The repeatability of the methods, as shown by figures 7.6 - 7.11, is generally good. The maximum difference between any two measurements is 0.24 (see figure 7.8). The vast majority of the points on all of the graphs fall within the +/- 2 standard deviations range and so, according to the criteria set out by Bland and Altman (1986 p308), the method can be said to be repeatable.

It is possible that the time lapse between the taking of each measurement set has had an impact on the repeatability of the method. If this were the case, it would be expected that a comparison of measurement sets A and B (taken consecutively) would show markedly different patterns to a comparison of measurement sets A and D (taken some period apart). It might be expected that the mean difference and the range of the differences between measurement sets would be smallest when they were taken consecutively rather than with a considerable time lapse between them. In this test, these variables do not seem to conform to this or any other obvious model.

A pattern that does emerge from figures 7.6 - 7.11 relates to both the time lapse between taking measurement sets and the magnitude of the measurements themselves. It can be seen in these figures that a comparison of measurement sets that were taken consecutively (A & B, B & C and C & D) produces differences that follow an approximately horizontal line. This implies that, for these comparisons, the values of the differences are not related to the "true" density. This contrasts with the differences between measurement sets that were taken some time apart (A & C and B & D) which show differences that have a more negative relationship with the average density (they are lower, when the average density is higher). The relationship between measurement sets A and D (those with the longest interval) is shown to be more strongly negative still. It seems, therefore, that some variability in the method, according to the interval between measurement sets, does exist. This variability is also partly dependent on the magnitude of the true density. Since cortical bone is known to be generally denser than trabecular bone, this pattern could in fact represent a difference according to bone type. It is impossible to speculate as to the nature of this variability, because, contrary to Altman and Bland's assumptions, the true density is not known. It is therefore impossible to be certain which measurement set is flawed and whether the values have been under estimated or overestimated.

Perhaps the most important features of figures 7.6 - 7.11 are the two outlying points on figures 7.8 and 7.11. Both of these points fall outside the line that represents -2 standard deviations, and, as such, indicate that the repeatability of these measurements is poor. Importantly, these two points both represent the measurement of the shaft of the metatarsal and both involve a comparison of measurement set D. It can be seen in table 7.9 that measurement set D produced a density value for the metatarsal shaft that was much higher than any of the other sets. It is this fact that is responsible for the outlying points in figures 7.8 and 7.11. The reason for this large degree of variation obtained for the metatarsal shaft from measurement set D is that the thickness values were difficult to define on the relevant radiograph. Since it was possible to predict at the measurement stage that this measurement would prove unreliable, the conclusion that the overall repeatability of the method is high has not been questioned. If this potentially erroneous result is removed from this test, all of the points on all of the graphs fall within the +/-2 standard deviations range. As a result of this test, any density values that were deemed potentially unreliable at the time of measurement were excluded from subsequent analyses.

In the following chapter it will become necessary to ascertain whether the difference between two measurements is the result of measurement error or not. The maximum difference between any measurements taken in this test was 0.24. Even though this large difference was predicted at the measurement stage, this figure will be used as the potential measurement error. Consequently, any differences between two measurements that do not exceed 0.24 will be assumed to be the result of measurement error.

By repeating a number of density measurements from a single animal and comparing the results, it has been possible to confirm that the intra-observer error in the methodology to be employed in this project is small. Furthermore, where intra-observer errors are likely to be substantial, these can be recognised at the measurement stage and affected data can be excluded from further analysis. The potential measurement error for the methodology has been defined as being 0.24.

7.4.3: The effects of organic material on the radiographically derived density

Some of the material used in this project is likely to contain a residue of organic material (chiefly bone grease and marrow that has not been completely removed during the preparation process). Since these residues are not completely radiolucent, they could potentially have the effect of increasing the overall radiodensity of a specimen. The grey level of a specimen containing organic matter will therefore be lighter than might otherwise be expected. Consequently, any density measurements derived from such a radiograph will be inflated (Ruff and Leo 1986 p182). In order to assess the impact of this organic material on radiographically derived density measurements, a simple test was carried out. A single fresh femur from an immature pig was radiographed and its density was measured according to the method outlined for this project. Measurements were taken at the proximal, distal and midshaft regions of the bone. Provided that all subsequent measurements were taken on exactly the same parts on the bone, the precise location of the three scan-sites selected was of little importance. In addition, this ensured that bone thickness remained constant and so could be excluded from the calculations. Consequently, the measurements obtained were in equivalent millimetres of aluminium.

