

[Supplementary material]

Redefining the timing and circumstances of the chicken's introduction to Europe and north-west Africa

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This section outlines the rationale for the samples selected for dating in this study. For publications relating to the specimens selected, please see Table S1 below.

Materials

Morocco

Evidence for the chicken's introduction and spread through Africa has been reviewed by a number of researchers (e.g. MacDonald 1992; Mwacharo *et al.* 2013; Woldekiros & D'Angela

2016; Peters *et al.* in press). It is generally accepted that whilst the birds were present in the Horn of Africa by the eighth century BC, it took approximately 1000 years for them to become established across the whole continent, especially the northwest. Several finds from Mogador in Morocco are of considerable interest, given their contextually assigned dates of mid-seventh century BC, with other specimens attributed to the first-third centuries AD. For this reason, four were selected for direct dating covering both proposed periods (see Table S1; Table 1; Figure 1).

Turkey

The original report for Korucutepe suggests that one chicken bone was found in Middle Bronze Age deposits, with a further 16 from Late Bronze Age layers (Boessneck & von den Driesch 1975). These finds have been cited as evidence that chickens entered Europe via the Turkish bridge (e.g. West & Zhou 1988) even though Boessneck & von den Driesch (1975) dismissed some of the specimens as intrusive. To test the status of these key specimens, two bones derived from contexts dated stratigraphically to *c.* 1800-1200 BC were selected for dating (see Table S1; Table 1; Figure 1).

Bulgaria

Chicken bones, dated to *c.* 5500-3550 BC by context and artefact association, have been reported for multiple Neolithic to Bronze Age sites (Boev 1993; 1995; 1996; 2004; 2006; 2009a; 2009b). Because so many sites appeared to have early chickens, these key specimens have been used to underpin models of the route/s by which chickens entered Eastern, Central and Western Europe (e.g. Kyselý 2010; Poole 2010). To test the validity of these models, four specimens were selected from the sites of Hotnitsa, Galabovo and Yabalkovo (see Table S1; Table 1; Figure 1).

Greece

Historical, iconographic and zooarchaeological records are in accord that chickens were present in Greece by the fifth century BC but there is less evidence that they were established before the ninth century BC (Homer, for instance, does not mention them but Theognis writing in the sixth century BC does (Richter 1968; Johansson 2012). A few excavations, such as that of Late Bronze Age Tiryns, have reported specimens dated by ceramic association to *c.* 1250-1100 BC (von den Driesch & Boessneck 1990) and whilst the authors exercised caution in interpreting

these, others (e.g. Halstead 2012: 23) cited them as confirmed specimens which have entered general narratives. Therefore, two of the 16 specimens noted for this site were selected for dating (see Table S1; Table 1; Figure 1).

Italy

Roman expansion is known to have encouraged the spread and uptake of chickens in western and northern Europe (e.g. Maltby 1997, 2016; Maltby *et al.* 2018), by which point, they were already established in Italy itself. The earliest chicken bones in Italy have been identified in a tenth/ninth-century BC cremation tomb (De Grossi Mazzorin 2005; De Grossi Mazzorin & Minniti 2019:10; date following the high Latial chronology, see van der Plicht *et al.* 2009; Guidi 2018) at Monte Cucco, Castel Gandolfo, and were recently re-examined by Albarella and Corbino to confirm their species ID (Corbino *et al.* in press). Unfortunately, although these appear to have secure stratigraphy, they were not available for direct dating. A small number have been reported at eighth century BC sites in Bologna and other sites in the Po Valley (De Grossi Mazzorin 2005; Trentacoste 2020). None of these early specimens could be accessed, but samples were acquired from two Etruscan sites: Forcello (Bagnolo San Vito) which produced (among other finds) a partial skeleton dated stratigraphically to the late sixth century BC, and Orvieto which yielded numerous specimens (at least 32) assigned the fifth century BC (George *et al.* 2017; C. Corbino & A. Trentacoste *pers. comm.*) (see Table S1; Table 1; Figure 1). From the mid-first millennium BC chicken remains become increasingly common in the Italian zooarchaeological record, including non-funerary contexts, and as such specimens from this period were not chosen for dating in the first instance (De Grossi Mazzorin 2005; Trentacoste 2014, 2020).

