Declarations

The following additional information is required for submission. Please note that failure to respond to these questions/statements will mean your submission will be returned to you. If you have nothing to declare in any of these categories then this should be stated.

Conflict of interest
All authors must disclose any financial and personal relationships with other people or organisations that could inappropriately influence (bias) their work. Examples of potential conflicts of interest include employment, consultancies, stock ownership, honoraria, paid expert testimony, patent applications/registrations, and grants or other funding.

Ethical Approval
Work on human beings that is submitted to Medical Engineering & Physics should comply with the principles laid down in the Declaration of Helsinki; Recommendations guiding physicians in biomedical research involving human subjects. Adopted by the 18th World Medical Assembly, Helsinki, Finland, June 1964, amended by the 29th World Medical Assembly, Tokyo, Japan, October 1975, the 35th World Medical Assembly, Venice, Italy, October 1983, and the 41st World Medical Assembly, Hong Kong, September 1989. You should include information as to whether the work has been approved by the appropriate ethical committees related to the institution(s) in which it was performed and that subjects gave informed consent to the work.

Competing Interests
The authors have performed research for the Ortho Kinematics Company, which is commercialising a version of this technology in the United States.

Please state any sources of funding for your research
No external funding was obtained for this research.

DOES YOUR STUDY INVOLVE HUMAN SUBJECTS? Please cross out whichever is not applicable.
Yes

If your study involves human subjects you MUST have obtained ethical approval. Please state whether Ethical Approval was given, by whom and the relevant Judgement’s reference number
Ethical approval was given by the National Research Ethics Service (REC reference 0/H0502/99).

This information must also be inserted into your manuscript under the acknowledgements section prior to the References.
Accuracy and repeatability of quantitative fluoroscopy for the measurement of sagittal plane translation and finite centre of rotation in the lumbar spine

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Abstract

Quantitative fluoroscopy (QF) was developed to measure intervertebral mechanics in vivo and has been found to have high repeatability and accuracy for the measurement of intervertebral rotations. However, sagittal plane translation and finite centre of rotation (FCR) are potential measures of stability but have not yet been fully validated for current QF.

This study investigated the repeatability and accuracy of QF for measuring these variables. Repeatability was assessed from L2-S1 in 20 human volunteers. Accuracy was investigated using 10 consecutive measurements from each of two pairs of linked and instrumented dry human vertebrae as reference; one which tilted without translation and one which translated without tilt. The results found intra- and inter-observer repeatability for translation to be 1.1mm or less (SEM) with fair to substantial reliability (ICC 0.533-0.998). Intra-observer repeatability of FCR location for inter-vertebral rotations of 5° and above ranged from 1.5mm to 1.8mm (SEM) with moderate to substantial reliability (ICC 0.626-0.988). Inter-observer repeatability for FCR ranged from 1.2mm to 5.7mm, also with moderate to substantial reliability (ICC 0.621-0.878). Reliability was substantial (ICC>0.81) for 10/16 measures for translation and 5/8 for FCR location. Accuracy for translation was 0.1mm (fixed centre) and 2.2mm (moveable centre), with an FCR error of 0.3mm(x) and 0.4mm(y) (fixed centre). This technology was found to have a high level of accuracy and with a few exceptions, moderate to substantial repeatability for the measurement of translation and FCR from fluoroscopic motion sequences.
Introduction

The *in vivo* measurement of intervertebral motion in the lumbar spine in individuals has been progressing. This information has traditionally been obtained as displacement on flexion-extension radiographs, however, this has been consistently found to be prone to large errors and variability between observers [1-5]. The method also suffers from the inability to detect the true end-range during motion and lack of standardised measurement methods [6].

Studies of quantitative fluoroscopy (QF) for measuring lumbar spine intervertebral kinematics using continuous motion tracking began in the 1980s [7]. QF measures continuous intervertebral motion and extracts end of range measurement from wherever it occurs in the bending sequence, giving a radiation dose similar to a conventional radiographic examination [8, 9]. Various iterations have been found to have good repeatability and accuracy for measuring intervertebral rotations at lumbar and cervical levels [5, 9-12]. However, excessive translation is thought to be more closely associated with back symptoms [13]. Translation also affects the finite centre of rotation (FCR) and the latter is an expression of the distribution of loading between the disc and facets during upright flexion-extension motion [14]. It is also said that the centre of reaction force (CR) can be extrapolated from the FCR [14].