The bone was then immersed in 50 cm^3 of biological washing powder ("Biotex") dissolved in 3 litres of freshly boiled tap water and simmered for 1 hour. Following this period of degreasing, the bone was removed, its density recalculated and returned to another 3 litres of freshly boiled Biotex solution. This process was repeated so that density data for the bone after 1, 2, 3, 4, 5, 6 and 12 hours of degreasing were produced. After 12 hours, the bone had adopted a dry, almost chalky appearance. Degreasing bone by simmering it in detergent is similar to the method used by English Heritage to degrease much of the bone that was used in this project (see page 7.2.3). The results of this test are shown in table 7.10 and figure 7.12.

	Location		
	Proximal	Shaft	Distal
Fresh	6.9	8.65	8.4
1 Hour	7.62	8.68	9.06
2 Hour	6.97	8.23	8.59
3 Hour	6.97	8.85	7.55
4 Hour	6.48	8.44	7.41
5 Hour	6.62	8.46	7.27
6 Hour	6.57	8.1	7.67
12 Hour	6.53	8.13	7.78

Table 7.10: Showing the bone density of the three scan-sites on the same pig femur (as measured by the methodology proposed by this project) containing various amounts of organic material.

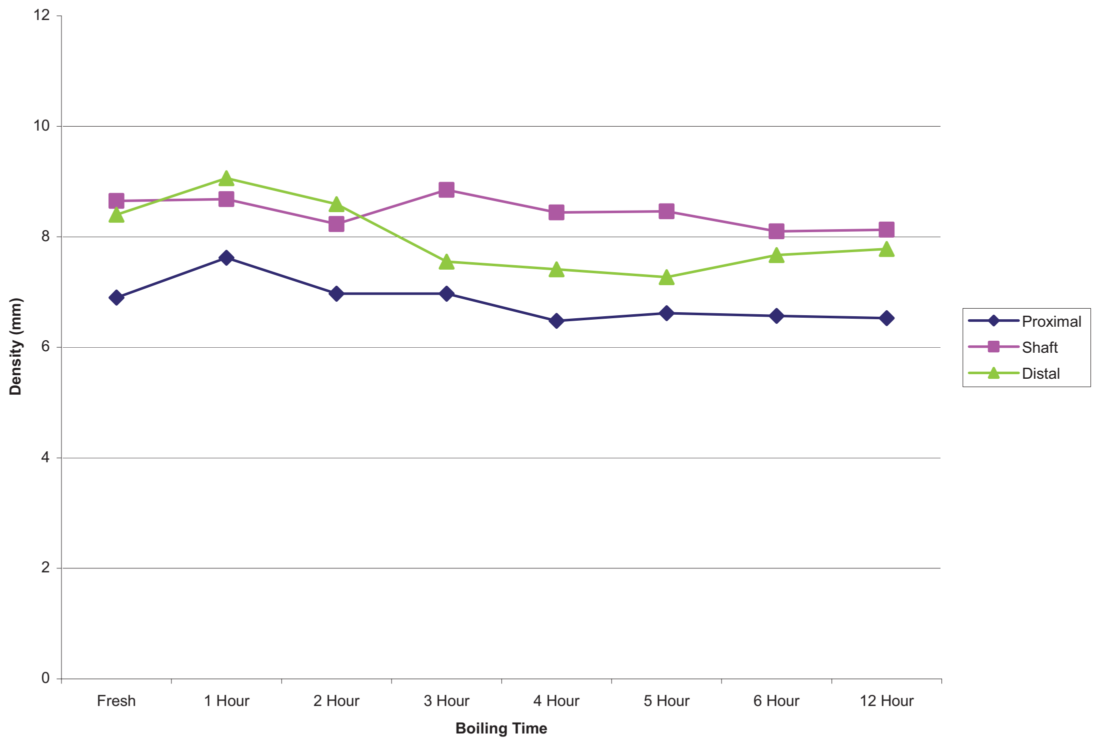

Figure 7.12: A line graph showing the changes in bone density of three scan-sites on a pig's femur after different periods of boiling.

