Chronology of the Yayoi skeletal remains from the Kanto district, Japan: a preliminary re-evaluation by radiocarbon dating of postcranial material

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Abstract Radiocarbon ages were determined for 25 samples of human skeletal remains previously assigned or attributable to the Yayoi period (tentatively considered ca. 500 BC to 300 AD) of the Kanto district, Japan. Suzuki (1969) and others, predominantly on cranial morphology, recognized that the Kanto Yayoi series contains three morphological groups: the native Jomon, transitional, and Kofun types. Morphological variation of the Kanto Yayoi people was considered to be chronologically based and formed the basis of Suzuki’s transformation hypothesis of the origin of the Japanese, which assumed limited influence, in eastern Japan, from continental immigrants. The 14C dates of the present study, determined for fragmentary postcranial elements of the same general assemblage, showed that these ‘Yayoi’ series, in part, contain skeletal remains of different ages, including those attributable to the Jomon and Kofun periods. All specimens from the Sano site that were examined were found to most likely derive from the Jomon period, while the dated Ourayama and Iwatsubo remains are best interpreted to be of Yayoi age. The Awajinsha and Bishamon samples were found to be of mixed chronological compositions. The implications of these results are discussed.

Key words: collagen, AMS, Japanese, Yayoi, population history

Introduction The aeneolithic Yayoi period (here tentatively considered to be ca. 500 BC to 300 AD, see below) is crucial in the reconstruction of Japanese population history. This is because a drastic change in skeletal morphology is thought to have occurred in or during the Yayoi period, after a longer Jomon time period (ca. 12000 to 500 BC) with relatively little morphological change.

Since the late 19th century, a number of hypotheses have been proposed regarding Japanese population history. The earlier hypotheses can be categorized into three groups: the ‘replacement theory’ (chikan-setsu, e.g. Morse, 1879; Tsuboi, 1887; Koganei, 1893), the ‘hybridization theory’ or ‘metization theory’ (konketsu-setsu, e.g. Kiyono, 1938, 1946, 1949), and the ‘transformation theory’ (henkei-setsu, e.g. Hasebe, 1940, 1951). Although these theories differed in the supposed impact of immigrants on Japanese population history, the lack of Yayoi skeletons precluded realistic discussions of immigration at that time. Subsequently the recovery of large numbers of Yayoi skeletons from the Yamaguchi prefecture and the northern Kyushu district (e.g. Ushijima, 1954; Zaitsu, 1956; Kanaseki et al., 1960), morphologically distinct from the Jomon people, led to the recognition of substantial immigrant-based population lineages in western Japan, presumably deriving from the Asian continent. This led to the more modern variant of the hybridization theory, the ‘immigration hypothesis’ (torai-setsu) of Kanaseki (1955, 1966, 1971, 1976), and other interpretations that emphasized the genetic contribution of the Yayoi immigrant and descendant populations to the modern Japanese gene pool (e.g. Howells, 1966; Turner, 1976; Brace and Nagai, 1982). However, other researchers have emphasized local morphological continuity and skeletal changes potentially induced by the combined effects of microevolutionary forces and changes in subsistence patterns, and adhere to what is known as the ‘microevolution hypothesis’ (shoshinka-setsu). This hypothesis can be considered a version of the transformation theory based on time-successive skull series of eastern Japan, including those of the Jomon, Yayoi, and historic periods (Suzuki, 1963, 1969, 1981).
logical influence apparently stemming from the Yayoi immigrant populations. This influence is considered to be variable in degree and timing, depending on the morphological features under consideration on the one hand, and geographic region on the other (Yamaguchi, 1982, 1985, 1986, 1987; Dodo, 1987; Hanihara, 1987; Mizoguchi, 1988; Dodo and Ishida, 1990; Kaifu, 1992; Nakahashi, 1993; Matsumura, 1995, 1998b; Nakahashi and Iizuka, 1998; Todaka et al., 2003; Okazaki, 2004; Watanabe et al., 2004). A more direct evidence of the genetic influence of continental populations has been provided by molecular studies of both modern humans (Omoto, 1978; Horai et al., 1991; Omoto and Saitou, 1997) and ancient skeletal remains (Oota et al., 1995). A recent synthetic hypothesis, the ‘dual structure model’ (nijukouzou-setsu), was proposed by Hanihara (1991). This model suggests that hybridization between pre-Yayoi local and immigrant Yayoi lineages resulted in the currently observed morphological and genetical clines within the Japanese archipelago, and that genetic admixture is still a continuing phenomenon (also see reviews by Mizoguchi, 1994; Yamaguchi, 1996; Hudson, 1999).

Despite the wide acknowledgment of the contribution of Yayoi immigrant populations to the formation of subsequent Japanese populations, aspects of the transformation hypothesis remain important both from a local perspective and in terms of the entire Japanese archipelago (e.g. Naito, 1981; Dodo, 1987; Hanihara, 1991; Aikens and Akazawa, 1992; Mizoguchi, 1994; Nagaoka, 2003). In particular, Suzuki’s (1969, 1981) transformation hypothesis was based mainly on his systematic study of secular changes in cranial morphology of the eastern Japanese (Kanto district) skeletal series. Suzuki (1969) identified two periods of rapid morphological change in the eastern Japanese cranial series, the Edo to recent and Yayoi to Kofun periods. Because Japanese populations were considered to have been affected minimally by external gene flow in the Edo to recent period, Suzuki interpreted the rapid change he observed in cranial morphology to stem predominantly from environmental factors related to modernization. Although the Kanto district Yayoi samples available to him were limited, he similarly interpreted the cranial variation to represent in situ transformation of the local Jomon population through the Yayoi period into the Kofun condition, mediated by a change of subsistence pattern.

