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Using Isotopic Evidence to Assess the Impact of Migration and the Two-Layer Hypothesis in Prehistoric Northeast Thailand

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KEY WORDS Ban Non Wat; strontium; carbon; Mun river valley; agriculture

ABSTRACT *OBJECTIVES:* The nature of the agricultural transition in Southeast Asia has been a topic of some debate for archaeologists over the past decades. A prominent model, known as the two-layer hypothesis, states that indigenous hunter-gatherers were subsumed by the expansion of exotic Neolithic farmers into the area around 2000 BC. These farmers had ultimate origins in East Asia and brought rice and millet agriculture. Ban Non Wat is one of the few archaeological sites in Southeast Asia where this model can potentially be tested. The site is located in the Mun River valley of Northeast Thailand, and divided into 12 phases that span over 2,000 years, from about 1750 BC to the end of the Iron Age (ca. 500 AD). These phases exhibit successive cultural changes, and current interpretation of the site is of an early hunter-gatherer population, with agriculturalists immigrating into the later phases.
METHODS: We analyzed strontium, oxygen, and carbon isotopes in tooth enamel from over 150 individuals, dating from the Neolithic to Iron Age, to assess extrinsic origins and differences in diet between early and later phases.
RESULTS: We find evidence of dietary and cultural differences between groups at Ban Non Wat during its early occupation, but little evidence for immigration from distinct environments beyond the Khorat Plateau of Northeast Thailand.
CONCLUSIONS: The lack of consistent isotopic differences between early and later Neolithic occupants at Ban Non Wat means that the site does not conclusively support the two-layer hypothesis. Am J Phys Anthropol 158:141–150, 2015.

INTRODUCTION

The prehistoric cultural sequence on the Khorat Plateau of Northeast Thailand from the initial settlement by rice farmers (1750 BC) to the end of the Iron Age (500 AD) was characterised by social change (Higham and Higham, 2009). At the site of Ban Non Wat, a sharp rise in mortuary wealth with the initial Bronze Age (from 1000 BC) was followed by a decline while burials in other sites of the region indicate that a further rise in social aggrandizement took place during the later Iron Age (O'Reilly, 2008). During the Iron Age, large settlements were the foci for agricultural intensification involving water control, ploughing in fixed fields and a massive increase in rice production, which underwrote the swift rise of early states (O'Reilly, 2008; Higham, 2014). In Cambodia and Northeast Thailand, this culminated in the kingdom of Angkor from the early 9th century AD (Higham, 2002).

Early archaeological research in Southeast Asia was conducted from the perspective that indigenous development of complex societies was highly unlikely, and that any cultural innovation was likely to stem from migration into the area or contact with more advanced peoples (Majumdar, 1944; Cœdès, 1969; Bayard, 1980). Though much has been done to redress this imbalance and recognize the agency of indigenous peoples (Hutterer, 1982; Stark and Allen, 1998; White and Hamilton, 2009; Cox et al., 2011), migration remains an important causative factor in several major societal changes. In Southeast Asia an example of this is the recently developed two-layer hypothesis which states that indigenous hunter-gatherers were replaced or subsumed by migrant agriculturalists who had their origins in East Asia, most probably the Yangtze river valley of China (Bellwood, 1993; Higham, 2013; Reid, 2013; Matsumura and Oxenham, 2014). Dates for this expansion are debated, but

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current genetic and morphological work on rice and millet (Fuller et al., 2010; Castillo, 2011), as well as archaeological evidence for changes in material culture (Rispoli, 2007) suggest rice appears in the lower reaches of major river valleys in mainland Southeast Asia by 2000 BC, with movement into the interior taking place gradually over the next 500 years (Zhang and Hung, 2010; Higham, 2013).

Extensive osteological analyses at the site of Man Bac in northern Vietnam have identified the contemporaneity of indigenous hunter gatherers and intrusive rice farmers (Matsumura et al., 2008a; Dodo, 2010; Oxenham et al., 2011; Matsumura and Oxenham, 2014). However, there has been little opportunity to test this "Two Layer" model elsewhere in Southeast Asia due to the paucity of sites with occupation layers spanning the Neolithic transition. Ban Non Wat in Northeast Thailand is potentially one of the few such sites. Here a phase of flexed burials, some of which are associated with unusual items of material culture, is interspersed with extended burials typical of early agriculturalists. This has been interpreted as evidence for early hunter-gatherer interaction with an incoming agricultural population (Higham, 2013).

Ban Non Wat, therefore, potentially offers a rare opportunity to test the model of agriculturalist migration into the Southeast Asian mainland using one of the most direct lines of evidence possible - isotopic analysis of the human remains. Strontium isotope analysis is well established as an indicator of past migration (Bentley, 2006; Schroeder et al., 2009; Montgomery, 2010; Price et al., 2015), and has been successfully employed in previous studies of Southeast Asian sites, albeit not covering the Neolithic transition, at Ban Lum Khao (Bentley et al., 2009) and Noen U-Loke (Cox et al., 2011) in the Upper Mun River valley. These studies have found little definitive evidence for long-distance immigration into the area during its prehistoric phase of increasing social complexity. Only two individuals analyzed from Ban Lum Khao (n = 27), and none from Noen-U-Loke (n = 34) could be confidently identified as migrants (Bentley et al., 2009; Cox et al., 2011).

