropogenic burning locally around the lake, associated with the establishment of agriculture.

Regionally, the L. Orícore record tells us that there was a decline in burning in northern Iténez from ~2200 BP, and an expansion of forest dated to ~2000 BP (Carson, et al. Chapter 3). This occurred in response to increased wetting across southern Amazonia, linked to the southward migration of the Intertropical Convergence Zone (Absy et al. 1991; Mayle et al. 2000; Burbridge et al. 2004). However, despite this regional transition from savannah to forest, LI remained an open grassland and continued to experience higher burning frequency/intensity, until 1240 BP (although the first appearance of Alchornea and Celtis in the record in zone 2 may reflect floristic changes driven by increasing regional precipitation). From 1240 BP, declining charcoal levels coincided with expansion of arboreal species, and the island transitioned from savannah to closed canopy forest. This suggests a similar history of land use to that practiced at the Bella Vista earthwork site, located 15 km north of LI (Carson et al. Chapter 3; Chapter 4). The inhabitants of LI appear to have held back the climatically driven expansion of forest, which occurred regionally ~2000 BP, through continued burning of the island and suppression of tree growth.

The dating of maize in the LL record demonstrates contemporaneous occupation with the ring ditch site at Bella Vista (Carson et al. Chapter 4), which was occupied from 2500 BP. This may indicate that the two sites were part of a wider population/culture, which lived in and built across the savannah-terra firme island landscape. However, the decline in burning and expansion of forest at LL dated to 1240 BP, occurs much earlier than the forest expansion recorded at Bella Vista, which was dated to 500 BP (Carson et al. Chapter 4). This disparity in vegetation history between the two sites may be explained by an earlier decline in intensity of land management at LI, in comparison to Bella Vista. It is also conceivable that LI served a different function during the latter stages of its occupation, perhaps being used for low intensity farming and resource gathering. This hypothesis is supported by Lombardo (Lombardo and Prümers 2010; Lombardo et al. 2011), who argue that forest islands in the Iténez province and the central Moxos, may have served as temporary slash-and-burn agricultural sites, rather than as settlements. This raises the important notion that, even within what appears to be a highly interconnected pre-
Columbian landscape in the Iténez province, there may have been heterogeneity of land-use between sites.

Unfortunately, there are no radiocarbon dates available to date the construction of the earthwork which encircles LI, and we cannot therefore confirm whether it was built at a time when the island was open. There is no evidence, however, of a clearance phase in our pollen record from LL. Since the earthwork lies around the edges of the island, it is possible that its construction took place post forest expansion, but did not involve major forest disturbance. In either case, our pollen data does not support the hypothesis that forest islands such as LI were cleared at any point during their use by pre-Columbian people, in order to build ring-ditches and palisades (Erickson 2010).

5.7.2. Agriculture and resource management

5.7.2.1. Maize

Maize was the only cultigen pollen type identified at La Luna. Maize requires well drained soils (Edwards et al. 1976) and is therefore unlikely to have been grown in the inundated grasslands without some form of hydrological engineering to improve drainage. There is no evidence in the area surrounding LI of the ridged agricultural fields which have been found in other parts of northern Iténez (Lombardo et al. 2013a). There are, however, paired causeway earthworks running across the inundated savannah between the forest islands, which may have be used to drain areas of wetland (Lombardo and Prümers 2010). However, when we consider that 1) there are no causeway earthworks immediately surrounding or that intersect with LL, 2) maize pollen is relatively poorly transported and will travel only short distances from the source plant (Lane et al. 2010), and 3) a non-forested LI would have provided an abundance of well-drained and open land, it seems much more likely that cultivation took place on the island itself. This is an example of early maize agriculturalists in southern Amazonia taking advantage of what was an open landscape, rather than growing in the slash-and-burn manner that is common across forested tropical regions today. This is also the earliest example of maize agriculture dated so far in the Bolivian Amazon, but together with other sites studied across the
region (Whitney et al. 2013; Whitney et al. 2014), it appears to support a broad introduction of maize to this region around 2000 BP.

**5.7.2.2. Agroforestry**

Our palaeoecological record has demonstrated that the earliest occupation of the LI site pre-dates the growth of forest on the island. As a result we have no truly “pre-anthropogenic” forest with which to compare the modern forest composition. Given that pre-Columbian people occupied the island during the transformation from savannah to forest, it seems likely that there was an anthropogenic influence on the development of species composition, but is this visible in the pollen record?

We attempted to test this by searching for the pollen of one of the key “orchard” species which is posited in the forest island landscape of north-east Bolivia, *Theobroma cacao*. However, despite increasing pollen counts to 1000 grains in both the lake surface sediment and the LB surface, *Theobroma* pollen was not found. *Theobroma*, like many Neotropical plant taxa, is an insect-pollinated (entomophilous) genus (Young 1994). Numerous pollen trap and lake sediment studies have demonstrated that insect propagation is no hindrance to preservation in the palaeoecological record (Bush 1991; Bush and Rivera 1998; Bush and Rivera 2001). We must conclude from its absence in the fossil pollen record, that *Theobroma* is either relatively unproductive of pollen, or has a peculiarly conservative pollination strategy, which means that it is extremely rare in the palaeoecological record and not useful as a “marker” of human forest management.

We can also note however, that other potential signs for a legacy of anthropogenic management of forest resources, are not borne out in the pollen record either. Palms for example, which are highly valued by modern indigenous groups for their fruits, the edible heart of palm, and for building materials (Posey and Balée 1989; Posey and Balick 2006), show no greater abundance in the surface sediments of either LL or LB, when compared to the pre-forest expansion environment documented in the LL core record. *Inga* sp. was also noted as a potentially important economic species in the modern botanical survey of LI. Most species of this genus produce edible seeds (Balée 1994; Clement 1999; Lentz 2000), and it has been identified as an important economic species from pollen records at another site in the western Beni (Whitney et al. 2014). *Inga* also produces large pollen grains, and
therefore, its recovery should have been improved by the enhanced sieving methodology (Whitney et al. 2012). Despite this, Inga does not appear in any significant abundance in the LL pollen record. The lack of any notable increase of these key economic taxa in the LL pollen record may indicate that their current abundance on LI is not attributable to planting by pre-Columbian people. However, as our palaeoecological record has shown that the forest on LI grew alongside human occupation, attempting to discern anthropogenic from natural drivers in the development of the LI forest community, may be an unanswerable problem.
5.8. Conclusions

This study has presented an informative palaeoecological record from an archaeological region with a paucity of such palaeoenvironmental data, and from a previously unstudied archaeological site. Pre-Columbian occupation of the La Luna earthwork site is shown to have begun at a time when vegetation on the island was open savannah, rather than closed canopy rainforest, as exists on the site today. While regionally the palaeoenvironment transitioned from savannah to rainforest from ~2000 BP (Carson et al. Chapter 3), the inhabitants at LI suppressed this climatically-driven rainforest expansion, maintaining an open landscape locally on the site. From 1240 BP, anthropogenic burning of the island declined and forest was allowed to expand, establishing the closed-canopy rainforest that exists on the island today. The afforestation of LI occurred earlier than that recorded at the nearby Bella Vista earthwork site (Carson et al. Chapter 4), indicating a probable earlier abandonment and/or decline in land use at LI. We found no evidence that, following the climate-driven afforestation of LI, there was any subsequent deforestation by pre-Columbian people. This contradicts the suggestion that earthwork construction on forest islands in this region necessitated extensive deforestation (Erickson 2010). The lack of evidence for pre-Columbian deforestation suggests that earthwork construction across the forest island landscape of north-east Bolivia, did not have profound impacts on biogeochemical cycling thorough biomass removal and increased carbon emissions (Dull et al. 2010; Nevle et al. 2011). Population estimates, based upon the labour required to clear the forest islands (Erickson 2010), should also be reconsidered in the light of this new data. The study has demonstrated further, that inferences about the environmental impact of Pre-Columbian earthwork builders should not be made based upon the state of the modern ecosystem.

We found the earliest example so far for the adoption of maize agriculture in the Bolivian Amazon, dated to 2090 BP. This provides a further example of early pre-Columbian agriculturalists taking advantage of an existing open landscape, rather than practicing shifting slash-and-burn agriculture. Our attempts to use the pollen of *Theobroma* as a possible marker of horticultural management revealed that the genus is poorly represented or silent in the pollen record. However, other supposedly key economic taxa, which were represented in the pollen record, such as palms and *Inga,*
did not show any increased abundance associated with pre-Columbian occupation, suggesting that these particular taxa did not form an important part of the resource management strategy employed at LI. The lack of observable changes in forest composition over the history of the site, may be due to the fact that humans occupied LI during its afforestation, and have been influencing the forests composition since the start. We therefore have no pre-anthropogenic “benchmark” forest composition with which to make comparison. This has important implications for the field of historical ecology, in which it has often been argued that anthropogenic influences formed an important driver in the development of Amazonian ecosystems (Posey and Balée 1989; Erickson and Balée 2006; Heckenberger et al. 2007; Clement and Junqueira 2010). This also has implications for ecological and conservation perspectives in this region, which lies within a protected area and would typically be viewed as “pristine”.

5.9. Acknowledgments:

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Chapter 6. General discussion

The results chapters of this thesis have been presented in journal format and address specific questions and/or discreet study sites in the Bolivian Amazon. However, as a body of work, these chapters represent a substantial contribution overall to palaeoecology and archaeology in the ring ditch region of Bolivian Amazonia and in the wider Amazon. The aims of the thesis can be described as addressing three broad issues in the study of pre-Columbian land use and impact in the Amazon, namely: 1) the Scale of past human impacts, 2) the Nature and Legacy of past land use practices, and 3) the adaptation and development of novel methodologies for investigating past Neotropical human-environment interactions. The work of the thesis is examined in detail below, under the umbrellas of these three themes, and discussed within the wider context of Amazonian human-environment interactions. Finally, the potential for extending this work in the future is discussed.