France

The presence of chickens from *c.* 600 BC (and particularly 500–400 BC) in France is widely accepted but the security of their date of introduction is unclear (Garcia-Petit 2002; Lignereux & Obermaier 2012; Seigle 2016; Peters *et al.* in press). Whilst there are several specimens identified as sixth century BC in France, at present none have been directly dated (Seigle n.d.). Chickens have been claimed in Late Bronze Age contexts at Boulancourt (Bălăsescu *et al.* 2008.) and their occurrence in sixth century BC assemblages from Marseille has also been noted (Seigle

2016, n.d). As representatives of the most northerly and southerly reaches of France, specimens from both of these sites have been selected (see Table S1; Table 1; Figure 1).

England

Contrary to popular belief, chickens were not a Roman introduction but rather appear to have been present in low numbers from the Early/Middle Iron Age (Maltby 1997; Kitch 2006; Hambleton 2008; Strid 2015). It has been argued that the earliest populations had special status and were not eaten, as their remains were often deposited as un-butchered articulated skeletons (Poole 2010; Sykes 2012). Three of these apparently early articulated specimens (Winklebury, Weston Down, and Houghton Down) were dated to assess their status. On re-examination during sample extraction, the metrics and morphology indicated that the Winklebury ABGs (associated bone groups) may be less discrete than initially thought and instead represent more than one individual. A further isolated specimen, from the Stonehenge Road improvement Scheme, was also selected (see Table S1; Table 1; Figure 1).

Scotland (including mainland and the Scottish Islands)

The northward dispersal of chickens to Scotland is known to have been delayed relative to their spread in southern Britain (Serjeantson 2013; Best 2014; Best & Mulville 2014). It has generally been accepted that they arrived in small numbers during the last few centuries of the Iron Age which spans c. 800 BC–AD 800. However, these early chickens come from stratigraphically complex sites. For mainland Scotland, a proposed Iron Age specimen was selected from Covesea Cave 2. In the Scottish Islands, the earliest possible chicken bones come from Orkney in the later Middle Scottish Iron Age (AD 200–400), with a small number reported at Late Scottish Iron Age sites (AD 400–800). A Middle and a Late Iron Age specimen were selected from the site of Howe (see Table S1; Table 1; Figure 1). Several of the proposed chicken finds from the Iron Age levels of this site were reidentified as red grouse.

Analytical methodologies

This section details the analytical methodologies employed in the study.

Radiocarbon dating

Twenty samples were submitted for analysis at the Oxford Radiocarbon Accelerator Unit, two were dated at Kiel AMS, and one at Beta Analytic. All samples produced results, which have been calibrated using OxCal 4.4.2 (Bronk Ramsey 2009). The IntCal20 calibration (Reimer *et al.* 2020) curve was used for all samples except: 4, 18 and 19. These three, being post-bomb, were calibrated using the Bomb13NH1 curve (Hua *et al.* 2013). A radiocarbon age was not available for recalibration of sample 22, and as such lab dates are quoted. The samples dated at ORAU were processed using the gelatinisation and ultrafiltration protocols described by Brock *et al.* (2010) and Bronk Ramsey *et al.* (2004a). They were then combusted, graphitised and dated by Accelerator Mass Spectrometry (AMS) as described by Brock *et al.*, (2010), Dee and Bronk Ramsey (2000), and Bronk Ramsey *et al.* (2004b). ORAU maintains a continual programme of quality assurance procedures, in addition to participation in international inter-comparisons (Scott *et al.* 2010), which indicate no laboratory offsets and demonstrate the validity of the precision quoted.