In using strontium isotope analysis to test the two-layer hypothesis we recognise that it is extremely unlikely that agriculturalists moved directly from their area of origin in China to Ban Non Wat in the space of a generation, instead migration is likely to have been short distance with successive generations moving into new areas gradually (Pinhasi et al., 2005; Fuller et al., 2010). Short-distance migration, however, may still be visible isotopically if the areas are geologically distinct, and it is also worth noting that while long-distance migration is less likely it is not impossible. There is increasing recognition of "Lévy flight" movement in human groups, where most movement is short-distance but also involves infrequent dramatic distance jumps (Brantingham, 2006; Brown et al., 2007; Raposo et al., 2009).

When over-simplified as a single generation of "founders" followed by generations of locally-born residents, the two-layer hypothesis would superficially suggest that only the first-generation of agriculturalists would be likely to be isotopic immigrants. The classic demic diffusion model (Ammerman and Cavalli-Sforza, 1971), however, predicts that people will continue to move between generations even after the initial colonisation wave has passed (Steele, 2009). The two-layer hypothesis therefore more properly predicts that earlier agricultural levels of the site will display higher mobility than later levels due to on-going influx of migrant peoples.

Carbon isotope evidence from Ban Non Wat may also help evaluate the two-layer hypothesis. If flexed burials are truly representative of hunter-gatherers and extended burials of agriculturalists then dietary differences between the two groups are likely. Carbon isotope analysis allows differentiation of dietary components, primarily relating to the photosynthetic pathway of plant resources consumed either directly by humans or by the animals they feed on (Schoeninger and DeNiro, 1984; Lee-Thorp et al., 1989). In the Upper Mun River Valley carbon isotope analysis has been previously employed at the agricultural sites of Ban Lum Khao (Bentley et al., 2009) and Noen U Loke (Cox et al., 2011) to reveal a relatively uniform carbon isotope signature associated with the populations dependant upon rice agriculture ($\delta^{13}C = {}^{-}14.5$–${}^{-}13.2\%$). The hunter-gatherer diet, on the other hand is likely to incorporate higher levels of C_4 plants or hunted meat/game, resulting in a shift to higher $\delta^{13}C$.

This study continues one of the largest isotopic projects to be undertaken in Southeast Asia, and Ban Non Wat has been extensively excavated to address problems of unintentional sampling bias. The sample of about 700 human burials is deemed large enough to represent a local population over two millennia (Higham and Kijngam, 2009), and isotopic information from a large subsample of this cemetery has now been obtained (King et al., 2013, 2014). In this article, we expand upon the existing isotopic dataset with new strontium isotope data, evaluate the importance of migrants in prehistory, and the possibility of an early hunter-gather population potentially validating the two-layer hypothesis.

BAN NON WAT: ARCHAEOLOGICAL CONTEXT

Site location and cultural sequence

Ban Non Wat is located in the Upper Mun River Valley, Nakhon Ratchasima province, Northeast Thailand (latitude 15°.16.02' longitude 102°.16.35'), shown on Figure 1. This area is recognized as an important archaeological landscape comprising multiple sites situated along paleochannels associated with the Mun River (McGrath et al., 2008; Boyd and Chang, 2010). Ban Non Wat was initially occupied about 1750 BC, and prehistoric occupation lasted until the end of the Iron Age, a span of about 2300 years (Higham and Higham, 2009), during which dramatic changes in social organisation and economic strategies can be observed (Boyd and Chang, 2010; Higham and Rispoli, 2014; O'Reilly, 2014).

The cultural sequence of Ban Non Wat begins with a phase of flexed burials interspersed with two Neolithic mortuary phases in which the dead were interred in an extended, supine position (Higham and Kijngam, 2011). The Neolithic cemeteries are overlain by six Bronze Age burial phases (beginning ca. 1050 BC). During the first two of these, there was a surge in mortuary wealth, including very rich infant burials (Higham and Kijngam, 2012a). The final three Bronze Age phases saw a sharp decline in the range and quantity of mortuary offerings. Iron Age occupation is also evident at Ban Non Wat (beginning ca. 420 BC). Three Iron Age mortuary phases have been identified (Higham and Kijngam, 2012b), though this article uses material from only the first two,