QF technology employs standardised image registration and analysis protocols with relatively straightforward and inexpensive hardware in contrast to specialist MR, CT or dual fluoroscopic systems which are not as readily available in hospital settings. However, the literature addressing the repeatability and accuracy of translation and FCR measurement from fluoroscopy is based on different techniques. For example, Cerciello et al determined the accuracy of measuring intervertebral rotation and FCR location in 2-D using stepped positions in a calibration specimen rather than from continuous motion [15]. Wang et al and Lin et al determined the accuracy of translation measurement in ovine specimens using 2D-3D dual fluoroscopic systems where the geometry was informed by magnetic resonance or CT-based vertebral models of the same participant rather than a calibrated reference [16, 17]. These studies also found excellent accuracy - and in the case of Wang et al good repeatability - for translation measurement. However, they involved greater radiation dose and expense, while Yeager et al found good repeatability for pooled vertebral levels using a less elaborate low-dose 2-D clinical QF system, but did not assess levels individually [5, 18].

The validation of QF technology for *in vivo* translation and FCR measurement from continuous motion sequences is therefore incomplete. The aim of this study was to determine the current accuracy and repeatability of 2-D QF for measuring lumbar inter-
vertebral translation and FCR location during motion using a standardised patient motion protocol. This research involved the use of two calibrated human cadaveric specimens to assess accuracy during sagittal plane motion in a prescribed pathway and repeatability in twenty volunteers executing a standardised bending protocol.

**Methods**

**Accuracy study**

Two sets of dry cadaveric vertebral pairs were used to provide reference data. Specimen A (Fig 1A) consisted of L4 and L5 vertebrae joined at their end-plate centres by a universal joint 4mm high, representing a fixed centre of rotation with zero translation. Specimen B (Fig 1B) comprised of L3 and L4 vertebrae. These were joined at their end-plate centres by a plastic linkage which allowed translation of the upper vertebra without rotation. It was driven by an actuator motor and controller (Arduino Software Ltd. UK – resolution 0.01mm) providing anterior to posterior translation across the lower vertebral end-plate during the rotation.

Both specimens were mounted on rigid bases and positioned 15 cm from a motion frame which incorporated a rotating disc (Fig 1 A and B). The central ray of a C-arm digital fluoroscope (Siemens Arcadis Avantic – Siemens GMBH, Germany) was positioned so as to pass through the centre of the disc space. A block of animal soft tissue was interposed between the X-ray source, the models and the fluoroscope’s image intensifier to degrade the images by generating soft tissue scatter.

The superior vertebra of specimen A was rotated to 18° of flexion and return representing an arbitrary physiological maximum measured using a tilt sensor (Axminster instruments UK– resolution +/- 0.002 degrees) [19]. This was done using a rod driven by a vertical rotating disc embedded in a vertical motion frame (Fig 1A). It was controlled and driven by a laptop computer using bespoke software (Daqfactory VSC – Heatherose Electronics Ltd. UK). The superior vertebra of Specimen B was translated posteriorly across 50% of the lower vertebral end-plate and back again. This was an arbitrary range designed to allow direct comparison between the reference and index values, which should apply, within reason, no matter how large or small the translation. Rotation was at 3°/sec and translation at 1.5mm/sec. These procedures were repeated 10 times for each specimen. Images were
recorded at 15 frames per second during the 10 sequences for each specimen. All image
sequences were analysed by one trained observer.

**Repeatability study**

Data were obtained from a parallel study of twenty volunteers being examined for passive
recumbent lumbar motion [9]. These were recruited using the eligibility criteria described in
Table 1 and following a favourable opinion from the National Research Ethics Service (REC
reference 0/H0502/99). Each participant was positioned in the lateral decubitus position on
a horizontal motion frame with the central ray of the fluoroscope positioned to pass through
the L4 vertebra (Fig 2). The inferior section of the motion frame was rotated through 40° of
flexion over a 12 second interval using the motion controller (Daqfactory VSC – Heatherose
Electronics Ltd, UK). This was immediately followed by 40° of extension. The effective
radiation dose for this procedure has been estimated as 0.24mSv [18].