6.1. The scale of pre-Columbian deforestation in Bolivia and the wider southern Amazon.

6.1.1. Anthropogenic impacts on biogeochemical cycling

The scale of deforestation and biomass burning associated with pre-Columbian cultures is a hotly contested issue in Neotropical archaeology, palaeoecology, and modern ecology. There is now widespread evidence for large, sedentary cultures in numerous regions of Amazonia, but whether the existence of these sites translates to evidence of extensive, contemporaneous deforestation, is disputed. Estimates of pre-Columbian population and scale of land use are often co-dependent. Assuming both large pre-Columbian populations and associated large-scale deforestation and biomass burning, a number of authors have modelled the role of Neotropical societies in driving pre-industrial, Anthropocene climate changes (Nevle and Bird 2008; Dull et al. 2010; Kaplan et al. 2010; Nevle et al. 2011). These authors have associated global declines in CO$_2$ recorded in Arctic and Antarctic ice
core records, and the Little Ice Age cooling experienced in the northern hemisphere, with native population collapse in the Americas from the 1500s. Dull et al. (2010), Nevle et al. (2011), and Neve & Bird (2008) compiled charcoal records from lacustrine and terrestrial archives across the Neotropics/Americas, and identified a substantial pattern of decline in burning through the 15th to mid-17th centuries. They equated this with a decline in anthropogenic burning, following the collapse of native societies due to myriad factors associated with the Columbian Encounter of 1492 AD and subsequent European colonization (Dobyns 1963; Denevan 1976; Denevan 1992b). The global decline in atmospheric CO₂ concentrations, they argue, is not accounted for by natural phenomena, such as volcanic activity and variations in solar irradiance. Instead they argue that post-collapse reforestation on a massive scale across the Americas resulted in a substantial carbon sequestration of 2 to 17 Gt carbon (Ruddiman 2007; Nevle and Bird 2008; Dull et al. 2010), equivalent to a reduction of atmospheric CO₂ by 6-10 ppm (Nevle et al. 2011). These figures were based on population estimates (Denevan 1992b), from which estimates of land use per person were derived, and therefore land cover area of post-collapse reforestation. Furthermore, it is posited that the lowland Neotropics, including the Amazon basin, contributed a substantial proportion (2-5 Pg) of this sequestration, because of its high pre-Contact population and high above ground biomass potential (Dull et al. 2010). All of these estimates of reforestation are based on the assumptions that 1) the palaeoenvironment surrounding pre-Columbian sites was forested during the mid-to-late Holocene, 2) human land use over this period involved large scale deforestation, and 3) an assumed carrying capacity of land per person, which rejects ideas of poor nutrient availability and protein limitation.

Some of the most compelling archaeological evidence for large-scale deforestation in lowland Amazonia comes from geometric earthwork or ring ditch sites across the southern sub-region. Over 400 “geoglyph” earthworks have now been documented in eastern Acre state (Schaan 2012; Schaan 2013), hundreds of ring ditches in north-east Bolivia (Prumers et al. 2006; Saunaluoma 2010; Erickson 2010), and a large network of circular ditched villages, connected by roadways, in the Upper Xingu (Heckenberger 2003). More earthworks continue to be discovered beneath intact forest through the use of remote sensing (Prümers 2012a; Prümers
and it has been estimated that those documented so far may represent as little as 10% of those that remain undiscovered beneath intact canopy (Parssinen et al. 2009). Heckenberger et al. (2008) links all of these sites culturally and historically as part of a great diaspora of the Arawak people, who spread the traditions of ring ditch building through southern Amazonia. This is not a universally agreed concept amongst archaeologists; however what does link these sites is their position today beneath closed canopy rainforest, in the seasonal southern Amazonian rainforest (SSAR) region. It is posited that the construction of these earthworks entailed widespread deforestation and the development of an open, highly anthropogenic landscape (Heckenberger et al. 1999, 2007; Heckenberger 2003; Erickson 2010). Pre-Columbian environmental impacts across the southern Amazon would therefore, have been substantial, with an equally large-scale forest recovery following indigenous collapse.

These conclusions regarding past human-environment interactions have been made within a context of sparse palaeoecological data and assume that the palaeoenvironment during construction of these earthworks, was the same as the modern closed-canopy forest landscape. However, palaeoecological studies from Lagunas Bella Vista, Chaplin (Mayle et al. 2000; Burbridge et al. 2004) and Yaguarú (Taylor et al. 2010) in the south-west Amazon, and Lake Carajas (Absy et al. 1991) in the south-east, along with several stable isotope studies from terrestrial soil transects (Pessenda et al. 1998a; Pessenda, et al. 1998b; Gouveia et al. 2002), have demonstrated that the forest-savannah ecotone across the southern Amazon has not been static over the mid-to-late Holocene. Drier-than-present climatic conditions during the mid-Holocene, as recorded in the Lake Titicaca precipitation record (Cross et al. 2000; Baker et al. 2001) and central South American Pantanal record (Whitney et al. 2011), meant that the rainforest ecotonal boundary was further north than its modern extent and dominant vegetation was a mix of more open, drought-tolerant, semi-evergreen tropical forest and savannah. From ~3000-2000 BP, there was a southward expansion of rainforest driven by increasing precipitation across the southern Amazon, linked to the southward migration of the Inter Tropical Convergence Zone (Mayle et al. 2000; Mayle and Power 2008). We therefore know that the climate and palaeoenvironment in southern Amazonia has not been static.
However, the spatial and temporal extent of this ecotonal movement, though it is known on a very broad, sub-continental scale from the records outlined above, is not well constrained. In the area of ~1,000,000 km$^2$ covered by what we have called the seasonal southern Amazonian Rainforest (SSAR), 4 lake records and 3 soil stable isotope records provide the only palaeoecological data on ecotonal movements. Furthermore, the position of geometric earthwork construction, both spatially and temporally, in the context of these biome shifts, has not previously been considered.

The pollen and charcoal record from Laguna Orícore (LO) (Chapter 3) has revealed that, in the Iténez region before ~2000 BP, the palaeoenvironment was characterised by savannah, with a higher incidence of burning in the landscape compared to present. After ~2000 BP, rainforest expanded in the region, most likely as a part of the time transgressive expansion of forest, which occurred across southern Amazonia, in response to the southward migration of the ITCZ. By 1700 BP, closed canopy forest, similar to that which exists today, had become established on the terra firme land of the pre-Cambrian Shield (PCS) in Iténez. Our local-scale records of palaeoenvironmental change and human land use on two ring ditch sites at Laguna Granja (LG) and Laguna La Luna (LL), have demonstrated that human occupation on both of these sites was established before this rainforest expansion. Once forest expansion began regionally in Iténez, the occupants around LG and on La Luna Island (LI) maintained an open landscape by suppressing forest growth locally around the sites. Afforestation did not occur on LI until 1240 BP and at LG until 500 BP, when intensity of land use appears to have declined on each site respectively. The construction period of the ring ditch earthworks around LG, dated from archaeological layers, falls between 800-500 years BP (Prumers et al. 2006; Dickau et al. 2012), and can clearly be seen as a period of intense burning in the LG palaeo record. These data demonstrates that earthwork construction took place in an open landscape, maintained since first occupation of the site, and therefore, contradicts the suggestion that construction of geometric earthworks in north-east Bolivia required massive deforestation.

The charcoal records from LG and LL indicate that pre-Columbian land use on those sites did entail greater frequency/intensity of burning than in the modern environment. Burning was likely an important tool, utilized to maintain an open
environment and possibly involved in the agricultural process. Furthermore, the LG and archaeological records from Bella Vista town (Prumers et al. 2006) indicate that land use declined on the site at ~500 BP, suggesting a possible role for the Columbian Encounter of AD1492 in precipitating a collapse of native populations/decline in land use in this region. This fits in with the model of a general decline in biomass burning across Amazonia, following European contact (Nevle and Bird 2008; Dull et al. 2010). However, comparison to our record from LO indicates that, on a regional scale, burning began to decline from ~2200 BP and by around 1200 BP, reached levels comparable to the modern, low frequency burning regime. The other Bolivian regional scale lake records from LCH, LBV, and Yaguaru, also show decline in burning earlier than 500 BP. We can surmise then, that the continued anthropogenic burning observed in the LG and LI records was limited to areas locally around the earthwork sites, and that regionally forest remained intact once established. We should also note that LL and other palaeoecological records from pre-Columbian sites in the Bolivian Amazon (Whitney et al. 2013; Whitney et al. 2014) show decline in anthropogenic burning well before 500 BP. This suggests that the timing of site decline/abandonment was not uniform across the Bolivian Amazon. An analysis of the patterning of charcoal decline throughout the Americas by Power et al. (2012) concluded that, on a global to hemispheric scale, burning declined as a result of reduced combustibility and was in fact driven by LIA cooling, rather than being a driver of global climate. Other global syntheses of past fire activity have concluded that the averaged biomass burning reconstruction curve from the Americas, is not well correlated with late-Holocene global CO₂ decline recorded in Antarctic ice cores (Carcaillet et al. 2002).

Crucially, the scenario of human-environment interaction demonstrated in the Iténez region, does not support the model of decline in regional scale anthropogenic burning, massive reforestation, and subsequent sequestration of atmospheric carbon after 500 BP. Instead, there was a gradual afforestation and burning decline across this landscape between ~2000-1700 BP, primarily driven by climatic factors. On a local scale, humans did impact the landscape through burning and agricultural activity, which may have meant modifying soils and maintaining open areas of land.
As has been mentioned, the spatial and temporal scale of late-Holocene
ecotonal movement across southern Amazonia are still not well constrained,
however, some clue to the possible extent of movement may lie in the modern
distribution of SSAR (fig. 6.1). This southern sub-region of the Amazon basin, which
now exists under a highly seasonal precipitation regime, would arguably have been
the most susceptible to changes in precipitation linked to the latitudinal position of
the ITCZ and to associated biome changes. The SSAR region also overlaps with the
distribution of geometric earthwork construction. We can hypothesise, therefore, that
the same scenario of human land use interacting with and adapting to natural
ecotonal movements, may apply to geometric earthwork sites across the southern
Amazon. This would imply that the net biomass increase occurring in the modern
forest environment across this region (Baker et al. 2004) is the result mainly of this
naturally-driven forest expansion, rather than a legacy of pervasive past
anthropogenic impact (Chave et al. 2008).
6.1.2. Population size, labour and tool use, inferred from scale of impact

The scale of pre-Columbian environmental impact is one of the parameters employed to estimate pre-collapse population size in Amazonia (Nevle and Bird 2008). In southern Amazonia, the existence of geometric earthworks covering a large area has been taken as evidence for large populations (Erickson 2000; Heckenberger 2003; Parssinen et al. 2009; Erickson 2010), based not only on the monumentality of the built landscape, but also on the labour required to deforest the land for construction (Erickson 2010). In the forest-island landscape of Iténez province, discussed in Chapter V, Erickson (2010) has inferred a large pre-contact population, based upon estimates of the labour required to clear the forest islands for earthwork construction. Similarly in the Upper Xingu, a pre-contact population of tens of thousands has been estimated, based upon the scale of the built landscape (Heckenberger and Neves 2009). In Iténez, it was assumed that the vegetation covering the PCS islands would represent “a highly anthropogenic version of the modern forest” (Erickson, 2010). Furthermore, ethnographic records of modern...
indigenous groups and colonial era accounts were referenced to infer that the whole area within a ring ditch would have been cleared of forest. Labour estimates where then derived using data from modern experiments with stone axes, which provided labour hours per tree felled (Denevan 1992a; Denevan 2001). The efficiency ratios of stone to steel axes was found to range from 10:1 with a 15cm diameter tree, up to as much as 60:1 for a large hardwood. Therefore, clearing of the areas of forest now known to be underlain by earthworks, would have been a highly laborious task.