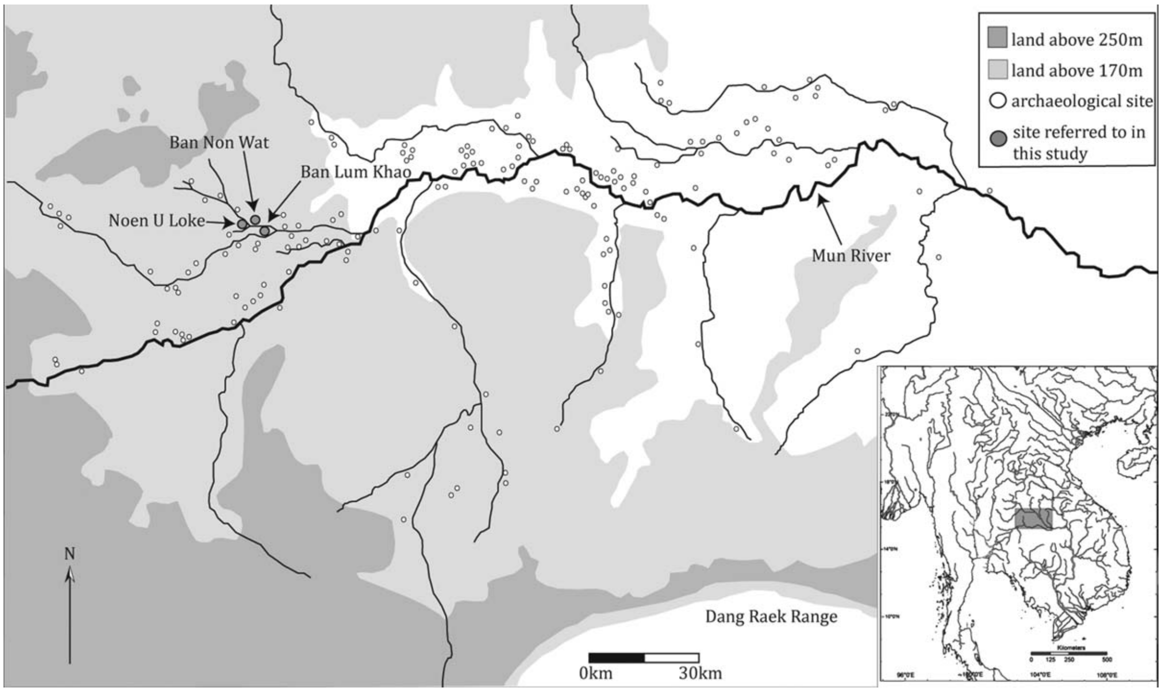

Fig. 1. The location of Ban Non Wat, shown in context with other archaeological sites of the Khorat Plateau.

due to small sample sizes in the later Iron Age. Mortuary phases are differentiated on the basis of stratigraphy, change in burial practice, orientation, and material culture (Higham and Kijngam, 2009).

Flexed cf. extended burial position in the Neolithic

Adult flexed burials at Ban Non Wat do not exhibit a uniform burial position. Some are lying entirely on their left or right side with upper and lower limbs flexed to some extent, while others have the upper body supine and the hips and knees flexed. There is no pattern to the placement of the upper limbs. Though most individuals within the phase are flexed in some way, there is one sub-adult classified as a flexed burial on the basis of burial depth, but laid supine in the grave. Adult burials from the other mortuary phases at the site are uniformly interred in an extended position, with the exceptions of two early Neolithic individuals who were interred seated and flexed within lidded mortuary vessels.

Most of the flexed burials are not associated with any mortuary offerings. Artifacts are found with only three burials; this material culture includes distinctive marine shell beads not found in later Neolithic contexts (*Tridacna* and *Anadara*) and fragments of a cord-marked pot and a globular pottery vessel dissimilar to ceramics defined as Neolithic. Other artifacts such as bivalve shells, an unshouldered stone adze-head, shell disc beads, and a pig skull are not unique to the flexed burials and are also found in those categorized as Neolithic 1 (Higham and Wiriyaromp, 2011). Ceramic vessels are the most common mortuary offering in the first Neolithic phase (Neolithic 1), and typical forms are described in detail elsewhere (Higham and Kijngam, 2011). Pig and fish bones, and adzes are also present, with shell jewellery quite rare. These fundamental differences in mortuary treatment are illustrated in Figure 2.

Interment in a flexed position is typical of hunter-gatherer burials across much of mainland Southeast Asia (Pookajorn, 1994; Shoocondej, 2004; Treerayapiwat, 2005; Oxenham et al., 2011), from Mesolithic cave sites through to the more recent Da Bhut culture of Vietnam (about 4500–2000 BC). Though it is most common for hunter-gatherers to be interred in a tightly flexed squatting position, as in Da Bhut (Oxenham et al., 2011), there is considerable variation in the burial position of hunter-gatherers (Zeitoun et al., 2013; Tayles et al., 2015). This variation includes examples of the burial position at Ban Non Wat, where the upper body is supine and lower limbs are flexed, for instance at Hang Cho in Vietnam (Matsumura et al., 2008b) and the Thai sites of Ban Rai (Pureepatpong, 2006) and Sai Yok (Jacob, 1969). Conversely the burials of Neolithic agriculturalists are almost universally found in an extended, supine position (Oxenham et al., 2011; Higham, 2013), with flexed burials such as that of Bronze Age Ban Chiang burial BCES B76 (Pietrusewsky and Douglas, 2002) being exceedingly rare.

The similarity in burial style, in conjunction with the distinctive mortuary offerings of the flexed burials, has led to the interpretation that these skeletons represent indigenous hunter-gatherers at Ban Non Wat, with extended burials being the first agriculturalists. Dating of the flexed burials indicates at least some of them overlap temporally with the extended Neolithic phases, indicating possible interaction between hunter-gatherer and agricultural groups (Higham, 2013).

GEOLOGICAL CONTEXT

This study uses strontium isotope ratios ($^{87}Sr/^{86}Sr$) in human skeletal tissues to establish prehistoric human mobility. These ratios are fundamentally derived from the underlying geology of an area, which contributes strontium to local soils, foods grown upon them, and

Fig. 2. Illustrating typical flexed (left = B454) and supine (right = B86) Neolithic burials. Scale bar = 20 cm.