After transfer of images from the fluoroscope to an image processing workstation, two
trained observers (a senior radiographer and a medical physicist) analysed the same 40
image sequences for inter-observer repeatability (two sequences per participant for the 20
participants). Five repeated mark-ups of flexion and extension images of intervertebral
levels from L2-S1 took approximately 20 minutes. Observers were blinded to each other’s
image registrations. The second observer also analysed each image sequence twice for
intra-observer repeatability.

**Kinematic data extraction**

The fluoroscopic sequences were transferred to a desktop computer and Image J (v 1.47 for
Windows OS) was used to separate the individual images from the digital sequences. The
images underwent user defined edge enhancement, after which templates were manually
placed five times around each vertebral body (L2–S1) in the first image. Bespoke software
written in Matlab (V R2007b, The Mathworks Inc.) used a cross-correlation method to obtain
automated frame to frame image tracking of the vertebral bodies in subsequent images [20].
Co-ordinates were placed on the vertebral body corners in the first image, linked to the
tracking templates and used to register the vertebrae in two dimensional space in each
Tracking was verified for quality assurance by viewing all sequences and repeating any tracking that failed.

The displacements between each pair of tracked positions were calculated using Distortion Compensated Radiographic Analysis [21]. These were averaged over 25 registration combinations and output as data series. (Fig 3). Each data series was inspected for tracking failure using video playback. Any failed tracking data were removed and if all templates failed, the data were not used in the analysis.

Translation calculation

Frobins method [21] for calculating translation (shown in Figures 4 and 5 A & B) is based on landmarks identified on the vertebral body ‘corners’. Vertebral midlines (Fig. 4) are defined as lines passing through the midpoints between corners 1-2 and 3-4 respectively.

Using the method depicted in Figure 5, a line is drawn from the centre of each vertebra to the coinciding bisectrix. These lines intersect the bisectrix at 90 degrees to the bisectors’ gradient.

Translation was calculated as the distance along the bisectrix between the points at which these two lines independently cross the bisectrix (Fig 5). To standardise this measurement this is given as a proportion of the mean vertebral body depth of the superior vertebra, where 1 VBU (vertebral body unit) is the mean of the upper and lower vertebral body end plate depth of the superior vertebra. For the in vivo studies VBU’s were converted to millimetres based on a standard vertebral depth of 35mm and for the specimens by their actual measurement.

FCR calculation
The FCR position and distance from the posterior superior corner of the inferior vertebral body was calculated by finding the least squares solution between the four corners and the corresponding co-ordinates on the subsequent image [22] (Fig 5 A and B).

The four corner reference template positions for two adjacent vertebrae were taken and repositioned so that the inferior vertebral position was superimposed. From these coordinate positions, the centre of rotation between the two images was calculated by finding the least squares solution between each of the four corners and their partners from the second image. The least squares solution was taken as described by McCane et al [22] which gives the Matlab script used to execute this calculation. The positions at which each of these least squares solutions meet was taken as the FCR for those two vertebrae between those two images. The axis of rotation was then displayed relative to the inferior vertebra in a pair as a function of the four-corner template on the inferior vertebra. The superior-posterior corner of the inferior vertebra was taken as the origin for this reference field where the X-axis is along the template on the superior vertebral border and the Y-axis perpendicular to the X-axis passing through the origin. The unit of distance used was the proportion of the average vertebral body depth of superior vertebra (due to the non-uniform shape of the sacral template) where the origin of this co-ordinate system is the anterior-superior corner of the inferior vertebra.

FCR positional data were calculated at the maximum rotation angle between any two template positions where the inter-vertebral angle was greater than 5 degrees as a cut-off - as when intervertebral rotation interval decreases, the variation in FCR position increases. This is a systematic error due to the way in which the FCR positions are calculated. FCR was measured continuously between the first frame of the image sequence and the image frame where angular rotation was at its maximum +/- 0.5°. The limit of +/- 0.5° was selected as this was the increment through which the tracking templates rotated when calculating vertebral body position within each image. The results were taken as the average position of the FCR in X and Y co-ordinates over the 5 trackings.

Statistical analysis

For the accuracy study, 10 sets of markings were performed for each specimen. Measured translation was compared with zero translation reference data in the fixed centre specimen (end plate depth 28.77mm) and with translation across 50% of the inferior end plate (depth 34.66mm) in the moveable centre specimen. Disagreement was expressed as the root-
mean-square (RMS) differences between measured and reference values for both translation and FCR. 95% limits of agreement (LoA) were calculated and expressed in VBU [23].