Suzuki’s (1969, 1981) Yayoi series were morphologically divided into three types, based predominantly on cranial and on some limited aspects of the postcranial evidence. The first type consisted of skeletal remains from the Sanoe site, which showed a strong similarity to the Jomon morphology. The second consisted of the Awajinsha (Awai shrine) site sample, which was interpreted as being intermediate between the Jomon and Kofun types. The third type of Yayoi morphology was found to be similar to that of the Kofun and later periods, which was recognized among skeletal remains of the Ourayama and Bishamon sites. Although there was insufficient chronological data available, this variability was interpreted as indicating a morphological transformation from the Jomon to Kofun conditions, in parallel with a subsistence change from hunting–gathering–fishing to rice cultivation and consumption.

Since in the Jomon period, eastern Japan saw greater cultural prosperity, and presumably larger population sizes, than western Japan (Koyama, 1978), cultural transition in the Yayoi period from hunter–gatherer–fisher to a rice-based agricultural society might have been relatively slow and gradual in eastern Japan (Akazawa, 1982; Aikens and Akazawa, 1992). It is thus possible that chronologically intermediate Yayoi people of eastern Japan had intermediate types of cranial morphology depending on their subsistence pattern. However, the same morphological variation might be explained not only by in situ microevolution and environmental factors in a continuous lineage, but also by the genetic influence of immigrants as suggested by Yamaguchi (1982, 1999).

In order to evaluate these possibilities, patterns of morphological variation through time and space in the Yayoi period must be established. However, the chronological context of the Yayoi skeletal series of the Kanto district has not been critically examined. It is therefore not clear whether the different types of Yayoi people identified by Suzuki (1969) actually form a chronological succession, or rather coexisted contemporaneously. The chronology of the Yayoi remains of the Kanto district is also important from an archaeological perspective. This is because a series of Yayoi cave sites showed a continuous archaeological sequence spanning the Jomon to Yayoi periods (Aikens and Akazawa, 1992), and provides important empirical evidence of subsistence pattern transformation from hunting–gathering–fishing to rice agriculture in eastern Japan. The relationship between the tempo and mode of Yayoi subsistence and cultural changes on the one hand, and morphological evolution or change on the other, should be thoroughly investigated.

The purpose of the present study is to review the contextual data of the Kanto district Yayoi skeletal series studied by Suzuki and others, and to provide new chronological perspectives based on direct radiocarbon dating of human skeletal elements. The Yayoi skeletal series considered in the present study are those from the Sanoe, Awajinsha, Bishamon, Ourayama, and Iwatsubo cave sites. Suzuki (1969) considered the former four site samples to represent three types of Yayoi morphology as mentioned above. The Yayoi remains from the Iwatsubo cave site were discovered subsequent to Suzuki’s initial studies, and were considered to represent the Jomon type (e.g. Kaifu, 1992). A total of 28 skeletal elements deriving from as many different individuals of the above five cave sites were directly measured for radiocarbon ($^{14}C$) dating using accelerator mass spectrometry (AMS). From curatorial considerations, we did not sample the diagnostic cranial material themselves. Rather we sampled and dated human postcranial elements of the same general assemblage. The implications of the chronology of these Yayoi skeletal remains are discussed.

Materials and Methods

In this study, samples were taken for $^{14}C$ dating from the skeletal remains of 28 human individuals excavated or collected from five Yayoi sites: Sanoe, Awajinsha, Bishamon, Ourayama, and Iwatsubo cave sites (Figure 1). The Awajinsha and Sanoe sites, and the Bishamon and Ourayama sites,
are coastal erosion caves located on the Boso and Miura peninsulas, respectively, in the Tokyo Bay area. The Iwatsubo cave is located about 100 km inland in the mountainous area (Figure 1). The skeletal remains of the Awajinsha and Ourayama cave sites were for the most part not found as individual burials, but with bony elements scattered largely without regard to anatomical position (Koganei, 1933; Suzuki, 1997). The Bishamon caves material includes both individual burials and skeletal remains without clear elemental associations (Akaboshi, 1953, 1970). The burial context of the Sano series is unknown because they were found and taken by road-workers (Yawata, 1925). The Iwatsubo remains were excavated as individual burials (Imamura, 1984). Aside from the Iwatsubo material, the sampled and dated postcranial elements are not individually associated with the morphologically more diagnostic cranial remains. However, they provide the first radioisotopic age assessments of the supposedly Yayoi human skeletal assemblages of each site. All skeletal remains are housed at the University Museum, the University of Tokyo.

Details of the Yayoi sites

The Sano cave site is located towards the southern end of the Boso peninsula in Tateyama city, Chiba prefecture. The skeletal remains of the Sano cave are some of the earliest discovered human remains of supposedly Yayoi age. They were found at the bottom of a small cliff during road construction in 1925, within landslide deposits caused by earthquake and rain (Yawata, 1925). Yawata inferred that the skeletal remains derived from an erosional cave (numerous in this area) that had collapsed, although no trace of the cave itself was discernible. According to Yawata (1925), three stone beads made of serpentinite and marble and a shell ornament were associated with the skeletal remains. However, the details of association and recovery conditions were not reported. Because similar stone beads were known from Yayoi sites of eastern Japan, and because over ten Yayoi cave sites were known in the southern Boso area, the Sano skeletal remains were assigned to the Yayoi period (Koike and Suzuki, 1955; Suzuki, 1964, 1969). However, the age of this material is ambiguous because of the entire lack of stratigraphy, and the absence of more diagnostic archaeological material such as pottery shards. The skeletal remains of the Sano cave site comprise over 20 individuals and were reported to be similar in morphology such as the glabellonasal root form, Suzuki considered the Sano remains to exhibit a typical Jomon morphology in spite of their assigned Yayoi age (Koike and Suzuki, 1955; Suzuki, 1964, 1969). In the present study, samples were taken from scapulae representing eight individuals.