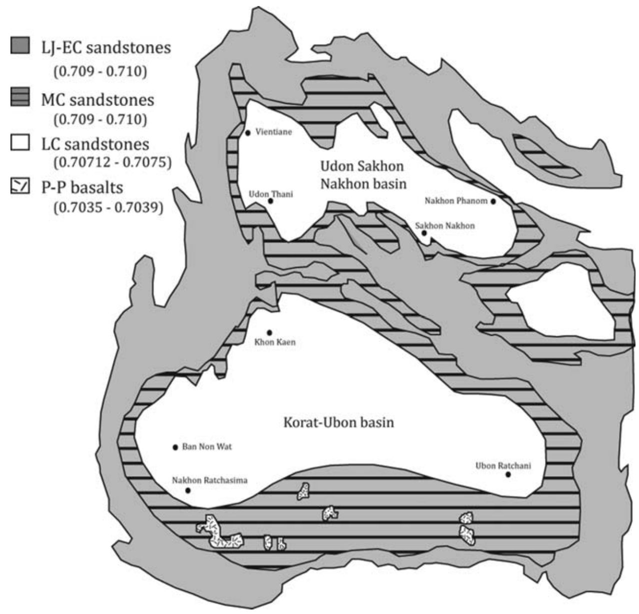

Fig. 3. Ban Non Wat and its geological context. LJ = Late Jurassic, EC = Early Cretaceous, MC = Middle Cretaceous, L-C = Late Cretaceous, P-P = Pliocene–Pleistocene (figure after Charusiri, et al., 2006). Figures given in legend indicate expected ^{87}Sr/^{86}Sr for different geological formations.

eventually the human skeleton during tissue mineralisation (Bentley, 2006; Knudson and Stojanowski, 2008; Montgomery, 2010). The Khorat Plateau is comprised of a number of sandstone and shale formations ranging in age from the Jurassic through to the late Cretaceous (Fig. 3). These include red arkose sandstones, which typically have ^{87}Sr/^{86}Sr ratios greater than 0.710 (Charusiri et al., 2006), evaporites with ^{87}Sr/^{86}Sr ratios reflecting Cretaceous seawater, from 0.707 to almost 0.708 (Timofeeff et al., 2006), and basaltic outcrops with ^{87}Sr/^{86}Sr ratios between 0.703 and 0.704 (Zhou and Mukasa, 1997). The Upper Mun River Valley will therefore have variable biologically available strontium depending on which of the rock types present are contributing most to the strontium found in soils and water sources. Studies conducted at nearby Noen U Loke and Ban Lum Khao have yielded a conservative local ^{87}Sr/^{86}Sr ratio range between 0.7096 to 0.710 (Bentley et al., 2009; Cox et al., 2011).

MATERIALS AND METHODS

Background to isotopic analysis

Geographic origins of individuals at Ban Non Wat were assessed using strontium (^{87}Sr/^{86}Sr) and oxygen (δ^{18}O) isotope analysis of dental enamel. Strontium isotopic ratios vary according to geologic substrate, so in theory, if an individual spent a childhood in an area with broadly different geology than the place of burial, then the isotopic signature of the place of origin in the dental enamel will be visible as an isotopic outlier. Various mixing and mobility scenarios, however, as well as non-geological sources of strontium, can cloud interpretations, and of course areas of origin with similar biologically available strontium will be indistinguishable using this technique (Bentley, 2006; Montgomery, 2010).

Oxygen isotope analysis provides an independent geographic proxy from tooth enamel. The ratio of ^{18}O to ^{16}O, reported as δ^{18}O, reflects the composition of water ingested during the period of enamel mineralisation, with isotopic ratios varying according to altitude, distance from large bodies of water and temperature. There is the potential, therefore, for considerable and significant variation between regions (Dansgaard, 1964; Gat, 1996). Previous isotopic studies in Southeast Asia have not so far shown particularly useful differences in oxygen isotope ratio between geographically distinct populations (Krigbaum, 2003; Bentley et al., 2005, 2007), δ^{18}O data are therefore viewed conservatively, and used mainly to identify outliers that evidence distinct natal origins.

Carbon isotope ratios in human dental enamel are traditionally used to distinguish the broad categories of food consumed, such as C_3 and C_4 plants, or marine versus terrestrial foods (Lee-Thorp, 2008), and have been explored elsewhere in this way (King et al., 2013, 2014). In this study, they are used to add to the picture of prehistoric mobility, as people from different natal groups may have had different dietary habits. For instance, parts of the nearby Khao Wong Prachan valley are geologically similar to the Khorat Plateau and individuals from this region would be indistinguishable on the basis of ^{87}Sr/^{86}Sr ratios. They do differ in terms of food source, however. Millet (a C_4 crop) was used as a staple in the Neolithic of the Khao Wong Prachan valley (Weber et al., 2010), but evidence for this is absent at Ban Non Wat. In this instance carbon isotope differences would indicate migration of an individual.

TABLE 1. Sample composition showing mortuary phase and sex of individuals analyzed

Mortuary phase	Male	Female	Unknown	Total
NF	5	4	0	9
N1	6	3	1	10
N2	5	8	1	14
BA1	1	1	0	2
BA2	8	6	2	16
BA3	11	12	2	25
BA4	25	28	2	55
BA5	5	5	2	12
IA1	4	8	3	15
IA2	1	1	2	4

N = neolithic, F = flexed, BA = bronze age, IA = iron age.

Isotopic sampling

One hundred sixty-two adult individuals were sampled from Ban Non Wat. The demographic composition of the sample is laid out in Table 1. The second molar was sampled preferentially, representing the period of life between 3 and 6 years (Hillson, 1996). Table 1 of the Supporting Information gives a full list of dental samples used. Five domesticated pig (*Sus scrofa*) samples were also analyzed. These were used as a proxy for the local signature, as in Southeast Asia pigs are commonly kept in close proximity to settlements (Griffin, 1998) and fed scraps reflecting the human diet (Bentley et al., 2005).

Isotopic methods

Two small chips (5–10 mg) of enamel were removed from each tooth mechanically using a surgical steel scalpel. Both chips were pretreated to remove diagenetic components by leaching for 4 h in 10% vol acetic acid followed by washing with MilliQ (Koch et al., 1997; Balasse et al., 2002). One chip was then prepared for Sr isotope analysis, the other for C&O analysis.