Figure 7.12 shows no dramatic reduction in the radiographically derived density of any of the three scan-sites on the test material. However, a close inspection reveals that there is a very slight reduction in radiodensity as boiling time progresses. This feature is more notable for the trabecular bone ends than for the bone shaft and may reflect the greater amount of organic material contained in the bone ends. It is therefore possible to confirm the assertion (Ruff and Leo 1986 p182) that organic material can potentially affect bone density measurements obtained through radiographic methods.

This impact of soft tissue on a bone's radiodensity is unlikely to affect the reliability of the data produced by this project. None of the material used in this project was "fresh". It all resembled bone that, according to this test, had been simmered for between 4 and 12 hours. This portion of the graph shows very little variation indeed. The maximum difference between two measurements of the same scan-site in this part of the graph is 0.51 (the difference between 5 and 12 hours simmering on the distal end of the bone). This is well within the intra-observer error described above and so can be attributed to general variation within the method.

The results of this test were confirmed in two ways. Firstly, variations in the radiographically derived density of groups of material prepared using different methods were examined (see section 8.5). No significant difference in density was observed between bones that had been subjected to different preparation methods. Also, each element examined was assigned a score according to how greasy it was (1 = dry and 5 = very greasy). At no stage in the analysis undertaken by this project was any variation according to this score noted.

This test has confirmed that the amount of organic matter retained within a bone will potentially have an impact on the radiodensity data obtained by this project. However, the material used by this project does not contain significantly different amounts of organic matter. Consequently, the bone density values will also not be significantly affected. This assertion has been confirmed by subsequent observations in this project.

7.4.4: Image magnification

X-radiography is known to produce images that contain certain distortions. One such distortion is the magnification of the object in the image. Figure 7.13 shows an object (XY) some distance (*t*) above the film. It is clear that the image (X'Y') will be subject to some degree of magnification.

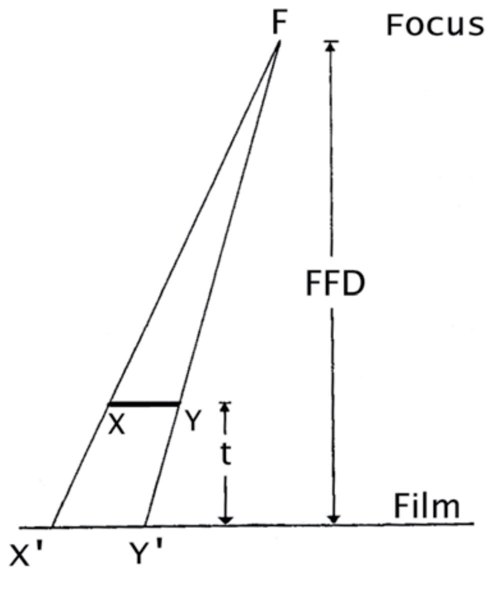

Figure 7.13: Showing how a linear object (XY) will appear as a magnified image (X'Y'). The degree of magnification is dependent on the length of* t *and the film to focus distance (FFD). After Wilks (1987 p31 fig3.3).

This magnification will be greatest when the object is some distance from the radiographic film, or when the distance from the radiographic film to the X-ray source is small. Since the magnification factor varies slightly according to the object to film distance, in the case of three-dimensional objects, the magnification of any particular *part* of a bone will depend on the distance from that *part* of the bone to the film. This is likely to result in an overestimation of bone size (thickness), the magnitude of which is dependent on the distance of the object from the film, but not on the position of the object on the X-ray cabinet shelf. The magnification factor can be calculated using the following equation:

$$M = \frac{\text{FFD}}{\text{FFD}-t}$$

Calculation of the magnification factor. Where* M *is the magnification factor, FFD is the distance from the film to the X-ray source (or focus), and* t *is the film to object distance. From Wilks (1987 p32).