The Awajinsha cave site is also located in Tateyama city,
Chiba prefecture. The site is presently located about 1 km away from the coast, and it is assumed to be a coastal erosion cave. The site was found in 1932 by well-sinkers, and excavated by Ohba (1933). The Awajinsha cave deposits revealed superimposed stratigraphy and archeological material including a complete pottery and 28 potsherds including both Jomon and Yayoi period shards (Ohba, 1977). The pottery associated with the human skeletal remains was described as “the Yayoi type in a broad sense which is similar to the Haji ware of the successive Kofun period” (translated from Ohba, 1977). In general, the age of the Awajinsha skeletal remains have been assigned to the Late Yayoi period, although this chronological attribution has not gone unchallenged. For example, the pottery referred to above by Ohba (1977) is today more generally considered to represent the Kofun period, which postdates the Yayoi. However, because the Awajinsha skeletal remains exhibit the ‘4I type’ pattern of anterior tooth extraction that was typical of the Latest Jomon people of central Japan (Aichi prefecture region), Harunari (1983, 2002) suggested that the Awajinsha people might have been early Middle Yayoi period migrants from central Japan. Similarly, Ishikawa (1988) speculated that the Awajinsha people might represent migrants from the Aichi area in the late Latest Jomon period because one potsherd found at Awajinsha was similar to those from the Latest Jomon period west of Aichi. The Awajinsha skeletal series were recovered as randomly scattered fragments, including at least 22 individuals according to the number of cranial bone pieces (Koganei, 1933). Suzuki (1969) regarded the Awajinsha materials as showing cranial morphology intermediate between the Jomon and Kofun conditions. In the present study, samples were taken from the first ribs of five individuals.

The site of the Bishamon caves is located at the southern end of the Miura peninsula in Miura city, Kanagawa prefecture. The Bishamon caves consist of four cave sites named A, B, C, and D. Human skeletons attributed to the Yayoi period were recovered from the C and D caves, excavated from 1949 to 1951 (Okamoto and Akaboshi, 1967). Up to four burials or clusters of skeletal remains of the C cave site were considered possibly to belong to the Yayoi period from stratigraphic interpretations, while other scattered human skeletal remains were considered to date mainly from the Kofun and successive periods (Akaboshi, 1953). The single sample from the D cave site was reported to have derived from a Yayoi period layer (Akaboshi, 1953, 1970). The archaeological material, including the Kugahara-type shards, suggested that the main Yayoi occupation of the Bishamon cave sites can be dated to the Late Yayoi period (Okamoto and Akaboshi, 1967; Akaboshi, 1970), although Suzuki (1969) considered the Bishamon crania to belong to the late Middle Yayoi period. Suzuki (1969) considered the morphology of the Bishamon crania to show a strong similarity to that of the Kofun people of the Kanto district. In the present study, humeral shaft portions of eight individuals were sampled; sample BHU-0008 was from the D cave and other remains were from the C cave.

The Ourayama cave is also a coastal erosion cave in Miura city, facing the entrance of the Tokyo Bay. Excavation at Ourayama started in 1949, and the human skeletal remains were recovered in 1962 and 1963 (Yokosuka Archaeological Society, 1997). Seven superimposed layers produced prehistoric and historic materials; the human skeletal remains were found in Layers 5, 6, and 7. The pottery shards from Layers 6 and 7 were assigned to Kugahara, Maenoocho (Late Yayoi), and Miyanodai (Middle Yayoi) types, although Layer 5 contained not only Yayoi pottery but also the Izumi-type Haji wear of the Kofun period (Akaboshi, 1967). The human skeletal remains of the Yayoi layers were considered to contain at least six males, five females, and two individuals of indeterminable sex; they were fragmented extensively prior to burial and found scattered widely in the cave (Suzuki, 1997). From bone damage and cut-mark patterns, Suzuki (1997) inferred that the human bodies had been systematically disarticulated by a method similar to animal slaughter of the Jomon period. Furthermore, from the imbalance of human bone elemental frequency, and the presence of auguring bones in the cave deposits, he suggested that portions of the deceased human body might have been carried into the cave and broken up for religious and witchcraft purposes (Suzuki, 1997). Although the skeletal remains of all individuals were partial and had lost their original associations, the cranial portions exhibit morphological conditions similar to those of the following Kofun period people (Suzuki, 1969). In the present study, samples were taken from five first ribs of the Yayoi layers, two of them (OC0-0002, -0005) deriving from Layer 6 and the others currently considered as undifferentiated Layers 5 to 7.

The Iwatsubo cave is an inland cave located in Tano county, Gunma prefecture, excavated in 1980 and 1982 (Imamura, 1984; Kaifu, 1992). Six Yayoi period skeletons, five adult and one child, were recovered from four burial places. Another infant skeleton attributed to the Yayoi period from a non-burial setting and one Early Jomon burial were reported as well. Stratigraphic superimposition was established, including levels with Early to Late Jomon pottery, and the early Middle Yayoi pottery determined as Nozawa I type (Imamura and Koizumi, 1984). The Iwatsubo skeletons represent the first well-preserved human remains of the Yayoi period from an inland site of the Kanto region. They exhibit cranial and dental features similar to the Jomon condition, but differ from the latter in a relatively high estimated stature and robust long bones (Kaifu, 1992; Matsumura, 1998a). In the present study, samples were taken from the ribs of the Yayoi period Iwatsubo individuals 1, 2, and 3. Rib samples from Iwatsubo individuals 5 and 6 were also taken, but these require further investigation due to preservation state of the samples, and possibility of skeletal element mixing due to the multiple burial situation of individuals 5, 6, and 7. Further sampling of these individuals has been planned and such results will be reported elsewhere.