The palaeoecological records from LL and LG, however, have demonstrated that land use on these sites did not involve major deforestation. Therefore, although calculations of labour required for the removal of earth during ditch construction may still be a valid parameter for population estimates in the ring ditch region, basing these estimates on labour intensive deforestation using the pre-Columbian technologies of stone tools, tree girdling, and fire, is not. The possibility, as suggested by the work in this thesis, that other geometric earthworks in southern Amazonia were constructed in a more open landscape than the modern environment, should be taken into account when estimating population sizes based on labour. Although the existence of a more open landscape during earthwork construction does not preclude the possibility that geometric earthwork building cultures were large, we can suggest that construction within an open landscape may have been achievable by smaller populations than those previously estimated, based on the labour required to clear dense-canopy forest.

6.2. The nature and legacy of pre-Columbian land use in Bolivian Amazonia

6.2.1. Agriculture

The basis of pre-Columbian subsistence in Amazonia is a topic of some debate, and is interwoven with issues of sustainable population size and social complexity (Roosevelt 1989; Meggers 1992a; Meggers 1992b; Heckenberger 1998). The old paradigm of pre-Columbian land use, based upon the lifestyles of modern indigenous groups, who are typically semi-nomadic and/or practice rotational slash-and-burn agriculture, held that poor soil quality in most regions of Amazonia was a
limiting factor for agricultural productivity and precluded sedentism (Meggers 1971; Meggers 1995). However, now that archaeological discoveries have provided ample evidence for sedentary societies in places such as the Bolivian Amazon, the debate has moved forward to ask what manner of subsistence supported these groups. Proponents of larger pre-Columbian societies have highlighted the rich protein resources available from aquatic animals (Posey and Balée 1989; Balée 1994; Erickson 2000) and proposed productive resource gathering strategies, which combined agriculture, agroforestry and aquaculture. The staple crop commonly grown in the Bolivian Amazon today is manioc or yuca (*Manihot esculenta*). Yuca is considered a hardier crop than other staples, such as maize, and better able to cope with less fertile soils (Rival and McKey 2008). Yuca has therefore been posited as the basis of early agricultural systems supporting large societies in the southern Amazon (Heckenberger 1998). Maize (*Zea mays* L.), which originated in Mesoamerica ~9000 BP (van Heerwaarden et al. 2011) and reached the lowland Amazon of eastern Ecuador as early as 6000 BP (Bush et al. 1989), is also a candidate for an early staple in the Bolivian Amazon, although it is typically considered a less hardy crop than manioc (Denevan 2001) and poorly suited to soils with high aluminium content, as are encountered in parts of the Beni (Lombardo et al. 2013a).

Research into the agricultural practices employed by pre-Columbian people in the Bolivian Amazon has, to date, consisted of only a few archaeobotanical and palynological studies (Dickau et al. 2012; Whitney et al. 2013; Whitney et al. 2014). Even in the wider Amazon basin, the number of palaeoecological studies which have presented evidence of cultivars, e.g. Bush et al. 1989; Piperno 1990; Bush et al. 2000; Bush et al. 2007a, is tiny compared to the size of the region and the complex diversity of cultures that inhabited it. Our palaeoecological records from LL and LG have revealed that maize agriculture was practiced in the northern ring ditch region of north-east Bolivia from ~2000 years BP. Despite the use of the enhanced sieving methodology described in Chapter 2 (Whitney et al. 2012), we found no pollen of other cultigens such as yuca or sweet potato. In the archaeobotanical study conducted by Dickau et al. (2012) at the Bella Vista site, the identifiable starch grains analysed from stone tools were almost exclusively from maize, leading the author to conclude
that this was the staple crop grown on the site. This however, was a tentative conclusion, due to the overall scarcity of phytoliths and starch grains recovered for analysis. Our pollen evidence from LG confirms that maize was likely the staple crop, and our record from LL further suggests that maize was an important crop grown across the ring ditch/forest island landscape. These are the first data of any kind on past agricultural practices from one of the natural forest island sites in north-east Bolivia. Other recent palaeoecological studies from the artificial monumental mounds region in the south-west *Llanos de Moxos* (Whitney et al. 2013), and from the raised agricultural field region (Whitney et al. 2014) in the central *Moxos*, also found maize pollen associated with the building and use of these pre-Columbian earthworks. These emerging data are beginning to suggest that maize agriculture played an important role in the subsistence of earthwork building cultures in the Bolivian Amazon, from a broad starting point at ~2000 years BP, and that the prevalence of yuca in modern cultivation, is a product of later, possibly post-European-contact events. However, the studies from the monumental mounds region also found phytoliths of squash (*Curcurbita* sp.), chilli pepper (*Capsicum* sp.), yam (*Discorea* sp.) and yuca (Dickau et al. 2012), and the raised fields appear also to have been used for sweet potato cultivation (Whitney et al. 2014), suggesting that agricultural subsistence may have been more varied in the monumental mounds and raised field regions, in comparison to the ring ditch region.

### 6.2.2. Agroforestry

Ethnobotanical studies and historical accounts of indigenous societies have shown that exploitation and management of non-domesticated, but edible or “economically useful” plant taxa, and some domesticated/semi-domesticated fauna, can constitute a large part of the subsistence regime of lowland Neotropical cultures (Balée 1994; Lentz 2000; Erickson and Balée 2006; Stahl 2008). Historical ecologists recognize the ability of naive Amazonians to be not just passive collectors of forest resources, but active in managing the landscape through selective encouragement of valued species. A key question is to what extent pre-Columbian societies practiced resource management or agroforestry as part of their subsistence, and further, what lasting legacy of impact this might have had on species
compositions in extant ecosystems (Clement et al. 2003; Chave et al. 2008; Clement and Junqueira 2010). The abundance of economically useful species in the modern environment has also been used as an indicator of historical anthropogenic activity, in places where artefactual evidence is scarce (Erickson 2008; Fraser et al. 2009; Balée 2010; Shepard and Ramirez 2011; Levis et al. 2012). However, there is disagreement over whether these patterns of resource-rich forest, recognized by historical ecologists, actually represent anthropogenic management, or whether they are better explained by natural environmental factors (Barlow et al. 2012). Monospecific stands of the palm *Mauritia flexuosa* (Buriti palm) for example, which are cited by historical ecologists as signs of pre-Columbian agroforestry, have been attributed to soil hydrology, as *Mauritia* inhabits wetland sites (Barlow et al. 2012). Similarly, monospecific stands of bamboo, liana and other species of palm, which are assigned an anthropogenic origin (Posey and Balée 1989), could alternatively be explained by natural forest disturbance or edaphic factors. There is also little account given to the influence of modern management of these supposedly pre-Columbian anthropogenic forests, many of which have been repopulated in modern times. Some of the forested monumental mounds in the central Moxos, for example, support modern settlements; yet historical ecological studies of these sites have assumed that forest composition is a remnant of pre-Columbian management, rather than resource exploitation in the modern era.

Palaeoecology has the potential to add a chronological perspective to this debate, and potentially identify the historical “moments” of vegetation compositional change associated with cultivation of particular species. This has certainly been demonstrated from palaeoecological lake records around Maya sites in Central America (Jones 1994; Rushton et al. 2012), where there is a clear signal of palm cultivation. In the Bolivian Amazon too, Whitney et al. (2014) identified a possible signal of *Inga* sp. cultivation in the raised fields region. However, the authors also noted that *Inga* is a common post-disturbance successional species, highlighting the difficulty of attempting to disentangle anthropogenic from naturally-driven palaeovegetation changes.

Botanical surveys undertaken in the forests and savannah across the different geoarchaeological regions of the Bolivian Amazon, have identified high occurrences
of economically useful tree species, such as Brazil nut (*Bertholletia excelsa*), the rubber tree (*Havea braziliensis*), *Inga* sp., and various species of palm (Posey and Balée 1989; Erickson and Balée 2006). These have been interpreted as the legacy of resource management by the pre-Columbian inhabitants of the Beni, who it is posited transformed their environment, not only on a local scale confined to habitation sites, but on a landscape scale. In the forest island sub-region of Iténez province, Erickson (2010) inferred a highly domesticated forest environment from the high densities of the chocolate tree *Theobroma cacao*, encountered on the islands around the town of Baures (Erickson 2010). Our attempt to identify *Theobroma* in the pollen record from LL and La Luna Bog (LB), in Chapter 5, revealed that it is a palynologically silent taxon. There was no evidence either of increased abundance of palms or of the tree genus *Inga* sp. (which occurs in the modern forest) associated with human occupation on the LI site, though their pollen was present in small abundance and identifiable in the LL record. The LG record also revealed no indication that wild tree species were cultivated around the Bella Vista site.

The results from Chapters 4 and 5 highlight three difficulties in utilising palynological analyses to identify economically useful taxa in the palaeoecological record. Firstly, there is the issue of silent or unproductive taxa. This potentially applies to other economic species that we may wish to use as markers of human influence. For example, Brazil nut is abundant in the *terra firme* forests in Pando and Acre states as a result, it has been suggested, of widespread planting by pre-Columbian peoples (Posey and Balick 2006; Shepard and Ramirez 2011). This species however, like *Theobroma*, is entomophilous (Nelson et al. 1985), and may therefore be poorly represented in the pollen record. Secondly, there is the issue of taxonomic resolution, i.e. being able to distinguish economic species from non-useful members of the same genus, or even from other genera. Most Neotropical plant taxa are identifiable only to genus level using their pollen and so distinguishing cultivated from non-cultivated types may not be possible. Thirdly, the array of economically useful taxa derived from ethnobotanical studies is expansive, making it possible that almost any species encountered in the forest may have some useful feature, and be described as economic. Therefore, deciding how to narrow these lists is a key consideration. Some methods have included targeting only species which have a
specific name linked to their use in a native language (Levis et al. 2012). In the case of plant genera or families which in general are exploited as economic types, such as Arecaceae (palms) and *Inga*, fine taxonomic identification may be less of an issue, and using a multi-evidence based approach combining pollen, charcoal and phytoliths to identify concurrent human activity, can provide greater confidence that an increase in an economic plant type is anthropogenic. In spite of this, no evidence of palm or *Inga* cultivation was found in the pollen records at LI or LG, leading us to conclude that, although they were very likely harvested by native people, their cultivation was not an important part of the subsistence strategy at these sites.

### 6.2.3. Legacy of land use

Although our palaeoecological records from LL and LG did not show any specific species compositional changes related to human occupation, in both cases they did demonstrated that human use of the sites predated the expansion of forest. We infer therefore, that anthropogenic influences may have formed an integral, but immeasurable, part of the ecosystem’s development. This agrees with one of the core arguments of historical ecologists, such as Erickson & Balée (2006), which posits that humans have been inhabiting and shaping Amazonian ecosystems, whether actively or passively, since the early-Holocene. Our record has not demonstrated such an ancient occupation in the Iténez region, but has shown that the current dominant rainforest ecosystem has never been without some degree of human impact.

This raises another interesting complication for palaeoecologists and conservationists. The aim of palaeoecological research, especially when its intention is to inform conservation practice, is often to identify ecological, pre-human-disturbance benchmarks. This has especially been the case in Amazonia, where the notion of the pristine pre-European human landscape is still prevalent. There has been strong argument, however, for the role of pre-Columbian peoples in shaping the development of ecosystems across Amazonia, and for this to be recognized in modern conservation practice (Heckenberger et al. 2007; Clement and Junqueira 2010). If further palaeoecological research produces records such as the ones presented here, demonstrating an ancient and continuous human impact in what is
superficially a “pristine” environment, greater credence may be lent to the model of parts of Amazonia as “cultural parkland” (Heckenberger 2003).