Using this equation, it is possible to calculate that an object with a depth of 45.7 mm (the maximum depth of any bones measured in this project) that is placed on the bottom shelf of the X-ray cabinet (556 mm from the X-ray source) would be magnified by a factor of no more than 1.11. Realistically, the actual magnification factor will be in the order of 1.05, because the distance from the film to the point at which measurements were taken (where magnification errors would have the most impact) was rarely more than 25 mm from the film.

The actual distortion produced by this phenomenon was tested by placing a wire mesh with an aperture of 7.9 mm above the film and radiographing it, with the distance “FFD” remaining constant, at 556 mm. Any distortions caused by magnification were apparent by comparing the aperture of the mesh as it appeared on the radiograph with its known actual aperture. When the mesh was suspended 48.6 mm above the film (t = 48.6), the aperture of the image was 8.7 mm (M = 1.12). When the mesh was suspended 41.8 mm above the film (t = 41.8) the mesh aperture on the image was 8.5 mm (M = 1.10). These results are very close to those predicted by the above equation. The distortion recorded in this image was constant no matter where on the film the measurements were taken.

In order to attempt to overcome the effects of this phenomenon on the results produced by this project, M was calculated for each of the elements under analysis (to achieve this, *t* was measured for every element in the experimental material). The results presented in this project have all been corrected for the effects of magnification.

The image of an object will be magnified to some degree. This effect increases with increasing image to film distance and decreasing film to X-ray source distance. The impact of this distortion on the radiographically derived densities produced in this project has been shown to be small and has been corrected in all of the experimental material.

7.4.5: Artificial density inflation at the film periphery

In addition to this, the depth of bone through which the X-rays travel will not always be equal to the true depth of that bone. Figure 7.14 describes this diagrammatically.

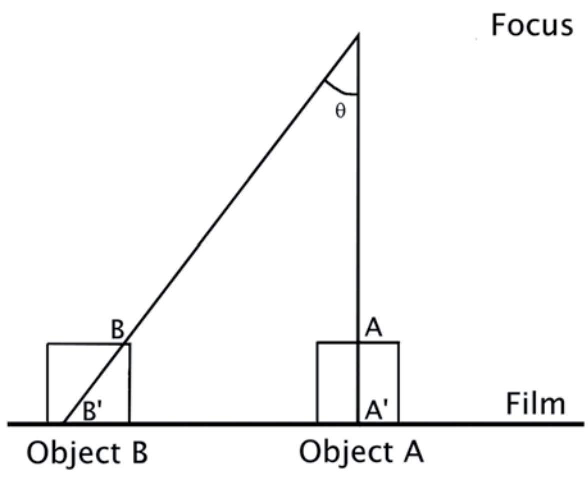

Figure 7.14: Showing two identical objects, A and B, resting on a film. It is clear that X-rays emanating from the focus will pass through a greater thickness of object B than object A. This difference can be described as the difference between A A' and B B' and can be calculated with reference to θ.

X-rays passing through an object on the edge of the radiographic film (E) will do so at an oblique angle (θ). They will therefore pass through a greater thickness of bone and so will suggest a more radiodense object than would be apparent if the object had been positioned immediately beneath the X-ray source in the centre of the film (M). This potential for error is dependent on the distance of the object from the *centre* of the film, rather than the film-object distance (or the depth of the object). The degree of distortion produced by this effect will not be constant for any given bone, but will be greater for the part of the bone further away from the centre of the film than for the part of the bone closest to the centre of the film. This effect can be calculated using the equation:

$$D = \frac{1}{\text{Cos } \theta}$$

Calculation of the "offsetting error". Where* D *is the ratio of the actual bone thickness to the thickness of bone through which the X-rays pass and θ is the angle from the centre of the film to the X-ray source to the object (see figure 7.15).

Using this equation, it is possible to calculate that X-rays passing through a bone placed at the edge of the radiographic film (the distance from the centre of the film to the object being 120mm, giving a θ of 12.18° if the film to X-ray source distance is 556mm) will pass thorough 1.023 times more bone than the measured bone thickness. In real terms this means that if a bone that is 25mm thick is placed at the very edge of the radiographic film, the X-rays will be passing through 25.57mm of bone.