Sampling method

Samples for radiocarbon measurements (Table 1) were selected considering two factors. The first was to choose portions of bone elements, such as of costae and scapula, which were expected to provide relatively little information for future anatomical studies. The second was to ensure that the same individual is not sampled multiple times. This was accomplished by sampling bone fragments that either over-
lap in skeletal element and position, or clearly represent different individuals based on age, size, and morphological considerations. Sampling numbers were arbitrarily given and do not correspond to individual burials or numbers assigned to crania in previous reports and population history studies (e.g. Koganei, 1933; Suzuki, 1969, 1997). The exception was the three Iwatsubo cave samples that come from individualized burial remains recognized at the time of excavation (Table 1). Pieces of bone were collected using a metal disk saw. Prior to sampling from each bone specimen, materials attached on the surface of the saw were removed by brushing and ultrasonic cleaning.

Radiocarbon dating
Collagen was extracted from bone by using an improved method of Longin (Longin, 1971; Yoneda et al., 2002a). After removing humic and fulvic acids by soaking in 0.1 M NaOH at room temperature, samples were crushed into fine powder with a freezer mill. The inorganic fraction of bone tissue, hydroxypatite, was dissolved with 1.2 M HCl in a cellulose tube. The remaining organic matter was gelatinized in pure water, overnight, at 90°C. Dissolved gelatin and residual deposition were separated by centrifuging and then freeze-dried. In order to investigate aspects of collagen preservation, the lyophilized gelatin, referred to as ‘collagen’ (with quotes) in this paper, was analyzed for carbon and nitrogen content. Typically, 0.25 mg of ‘collagen’ was analyzed by an elemental analyzer (Carlo Erba NA1500™). The CO₂ produced was then automatically fed into an isotope ratio mass spectrometer (Finnigan MAT 252™). δ¹³C values were measured and used to correct for isotopic fractionation and to estimate protein source (Chisholm et al., 1982; Arneborg et al., 1999; Yoneda et al., 2002a). For the ¹⁴C analysis, 2.5 mg of ‘collagen’, containing about 1 mg of carbon, was oxidized to CO₂ within evacuated tubes with copper dioxide at 850°C, and then CO₂ was purified cryogenically in a vacuum system (Tanaka et al., 2000). Some of the samples were treated with an elemental analyzer for production and purification of CO₂ (Yoneda et al., 2004). CO₂ was reduced to graphite with hydrogen and iron powder catalysis for accelerator mass spectrometry (Kitagawa et al., 1993). Pretreatment of graphite and ¹⁴C measurements were conducted at NIES-TERRA (Tanaka et al., 2000). At least two kinds of standard materials were analyzed by AMS contemporaneously with unknown samples to calibrate the ¹⁴C contents. Usually we put new NBS oxalic acid (NIST SRM-4990C) and ANU sucrose (IAEA-C6), which gave an uncertainty of around 40 ¹⁴C years in typical measurements.

Calibration of ¹⁴C dates
In order to compare the ¹⁴C results with the generally accepted Yayoi chronology based on calendar ages of Chinese history, conventional ¹⁴C ages must be transformed into calibrated ¹⁴C ages. This is not only because conventional

Table 1. Elemental composition of the Yayoi human bones from the Kanto district

<table>
<thead>
<tr>
<th>Site</th>
<th>Sample number</th>
<th>Element</th>
<th>C/N</th>
<th>%Col</th>
<th>%C</th>
<th>%N</th>
<th>Excavation record</th>
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</thead>
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</tr>
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<td>ICO-0002</td>
<td>costa</td>
<td>3.1</td>
<td>18.3</td>
<td>43.8</td>
<td>16.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ICO-0003</td>
<td>costa</td>
<td>3.2</td>
<td>14.3</td>
<td>44.2</td>
<td>16.3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Altered C/N ratios show that, due to diagenetic effects, these samples might not preserve sufficient biogenetic signals.

Bishamon-C4 and -C3 are not individual burials but small bone clusters that include elements from multiple individuals.
14C was defined using Libby’s 14C half-life (5568 yr) instead of the more accurate half-life (5730 yr), but also because radiocarbon activity in atmospheric CO2, which was assumed to be a stable point of reference, is known to have substantially fluctuated in the past (Stuiver and Polach, 1977; Stuiver and Braziunas, 1993).

Furthermore, the tissues of omnivorous human beings may contain carbon atoms that originated from both atmospheric/terrestrial and marine carbon reservoirs that are not in isotopic equilibrium. Surface ocean water, for example, is on average about 400 14C years older than the contemporaneous atmosphere because of this ‘marine reservoir effect’ (Stuiver et al., 1986). A calibration curve mixing the atmospheric (INTCAL98; Stuiver et al., 1998a) and marine data sets (MARINE98; Stuiver et al., 1998b) should be used for each individual sample, depending on the estimated percentage of marine carbon in its collagen (Arneborg et al., 1999; Yoneda et al., 2002a; Bayliss et al., 2004).

Arneborg et al. (1999) estimated the percentage of marine food in prehistoric diet using a linear mixing of 14C value to be between −21.0% and −12.5‰, which represent values for pure terrestrial (C4) and pure marine consumers, respectively. Although these end-points were based on the values observed among human populations in northern Europe, a similar estimation can be performed for Japanese populations as well. For example, the Kitakogane population in the early Jomon period, who were marine mammal hunters in Hokkaido, showed an average of −14.1 ± 0.3‰ (n = 10), corresponding to a diet consisting of 82% marine food by this estimation. This is very similar to another estimate of 79 ± 6% derived from apparent 14C age-differences between human and marine mammal and terrestrial mammal ages (Yoneda et al., 2002b). Furthermore, in the case of the Earliest Jomon population of the Tochibara site in inland Nagano, an average 14C of around −19.9 ± 0.3‰, corresponding to a diet of 13% marine food, was consistent with a significant correlation between 14C and δ15N, which suggested that this population exploited not only terrestrial but also other resources with higher isotopic values such as anadromous salmon (Yoneda et al., 2002a). The lowest 14C among the Tochibara population at −20.4‰ suggests that the end-point at −21‰ for the C4 consumer is reasonable for Japanese populations as well. Hence, with an uncertainty of 10% (Arneborg et al., 1999), we think that this estimation based on 14C can apply to the Japanese populations as well.