Strontium isotope analyses were obtained on a ThermoFisher Neptune Multi Collector Inductively Coupled Plasma Mass Spectrometer (MC-ICP-MS) at the Northern Centre for Isotope Analysis and Elemental Tracing, University of Durham. Strontium was purified through established methods of column chemistry involving sample dissolution in 3N HNO_3 and running of solution through Sr Spec resin (Charlier et al., 2006). Sr ratios were normalized using repeated measurements of the NBS 987 standard ($^{87}Sr/^{86}Sr = 0.710240$) in each session of analysis (n = 90). The average values for the standard in each session are reported in Table 1 of the Supporting Information. Total procedural blanks for each batch of chemistry were analyzed alongside tooth samples with none exceeding 75 pg of Sr, and a mean concentration of 29 pg. It should be noted that even the highest concentration blank is insignificant given the very high concentrations of strontium retrieved from the samples.

Carbon and oxygen analyses were obtained in the Stable Isotope Laboratory, Department of Earth Sciences, Durham University. Each analysis used approximately 4.5 mg of crushed sample dissolved in 6 mol l^{-1} phosphoric acid. Carbon dioxide was separated using a GasBench II chromatograph, and isotopic ratios were measured using a using a Thermo Electron MAT253 mass spectrometer, with results normalized to standards NBS 19 and LSVEC, and standardized using repeated measurements of internal standard DCS01. Samples were run in several batches, with the running averages of DCS01 reported in Supporting Information Table 1. We do not use collagen analysis in this study simply because earlier investigations have shown it is not often preserved at Ban Non Wat (King et al., 2011).

RESULTS

Isotopic data obtained from the sample (Supporting Information Table 1) are plotted with $^{87}Sr/^{86}Sr$ versus $\delta^{13}C$ (Fig. 4) and $^{87}Sr/^{86}Sr$ versus $\delta^{18}O$ (Fig. 5). Both of these figures show that the majority of isotopic outliers are from Neolithic phases, though it is apparent that there is little correlation between strontium and oxygen results, confirming the suspicion that oxygen isotope variation in the area is very complex, and of little use in elucidating geographic origins.

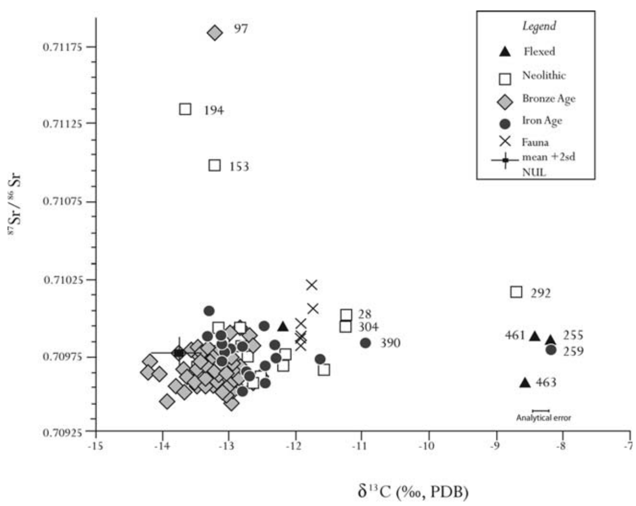

Fig. 4. $^{87}Sr/^{86}Sr$ vs. $\delta^{13}C$ isotopic results. Analytical error is given for $\delta^{13}C$ results, but $^{87}Sr/^{86}Sr$ error lies within the area of the symbols used.

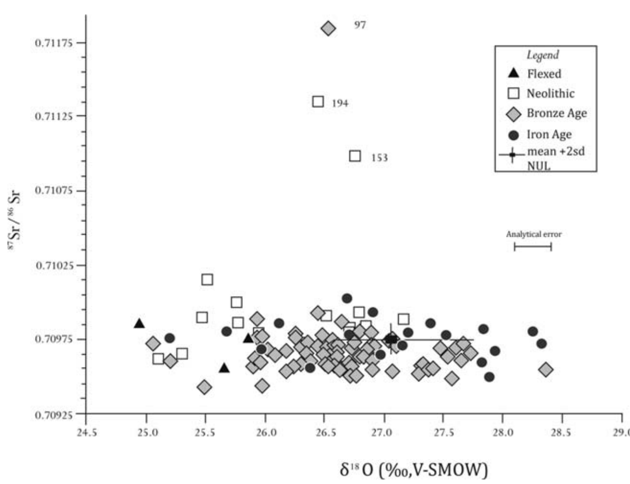

Fig. 5. $^{87}Sr/^{86}Sr$ vs. $\delta^{18}O$. Note that three Sr outliers with ratios above 0.7110 (shown on Fig. 4) are excluded from this plot, and one faunal $\delta^{18}O$ outlier at 22.78 in order to better show the main body of data. Error is given for $\delta^{18}O$ analyses, Sr error is contained within the area of the symbols used.