Zooarchaeological analysis

Samples 1–15 were identified to species using the Bournemouth University reference collection and recorded following the protocols outlined by Cohen and Serjeantson (1996). The methods outlined in MacDonald (1992) and Tomek and Bochenski (2009) were used to aid species level identification, and to exclude other galliform species. Where possible bones were measured to an accuracy of 0.01mm. Ageing was assigned following Thomas *et al.* (2014) and all evidence of butchery type and location, rodent and carnivore gnawing, burning, root etching, weathering, and other modifications was recorded. Where possible ABGs (associated bone groups) were targeted since these are less likely to have become stratigraphically displaced than isolated remains and provide more data on the individual bird. Where selected specimens were part of an ABG all other remains were also recorded following these conventions. Medullary bone, an endosteal layer of bone which serves as a rapidly mobile calcium reservoir during egg laying, is a reliable indicator of female sex and was recorded by macroscopic analysis. Spurs, spur scars, and spur shields were recorded and considered a probable, but not definite, indicator of male sex.

Genetic analysis

A programme of genetic analyses was also run, both for sample specific data, but to also confirm species identification of one specimen that was very large and could not be identified morphologically (4: Hotnitsa). Consequently, prior to dating, DNA analysis was conducted to confirm that this specimen was not a large wild galliform. The surface of each sample was removed via surface sanding and bone powder was obtained using a mikrodismembrator (Sartorius). 0.05 g of bone powder was then incubated overnight at 50°C with 1mL of extraction buffer (0.5 M EDTA at pH 8.0, 0.5% SDS and 0.5 mg/mL proteinase K) in a 1.5mL tube. DNA was extracted using a QIAquick purification kit™ according to manufacturer's instructions. Precautions to avoid contamination were taken during every stage of aDNA extraction and PCR set up, which took place in a separate laboratory dedicated to ancient DNA research free from contemporary DNA or PCR product. No laboratory materials or clothing were transferred from the post amplification rooms to the ancient laboratory. All work surfaces and equipment were thoroughly cleaned with 10% bleach (sodium hypochlorite) followed by 70% ethanol. Surfaces, equipment, and solutions were also routinely exposed to UV light for at least 10 minutes. All extractions and PCR setup was carried out in class II PCR hoods. Negative extraction and PCR controls (1 sample in every 5) were included to detect potential contamination in reagents and cross contamination between samples. Fifty per cent of samples were replicated by extracting twice from independent samples of the same bone followed by PCR amplification and DNA sequencing.

Isotope analysis

The $^{12}\text{C}/^{13}\text{C}$ ($\delta^{13}\text{C}$) and $^{14}\text{N}/^{15}\text{N}$ ($\delta^{15}\text{N}$) isotope values presented in this paper were analysed alongside the radiocarbon analysis, in the laboratories detailed in Table 2 following their collagen extraction protocols. In general, two $\delta^{13}\text{C}$ values are measured for ^{14}C analysis: the Accelerator Mass Spectrometer (AMS) value, used to correct for isotopic fractionation of the ^{14}C value, and the Isotope Ratio Mass Spectrometer (IRMS) value which is representative of the $\delta^{13}\text{C}$ of the sample, and the point at which $\delta^{15}\text{N}$ values are also reported. It is the IRMS values that are investigated as dietary indicators in this paper. Carbon and nitrogen isotope values ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) are reported per mil. (‰) relative to VPDB and AIR, respectively. Samples CKN1 to CKN21 produced C:N ratios between 3.2-3.4, indicative of well-preserved collagen (DeNiro 1985; Ambrose 1990; van Klinken 1999). C:N ratios were not generated for samples CKN22 and

CKN23 due to the graphitisation process during AMS analysis at the Leibniz Lab for Radiometric Dating (KIA). Due to the isotopic fractionation resulting from this process, these samples were omitted from the stable isotope analysis (see Figure 4).

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Table S1. Specimens selected presented by country and proposed date, with publication, context and zooarchaeological information, where available.