A possible regional variability of marine reservoir age has to be considered for these samples, but the regional correction value for the marine reservoir effect (ΔR value) at Tokyo Bay is not available. Some studies on pre-bomb shells and modern coral skeletons showed that water-mass originating from the Kuroshio current had small ΔR values (Yoneda et al., 2000; Hideshima et al., 2001). Because the Miura and Boso peninsulas are located adjacent to the Kuroshio current, we applied the null ΔR value to our 14C results for the time being. The validity of the above procedures should be re-examined when better estimates of individual marine food intake and/or ΔR values at Tokyo Bay become available. Calculation was performed by Oxcal v. 3.9 software (Bronk Ramsey, 1995, 2001), which creates calibrated curves by mixing the two different datasets with arbitrary percentages.

Results

The preservation state of bone material is important in assessing the reliability of 14C results. Table 1 shows the percentage of ‘collagen’ in bone (%Col), carbon and nitrogen content in ‘collagen’ (%C and %N), and atomic C/N ratio. The C/N ratio is the most common indicator of diagenetic effects on collagen because collagen has a significantly lower C/N value (3.2) than soil organic matter derived from plant material. Except for those from the Sano site, samples analyzed in this study showed C/N ratios between 3.1 and 3.3, well within the known range of modern and well-preserved ancient collagen between 2.9 and 3.6 (DeNiro, 1985; Ambrose, 1990; van Klinken, 1999). Although it has been suggested that NaOH treatment before decalcification does not sufficiently remove exogenous carbon in the case of heavily contaminated material (Minami and Nakamura, 2000; Minami et al., 2004), we operationally consider results of samples with C/N ratios of lower than 3.6 as potentially valid ages. According to Minami and Nakamura (2000), a sample with an atomic C/N ratio of 3.5 showed little difference in 14C dates before and after additional purification with XAD-2 resin, while this further treatment apparently removed some amount of remaining exogenous contamination in samples with C/N ratios between 3.7 and 4.0. Again, except for the Sano samples, most of the samples had C/N values of 3.1 or 3.2, indicating that exogenous organic matter is unlikely to have affected biogenic signals, including 14C contents, to any significant degree. On the contrary, the diagenetic effect apparently altered the chemistry of ‘collagen’ in three samples (SSC-0005, -0007, and -0008) from the Sano cave site. Values of %Col, %C, and %N in ‘collagen’ also indicate the status of protein in ancient bone (Ambrose, 1980, 1990; van Klinken, 1999). In an analysis of samples from tropical Africa, Ambrose (1990) classified bone with %Col, %C, and %N greater than 3.5%, 13%, and 4.8%, respectively, as well preserved. Van Klinken (1999) confirmed that these elemental indicators were also applicable to 14C dating of bone material from temperate European sites. He also suggested that even bone with %Col as low as 1% could be used for isotopic research because temperate European material showed alteration by contamination rather than degradation of collagen, which was dominant in the African material. Table 1 shows that samples from the Awajinsha, Bishamon, Ourayama, and Iwatsubo cave sites had well-preserved collagen according to the above criteria. On the other hand, scapular samples (SSC-0001, -0002, -0004, -0005, -0006, -0008) from six Sano individuals showed relatively low %Col values, but these values were still higher than 1%. It was suggested again by %Col that collagen preservation at the Sano site was significantly worse than at the other sites. However, from the criteria of %C > 13% and %N > 4.8% (Ambrose, 1990), only sample SSC-0007, which had the most altered C/N ratio, is considered to show clear signs of possible diagenetic alteration. From the above combination of criteria, five Sano samples and all samples from Awajin-
sha, Bishamon, Ourayama, and Iwatsubo were considered relatively reliable, while three Sano samples (SSC-0005, -0007, and -0008) were omitted from further consideration.

The elemental indicators discussed above are sensitive to degradation of collagen but are not necessarily sensitive at detecting the uptake of small amounts of exogenous carbon contaminants that may influence $^{14}$C dates (van Klinken, 1999). Higham et al. (2004) noted that a C/N ratio of 3.6 might be taken as evidence for the presence of ~5–20% added contaminant carbon. Figure 2 and Figure 3 illustrate relations between parameters in a further attempt to detect potential contamination. Contamination from humic material, which mainly derives from plants, generally results in a higher C/N ratio, and higher %C and more negative $^{13}$C values in temperate zone bone (van Klinken, 1999). If contamination is significant in ‘collagen’, and the $^{13}$C value of the contaminants is different from that of bone collagen, significant correlations are expected between $^{13}$C and the parameters potentially related to carbon contamination (C/N or %C). In our data set, there were no suggestions of significant correlation of these parameters with $^{13}$C (at the 5% level).

Correlations between %C and C/N, and between %N and C/N, are positive in the Awajinsha series, although they are not statistically significant. This might suggest the existence of exogenous contaminants with higher %C and %N, but lack of correlation with $^{13}$C does not suggest such an influence. Figure 3 shows that the %C and %N values of the Sano samples correlate significantly with the C/N ratio ($r = -0.787$ and $-0.895$, $P = 0.020$ and 0.003, respectively), but this negative correlation is better considered to relate to collagen degradation rather than exogenous contamination. Although, taken together, the above considerations are still not fail-safe against very low levels of contamination, it is possible to conclude that the ‘collagen’ of samples from the Awajinsha, Bishamon, Ourayama, and Iwatsubo sites is suitable for analysis and interpretation.