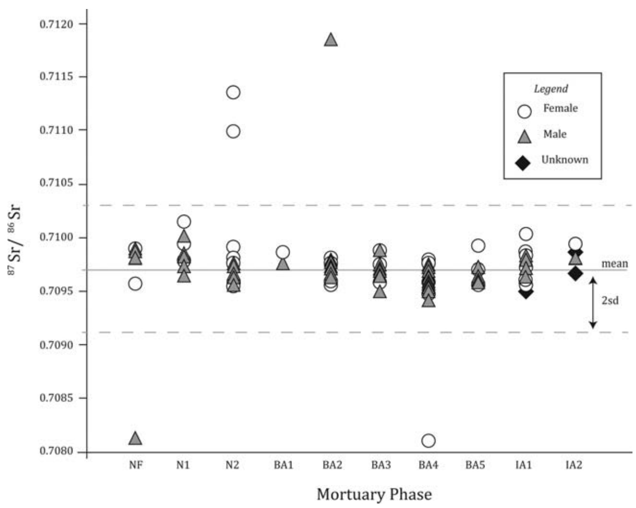

Fig. 6. Showing $^{87}Sr/^{86}Sr$ results with reference to mortuary phase and sex.

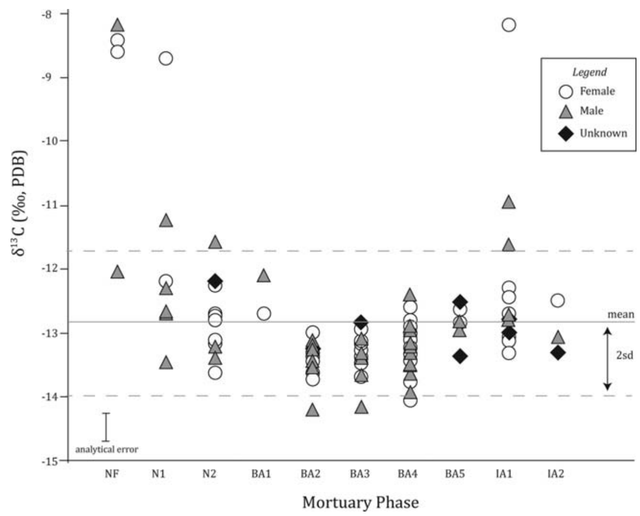

Fig. 7. $\delta^{13}C$ results presented in terms of sex and mortuary phase.

Because certain sampled individuals yielded insufficient material for both strontium and light isotope analysis, those with just one set of data do not appear on all of these figures, but are shown on Figures 5 and 6 which separate the individuals according to mortuary phase and Sr ratio (Fig. 6) and $\delta^{13}C$ (Fig. 7).

The mean strontium isotope ratio for all human individuals at the site is 0.70970 ± 0.0006 (2 SD $n = 145$), and the majority (96%) of individuals fall between 0.7094 and 0.7102. This is therefore considered the more conservative local isotopic range. Faunal samples ($n = 5$) give a mean strontium isotope ratio of 0.70996 ± 0.0003 (2 SD), but the removal of one outlier ($^{87}Sr/^{86}Sr = 0.71021$, $\delta^{18}O = 22.78$) yields a revised mean value of 0.70990 ± 0.00007 (2 SD). The clustering of faunal samples within the main body of human isotopic data supports the idea that this is indeed the local range.

There are five obvious outliers present in the strontium isotope dataset; a young male from the early flexed group of burials (B296, $^{87}Sr/^{86}Sr = 0.70813$), a mid to old female (B153, $^{87}Sr/^{86}Sr = 0.71099$) and a middle-aged female (B194, $^{87}Sr/^{86}Sr = 0.71136$) from Neolithic 2, a middle aged adult male from Bronze Age 2 (B97, $^{87}Sr/^{86}Sr = 0.71189$), and an old female from Bronze Age 4 (B440, $^{87}Sr/^{86}Sr = 0.70810$). These individuals fall well outside the local range defined for Ban Non Wat, and are prime candidates for immigrants from outside the immediate area.

Whereas two of these immigrants (B296 and B440) identified by $^{87}Sr/^{86}Sr$ unfortunately lacked enough material for carbon or oxygen isotopic analysis, the other $^{87}Sr/^{86}Sr$ outliers do not stand out in terms of $\delta^{13}C$. This indicates that despite different origins, their diet is likely to have been similar to that of the local population.

Carbon isotope analysis shows that there are individuals who fall within the "local" Sr bracket who appear to have had very different diets. These are; B461 and B463 (both middle aged females, flexed) B255 (young male, flexed), B292 (young-mid female, Neolithic 1), and B259 (young female, Iron Age 1). In addition to these obviously different individuals, there are four whose isotopic ratios are over 2 standard deviation above the site's mean, these are; B28 (old male, Neolithic 1), B304 (middle aged male, Neolithic 2), B211 and B390 (both approximately middle-aged males, Iron Age 1). The more positive $\delta^{13}C$ of these individuals indicates more marine/C_4 crop/meat input into the diet than is "normal" at Ban Non Wat. Without nitrogen isotope data, which requires collagen that is unfortunately not preserved at Ban Non Wat (King et al., 2011), it is impossible to say definitively which of these sources is responsible for the enrichment.

In terms of mortuary wealth, the outlier individuals are, for the most part, indistinguishable from the local population; neither unusually wealthy nor poor in terms of absolute number of grave offerings, and without unusual items for Ban Non Wat (Supporting Information Table 2, see Higham and Kijngam, 2009 for detail). An exception is burial B461 from the flexed phase and both outliers in Neolithic 1. Burial B97 is one of the poorest individuals in BA2, but this burial was excavated from an area outside the main excavation square, where contemporary burials are equally poor.

B461 was buried with crudely fashioned *Anadara* shell beads, ornaments absent from any other burial at Ban Non Wat (Higham and Kijngam, 2011). The outlier individuals in Neolithic 1, B28 and B292, are extremely unusual as they are the only two adult jar burials (Fig. 8). This kind of interment is the norm for infant burials at this site and elsewhere in the Khorat Plateau, but, aside from the flexed burials at Ban Non Wat, adults are buried in a extended position (Higham and Kijngam, 2011).