Our data do not agree, however, with the statement that archaeological evidence for large pre-Columbian societies is interpretable as an indication of the resilience of rainforest ecosystems to large-scale deforestation (Meggers 1992b; Willis et al. 2004; Bush and Silman 2007). We have demonstrated that local scale patches of open land were maintained within forest around earthwork sites until as recently as 500 years BP and subsequently became forested following site decline. However, the LO record shows that deforestation did not occur in the north-east Bolivian Amazon on a regional scale, and that pre-Columbian land use did not involve forest clear-cutting. Forest in this region cannot, therefore, be said to have recovered from large-scale deforestation in the past.

6.2.4. Heterogeneity of land use

There are many dichotomous debates over the nature of pre-Columbian land use in Amazonia (Roosevelt 1991; Meggers 1992b; Heckenberger 1998; Meggers and Brondizio 2003) and on either side of these, the tendency has been to posit a homogeneous model of land use in the Amazon basin, whether advocating large (Heckenberger 2003; Erickson 2008) or small-scale impacts (Bush and Silman 2007; McMichael, et al. 2012b) (fig. 6.2). However, perhaps inevitably as new data emerges from the fields of archaeology and palaeoecology, a more spatially diverse picture of pre-Columbian land use is being formed (Arroyo-Kalin 2012; McMichael et al. 2014).
In our analysis of the LL and LG records, we demonstrated that land use on the geometric earthwork sites in the Iténez region employed much more intensive/frequent burning of the *terra firme* landscape than is practiced in the region today, and that maize agriculture was likely an important part of the subsistence strategy. However, even within the relatively small area encompassed by our study sites, which given our dating of their occupation from the charcoal and maize records, were probably being used by the same or related population(s), there is notable variability in land use. The LL record shows a much earlier decline in intensity of land use (1240 BP), as indicated by charcoal decline and afforestation of LI, than LG (500 BP). After 1240 BP, LI is apparently still being cultivated, but under a less intensive regime, while LG continues to be burned and maintained as an open site. As suggested in Chapter 5, this may reflect the LG site’s use for occupation, while LI in the latter stages of its use was a temporary agricultural site (Lombardo and Prümers 2010; Lombardo et al. 2011). In southern Iténez, archaeological evidence for fish weir construction in the savannahs (Erickson 2000) and the presence of dark earth anthrosols (Hastik et al. 2013) suggest different forms of land use to those practiced in the north, even though the similarity of earthwork design and ceramics suggests that these were the same culture (Erickson 2010; Prümers 2012b; Lombardo et al. 2013a).

Extending further to the *Llanos de Moxos* as a whole, when we compare our palaeoecological records to records of contemporaneous societies in the central and western *Moxos* (Whitney et al. 2013; Whitney et al. 2014), we find further evidence of land use heterogeneity. As has already been discussed, while maize is grown on...
all sites, agriculture in the mounds and raised field regions involved a greater diversity of cultivars than in the ring ditch region. Intensive burning seems also to have been an important management tool in the central Moxos. However, in the raised field culture, it is suggested that there was a switch in land use practices to active suppression of burning during the latter half of the site’s occupation (Whitney et al. 2014). This practice of fire suppression is not evident in the ring ditch or mound regions. It is also suggested that prescribed burning in the mounds region was confined to the savannahs, and that there may have been considerable deforestation locally around the occupation sites. This contrasts with the system of land use in the ring ditch region, which we infer employed burning and cultivation of the terra firme as well as the savannah, and where a system of forest suppression rather than deforestation was practiced. However, it should be noted that neither of the records from Whitney et al. are as long as the LL or LG records (both are ≤2000 years BP) and therefore, give no comparable account of anthropogenic land use in the central Moxos before or during the regional forest expansion ~2000 BP.

6.3. Advancing knowledge through suitable methodological approach

The final major theme of this thesis has been the development of novel methodologies and adaptation of methodological approaches, to address the specific questions facing Neotropical palaeoecology/archaeology, and the environments in which we work. These include, for example, 1) recovery of rare cultigen pollen from large, flat, lake basins, 2) determining the spatial scale of past environmental changes, and 3) separating human from natural forcing of environmental change. The question of scale of impact is a central one and highly contentious within our field. So far, this debate has continued largely in the absence of palaeoenvironmental data or palaeoecological studies with the specific aim of investigating past anthropogenic impacts. Archaeologists have been heavily criticised by ecologists and other archaeologists/anthropologists, for projecting evidence of local-scale human impact at discreet archaeological sites into the wider landscape, with little basis (Bush and Silman 2007; Barlow et al. 2012; McMichael et al. 2012b).
Palaeoecological studies in the Ecuadorian Amazon used collections of neighbouring small lakes, some with known pre-Columbian occupation histories and others without, to determine the spatial extent of past impacts (Bush et al. 2000; Bush et al. 2007; Urrego et al. 2013). This approach, however, bore no information about impacts in the wider landscape, away from the lake sites. The approach of McMichael et al. (2011; 2012a; 2012b) to determining the spatial scale of pre-Columbian human impacts in the western and central Amazon was to use transects of soil auger cores, radiating out from known or suspected occupation sites. Phytoliths and macroscopic charcoal were then analysed from these cores to reconstruct vegetation and burning history. From this, the authors inferred decreasing human impact further from the floodplain zones and very little impact in general in the interfluvial environs of the western and central Amazon. Although this study covered a greater land area and more localities than any previous studies, it still involved extrapolating from point data (each soil core only representing a minute, local point, within the vast region sampled) and largely ignored archaeological evidence of large scale human habitation in the interfluvial zones, such as the geometric earthworks in Acre and north-east Bolivia. Our approach to investigating the scale of human impacts in the ring ditch region was to combine palaeoecological reconstructions from both large and small lakes, with differently sized pollen catchment areas. We have demonstrated that selecting lakes with overlapping pollen catchment areas is an effective way to discern local from regional-scale environmental changes within the same geoarchaeological landscape. Importantly, this also allowed us to describe the interaction between natural biome-scale changes and human land use, which was key in a region that has undergone considerable naturally-driven ecosystem changes.

On a local scale, our data from Chapter 3 has demonstrated that obtaining both pollen and phytolith data from a lake sediment core is highly useful, both for corroborating the presence of cultivars and for reconstructing the spatial variability of land use locally on an archaeological site. Whitney et al. (2013a) had previously demonstrated this spatial differentiation of source area between pollen and phytoliths in a large lake basin in the central Moxos. Our study from LG demonstrates that, even from a small oxbow lake, there is a useful spatial variability in the source areas of pollen and phytolith microfossils.
Finally, we demonstrated that we can improve the detection of human land use in the palaeoecological record by developing novel laboratory techniques, adapted to the peculiarities of the Neotropical flora and lake basins with which we work. The new sieving methodology described in Chapter 2 of the thesis, was not only integral to the success of the research presented in the subsequent chapters, but is a contribution to palaeoecology in the wider Neotropics, where the identification of cultigen pollen is also a central tool for identifying human activity in the palaeoecological record (Whitney et al. 2012). The improved sieving methodology has been applied to recover evidence of cultivation in the raised field regions of the central Moxos (Whitney et al. 2013; Whitney et al. 2014) and coastal French Guiana (Iriarte et al. 2012a), at an ancient Maya site in Belize (Rushton et al. 2012), and also in the L. Chaplín lake record (unpublished data).
6.4. Potential for future work

When compared to the state of knowledge in the Old World, Neotropical palaeoecology and Amazonian archaeology remain in their infancy. Though palaeoecology has been established in Amazonia since the early work of Paul Colinvaux in the 1980s, and new studies are continually being published, palaeoecological archives remain sparse across this vast region. When we narrow our search to palaeoecological studies that integrate their data with archaeological records, or that specifically aim to investigate pre-Columbian land use, this drops away to only a handful of examples. Yet these are arguably some of the most pressing and contentious debates in Amazonian science, with relevance across disciplines, in one of the most important ecosystems in the world today.

The work presented in this thesis has introduced an important new scenario of human-environment interaction, which entailed lesser environmental impacts and less labour for pre-Columbian earthwork construction in the northern Iténez region. We also discussed the potential applicability of this scenario to the wider southern Amazon, based upon current knowledge of past savannah-rainforest ecotonal dynamics and modern precipitation patterns. This leaves us with a testable hypothesis, which we can investigate through further lake sampling in the other known geometric earthwork regions.

A simple survey using Google Earth imagery reveals potential target lakes located close to archaeological sites around Baures in the south of Iténez province and close to the Riberalta site in Pando state (figs. 6.3 and 6.4). Analysis of lake cores from these regions could potentially elucidate not only the extent of savannah-forest biome shifts in the late-Holocene, but also whether the type of land use which we have observed from our records in northern Iténez, was practiced more widely across geometric earthwork cultures. Constraining spatially and temporally the movement of this ecotone will have implications also for Amazonian biogeography and our understanding of climate-vegetation responses.
There remains great potential for further refinement and improved understanding in the use of Neotropical palynology as a tool to investigate anthropogenic impacts. Our investigation into the detectability of *Theobroma* pollen, though it produced a negative result, is an example of the type of work that will be necessary in Amazonian palaeoecology to build a “toolbox” of palynological markers of anthropogenic activity. Our laboratory techniques could also be further enhanced to detect agroforestry in the pollen record. The use of fine sieving, for example, in our sample preparations, could be a method for isolating/concentrating...
rare pollen grains from economic taxa. Some economic taxa, such as *Discorea* (yam), have relatively small pollen grains, and could potentially be isolated/concentrated using an appropriately sized sieve. Although the introduction of such a stage would not be an efficient use of sediment, in circumstances where the aim of the research was to investigate pre-Columbian management of specific economic taxa/taxon, the conservation of sediment may be deemed less important.

Improving our understanding of the spatial representation of lake pollen records in the Neotropics, will be another key development in Amazonian palaeoecology. The work in this thesis has demonstrated the efficacy of combining pollen records from lakes with differently sized pollen catchments, to discern palaeoenvironmental changes and human impacts on local and regional scales. However, this approach gives an indication only of the relative scales of change in a landscape. In order to move from a qualitative to a quantitative discussion of the scale of late-Holocene vegetation change and pre-Columbian human impacts, we will need to have quantitative estimates of the pollen catchment area of Neotropical lakes. Pollen catchment modelling in temperate landscapes is well developed (Bunting et al. 2004; Sugita 2007a; Sugita 2007b) and offers a potential basis for modelling of Neotropical systems, however at the same time, the Neotropics offer their own unique challenges. Unlike temperate flora, the majority of Neotropical plant species are not anemophilous, but insect or animal pollinated. This means that a proportion of the pollen signal may not be delivered to lake basins by wind transport, but perhaps by rewashing and water movement over land surfaces. Neotropical pollen also encompasses a greater degree of size ranges than temperate flora, with grains ranging from 5 µm to over 100 µm. This means incorporating greater variability of pollen grain fall speeds and source-to-sink travel distances. The enormous diversity of plant species in the Neotropics and poor floristic resolution of most pollen identifications further complicates trying to link pollen assemblages to percentage land cover.