In order to assess how these two features of X-radiography might affect the density values returned in this project a simple experiment was devised. The step wedge was radiographed at four different positions on a sheet of film (one at the centre of the film as well as at 40, 80 and 100mm from this centre point). The densities for each step of these four images were calculated in the usual manner. It would be expected for all of the steps to have radiographically derived densities of 1.0, and any distortion of the type described above will result in deviations from this figure. The results of this experiment are shown in table 7.11 and displayed graphically in figure 7.15.

Actual Thickness of Wedge (mm)	Distance to the Film Centre (cm)			
	0	4	8	10
1	0.83	0.83	0.83	0.83
2	1.03	1.03	1.03	1.03
3	1.03	0.98	1.06	1.02
4	0.99	0.97	1.03	0.97
5	0.98	0.98	0.99	0.98
6	0.99	0.98	1.01	0.99
7	1.00	0.99	1.00	1.00
8	1.01	0.98	1.02	1.00
9	0.99	0.98	1.00	0.99
10	1.00	0.98	1.04	1.00
11	1.00	0.98	1.04	1.00
12	1.00	1.00	1.01	1.01
13	1.01	1.00	1.05	1.05
14	0.98	0.98	1.00	0.99
15	0.95	0.92	0.96	0.95

Table 7.11: Showing how the bone density measurements produced by this project will alter according to the location of the object on the radiographic film.

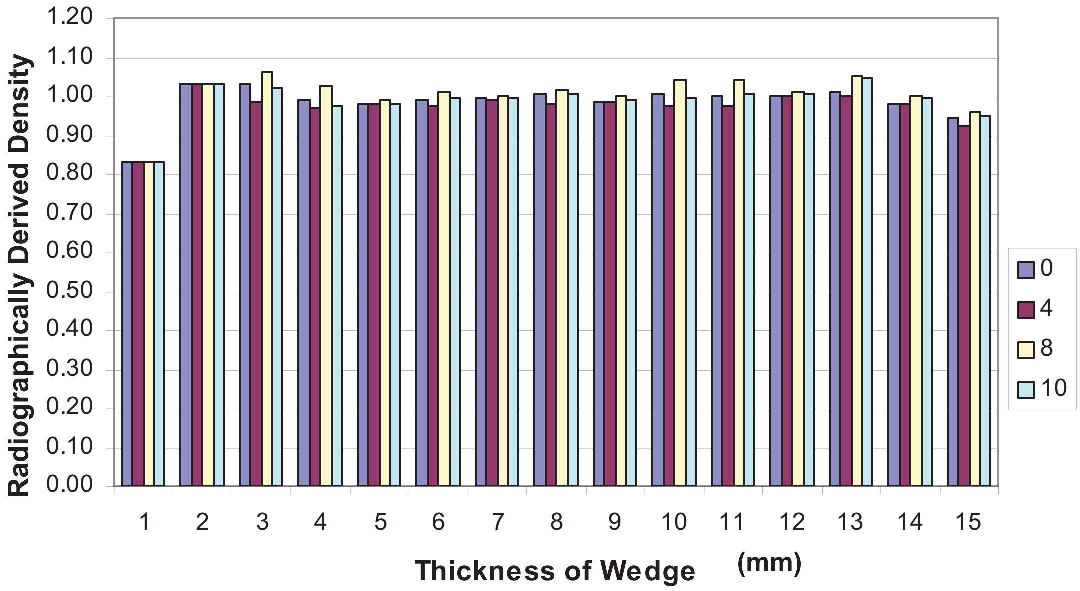

Figure 7.15: Using the data presented in table 7.11, showing graphically how the density values returned using the methodology used by this project will vary according to their position on the radiographic film. The legend refers to the distance of the object from the position directly beneath the X-ray source, while the x-axis denotes increasing object size.

Figure 7.15 shows almost all of the results closely clustered around 1.0 and did not increase as the distance of the object from the centre of the film increased. This suggests that the impact of the position of the object on the film will have little or no impact on the results obtained by this project.