There is no significant difference in mean between males and females in $^{87}Sr/^{86}Sr$, $\delta^{13}C$ or $\delta^{18}O$ ($p = 0.992$, 0.585, and 0.423, respectively; two-tailed t-test), or in the variance of these values ($p = 0.146$, 0.266, and 0.977; Levene's test). There is a significant difference in variance among mortuary phases ($p = 0.001$, ANOVA), but this is likely to be a function of difference in sample size, as post hoc testing (Tukey's test) shows it is BA4 and BA1 (our largest and smallest subsamples) which have significantly different variance.

DISCUSSION

Migrants in the Mun River Valley

The results harmonize with previous studies in the area (Bentley et al., 2009; Cox et al., 2011), which have

Fig. 8. B28, jar burial from the Neolithic 1 mortuary phase.

found little evidence for long distance migration. Of the 127 Ban Non Wat individuals analyzed, the majority yielded ^{87}Sr/^{86}Sr ratios within the range defined by local domestic pigs, and only five had ^{87}Sr/^{86}Sr ratios consistent with origins outside the Upper Mun River Valley. So far, the fraction of probable immigrants during the prehistoric precursors to state society is less than 5% at Ban Non Wat, Noen U Loke, and Ban Lum Khao, and over half of the excavated individuals have been analyzed isotopically (Bentley et al., 2009; Cox et al., 2011). It therefore seems unlikely that immigration from outside the URMV was the driver of social change. While migrant individuals may be social innovators and influential members of society despite their low numbers, at Ban Non Wat we see no evidence for this kind of social differentiation. The richest burials belong to local individuals, and the mortuary ritual associated with migrant individuals is in no way unusual, except in the very earliest phases of the site.

^{87}Sr/^{86}Sr ratios are the culmination of a range of factors and so cannot be used to pinpoint geographic origins, but it is notable that there are areas within the Khorat Plateau that would give ratios similar to those seen in our "migrants." Burials 153, 194, and 97, have ^{87}Sr/^{86}Sr ratios near 0.711 and are within the range we would expect to see in the Songkhram Basin (Bentley et al., 2005), north of the UMRV. The two outliers which fall around 0.708 could derive from the evaporite formations, which are present within the sandstones of the region, and stretch across the Khorat Plateau. In other words even the few, definitive immigrants among the Ban Non Wat sample may well be from within the Khorat plateau. This is perhaps unsurprising, as migration is most likely to occur over short distances between habitually interacting social groups (Lewis, 1982; Anthony, 1990; Fix, 2004). This is likely to have been the case in prehistory, and it seems far-fetched to assume that the settlements of the UMRV were socially and reproductively isolated from one another. We find continuity in terms of material culture and mortuary traditions between Ban Non Wat and contemporaneous phases of nearby sites (Higham and Kijngam, 2009), which suggests contact and/or trade. This could also include marriage exchanges.

Are the extended Neolithic burials migrant agriculturalists?

Our results provide little evidence for long-distance migration driving the long-term agricultural transition or corresponding social development. The only isotopically unusual individuals in the extended Neolithic phases are the two adults interred in lidded ceramic vessels, both being carbon isotope (rather than ^{87}Sr/^{86}Sr) outliers. On the face of it, these early agriculturalists appear to have moved short distances, driving demic diffusion over many generations rather than through long-distance migrations within individual lifetimes.

In the Upper Mun River Valley, however, the geology is homogenous enough that migrations of hundreds of kilometres might not be distinguishable in terms of ^{87}Sr/^{86}Sr. This unfortunately means that the majority of the Neolithic phase cannot be distinguished isotopically from either the contemporary putative hunter gatherers or the later Neolithic and Bronze Age phases. Exceptions to this are the two jar burials, whose unusual carbon isotope ratios suggest external origins. The enriched δ^{13}C values of these individuals are consistent with more marine or C_4 crop input into the diet, food resources not found in the Mun River Valley. Adult jar burial is not recorded elsewhere in prehistoric Thailand, although there is one record of a flexed adult individual buried completely covered with large pots at the coastal site of Nong Nor (Higham and Thosarat, 1998). It is possible that the jar burials represent early migrants who retained the mortuary traditions of their natal settlements, setting them apart from their contemporaries in death.

It is also possible, however, that these jar burials could belong to the flexed burial group. Their carbon isotope values align well with many individuals in the flexed phase, and it is possible that jars are simply an extension of the baskets or matting which would have been necessary to keep flexed burials in position during burial (following the ideas of Duday and Guillon, 2006). Both individuals (B28, B292) interred in the jar burials also exhibit dental ablation in the form of removed maxillary lateral incisors and canines, a dental modification shared by members of the Neolithic flexed phase (B438, 461, 626, and 633), but no other individuals at the site. The jar burials were assigned to the Neolithic 1 phase because the ceramic burial vessel of B292 is clearly a form 4 pot, characteristic of the Neolithic 1 phase (Higham and Wiriyaromp, 2011). If, however, the flexed and extended phases represent contemporary groups, they may have shared material culture.

Were the flexed burials hunter-gatherers?

The flexed phase comprises 14 individuals of which nine have been analyzed isotopically. Only one is a strontium isotope outlier, but three of the nine are carbon isotope outliers, meaning that of the analyzed subsample, 44% are isotopically unusual. This is a significantly larger proportion than in any other mortuary phase at Ban Non Wat.