In our research, we have analysed sediments from oxbow lakes. These lakes have typically been treated with caution by palaeoecologists, because of their dynamic nature and tendency to reconnect to river systems during times of flood. However, in lowland South America, they are often the only type of lake available
for coring, and have proved to be reliable archives of palaeoenvironmental change, at least over Holocene timescales (Behling 1995; Behling et al. 2000; Behling 2001; Whitney et al. 2014). Our understanding of the influence of river-transported pollen to lake sediment archives could be improved, for example, by utilizing surface sediment traps from oxbow lakes/swamps in different stages of development and analysing the pollen collected in these, or by taking water/sediment samples from river channels and analysing their pollen content.

In summary, there is a great deal still to be learned about pre-Columbian land use and environmental impact in Amazonia through the integration of palaeoecological and archaeological data. The work presented here is hopefully only the beginning of an improved understanding of human-environment interactions in one region of a rich historical landscape, which will form the basis of further investigations and a better understanding of pre-Columbian Amazonian cultures as a whole.
Chapter 7. General conclusions

The original aims of each of the four journal style chapters of the thesis are outlined below, along with the main findings of each paper. General conclusions from the overall work of the thesis are then discussed below.

Chapter 2 began with the initial problem of detecting historical cultivation using pollen in the palaeoecological record. It discussed the development of a new laboratory technique to improve the recovery of large cultigen pollen grains from Neotropical lake sediments. The aim was to test the efficacy of this new technique, using sediments from lakes in the Bolivian Amazon. The principal findings where that:

- The use of a 53 μm sieve is an effective way to isolate and concentrate large pollen grains, and importantly the grains of Zea mays, Ipomoea sp., Curcurbita sp. and Manihot sp.
- Through experimentation with the lab protocol, we determined that three factors were important for successful fractionation: 1) proper removal of clays through use of deflocculating chemicals and digestion using HF, 2) use of 15 ml centrifuge tubes, and 3) increasing spin speed to 3500 rpm, to ensure retention of small pollen grains.
- The technique was used to improve the recovery of large cultigen grains from two large lakes in the Bolivian Amazon, and has been successfully applied to sites in French Guiana and Belize.

Chapter 3 investigated the scale of environmental impact associated with geometric earthwork construction in the Bolivian Amazon and the wider southern Amazon. The principal findings where that:

- Earliest occupation of the earthwork site occurred when the landscape regionally was an open environment, under drier-than-present climatic conditions.
Following the climatically-forced regional rainforest expansion from ~2000 BP, the earthwork site was maintained open locally by its inhabitants through suppression of tree growth.

The geometric earthworks around LG were constructed in a maintained open landscape, reducing the need for labour-intensive deforestation and possibly requiring a smaller population.

Modern precipitation patterns across the Southern Seasonal Amazonian Rainforest (SSAR), suggest that this scenario of earthwork construction in a more open landscape, may apply to geometric earthworks across the southern Amazon.

These findings suggest that assumptions about large-scale deforestation associated with geometric earthwork construction, may not be correct, and that these pre-Columbian societies did not have large impacts on biogeochemical cycling through deforestation.

Chapter 4 discussed in more detail the pollen and charcoal records from Laguna Granja (LG) and their integration with phytolith and archaeological data. The chapter focused on the pre-Columbian land use/subsistence practices of the inhabitants of the Bella Vista earthwork site. It also discussed the advantages of using a core from a small lake situated in close proximity to an earthwork site. The principal findings where that:

- Evidence of anthropogenic burning and maize cultivation indicate that human activity began on the site ~2500 BP, much earlier than previously dated from archaeological layers.
- Land use locally around the site involved more frequent/intensive burning than that which occurs today.
- The scale of open land maintained around the site in pre-Columbian times is greater than the scale of modern clearance.
- Maize was the principal agricultural crop grown on the site.
- The integration of pollen, charcoal, and phytoliths from the same lake core, yields useful information on the spatial variability of anthropogenic impacts over time.
• Anthropogenic activity declined on the site ~500 BP, suggesting possible population decline linked to the Columbian Encounter of AD1492.

• The selection of a small lake located in close proximity to the earthwork site for coring, provides a local scale palaeoenvironmental reconstruction, which can be directly related to specific earthworks.

Chapter 5 discussed a local-scale palaeoenvironmental reconstruction from an earthwork site on a forest island in the Bolivian Amazon. The principal aims were to determine the land use practices and environmental impact associated with earthwork construction on the site. The principal findings where that:

• First occupation of the forest island site was dated to 2100 BP by the first appearance of maize pollen in the palaeoecological record.

• As at the La Luna Island (LI) site, occupation began before the regional forest expansion, when the island was not forested.

• We found no evidence that construction of the ring ditch earthwork on the island involved deforestation of the site.

• Population estimates, based on estimates of labour required for deforestation of the forest island landscape, are not reliable.

• The economically useful plant species *Theobroma cacao*, though prevalent on the forest island today, is not detectable in the pollen record.

• We found no evidence in the pollen record to indicate planting of particular economic species, such as palms, by pre-Columbian people. However, the fact that people used the island site before and during forest expansion, suggests that anthropogenic influences where important in the forest’s development.

In general, the work of this thesis provides palaeoecological data in a region where, to date, such information has been lacking. The pre-Columbian built landscape of the Bolivian Amazon is impressive and has revealed a cultural complexity which, only decades ago, was thought improbable in the Amazon basin. However, a great deal remains unknown about these cultures and their potential impact and environmental legacy. Here, we have demonstrated the utility of
palaeoecological data in providing information on land use practices, chronology, scale of anthropogenic impacts, and the role of human-environment interaction in the development of late-Holocene Amazonian ecosystems. Some general conclusions are that:

- The presence of extensive earthworks beneath modern rainforest should not be assumed to indicate large-scale pre-Columbian deforestation. Climate and vegetation in the South American tropics have not been quiescent over the Holocene. Establishing a palaeoenvironmental context is therefore vital to investigating past human land use and impact in the Amazon.

- Maize agriculture was an important part of the subsistence strategy of early agriculturalists in the Bolivian Amazon, and may have been introduced to this region as a whole ~2000 BP.

- Land use practices were heterogeneous between the archaeological sub-regions of the Bolivian Amazon and more widely between sub-regions of the Amazon basin. The pristine wilderness versus cultural parkland debate should, therefore, move beyond simplistic, basin-wide descriptions of human impact, irrespective of whether one supports a model of pervasive or limited impact.

- Human occupation of ring ditch sites in the Iténez region was long-lived and contemporaneous, demonstrating that these earthwork structures were likely built by the same culture, or related populations.

- Human occupation in Iténez also pre-dated the arrival of the closed-canopy, evergreen rainforest, which inhabits the terra firme today. We infer, therefore, that these forests developed alongside anthropogenic impacts, and could be described, to some degree, as anthropogenic. Further work is required however, to determine more about the nature of this influence.
A Appendix

A.1 Evidence for maize cultivation in raised fields region of south-west *Llanos de Moxos*: data from Laguna Isireri

In the early stages of this research project, one of the aims was to investigate the pre-Columbian land use history of the south-west region, in the *Llanos de Moxos* (See map fig 1.1, main thesis introduction). This region is distinct in its archaeology, being characterised by ridged agricultural fields, canals, causeways and some anthropogenic forest islands (Denevan 1966; Mann 2008; Saavedra 2009; Lombardo et al. 2011). Regionally, the vegetation consists mostly of inundated savannah, created by seasonal flooding of the impermeable clay soils (Clapperton 1993), with gallery forest occurring on river levees, lake shores, and other raised features (Navarro and Maldonado 2005; Erickson 2008). It has been suggested that this and other geoarchaeological regions of the Moxos represent an anthropogenic landscape, which has been heavily altered by human management throughout the mid-to-late-Holocene (Erickson and Balée 2006; Erickson 2008). We aimed to investigate the environmental context of earthwork construction in this region and to determine the type of agriculture and land use practices associated with the raised agricultural fields, using a palaeoecological record from Laguna Isireri (LIS). However, subsequent radiocarbon dating revealed that sediments in the top two-thirds of the surface core have probably undergone bioturbation or sediment overturning, and cannot therefore provide a chronologically sound record of vegetation change. The lake did produce pollen evidence of *Zea mays* L. (maize) cultivation, dated by a more congruous radiocarbon date at the base of the surface core. A fourth radiocarbon date, taken at 85 cm depth, shows an age reversal and was dated to 2159 ±40 radiocarbon years BP. The details of the study are briefly outlined below.

**A.1.1 Study site**

The lake selected for coring was LIS (14°59’16”S, 65°41’04”W), a large (19 km²), shallow (1.5 m), flat bottomed lake, located next to the modern town of *San Ignacio de Moxos* (fig. A.1 below), in the forest-savannah mosaic of the south-west Moxos. The site was selected for its close proximity to raised agricultural fields,
which have been found just a few hundred metres from the southern lake shore (per comms. Machicado Murillo, E. P.). A large lake basin, with a potentially large pollen catchment-area, was selected to gain a regional-scale record of anthropogenic impact and environmental change.

**Figure A.1 Google Earth® satellite image of L. Isireri**

**A.1.2 Sediment acquisition**

Coring of the lake was performed during fieldwork in June/July 2010. Surface sediments were extracted using a 5 cm diameter Perspex® tube and piston from a floating platform. Deeper sediments were taken using a drop hammer Colinvaux-Vohnout modified Livingstone piston corer (Wright 1967; Colinvaux et al. 1999). In total, a 110 cm sediment core was recovered, including a 51 cm surface core. The surface core was extruded in consecutive 0.5 cm increments in the field into sealed plastic bags or screw-lid bottles, and shipped to the University of Edinburgh. The Livingstone section of the core was shipped back to the UK in its aluminium core casing. Extrusion of the Livingstone core was done in the UK and samples stored in rigid plastic guttering and watertight plastic packaging. All samples were stored at 4°C. The sediments from Isireri are clay-rich and contain very little organic matter (< 5%), as estimated through loss-on-ignition at 550°C.
**A.1.3 Results**

Initial coarse-resolution pollen analysis revealed that abundant identifiable fossil pollen was present in the core from 0-50 cm depth. Below 50 cm depth pollen was not preserved. Pollen sampling resolution was increased to 3-3.5 cm intervals and macroscopic charcoal analysis performed at 0.5-cm intervals throughout the entire core.