Table 2 shows results of conventional $^{14}$C dates, estimated percentages of marine food in total protein intake, and calibrated $^{14}$C dates. Error margins of the $^{14}$C ages are shown by one standard deviation ($1\sigma$). It is evident that some of the ages lie outside the range of the Yayoi period. All $^{14}$C dates of the Sano samples are considerably older, even assuming a strong marine reservoir effect, with $^{13}$C values suggesting intake of more marine protein than the other populations. The conventional and calibrated $^{14}$C ages show that the dated Sano specimens do not originate from the Yayoi period but belong to the Jomon period. Two humeri of the Bishamon caves (samples BHU-0006 and -0007) are significantly younger than the other Bishamon samples. They appear to belong to recent or modern skeletons.

The other conventional $^{14}$C dates of the present study are
within the broad range of previously reported Yayoi period radiocarbon dates of 2500 to 1200 BP (Watanabe, 1966). However, conventional \(^{14}\)C dates must be evaluated by means of calibrated \(^{14}\)C ages with marine reservoir correction. This is especially the case in the present study, because stable isotopes suggest that marine resources were probably an important protein source in all sampled individuals (Table 2), even of the inland-located Iwatsubo cave. With the calibrated ages, it can be seen that samples from the Ourayama and Iwatsubo sites show good agreement with the conventionally accepted range of the Yayoi period of 300 BC to 300 AD (Kobayashi, 1951; Uno, 1989), but some of the Awajinsha and Bishamon material might belong to the Kofun period. A careful evaluation of each site is given below.

**Discussion**

The chronological framework of the Yayoi period has conventionally been considered to range from 300 BC to 300 AD based on pottery typology and comparison of metal ware excavated mainly from western Japan with those from China and Korea (Kobayashi, 1951; Uno, 1989). Although an earlier radiocarbon dating study suggested a wide chronological range of 2500 to 1200 BP for Yayoi period materials (Watanabe, 1966), until recently, in the context of Chinese history and calendar ages, a chronology that considers the Yayoi period to start from 300 BC has generally been accepted (Kobayashi, 1951; Sahara, 1975; Uno, 1989). However, recent application of dendrochronology in Japan is enabling a new look at Yayoi chronology; for example, a wooden coffin found at the Higashi-Muko site in Hyogo assigned to the Early Yayoi indicated an age of 445 BC (Mitsutani, 2000). Furthermore, recent studies of radiocarbon dating of charred material adhering to Early Yayoi pottery shards of northern Kyushu resulted in calibrated ages of between 900 and 800 calBC (Harunari et al., 2003). In the present study, we tentatively assumed a Yayoi period range of 500 BC to 300 AD, based on the recently emerging archaeological framework (Morioka, 2001); however, as outlined above, it should be noted that this estimate may contain an uncertainty of several hundred years.

All samples from the Sano cave showed substantially older calibrated \(^{14}\)C ages than the range of the Yayoi period, regardless of the Yayoi chronological uncertainties summarized above. The calibrated \(^{14}\)C ages of around 5000 to 4500 calBC, after accounting for the marine reservoir effect on human collagen, cannot fall within the Yayoi period. These skeletal remains probably date to the Early Jomon period, generally considered to be between 6300 and 4800 BP (5300 and 3630/3550 calBC) in the Kanto and Chubu districts.

Figure 3. Correlation among C/N ratio, %C, %N, and \(\delta^{13}\)C values of the Sano human remains.
Table 2. Conventional and calibrated radiocarbon ages of the human bones