For the repeatability studies, 4 intervertebral levels (L2-S1) were analysed for both flexion and extension translation for each of the 20 participants. For FCR location, data were removed from FCR analysis when rotation did not reach 5°. This range has been suggested as the lowest over which intervertebral FCRs should be calculated from radiographs without unacceptable error [24]. Therefore, in anticipation that not all levels would reach the necessary 5°, the levels were pooled to give a maximum possible 80 observations for each of flexion and extension. Intra and inter-observer reliability were expressed as intraclass correlation coefficients (ICC_{consistency,3,1}) using adjectives proposed by Shrout and Fleiss and revised from the original scale of Landis and Koch [25, 26]. In the Shrout and Fleiss scale, reliability as denoted by an ICC of 0.00-0.01 is considered as “virtually none”, 0.11-0.40 “slight”, 0.41-0.60 “fair”, 0.61-0.80 “moderate” and 0.81-1.00 “substantial”.

Results

Accuracy

The proportion of vertebral body depth that was translated in the moveable centre specimen as measured by the actuator motor was 0.52 VBU (17.95mm). Table 2 shows the RMS differences and 95% LoAs between the reference and measured translation and FCR locations.

For the fixed centre of rotation specimen, the average discrepancy (RMS) in translation range between reference and image data was 0.004 VBU (0.10mm) (LoA 0.01mm). For the translating specimen, the discrepancy when the superior vertebra was translated across 50% of the end-plate of the lower one was 0.062 VBU (2.16mm) (LoA 0.52mm). For FCR, the RMS x and y co-ordinate location differences between the reference and measured locations in the fixed centre specimen were 0.009 VBU(x) or 0.25mm (LoA 1.30mm) and for 0.014 VBU(y) or 0.40mm (LoA 1.20mm). (Table 2). Bland-Altman plots for these are shown in Fig 7 (A-D).

Fig 7 about here
Repeatability

The participant sample was made up of 9 females and 11 males aged 26 to 46 (mean age 35.7, SD 7.20). Their mean body mass index was 24.71 (SD 2.22).

Between 6 and 14 observations for each level in the 20 subjects were visible and tracked successfully for translation. Not all levels and directions were visible or trackable in all subjects. Artefacts due to the movement of bowel gas across images and tall patients whose upper vertebral levels did not fit the image field) were the main causes of this. Intra and inter-observer repeatability for each intervertebral level are shown in Table 3. All levels and directions showed at least fair agreement and reliability. The best agreement was between observers at L2-3 in extension (SEM=0.17mm) and the worst within observers at L5-S1 in extension (SEM=1.14mm). The best reliability was within observers at L2-3 in flexion ((ICC=0.998 (0.958-0.997)) and the worst within observers at L3-4 in flexion ((ICC=0.533 (0.406-0.849)).

Repeatability results for FCR are shown in Table 4. Five degrees of rotation was reached by 30 intervertebral pairs. For both translation and FCR location, within observer disagreement did not exceed 2mm for either flexion or extension. Inter-observer disagreement was high for FCRy in extension (5.67mm). All directions otherwise showed moderate to substantial reliability, the smallest ICC being 0.621 (0.429-0.813) for FCRx flexion between observers.

Discussion

Where mechanical impairment of intervertebral motion in the spine is at issue, its assessment will depend on the availability of technology with which to perform standardised measurements in patients during motion and to provide reference values and error estimates for the various parameters. This study is the first to assess the accuracy and level by level repeatability of the measurement of sagittal plane translation and FCR location from moving vertebral images using low dose 2-D QF. Its results indicate where the current strengths and weaknesses in the technique lie when reporting results of patient studies to clinicians.

The accuracy of techniques for radiographic measurement of intervertebral kinematics has been determined using calibration models for roentgen stereophotogrammetry, (which although highly invasive, is sometimes considered the gold standard), biplanar radiography
and QF [10, 15, 27, 28]. In this study, idealised conditions were also avoided by degrading the images with animal soft tissue and in the upright position, although it is not uncommon for such studies to be undertaken with no loading or in an animal model with no tissue degradation [16, 29, 30].