Sample no	Archaeological site	Country	Proposed date	Refs (if any)	Context information	Zooarchaeological Info
CKN1	Yabalkovo	Bulgaria	4500 BC	Boev (2009b)	Settlement site	Isolated adult humerus
CKN2	Galabovo	Bulgaria	3550 BC	Boev (2004)	Settlement site	Isolated adult femur
CKN3	Galabovo	Bulgaria	3550 BC	Boev (2004)	Settlement site	Isolated adult femur. Female (medullary bone). Slightly greasy appearance.
CKN4	Hotnitsa	Bulgaria	5500 BC	Boev (2009a)	Settlement site	Isolated adult tibiotarsus. Very large. Cuts and carnivore gnawing on distal end.
CKN5	Forcello (Bagnolo San Vito)	Italy	530–520 BC	Trentacoste (2014)	Use level of outdoor artisan working area (Context 1118: Phase H3)	Femur from a juvenile articulated skeleton.
CKN6	Orvieto	Italy	500–400 BC	George <i>et al.</i> (2017)	Fill of disused quarry	Isolated adult femur. Female (medullary bone)
CKN7	WA50157: A303 Stonehenge	England	800–100 BC	Grimm (2008)	Pit deposit (Context: 530)	Isolated adult tarsometatarsus. Pathological & eroded.
CKN8	Weston Down	England	400–100 BC	Gibson & Knight (2007)	Pit deposit (5360)	Femur from articulated skeleton. Female (medullary bone). Pathological.
CKN9	Houghton Down	England	470–360 BC	Hamilton (2000)	Pit deposit (340). Layer 6	Tibiotarsus from articulated skeleton. Spur (probably male)
CKN10	Winklebury	England	800–100 BC	Jones (1997)	Hillfort. Context: 987. Feature: 986.	Tibiotarsus from supposedly articulated skeleton. Female (medullary bone). Large.
CKN11	Howe, Orkney, Scotland	Scotland	AD 0–400	Bramwell (1994)	Rubble layer in settlement: 3337	Isolated adult femur. Large. Cut. Possible insect modification.
CKN12	Howe, Orkney, Scotland	Scotland	AD 400–800	Bramwell (1994)	Rubble layer in settlement	Isolated adult tibiotarsus. Greasy surface appearance.
CKN13	Boulancourt Le Chatelet	France	920–800 BC	Bălăşescu <i>et al.</i> (2008)	Internal ditch of fortified hill settlement	Isolated adult femur. Female (medullary bone). Root etched and abraded.

CKN14	Marseille	France	580–560 BC	M. Seigle (<i>pers. comm.</i>)	House of the Greek colony of Massalia	Isolated adult ulna with mild root etching.
CKN15	Covesea Cave 2, Moray	Scotland	800 BC–AD 800	Büster & Armit (2016)	Cave layer (Context 248)	Isolated adult tarsometatarsus. Spur (probably male)
CKN16	Korucutepe/Elazig	Turkey	1400–1200 BC	Boessneck & von den Driesch (1975)	Settlement mound	N/A
CKN 17	Korucutepe/Elazig	Turkey	1800–1600 BC	Boessneck & von den Driesch (1975)	Settlement mound	N/A
CKN18	Mogador	Morocco	650 BC	Becker <i>et al.</i> (2013)	Settlement refuse	Isolated carpometacarpus. (RB586 in Loog <i>et al.</i> 2017)
CKN19	Mogador	Morocco	AD 0–300	Becker <i>et al.</i> (2013)	Settlement refuse	Isolated carpometacarpus. (RB579 in Loog <i>et al.</i> 2017)
CKN20	Mogador	Morocco	700–400 BC	Becker <i>et al.</i> (2013)	Settlement refuse	Isolated adult coracoid
CKN21	Mogador	Morocco	700–400 BC	Becker <i>et al.</i> (2013)	Settlement refuse	Isolated adult tibiotarsus (thought to be RB582 in Loog <i>et al.</i> 2017)
CKN22	Tiryns	Greece	1250–1100 BC	N/A	Settlement mound	N/A
CKN23	Tiryns	Greece	1250–1100 BC	N/A	Settlement mound	N/A