<table>
<thead>
<tr>
<th>Site</th>
<th>Sample number</th>
<th>(^{14}C) age (BP)</th>
<th>(^{14}C) (‰) marine</th>
<th>Range of calibrated age</th>
<th>Lab. code</th>
<th>Excavation record</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sano</td>
<td>SSC-0002</td>
<td>5039 ± 61*</td>
<td>-12.2</td>
<td>3520 calBC ~ 3350 calBC</td>
<td>TERRA-112103a06</td>
<td></td>
</tr>
<tr>
<td></td>
<td>SSC-0004</td>
<td>5823 ± 66*</td>
<td>-16.4</td>
<td>4520 calBC ~ 4360 calBC</td>
<td>TERRA-052103a08</td>
<td></td>
</tr>
<tr>
<td></td>
<td>SSC-0006</td>
<td>5960 ± 58*</td>
<td>-14.1</td>
<td>4580 calBC ~ 4400 calBC</td>
<td>TERRA-112103a14</td>
<td></td>
</tr>
<tr>
<td></td>
<td>SSC-0003</td>
<td>6332 ± 53</td>
<td>-11.7</td>
<td>4920 calBC ~ 4760 calBC</td>
<td>TERRA-052103a07</td>
<td></td>
</tr>
<tr>
<td></td>
<td>SSC-0001</td>
<td>6332 ± 52</td>
<td>-12.5</td>
<td>4920 calBC ~ 4760 calBC</td>
<td>TERRA-052103a06</td>
<td></td>
</tr>
<tr>
<td>Awajinsha</td>
<td>ACO-0002</td>
<td>1745 ± 58</td>
<td>-15.4</td>
<td>460 calAD ~ 620 calAD</td>
<td>TERRA-082699a26</td>
<td></td>
</tr>
<tr>
<td></td>
<td>ACO-0005</td>
<td>1835 ± 67</td>
<td>-16.5</td>
<td>260 calAD ~ 480 calAD</td>
<td>TERRA-082699a29</td>
<td></td>
</tr>
<tr>
<td></td>
<td>ACO-0003</td>
<td>2136 ± 55</td>
<td>-16.9</td>
<td>50 calBC ~ 120 calAD</td>
<td>TERRA-082699a27</td>
<td></td>
</tr>
<tr>
<td></td>
<td>ACO-0004</td>
<td>2237 ± 52</td>
<td>-15.9</td>
<td>120 calBC ~ 50 calAD</td>
<td>TERRA-082699a28</td>
<td></td>
</tr>
<tr>
<td>Bishamons C</td>
<td>BUH-0006</td>
<td>77 ± 44</td>
<td>-20.4</td>
<td>1880 calAD ~ 1950 calAD</td>
<td>TERRA-052103a28</td>
<td></td>
</tr>
<tr>
<td></td>
<td>BUH-0007</td>
<td>185 ± 43</td>
<td>-19.3</td>
<td>1700 calAD ~ 1950 calAD</td>
<td>TERRA-052103a29</td>
<td></td>
</tr>
<tr>
<td></td>
<td>BUH-0004</td>
<td>1514 ± 44</td>
<td>-16.3</td>
<td>600 calAD ~ 695 calAD</td>
<td>TERRA-052103a26</td>
<td></td>
</tr>
<tr>
<td></td>
<td>BUH-0001</td>
<td>1639 ± 44</td>
<td>-16.3</td>
<td>460 calAD ~ 600 calAD</td>
<td>TERRA-052103a23</td>
<td></td>
</tr>
<tr>
<td></td>
<td>BUH-0005</td>
<td>1774 ± 44</td>
<td>-18.4</td>
<td>260 calAD ~ 430 calAD</td>
<td>TERRA-052103a27</td>
<td></td>
</tr>
<tr>
<td></td>
<td>BUH-0002</td>
<td>1779 ± 58</td>
<td>-18.7</td>
<td>410 calAD ~ 570 calAD</td>
<td>TERRA-052103a24</td>
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<td></td>
<td>BUH-0003</td>
<td>1828 ± 45</td>
<td>-17.9</td>
<td>320 calAD ~ 480 calAD</td>
<td>TERRA-052103a25</td>
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<tr>
<td>Bishamons D</td>
<td>BUH-0008</td>
<td>1864 ± 45</td>
<td>-18.4</td>
<td>260 calAD ~ 430 calAD</td>
<td>TERRA-052103a30</td>
<td></td>
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<tr>
<td>Ouryama</td>
<td>OCO-0005</td>
<td>2192 ± 53</td>
<td>-18.5</td>
<td>200 calAD ~ 0 calAD/BC</td>
<td>TERRA-082699a39</td>
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<tr>
<td></td>
<td>OCO-0001</td>
<td>2232 ± 53</td>
<td>-16.5</td>
<td>180 calAD ~ 50 calAD/BC</td>
<td>TERRA-082699a30</td>
<td></td>
</tr>
<tr>
<td></td>
<td>OCO-0004</td>
<td>2277 ± 63</td>
<td>-16.4</td>
<td>210 calAD ~ 0 calAD/BC</td>
<td>TERRA-082699a38</td>
<td></td>
</tr>
<tr>
<td></td>
<td>OCO-0003</td>
<td>2296 ± 108</td>
<td>-15.9</td>
<td>340 calAD ~ 30 calAD</td>
<td>TERRA-082699b37</td>
<td></td>
</tr>
<tr>
<td></td>
<td>OCO-0002</td>
<td>2318 ± 96</td>
<td>-17.4</td>
<td>360 calAD ~ 110 calAD</td>
<td>TERRA-082699a36</td>
<td></td>
</tr>
<tr>
<td>Iwatsubo</td>
<td>ICO-0003</td>
<td>2155 ± 53</td>
<td>-17.9</td>
<td>120 calBC ~ 50 calAD</td>
<td>TERRA-082599a37</td>
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</tr>
<tr>
<td></td>
<td>ICO-0001</td>
<td>2164 ± 60</td>
<td>-16.2</td>
<td>50 calBC ~ 120 calAD</td>
<td>TERRA-082599a29</td>
<td></td>
</tr>
<tr>
<td></td>
<td>ICO-0002</td>
<td>2174 ± 56</td>
<td>-16.3</td>
<td>90 calBC ~ 90 calAD</td>
<td>TERRA-082599a30</td>
<td></td>
</tr>
</tbody>
</table>

* Sample sizes were smaller than 1 mgC (0.58 mg and 0.14 mg for SSC-002 and 006, respectively).

\( \text{A}_{\text{overall}} \) is the overall agreement index for multiple dates within a series of \(^{14}C\) dates. The value should be greater than 60% for the results to be significant (Bronk Ramsey, 1995). It is apparent that the estimated ages of these specimens coincide with the archaeological timeframe of the Yayoi period. Although the excavated archaeological materials suggest that the entire Ouryama Yayoi series may contain some time depth (Yokosuka Archaeological Society, 1997), given the unique circumstances of human bone breakage and burial, it is possible that the human remains all derive from a single, relatively narrow time interval. The Iwatsubo specimens were associated with a single Yayoi period pottery phase (Nozawa I) (Imamura, 1984).

On the other hand, the Awajinsha and the Bishamons samples of the present study consist of individuals from different time periods. In the case of Awajinsha, the agreement among the four probability distributions is poor (\( \text{A}_{\text{overall}} = 0.1% \)), and the dated specimens can be divided into two groups. The older two dates are tentatively combined into a 50 calBC to 60 calAD range (\( \text{A}_{\text{overall}} = 111.0% \)), which lies within the Yayoi time period. The younger two dates of the Awajinsha site, however, showed a combined probability of around 430 to 540 calAD (\( \text{A}_{\text{overall}} = 79.7% \)). These specimens may belong to the succeeding Kofun period. Although the human skeletal material from the Awajinsha cave site includes more than 20 individuals (Koganei, 1953), the present study shows that these samples probably combine material from two or more different periods. Suzuki (1964, 1969) described the Awajinsha cranial material as exhibiting a
range of morphologies and interpreted the series to represent a population that shows a transitional morphology between the native Jomon and Kofun types. The present study shows that this interpretation needs to be re-evaluated from future age estimations of the relevant cranial remains themselves.