In this study, we compensated for radiographic image distortion using distortion-compensated roentgen analysis and used an image intensifier that incorporated automatic distortion correction [21]. Measurement is virtually independent of distortion of the radiographic image resulting from central projection, axial rotation, lateral tilt, and off-centre position with an error for translation of between 0.4 and 0.8 mm. Measurement of translation was determined from the vertebral body centres, making it independent of rotation. Previous QF studies have also shown that degrading the alignment by axially rotating it 10° out of plane and inclining the X-ray beam inclined 10° inferiorly results in minimal loss of accuracy in rotational studies [10]. Thus the technique should be sufficiently accurate to give useful information about ranges and motion patterns. However, this technique is not thought to be possible in scoliotic spines due to failure of image tracking.

This study found the current QF method to have fair to substantial repeatability for all levels and directions using the current protocol. It also found acceptable accuracy in vitro for the measurement of FCR location and translation during continuous spinal motion. Reliability was mainly good, but at some levels and directions suggests that training and quality assurance are needed when applying the measurement to comparisons between individuals and reference standards [31].

The inter-observer y-error in determination of FCR in extension (5.67 mm) and the intra-observer ICC (0.644) for extension translation at L5-S1 point to a need for caution. Closer inspection of the data revealed that the former was also greatest at L5-S1, where image quality and consequently co-ordinate placement may be rendered problematical by the super-imposition of the ilia and/or lack of perfect orthogonal alignment of the central X-ray beam with the vertical axis of the vertebrae. Previous work found radiographic positioning to be more important than tracking accuracy as a contributor to the variability in measurement of angular position, but that this does not preclude high repeatability and accuracy of measurement of rotation [19, 48]. However, for translation and FCR this may be more critical.

FCR was once thought to be promising as a way of assessing abnormal loading during intervertebral motion in patients [32, 33] but fell out of favour owing to high errors in measurement and the intrinsic computational errors that occur when rotational range is low.
The suggestion that it might be used to measure stability has therefore also not generally been taken up [14]. However, the present study has shown that despite the use of continuous motion data, as is necessary in patient studies, greater accuracy was achieved for determining the FCR (average error 0.3mm$_x$, 0.4mm$_y$) than was found in a previous study with such a specimen that used stepped rotation positions (average error 2mm)[15].

The repeatability study utilised information from participants undergoing passive recumbent and not weight bearing motion. It may be thought that weight bearing information would have been preferable to study the repeatability of translation and FCR measurement. However, this would have meant irradiating additional participants to obtain the same data and differences in motion patterns associated with weight bearing should not affect their measurement. Indeed, Wood concluded that the lateral decubitus position was superior for the detection of instability in patients with spondylolisthesis and Yeager et al used these interchangeably for their repeatability analysis of rotation and translation at pooled levels [37][5].

FCR, at least in the sagittal plane, could therefore be used to inform both patient care and patient-specific mathematical models. However, further studies are needed to establish normative *in vivo* reference standards at individual levels using QF. It would also be beneficial to explore the effects of spinal geometry and muscle contraction on FCR location, to add coronal plane validation and to confirm whether the FCR locus might be used to assess relationships between structural change and the *in vivo* biomechanical performance characteristics of discs under load. Finally, rotational cut-offs for accurately locating the FCR should be revisited in the light of the greater standardisation offered by QF protocols.

Diagnostic advances in spine biomechanics have also been made using kinetic MRI [37-41] and SPECT-CT imaging [42, 43]. However, although kinetic MRI locates points of encroachment on neural tissues and SPECT-CT contributes to the identification of potential sites of pain generation, neither can extract end-range or continuous inter-vertebral motion. In addition, the radiation dosage from SPECT-CT is considerably larger than that of QF.

Improvements in repeatability and accuracy are ongoing requirements for any diagnostic test, which means that reference standards will always be imperfect. Validation of QF will therefore require that scientists and practitioners also examine the extent to which test results are meaningful in practice [44]. This may be appreciated from patient register data. In parallel with this, technology development should address any measurement deficiencies.

**Limitations**
Participants with a BMI over 31 or aged over 51 were excluded from the study and none had osteoporosis, osteoarthritic change, vertebral deformities or curvatures; which may precipitate tracking failures. In the accuracy study, the translation error was considerably higher (2.10mm) in the translating specimen than in the fixed specimen (0.10mm). This may have been due to the resolution of the actuator motor in the latter (0.01mm), or by a small amount of out of plane motion due to imperfections in the mechanical linkage of this specimen. However, this discrepancy is well below the generally accepted cut-off of 4mm for excessive translation [45-48].