The results obtained deviate from the expected results on two occasions (where the thickness of the wedge is 1mm and 15mm) and this should be explained. Radiographic film is only able to react to exposure to X-rays when the intensity of this exposure is within certain limits. When the exposure is very low, the emulsion will not be affected and when it is very high, the emulsion will have reacted completely and so will be unable to react to further increases in exposure. At these extreme ends of the step wedge, the radiographic film received an exposure of X-rays that is so low or so high that it does not fall within the effective working range of the film. This does not relate to the distortion described above, but does indicate that areas that appear very dark or very light on the radiograph are likely to produce erroneous radiographically derived densities. No results from such images were used in this project.

The radiodensity of an object will appear higher as the distance between the object and the centre of the film increases or as the distance between the object and the X-ray source decreases. Experimentation has shown that the effect of this phenomenon on the radiographically derived densities of material used in this project is negligible. It also demonstrated that such data from images that appear either very light or very dark on the film should be treated with caution or ignored.

7.4.6: The representativeness of the scan-sites

The use of single points as scan-sites represents a deviation from previous studies of this type, all of which used linear scan-sites. It is necessary to ensure that the innovation of using individual points as a basis for bone density measurement will not impair the validity of the density measurements produced. The choice of scan-site locations for this project is intended to mirror the location of predefined diagnostic zones (DZs) on each bone (see section 7.1.3). Unlike the scan-site locations, DZs take the form of linear slices across the entire width and depth of each bone. These DZs will later form the basis of bone quantification, when the density values obtained by this project are applied to an archaeological assemblage (see chapter 9). When more than 50% of an individual DZ is present the corresponding bone part will be said to be present, otherwise it will be recorded as being absent. Consequently, if (as is the case) the densities of single points on bones are used to explain the presence or absence of larger (linear) areas of bone, it must be ensured that the density of each scan-site truly represents the density of each DZ as a whole.

In the next chapter, the variation of bone density according to a number of factors will be explored. This will not involve the need to compare bone density values to the presence or absence of DZs. Consequently, the need for the scan-sites to be representative of the DZs as a whole is of lesser importance. However, since, in this case, the density of scan-sites will be compared with *each other* it is imperative that the scan-sites from different animals are comparable. This can only be achieved by ensuring that the scan-sites are always positioned in exactly the same position on different individuals. Every effort has been made to ensure that this is the case. The fact that the intra-observer error has shown that the method is repeatable suggests that the placement of scan-sites can also be reliably repeated (see section 7.4.2).

The extent to which the scan-sites represent the density of the relevant DZs is a more important consideration when the data derived from the scan-sites are compared with the presence or absence of DZs in an archaeological assemblage (see chapter 9). A notable feature of the archaeological faunal assemblage that is examined in chapter 9 is that, when more than 50% of a DZ is recorded as being present, the part of the bone that includes the scan-site is also invariable present. This is because DZs tended to be represented by entire bone ends. It is for this reason that it is possible to assume that this project is not comparing the density of scan-sites with the presence or absence of DZs, but instead is comparing the density of scan-sites with the presence or absence of scan-sites.

Regardless of the validity of this assumption, it is should be noted that density values derived from individual points (scan-sites) have already been shown to be comparable with density values for larger, linear slices through a number of bones (DZs). This comparison is described in section 7.4.1.

In this project, the choice of single points as the basis for scan-sites was made in order to overcome the methodological problems (associated with determining the cross-sections of the linear scan-sites) encountered by previous researchers. In following this course of action it may be arguable that this project will encounter problems associated with the ability for the scan-sites to represent accurately the density of the DZs to which they relate. However, even if this is the case, the ability to calculate reliably the size of the specimens being measured will more than compensate for any shortcomings in the representativeness of the scan-sites.

This chapter has described in detail the attributes of the experimental material to be used in this project. It has also described the experimental procedures that will be used to obtain density data from the material. Finally, a series of methodological tests have demonstrated that the methods used here (and so, the data derived from them) are reliable and relevant to the questions posed in this project.