**A.1.4 Radiocarbon dating**

A total of 4 AMS $^{14}$C dates were analysed from the core. Sieving did not produce sufficient plant macrofossil or macroscopic charcoal material for dating from samples 11.5 cm, 27.5 cm, and 48.5 cm, and so all these dates were from bulk sediment samples. A sample was also taken from 86.5-88 cm. The sediment in this depth range contained some of the highest abundance of macroscopic charcoal particles in the record, but overall particle sizes were small ($\leq$ 5 mm), and so macroscopic charcoal was extracted and amalgamated from across the depth range, to meet the sample size requirement for dating.

The samples taken from 11.5 cm and 27.5 cm depth both produced modern dates. This likely reflects contamination from “young” carbon as a result of bioturbation or sediment mixing. The sample from 48.5 cm depth produced a radiocarbon age of 2254 ± 37 radiocarbon years BP. This date seems congruous given its depth in the core and when we compare it to dates obtained from similar depths from lakes used in other palaeoecological studies in the *Llanos de Moxos*, for example L. Frontera (LF) (Whitney et al. 2014). Dating of the first appearance of maize at LIS at ~ 2000 BP, would also match broadly with the dating of the first maize pollen grains identified at LF, L. Granja (Carson et al. 2014b), and L. La Luna (Carson et al. 2014c). It is possible that lake sediments at this depth in LIS were stiff enough not to be effected by overturning caused by modern activity on the lake.

The fourth sample taken from 86.5 cm depth produced an anomalously young radiocarbon age of 2159 ± 40 years BP. The radiocarbon analysis report from the NERC East Kilbride facility explained that, during initial stages of treatment, this sample was found to contain <500 µg C. This required specialised AMS analysis at
low current and the preparation of additional standards which matched the sample carbon weight, in order to estimate the most appropriate $\delta^{13}C_{VPDB}(\%)$ for normalization of $^{14}C$ data. This value is shown in brackets below and does not necessarily represent actual $\delta^{13}C$ values in the original sample. The value shown without brackets is representative of $\delta^{13}C$ values in the original sample. The lack of pollen in the record from sediments below 50 cm depth, suggests that sediments below this point represent the ancient land surface, before lake formation and sedimentation.

Table A.1 $^{14}C$ AMS dating results from Laguna Isireri. Ages are shown as un-calibrated radiocarbon years. The % of modern $^{14}C$ enrichment is also presented.

<table>
<thead>
<tr>
<th>Sample identifier</th>
<th>Publication code</th>
<th>Depth below sediment-water interface (cm)</th>
<th>$^{14}C$ enrichment (% modern ± 1 $\delta$)</th>
<th>Conventional $^{14}C$ age (years BP ± 1 $\delta$)</th>
<th>$\delta^{13}C_{VPDB}(%)$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isireri 2: 11.5-12</td>
<td>SUERC-40051</td>
<td>11.5</td>
<td>105.80±0.28</td>
<td>modern</td>
<td>-23.6</td>
</tr>
<tr>
<td>Isireri 2: 27.5-28</td>
<td>SUERC-40052</td>
<td>27.5</td>
<td>105.33±0.31</td>
<td>modern</td>
<td>-22.0</td>
</tr>
<tr>
<td>Isireri 2: 48-48.5cm</td>
<td>SUERC-34152</td>
<td>48 cm</td>
<td>75.53±0.35</td>
<td>2254 ± 37</td>
<td>-25.0</td>
</tr>
<tr>
<td>Isireri 2: 86.5-88cm</td>
<td>SUERC-40500</td>
<td>86.5</td>
<td>76.43±0.38</td>
<td>2159±40</td>
<td>-15.4</td>
</tr>
</tbody>
</table>

*on-line $\delta^{13}C$ value made at SUERC AMS during $^{14}C$ determination. This value is not representative of the $\delta^{13}C$ of the original pretreated material.

A.1.5 Pollen and charcoal

The two samples at the base of the surface core (46 and 49.5 cm) showed slightly higher Poaceae levels (20-24%) and lower proportions of evergreen tree taxa such as Moraceae/Urticaceae (20%) and Cecropia (3-9%), in comparison to the rest of the record. A single Zea mays L. pollen grain was recovered from each of these horizons. Apart from these samples, the remainder of the core shows consistent abundances of evergreen trees vs. grasses, reflecting the savannah-forest mosaic that exists regionally around the site. However, whether this apparent stability of vegetation in the landscape reflects reality, or is the result of sediment overturning, we cannot be sure. The charcoal record throughout the core shows little variation, apart from between 80 and 110 cm, where there is a substantial spike in macroscopic charcoal. Pollen and charcoal results are presented in figure A.2 below.
Figure A.2 Pollen and macroscopic charcoal results from Laguna Isireri. Pollen is presented as percentage abundance per cm$^3$ with the exception of Z. mays which is presented as presence/absence. Macroscopic charcoal is presented as particles per cm.
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An improved methodology for the recovery of Zea mays and other large crop pollen, with implications for environmental archaeology in the Neotropics

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Abstract
We present a simple sieving methodology to aid the recovery of large colinegen pollen grains, such as maize (Zea mays L.), manioc (Manihot esculenta Crantz), and sweet potato (Ipomoea batatus L.), among others, for the detection of food production using fossil pollen analysis of lake sediments in the tropical Americas. The new methodology was tested on three large study lakes located next to known and/or excavated pre-Columbian archaeological sites in South and Central America. Five paired samples, one treated by sieving, the other prepared using standard methodology, were compared for each of the three sites. Using the new methodology, chemically treated sediment samples were passed through a 53 μm sieve, and the residue was recovered, mounted in cloisonné, and counted for large colinegen pollen grains. The counts were measured and analyzed for pollen according to standard palynological procedures. Zea mays (L.) was recovered from the sediments of all three study lakes using the sieving technique, where no colinegen pollen had been previously recorded using the standard methodology. Confidence intervals demonstrate there is no significant difference in pollen assemblages between the sieved versus unseived samples. Equal numbers of colinegen spores added to both the sieved and unseived parts produce the same results. This allows for direct comparison of colinegen pollen abundances with the standard terrestrial pollen record. Our technique enables the isolation and rapid screening for maize and other colinegen pollen in lake sediments, which, in conjunction with chemical and pollen records, is key to determining land-use patterns and the environmental impact of pre-Columbian societies.

Keywords
colinegen pollen, palynology, pollen preparation techniques, pre-Columbian agriculture, pre-Columbian archaeology, Zea mays

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Introduction
The pollen of domesticated plants, including maize (Zea mays L.), are key indicators of pre- and post-Columbian food production in palaeoecological records throughout South and Central America (Belling et al., 1988; Borja et al., 2004, 2005; Bush et al., 2007; Bush et al., 1989, 2007; Clement and Horn, 2001; Gossert et al., 2011; Isobe et al., 1996; Kennedy and Horn, 2006; Niemann and Belling, 2009; Northrop and Horn, 1996; Piperno, 2006a). Maize pollen is particularly advantageous for the detection of food production in the paleoecoregion because, although it has been shown to overlap in morphology, routinely size, with its ancestor, Ibanac teocintle (Zea mays subsp. parviglumis) (Hold et al., 2007; Mommsen et al., 2005; Piperno et al., 2004, van Hoevevaarden et al., 2011), maize pollen can be readily separated from that of other wild species by its size, surface sculpturing, and conical structure (Hold et al., 2007). However, maize pollen is poorly dispersed from the source plants because of its size (55-120 μm), as shown in both field and laboratory settings (Janssens et al., 2003; Rayner et al., 1992), as well as in modern lacustrine environments (Lame et al., 2010). Additional staple food crops common in the Neotropics, such as cassava (Manihot esculenta Crantz), sweet potato (Ipomoea batatas L.), and squash (Cucurbita spp.), can also be useful indicators of food production, although, at present, insufficient studies have been conducted to distinguish the pollen of these domesticated varieties from their wild relatives. Similar to maize, peanut (Arachis hypogaea L.), and Myrtus comosa produce large pollen grains (88-250 μm) (Herrera and Uriega, 1996), and because they are insect-pollinated (Hold et al., 1971; Reed, 1985; Reed and McCay, 1986; Rogers, 1995), these crops also yield low quantities of pollen.

Thus, key indicators of cultivation are generally rare within lacustrine fossil pollen records (Belling et al., 1998; Bush et al., 1995; Niemann and Belling, 2009), often represented by single grains in non-contiguous horizons, but because of their poor dispersal, large colinegen pollen are considered to be strong indicators of local food production (Bryant and Hall, 1993; Bush et al., 2007; Isobe et al., 1996; James, 1994; Lane et al., 2010; Pollard et al., 1996). Palaeoecologists and archaeologists have therefore inferred cultivation from simple presence/absence of these rare...
pollen grains, rather than quantified variations in abundance afforded by more common pollen types.

Given that it can be identified to species level, maize pollen is considered to be the strongest evidence of local food production in pre-Columbian archaeological and palaeoenvironmental studies (Clement and Horn, 2001). To increase the likelihood of its detection, palynologists often count outside the standard terrestrial pollen sum (typically 500 grains) (Achdut et al. and Horn, 2005; Bush et al., 1989; Clement and Horn, 2001; Horn and Kennedy, 2001; Kennedy and Horn, 2008), which can be a laborious and inefficient task. Even for a lake surrounded by cultivated maize fields, Zea pollen is either rare (< 80 grains/cm²), or even absent, from standard terrestrial pollen sums (Lane et al., 2010). Also, the number of slides required to be scanned for maize pollen is likely to increase with lake area and distance from the site of cultivation, given these factors are negatively correlated with maize concentrations in lacustrine sedimentary environments (Lane et al., 2010).

A better method that increases the probability of retrieving maize and other large eutin pollen involves the concentration of these grains using a fine sieve. A detailed methodology has been described for the retrieval of cereal grains in northern Europe (Bowler and Hall, 1989), using a 30 µm aperture sieve. However, this methodology suffers from the disadvantage of requiring two sediment samples from a given stratigraphic horizon—one for the standard pollen preparation, and the other specifically for sieving for large eutin pollen grains. Not only is this double-sample method time-consuming compared with analysis of a single sample, it is also wasteful of sediment (a sparse commodity from a typical 5 cm diameter core) that could potentially be used for additional palaeoenvironmental proxy analyses.

**Aims**

Here, we present a simple sieving methodology for detection of maize and other large eutin pollen that does not suffer from the drawbacks of a double sampling protocol (Bowler and Hall, 1989). Our method incorporates sieving as an extra key step within a standard chemical digestion pollen protocol (Fagri and Iverson, 1989), which markedly increases the likelihood and ease of recovery of maize and other large pollen from staple crops of South and Central America from lake sediments.

**Study sites**

We tested our methodology on sediments of three lakes from two distinct regions in Latin America, each located near known archaeological sites (Figure 1). Lake data are summarized in Table 1.