Providing this pattern is not a function of unintentional bias, there are several possible explanations for the isotopic differences between the flexed and other mortuary phases at Ban Non Wat. Higher $\delta^{13}C$ may be due to a number of factors; increased use of marine resources, terrestrial meat or C_4 crops (Schoeninger and DeNiro, 1984; Lee-Thorp, 2008). Increased use of marine resources seems unlikely due to the inland location of Ban Non Wat inland location, but it is possible that these people originated in coastal areas of similar geology (and thus similar strontium isotope ratio), and in due course moved to Ban Non Wat.

If, on the other hand, increased use of terrestrial meat sources/C_4 plants is responsible for the higher carbon isotope ratios then the hypothesis of these people representing an early hunter-gatherer population (Higham, 2013) could be supported. Hunting/gathering behavior would lead to a diet incorporating wild game and naturally occurring C_4 resources, and higher $\delta^{13}C$ than the rice-dominated diet of later agriculturalists.

It is, however, equally possible that the flexed burials are not hunter-gatherers but instead agriculturalists who were raised in a community focused on millet, as their primary crop, shifting to rice agriculture later in prehistory as environmental conditions changed. The consumption of domesticated animals, as opposed to wild game, is also not excluded by these results.

The early use of millet certainly seems to have been the case in the nearby Khao Wong Prachan valley (Weber et al., 2010) Lopburi to the east, and Khao Sam Kaeo on the Thai peninsula (Castillo, 2011). Millet has also been suggested as a food source at Ban Chiang to the north (King and Norr, 2006). Though paleobotanical work has been conducted at Ban Non Wat, and confirms the presence of domesticated rice in later extended phases (Castillo, 2011), there is no occupation material definitely associated with the flexed burials, and so the presence of millet and domesticated animals cannot be evaluated with these isotopic data.

Other evidence for different economic strategies between the flexed and extended burials

The two layer model has been most rigorously tested at the site of Man Bac in northern Vietnam, using an extensive suite of osteological and chemical techniques (Matsumura et al., 2008a; Dodo, 2010). Though isotopic evidence suggests that migration is not common into early levels of Ban Non Wat, and that subsistence differences are not consistent between early and later levels, osteological evidence may highlight genetic differences between populations that are invisible to isotopic analyses.

Osteological evidence from Ban Non Wat, however, has so far failed to identify any differences between the flexed and extended burials that might indicate they had different biological affinities. A study of musculoskeletal activity markers in a subsample from Ban Non Wat highlighted only two unusual individuals, both belonging to the second Neolithic phase (Foster, 2011). Nonspecific indicators of health, such as dental enamel defects in the form of linear enamel hypoplasia, and subperiosteal new bone formation may also relate to differences in subsistence strategy. These variables have been examined but no discernable difference between the flexed and extended groups has been found (Clark et al., 2014). Unfortunately sample sizes from these studies are too small to discount the possibility that osteological differences exist but cannot be discerned.

As previously mentioned, dental ablation is a feature of the flexed group but is not practiced in the extended phases (excluding the jar burials). The removal of certain teeth is obviously not an indication that the two groups are biologically distinct, but may be evidence for different cultural practices. The practice is also in evidence at the Thai site of Khok Phanom Di (Tayles, 1996), a site where domesticated rice is in evidence (Thompson, 1996), but perhaps not cultivated at the site and where marine hunting/gathering was arguably more important (Higham and Thosarat, 2005). Dental ablation is by no means restricted to hunter-gatherer contexts, being also recorded in nearby Noen U-Loke (Nelsen et al., 2001), an Iron Age site, and Phum Snay in Cambodia (Domett et al., 2013), another Iron Age settlement.

CONCLUSION

This isotopic study suggests that long-distance migrants have not played a significant role in the development of society in the Upper Mun River Valley. Short-distance migration cannot be ruled-out, however, due to the geological homogeneity of the surrounding region.

The "two-layer hypothesis" of indigenous hunter-gatherer replacement by agricultural populations, observed at Man Bac in northern Vietnam, cannot be definitively extended to Ban Non Wat on the basis of isotopic work alone. The Neolithic flexed burials include a high proportion of isotopic outliers, but equally 56% of individuals are indistinguishable from the majority of later inhabitants at Ban Non Wat. The two groups are not, therefore, completely discrete in terms of diet. Those differences which are observed isotopically between members of the flexed burials and extended Neolithic rice consumers may be evidence of hunting and gathering behavior, but may equally well reflect early cultivation of millet.

The flexed burials and extended burials are also not significantly distinguished by skeletal activity markers or nonspecific health indicators. In Vietnam the two-layer interpretation is based on craniometric analysis, ancient DNA and dental nonmetric trait recording, these analyses have not yet been undertaken, but would supplement the isotopic evidence at Ban Non Wat.

Perhaps neither the "two-layer" nor the opposing "regional continuity" model is sufficient to explain the complexity of Southeast Asian prehistory. Human biology and behavior is rarely as simple as these models imply, and the introduction of agriculture to specific communities in Southeast Asia may have involved a complex mix of long-distance and short-distance migration, possibly combined with some indigenous innovation. It is also very possible that both gene flow and cultural exchange between hunter-gatherers and agriculturalists occurred, and human biology and culture will both have been affected by external factors such as environmental change. This means it is unlikely that archaeological evidence will fit neatly into one of the two possible models, and may explain why the flexed population at Ban Non Wat cannot be easily distinguished from the extended.

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