The occurrence of prominent Jomon-like nasal root morphology (Suzuki, 1964, 1969), anterior tooth extraction (Koganei, 1933; Suzuki, 1969), and Jomon-like tooth size proportions (Matsumura, 1998b) in the Awajinsha sample suggest either a chronologically mixed assemblage, perhaps even including some Jomon-period specimens, or a Yayoi population with Jomon-like or intermediate morphological features.

The Bishamon specimens showed wider variability with three clusters: two recent to modern dates of 77 and 185 BP, respectively; one sample with a calibrated age of 600 to 695 calAD; and the five other samples that combine into a distribution of around 400 to 465 calAD with 1σ probability. The latter combined age approximates the end of the Yayoi period. It is possible that these specimens correspond to the scattered skeletal remains attributed to the Kofun period (Akaboshi, 1953). Matsumura (1998b) suggested that the

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**Figure 4.** Probability distributions of calibrated 14C dates of Awajinsha, Bishamon, Ourayama, and Iwatsubo samples. Shadowed area indicates the tentative estimation of the Yayoi period (500 BC to 300 AD). See Discussion for further details regarding the present uncertainties surrounding this chronology.
Bishamon material included both ‘native’ and ‘migrant’ conditions of tooth size proportions as demonstrated by discriminant function analysis. The Kofun-age estimation of most of the Bishamon samples of this study is in concordance with Matsumura’s (1999b) evaluation of the Bishamon tooth dimensions of two of three individuals to show the ‘migrant’ lineage condition. Further evaluations of both chronology and morphology are needed for a fuller understanding of the Bishamon series.

A comparison of the combined dates of the Ourayama (170 to 50 calBC) and Iwatsubo (45 calBC to 55 calAD) samples appears to suggest that the Ourayama population preceded the Iwatsubo population, although the associated pottery type of the latter (Nozawa I type) is considered to precede that of Ourayama (Miyanodai type) (Ishikawa, 2003). It is important to determine the time relationship of the Ourayama people with Kofun-like cranial morphology and the more Jomon-like Iwatsubo people.

In these and previous detailed considerations of estimated ages, one source of remaining uncertainty is the possibility of contamination from small amounts of extraneous organic material, despite the precautions taken as outlined above. Additional uncertainties concerning age calibration of human bones furthermore make it difficult to determine their age contexts conclusively. For example, the percentage of marine carbon might be overestimated for the Iwatsubo samples, an inland population, if they were accessing C₃ crops. If such was the case, without marine reservoir correction, the Iwatsubo series would show combined calibrated ¹⁴C ages of 360 to 100 calBC, which predates the Ourayama series. This should be further discussed in conjunction with other indicators including nitrogen isotopic ratio and analyses of faunal and plant remains.

The present study, however, demonstrates the likelihood that some of the Ourayama and Iwatsubo people lived broadly contemporaneously. It also suggests that the Kofun-like Ourayama morphology existed fairly early in the Yayoi chronology of eastern Japan. Following Suzuki’s hypothesis, one possibility would be that the morphological differences between the two series may reflect difference in subsistence between the coastal Ourayama and mountainous Iwatsubo locations (e.g. Kaifu, 1992). One focus for future studies would be to investigate whether the Awajinsha and/or other cranial remains considered to be of intermediate morphology actually preceded the Ourayama series in age, corresponding with the time and location of actual changes in subsistence patterns. Otherwise, these two morphological types of the Kanto Yayoi may indicate lineage differences.

In this latter scenario, the coastal area would be dominated by immigrant-based populations, while the inland mountains would largely be populated by people of local Jomon descent, as is similarly considered to be the case in the Yayoi populations of the Kyushu district where the Kofun-like and Jomon-like populations inhabited the northeast plains and northwest coastal areas, respectively (Naito, 1971, 1981; Nakahashi, 1993; Nakahashi and Iizuka, 1998).

Conclusions

Taking into account the marine reservoir effect of each sample, calibrated ¹⁴C ages were determined for 25 skeletal remains previously assigned or attributable to the Yayoi period. The results suggest that the dated Ourayama and Iwatsubo samples originate from the Yayoi period, while the Awajinsha, Bishamon, and Sano series include individuals from other periods. In particular, all of the Sano specimens examined were dated to the Early Jomon period. The Awajinsha specimens appear to include both Yayoi and Kofun period elements, and the Bishamon series is shown to additionally contain human remains of the Kofun and recent/modern periods.

Suzuki (1969) believed that environmental factors had changed the skeletal morphology of the Yayoi people of the Kanto region, from the Jomon-like type (Sano) through an intermediate condition (Awajinsha) to the Kofun-like type (Bishamon, Ourayama). The ¹⁴C ages of the present study suggest that his Yayoi cranial series most likely included material both preceding and postdating the Yayoi period. However, since the present study did not directly date the cranial material itself, the most diagnostic in Suzuki’s (1969) transformation hypothesis, further conclusions must await forthcoming studies dealing with both chronology and morphology of the relevant cranial material.

Meanwhile, we have demonstrated that there were almost certainly at least two different types of morphology among the Yayoi populations of the Kanto region. Most importantly, the Iwatsubo individuals with Jomon-like cranial morphology and the Kofun-like Ourayama series were shown to be broadly contemporaneous. The morphological differences between the two series may reflect differences in subsistence between the coastal and inland mountainous locations. Alternatively, the two morphological types of the Kanto Yayoi, suggested above to be contemporaneous, may indicate lineage differences.

We have shown that the Awajinsha series, considered by Suzuki (1969) to be of intermediate morphology, is likely to be chronologically heterogeneous. It will be important to document, within the chronologically defined Yayoi specimens, the morphological characteristics of the Awajinsha material on the one hand, and to derive precise age estimates of those specimens on the other. Such combinations of morphological and chronological resolution, as well as future new finds of Yayoi period skeletal remains, will enable further evaluation of the population history of the Japanese.

Acknowledgments

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