**Region 1: Northern Belize**

Belize is located in the northeastern part of the Yucatan peninsula, bordered by Mexico to the north, Guatemala to the south and west, and the Caribbean Sea to the east. This country is characterized by extensive pre-Columbian Maya occupation, including Lamanai, a site located in the north-central part of Belize where archaeological evidence points to continuous occupation from 300 BC to AD 1675 (Graham, 2001, 2004; Pendergast, 1986). The Maya settlement at Lamanai is located on the west shore of our study site, New River Lagoon (NRL), a substantial open-water body 60 km from the estuary of New River, one of the largest rivers in northern Belize (Table 1). The vegetation at Lamanai is characterized by lowland evergreen broadleafed forest (Bridge et al., 2012). A large swatch of marshy vegetation forms the eastern boundary of the lagoon (Micciche and Salvado, 2001; Metcalfe et al., 2009). Although a lake-level reconstruction from NRL points to shifts in the late-Holocene precipitation regime in northern Belize (Metcalfe et al., 2009), there is no archaeological or palaeolimnological evidence of Terminal/Late Classic collapse (c. AD 750–1050) (Graham, 2004; Metcalfe et al., 2009), unlike...
Table 1. Site information for the three study lakes, coring location, sediment type, and key archaeological features

<table>
<thead>
<tr>
<th>Location</th>
<th>Core location</th>
<th>Area (km²)</th>
<th>Water depth (m)</th>
<th>Sediment type</th>
<th>Archaeology</th>
<th>Distance of core to lake shore (m)</th>
<th>Distance of core to nearest archaeological feature (m)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laguna San José</td>
<td>14°56'58&quot;S</td>
<td>14.4</td>
<td>1.0</td>
<td>Clay</td>
<td>Neolithic</td>
<td>1500</td>
<td>&lt; 100</td>
</tr>
<tr>
<td></td>
<td>64°29'42&quot;W</td>
<td></td>
<td></td>
<td></td>
<td>mound</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Laguna Verde</td>
<td>14°59'10&quot;S</td>
<td>19.0</td>
<td>1.5</td>
<td>Clay</td>
<td>Raised fields</td>
<td>1000</td>
<td>Several kilometres</td>
</tr>
<tr>
<td></td>
<td>65°14'04&quot;W</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>New River Lagoon</td>
<td>17°40'19&quot;N</td>
<td>13.5</td>
<td>2.0</td>
<td>Organics</td>
<td>Muis settlement</td>
<td>&lt; 10</td>
<td>&lt; 10</td>
</tr>
<tr>
<td></td>
<td>88°39'16&quot;W</td>
<td></td>
<td></td>
<td>in carbonate</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

neighbouring archaeological sites in Belize (Alcala-Herrera et al., 1994; Jacob and Halmark, 1996; Guatemala (Hodell et al., 2001) and Mexico (Curtis et al., 1997; Hodell et al., 2003).

Region 2: Llanos de Moxos, Bolivia
The Llanos de Moxos is a large hydrological basin (130,000 km²) in SW Amazonia, situated in the Beni department, lowland Bolivia. The Llanos de Moxos is characterized by a mosaic landscape that consists predominantly of seasonally inundated savannahs, interspersed with forested mounds and river levees (Ortolana et al., 2004). Larger-scale pre-Columbian earthworks, including habitation mounds, raised fields, canals, causeways and fish weirs (Denevan, 1966; Erickson, 2000; Lombardo and Primors, 2010; Lombardo et al., 2011; Mann, 2008) have been identified throughout the region. Approximately 50 km east of Trinidad, the capital of the Beni department, the pre-Columbian earthworks consist of extensive large habitation mounds (> 100), canals and causeways (Lombardo and Primors, 2010). Situated among these archaeological features, Laguna San José is one of many flat-bottomed and shallow rectangular lakes dispersed across the Llanos de Moxos (Clapperton, 1993). Two large habitation mounds, Loma Salvatierra and Loma Mendoza, lie within 2 km of Laguna San José, and have been extensively excavated by archaeologists (Dickson et al., 2012; Lombardo and Primors, 2010; Primors, 2008, 2009a, 2009b). Several additional pre-Columbian earthworks surround the lake, including a canal located within 100 m of its southwestern shore (Lombardo and Primors, 2010). Dominant vegetation around Laguna San José includes seasonally inundated savannahs, some of which are used for cattle pasture. Forested patches are located near the lake shore. Situated approximately 80 km west of Trinidad, Laguna Eiriri is another large, rectangular lake adjacent to the town of San Ignacio de Moxos. Typical of the northern and western region of the Llanos de Moxos, evidence of pre-Columbian land use in this area consists of raised field cultivation (Mann, 2008), and several raised fields have been identified adjacent to Laguna Eiriri (Erickson, 1995; Suevedra, 2009). Although the local vegetation has been heavily modified in recent centuries, the dominant vegetation in the region is seasonally inundated savannah, with forest fringing the lake shore.

Methods
Sediment collection
Belize. The NRL core was taken in 1999 from a jetty adjacent to the Lamanai settlement using a square-ended modified Livingstone piston core. Four consecutive cores, totalling 510 cm of sediment, were recovered. Cores were shipped in plastic piping and stored at 4°C. Details of six radiocarbon dates, reported in

Metcalfe et al. (2009), demonstrate the core spans 1800 BC to AD 1500. Sediments from the NRL core comprise organic layers within a calcareous matrix, with increasing abundance of silts and clays down core.

Bolivia. Surface-sediment coring of the two lakes was performed in June and July 2010 using a 5 cm diameter Perspex® tube and piston from a floating platform. Surface cores of 31 cm (San José) and 51 cm (Isibiri) were extruded in consecutive 0.5 cm increments in the field into sealed plastic bags or screw-ad bottle, and shipped to the University of Edinburg where they were stored at 4°C. Sediments from both cores are clay-rich and contain very little organic matter (< 3%), as estimated through loss-on-ignition at 550°C.

Summary of protocol
For each of the three study lakes, five pairs of 1 cm² sediment samples were prepared. For each pair, both samples received identical chemical treatments, but one sample was treated with an extra sieving stage (53 μm), while the other core sample, taken from the same stratigraphic horizon, was not sieved.

Standard chemical digestion protocol was used for the preparation of all fossil pollen samples (Bennett and Willis, 2001; Faegri and Iversen, 1989), including hot 10% NaOH, 40% HF (with the exception of NRL), and acetylation treatments. However, the lengths and types of treatment differed somewhat between lakes, according to differences in sediment lithology. The sequence of chemical treatments performed for each study lake is outlined in Figure 2. In clayey sediments from the two Llanos de Moxos lakes, sample preparation began with a hot 5% sodium pyrophosphate treatment to disaggregate clays (Bates et al., 1976), followed by repeated rinses with water until the supernatant became clear. Hydrofluoric acid and acetylation treatments followed, after which the samples were passed through a 53 μm sieve (details below). The predominantly calcareous sediments from NRL in Belize were initially treated with 40% HCl until the sample stopped effervescing. New River Lagoon samples were not treated with HF, but instead were treated with hot Calgon®, and repeatedly rinsed until the supernatant was clear of suspended clays. Acetylation followed the removal of clays. After the chemical treatments detailed above had been completed, samples from all three sites were passed through a 53 μm sieve, as described below, to isolate the large euglenin pollen grains.

Isolation of large pollen grains
We used a 53 μm aperture sieve to isolate maize and other large euglenin pollen, based on the minimum size of maize pollen in Mexico determined by Holst et al. (2007). Also, pollen of
Figure 2. Flowchart of the pollen preparation method used for each study site, and incorporating the sieving stage.
Table 2. Results of the coarse (> 33 µm) fraction for each sample analyzed, including number of slides scanned for cuticle pollen, the proportion of small (< 53 µm) pollen grains inadvertently retained within the coarse fractions, and number and type of cuticle pollen recovered. The equivalent terrestrial pollen count refers to the number of pollen grains that the volume of residue scanned would have contained were it unaltered. See 'Statistics' for further details.

<table>
<thead>
<tr>
<th>Sample</th>
<th>No. of slides scanned for cuticle pollen</th>
<th>Equivalent terrestrial pollen count</th>
<th>No. of small (&lt; 53 µm) pollen counted on slides from coarse fraction</th>
<th>% small pollen on coarse fraction slides, relative to standard count</th>
<th>No. of cuticle pollen grains recovered</th>
</tr>
</thead>
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<tr>
<td>NRL</td>
<td>2</td>
<td>1096</td>
<td>3</td>
<td>0.2%</td>
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</tr>
<tr>
<td>1</td>
<td>1</td>
<td>640</td>
<td>0</td>
<td>0</td>
<td>nil</td>
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<tr>
<td>2</td>
<td>2</td>
<td>1116</td>
<td>5</td>
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</tr>
<tr>
<td>3</td>
<td>1</td>
<td>505</td>
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<td>0.8%</td>
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</tr>
<tr>
<td>4</td>
<td>2</td>
<td>776</td>
<td>8</td>
<td>1.0%</td>
<td>nil</td>
</tr>
<tr>
<td>L. san josei</td>
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<td>2976</td>
<td>2</td>
<td>0.07%</td>
<td>nil</td>
</tr>
<tr>
<td>7</td>
<td>3</td>
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</tr>
<tr>
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<tr>
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</tr>
<tr>
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<tr>
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</tr>
<tr>
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<tr>
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<td>7</td>
<td>1891</td>
<td>4</td>
<td>0.2%</td>
<td>nil</td>
</tr>
</tbody>
</table>

Pollen identification and counting

Cucurbita sp., Ipomoea batatas, and Manihot esculenta all have diameters > 80 µm (Herrena and Urrego, 1996). Preparations for sieving were begun by suspending the pellet in 10 ml of 10% NaOH and heating in a boiling water bath for 5 min, stirring occasionally to disaggregate the sample. Samples were transferred to a small beaker, using deionized water to rinse the contents of the tube into the beaker. Approximately 30 ml of water was used to transfer the sample.

The sample was passed through a 53 µm brass sieve with a few short blasts of deionized water from a wash bottle, and in doing so, we tried to keep the total volume of filtrate < 100 ml. The filtrate was saved for the standard terrestrial pollen count, and the residue was easily washed off the sieve into a 15 ml centrifuge tube, reserved for large (>53 µm) pollen grains. The filtrate was concentrated by centrifuging in 15 ml tubes at 3500 rpm, decanting, and topping up with additional filtrate until all of the fine fraction was contained in the pellet. All 15 ml tubes, now double the original number, were centrifuged and decanted.

At this stage, we checked to ensure whether the sieving had been successful (Figure 2). Selected tubes containing the coarse fraction (>53 µm) of a sample were wholly mixed, and, using a clean pipette, a small aliquot of the sample was transferred onto a slide and scanned at 100× magnification to check if the sample contained small (<53 µm) pollen and/or 'chopped' organic material which binds small pollen and prevents it from passing through the sieve. Although this was rarely the case, if the sieving stage was found to be ineffective, the coarse fraction (residue) can be re-suspended in 10% NaOH and re-sieved. The resulting filtrate can be concentrated as above, and combined with the fine fraction from the first sieving attempt.

After the sieving stage, an equal number of Lyophilium tablets (Stockmann, 1971) were added to each tube containing the residue and filtrate. The separated coarse and fine fractions were then dehydrated in tertiary-butyl alcohol and mounted in silicone oil for analysis.
found that only a negligible number of small pollen grains were inadvertently caught in the coarse fraction (Table 2).