Distortion that changes during motion is not correctable if the templates that track the images from frame to frame do not change to accommodate it. In the future, this could be provided by adaptations to the tracking codes [8]. The US versions of this technology image the upper and lower lumbar levels separately to minimise out of plane images and ensure inclusion of all lumbar levels. While this increases the X-ray dose, it also makes for better reliability in the measurement of translation than was found here [5].

Future studies of accuracy and repeatability are needed to substantiate the present work. These could use a larger number of examiners, a range of rotational angles for FCR accuracy and a more elaborate calibration set up that combines rotation and translation. A larger number of human participants would overcome the problem of low angles of rotation and enable determination of the level by level repeatability of FCR location at 5° and above. For example, poorer agreement was found at L5-S1 than other levels, possibly owing to lower image quality resulting from superimposition of both ilia on the vertebral images.

**Conclusion**

Quantitative fluoroscopy was found to have a high level of accuracy as well as moderate to substantial observer agreement and reliability for the measurement of FCR and translation. Exceptions were in the reliability of measuring translation at L3-4 and agreement between observers in locating the FCR in extension. The development of reference standards and analysis quality assurance measures will be essential for optimal clinical use [6].
List of Figures

Figure 1. Lumbar intervertebral motion specimens. (A) Fixed centre specimen (B) Movable centre specimen

Figure 2. Diagram of patient positioning for fluoroscopic imaging (Ortho Kinematics Inc., with permission)

Figure 3. Example of translation data for extension at L5-S1 (live participant). Solid line shows filtered average of 25 trackings. Shaded area represents all data.

Figure 4. Graphical representation of two lumbar vertebrae undergoing extension in the sagittal plane with a four-point reference template marked on the corner of each vertebra to calculate the bisectrix. The bisectrix is to be used as a basis of calculation of translation changes.

Figure 5 A and B. Depiction of translation measurement calculation between two adjacent lumbar vertebrae in (A) full extension (B) full flexion

Figure 6 A and B. Examples of computer-generated measurements of: (A) FCR in fixed centre specimen (B) translation in movable centre specimen

Figure 7 A to D. Bland-Altman plots: (A) Translation in fixed centre specimen (B) Translation in movable centre specimen (C) FCRx in fixed centre specimen (D) FCRy in fixed centre specimen
Acknowledgements

This article presents independent research and the views expressed are those of the author(s) alone. The authors wish to acknowledge the contributions of the participants and of Fiona Mellor for sharing some of her participant image sequences and contributing to the participant image registration.

Competing Interests

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Accuracy and repeatability of quantitative fluoroscopy for the measurement of sagittal plane translation and finite centre of rotation in the lumbar spine

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Abstract

Quantitative fluoroscopy (QF) was developed to measure intervertebral mechanics in vivo and has been found to have high repeatability and accuracy for the measurement of intervertebral rotations. However, sagittal plane translation and finite centre of rotation (FCR) are potential measures of stability but have not yet been fully validated for current QF. This study investigated the repeatability and accuracy of QF for measuring these variables. Repeatability was assessed from L2-S1 in 20 human volunteers. Accuracy was investigated using 10 consecutive measurements from each of two pairs of linked and instrumented dry human vertebrae as reference; one which tilted without translation and one which translated without tilt. The results found intra- and inter-observer repeatability for translation to be 1.1mm or less (SEM) with fair to substantial reliability (ICC 0.533-0.998). Intra-observer repeatability of FCR location for inter-vertebral rotations of 5° and above ranged from 1.5mm to 1.8mm (SEM) with moderate to substantial reliability (ICC 0.626-0.988). Inter-observer repeatability for FCR ranged from 1.2mm to 5.7mm, also with moderate to substantial reliability (ICC 0.621-0.878). Reliability was substantial (ICC>0.81) for 10/16 measures for translation and 5/8 for FCR location. Accuracy for translation was 0.1mm (fixed centre) and 2.2mm (moveable centre), with an FCR error of 0.3mm(x) and 0.4mm(y) (fixed centre). This technology was found to have a high level of accuracy and with a few exceptions, moderate to substantial repeatability for the measurement of translation and FCR from fluoroscopic motion sequences.
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Studies of quantitative fluoroscopy (QF) for measuring lumbar spine intervertebral kinematics using continuous motion tracking began in the 1980s [7]. QF measures continuous intervertebral motion and extracts end of range measurement from wherever it occurs in the bending sequence, giving a radiation dose similar to a conventional radiographic examination [8, 9]. Various iterations have been found to have good repeatability and accuracy for measuring intervertebral rotations at lumbar and cervical levels [5, 9-12]. However, excessive translation is thought to be more closely associated with back symptoms [13]. Translation also affects the finite centre of rotation (FCR) and the latter is an expression of the distribution of loading between the disc and facets during upright flexion-extension motion [14]. It is also said that the centre of reaction force (CR) can be extrapolated from the FCR [14].