Large non-cultigen pollen grains that palaeoecologists would usually include as components of the standard terrestrial pollen count, for example *Inga* spp. and *Anona* spp., are also concentrated in the coarse fraction. These grains usually comprise a very low proportion of the pollen sum, but their presence can be of high ecological value for the interpretation of the pollen signal. An exception, however, is *Pinus* pollen (> 53 µm), which are highly abundant in pollen assemblages from the Yucatan (Bhattacharya et al., 2011), where *Pinus caribaea* is an ecological dominant of Bajaian lowland savannas. In samples from NRL, this taxon comprises a significant proportion of the terrestrial pollen sum. However, here we included *Pinus* in the main sum by scanning the coarse fraction for *Pinus* until the number of *Lycozopodium* spores encountered on the slide was equal to the number counted in the fine fraction for the standard terrestrial pollen count. Given that we found an equal count of *Lycozopodium* spores was usually encountered in the first few transects across the coverslip, this is not a time-consuming task. Percent abundance of *Pinus* determined using this methodology for the sieved NRL samples are compared with standard non-sieved counts in Figure 3.

**Statistics**

Confidence intervals (95%) were calculated using the modification of Mather's lognormal distributions method in Paimpoll (Bennett, 2007; Mather, 1972). To relate the volume of sample
analyzed from the coarse fraction to that of the standard terrestrial pollen count, or the ‘count equivalent’ (Table 2), the number of Lycopodium spores found on the coarse fraction slide was expressed as a proportion of the Lycopodium count of the main pollen sum. For example, if 100 Lycopodium spores were encountered in the standard terrestrial pollen count and 10 spores were counted on the coarse fraction slide, then the volume of material on the coarse fraction slide represents 10% of the amount of sample analyzed for the standard terrestrial pollen count.

To determine what proportion of small pollen grains (<53 μm) were unintentionally retained in the coarse fraction (sieve residue) during preparation, the number of small grains that were encountered in the coarse fraction slide were expressed as a percentage of the main pollen sum using the aforementioned ‘count equivalent’. Expanding on the above example, and assuming a standard count of 300 grains, the amount of residue scanned in the coarse fraction is equivalent to a count of 3000 pollen grains. Thus, if 30 small pollen grains were found on the coarse fraction slide, we can estimate that this equates to approximately 1% of the 300-grain standard pollen sum (Table 2).

Results and analysis

Our results demonstrate that sieving at 53 μm successfully isolates large colutein pollen grains such as Zea mays (Table 2).

Although this is the only colutein pollen type found in the test samples, subsequent use of this technique has yielded Cucurbita (squash) pollen at NRL, and Manihot (manioc) and Ipomoea batatas-type (sweet potato) pollen grains in sediments from an additional study site in French Guiana (Briat et al., unpublished data).

All three study sites are substantial water bodies and cores from Laguna Hietz and San José were taken 1000 m and 500 m from the lake shore, respectively, to ensure the recovery of a continuous sediment sequence in case of lower lake levels due to past drought (the NRL core was taken within a few metres of the shore, adjacent to the Maya settlement Lamanai). Despite the considerable distance of the core location from shore, which is shown to reduce marine pollen concentrations in the sediment (Lane et al., 2010), we found no marine pollen in all samples analyzed from the San José core. Furthermore, the concentration of large pollen (<53 μm) by sieving was particularly successful for this site, where in some horizons, the proportion of 1 cm³ sediment sample examined for the coarse fraction equated to over 100× the proportion of the sample examined for the standard terrestrial pollen sum (Table 2). It follows then, that even if the samples were scanned outside the standard terrestrial pollen sum, it is highly improbable that any Zea mays grains would be found at Laguna San José and Hietz if this additional sieving stage had not been employed.

Zea mays pollen recovery was higher for sediments from NRL, which is unsurprising given the proximity of the core to Lamanai. However, analysis of previously unsieved preparations from NRL revealed no colutein pollen within the standard terrestrial pollen count, which meant that additional steps were necessary to isolate Zea mays from this site. This could have been achieved by scanning the unsieved preparations outside the pollen sum, but this too would have been very time-consuming, requiring the scanning of numerous additional pollen slide preparations.

In contrast, maize pollen was encountered in the first coarse-fraction slide analyzed in our new sieving methodology, thereby considerably reducing the amount of time an analyst needs to invest in searching for evidence of pre-Columbian cultivation.

Confidence intervals calculated for the paired samples (Bennet, 2007) demonstrate that the added sieving stage does not concentrate small pollen grains such as Cerealia or reduce the relative abundance of larger grains such as Poaceae. Furthermore, whatever small (<53 μm) grains are caught in the coarse fraction, they are not disproportionately represented by any particular pollen type. However, the number of small grains trapped in the coarse fraction is negligible (<1%) if care is taken to properly digest the sediments prior to sieving. Moreover, the 95% confidence intervals show tight overlap among the sieved and unsieved pollen percentage values for Pinus in the NRL record, which means that large (<53 μm) pollen grains can be confidently incorporated into the terrestrial sum using equivalent Lycopodium counts on both fine and coarse fraction slides.

A further key advantage to including our coarse-sieving stage is that the pollen assemblages in the fine fraction are more concentrated, and comprise ‘cleaner’ preparations, than those for non-sieved samples, making the process of pollen counting significantly faster and easier. To reach a count of 300 terrestrial grains can be relatively time-consuming in the analysis of largely inorganic sediments from large lakes in the Bolivian lowlands where pollen concentrations are often relatively low. Analysis of unsieved samples from NRL was particularly laborious (HF was not used in the preparation of these samples), and in most horizons, an average of seven slides were required to achieve a sum of near to 300 terrestrial grains. However, incorporating our sieving stage (53 μm) concentrated the samples such that full counts were reached in only two slides.

Recommendations for best sieving practice

The most important factors in ensuring a good recovery of small (<53 μm) pollen grains in the filter were: (1) choosing the appropriate chemical treatments for each sample type, and (2) sufficient rinsing with deionized water to remove chemical residues after each treatment, particularly HF. Standard chemical digestion protocol (Bennett and Willis, 2003; Fagert and Iversen, 1989) describes a set sequence of stages for the preparation of fossil pollen, often beginning with the hot NaOH (or KOH) treatment. Although this sequence works well for organic samples, such as peat and garbage, in the case of tropical sediments containing high proportions of clays and silts, the analyst is best served by first tackling the removal of organic matter to facilitate further chemical treatments. Second, intensive HF treatments (two treatments of 40% HF for 30 min in a hot water bath) are frequently required for lake sediments from tropical regions where the abundance of very small pollen (e.g. Cerealia and Minaea, <5 μm diameter) precludes the possibility of fine sieving (Gwynn et al., 1979) for the removal of clays and fine silts. However, the chemical residues from the digestion of silica (flouritecates), resulting from intensive HF treatment, often create a sticky black pellet that does not easily dispense in liquid. Although a common solution to the problem of fluoritecates buildup is the use of a hot 10% HCl treatment (Bennett and Willis, 2003), we found that 3-4 rinses in deionized water following the HF treatment removed the chemical residues and allowed the pellet to disperse in liquid. Further care was taken to ensure that all chemical residues were removed after each treatment, thereby ensuring the pellet was not clumped before we proceeded with the sieving step.
We advise against the use of 50 ml centrifuge tubes for concentrating the filtrate: without further experimentation to adjust the speed and/or time of centrifugation. Initially, we sieved at 53 µm using generous volumes of water (400–500 ml) to get the best pollen recovery in the fine fraction, but this practice dilated the fine fraction to such an extent that concentrating the pellet was difficult. After several rounds of centrifugation in 50 ml tubes, the fine organic material (and with it, small pollen) gathered on the angled sides at the bottom of the tube instead of forming a discrete pellet. This made decanting very difficult, and we lost a sizeable proportion of fine pollen grains, in particular, Cocos nucifera, experimenting with this method. Instead, we found the judicious use of ~ 100 ml of water, which resulted in a volume of filtrate that can be reasonably concentrated in 15 ml tubes, gave the best results. Also, increasing the centrifuge speed from 3000 to 3500 rpm ensures a better recovery of small grains in the filtrate.

Implications for palaeoecology and archaeology in the Americas

Where palaeoecological studies rely on terrestrial pollen and charcoal records from lake and bog sediments to demonstrate evidence of past land-use practices in archaeological contexts, changing climate can present a complicating story, particularly in regions such as the Yukon Peninsula, which has experienced large shifts in its precipitation regime over the past few millennia (Curris et al., 1990; Hodell et al., 1995, 2001; Metcalfe et al., 2009), and in the Southern Hemisphere tropics of South America, where rising precipitation in the late Holocene (Baier et al., 2001; Mayle and Power, 2008) has resulted in shifting Amazon forest-savanna boundaries (Burbridge et al., 2004; Mayle et al., 2006).

Thus, disentangling the relative impacts of changes in human land use, climate, and fire regime, upon past vegetation can prove problematic. Crucially, direct evidence of crop cultivation is required to unequivocally demonstrate human land use in the palaeo-record, but the recovery of large pollen grains has often proved difficult because of its rarity in pollen assemblages. Starch grain and phytolith analyses from selected archaeological features and residues from food containers (ceramic, bottle gourds) and plant processing tools (manos, edge ground cobbles, ceramic grinders), in addition to combined analysis of phytoliths, pollen and charcoal from lake and wetland sediments, have been successful in addressing this issue (Dickau et al., 2012; Duncan et al., 2009; Irrani et al., 2004; Pearsall et al., 2003; Piperno et al., 2007, 2009).

However, some important food crops, notably Manihot esculenta and Ipomoea batatas, do not produce diagnostic phytoliths (Piperno, 2006). The combination of all three techniques from archaeological contexts and lake sediments provides the most complete picture of the natural environments and plant associations in which the first farming arose (Denham et al., 2003; Piperno et al., 2007, 2009; Ramée et al. 2009) and assessing pre-Columbian land use in palaeoenvironmental studies (Pearsall, 2000), particularly as a lack of comparative palynological morphology studies means that present, cultivated varieties of domesticated plants (Ipomoea batatas, Manihot esculenta) are palynologically indistinguishable from some of their wild relatives. However, in the absence of these multiproxy analyses, cultivar pollen types in conjunction with evidence of forest clearance and/or disturbance, along with charcoal evidence for anthropogenic fires, is strong evidence of local human activities. Among many research topics, this improved technique for recovering cultigen pollen will allow palaeoecologists and archaeologists alike to maximize their chances of detecting early food-production and assess past human impact on the Neotropics, even in regions where archaeology and archaeobotany are at a very early stage of development. Similarly, it will prove crucial to documenting the practice of early slash-and-burn agriculture (e.g., Piperno et al., 1981), which unlike irrigation canals, raised fields, and agricultural terraces, does not leave visible imprints on landscapes. Lastly, it will help better understand arguably the most dramatic changes in land-use practices since the Pleistocene-Holocene transition brought by the 1492 Columbian Encounter (Turner and Butzer, 1992).

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References