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recorded at 15 frames per second during the 10 sequences for each specimen. All image sequences were analysed by one trained observer.

Repeatability study

Data were obtained from a parallel study of twenty volunteers being examined for passive recumbent lumbar motion [9]. These were recruited using the eligibility criteria described in Table 1 and following a favourable opinion from the National Research Ethics Service (REC reference 0/H0502/99). Each participant was positioned in the lateral decubitus position on a horizontal motion frame with the central ray of the fluoroscope positioned to pass through the L4 vertebra (Fig 2). The inferior section of the motion frame was rotated through 40° of flexion over a 12 second interval using the motion controller (Daqfactory VSC – Heatherose Electronics Ltd, UK). This was immediately followed by 40° of extension. The effective radiation dose for this procedure has been estimated as 0.24mSv [18].

Table 1 about here

Fig 2 about here

After transfer of images from the fluoroscope to an image processing workstation, two trained observers (a senior radiographer and a medical physicist) analysed the same 40 image sequences for inter-observer repeatability (two sequences per participant for the 20 participants). Five repeated mark-ups of flexion and extension images of intervertebral levels from L2-S1 took approximately 20 minutes. Observers were blinded to each other’s image registrations. The second observer also analysed each image sequence twice for intra-observer repeatability.

Kinematic data extraction

The fluoroscopic sequences were transferred to a desktop computer and Image J (v 1.47 for Windows OS) was used to separate the individual images from the digital sequences. The images underwent user defined edge enhancement, after which templates were manually placed five times around each vertebral body (L2–S1) in the first image. Bespoke software written in Matlab (V R2007b, The Mathworks Inc.) used a cross-correlation method to obtain automated frame to frame image tracking of the vertebral bodies in subsequent images [20]. Co-ordinates were placed on the vertebral body corners in the first image, linked to the tracking templates and used to register the vertebrae in two dimensional space in each
frame. Tracking was verified for quality assurance by viewing all sequences and repeating any tracking that failed.

The displacements between each pair of tracked positions were calculated using Distortion Compensated Radiographic Analysis [21]. These were averaged over 25 registration combinations and output as data series’ (Fig 3). Each data series was inspected for tracking failure using video playback. Any failed tracking data were removed and if all templates failed, the data were not used in the analysis.

**Translation calculation**

Frobins method [21] for calculating translation (shown in Figures 4 and 5 A & B) is based on landmarks identified on the vertebral body ‘corners’. Vertebral midlines (Fig. 4) are defined as lines passing through the midpoints between corners 1-2 and 3-4 respectively.

The average gradient and y axis crossover of the two midlines are calculated for a vertebral pair. The resultant line is called the bisectrix and normally passes through the inter-vertebral disc space.

Using the method depicted in Figure 5, a line is drawn from the centre of each vertebra to the coinciding bisectrix. These lines intersect the bisectrix at 90 degrees to the bisectors’ gradient.

Translation was calculated as the distance along the bisectrix between the points at which these two lines independently cross the bisectrix (Fig 5). To standardise this measurement this is given as a proportion of the mean vertebral body depth of the superior vertebra, where 1 VBU (vertebral body unit) is the mean of the upper and lower vertebral body end plate depth of the superior vertebra. For the *in vivo* studies VBU's were converted to millimetres based on a standard vertebral depth of 35mm and for the specimens by their actual measurement.

**FCR calculation**
The FCR position and distance from the posterior superior corner of the inferior vertebral body was calculated by finding the least squares solution between the four corners and the corresponding co-ordinates on the subsequent image [22] (Fig 5 A and B).

The four corner reference template positions for two adjacent vertebrae were taken and repositioned so that the inferior vertebral position was superimposed. From these coordinate positions, the centre of rotation between the two images was calculated by finding the least squares solution between each of the four corners and their partners from the second image. The least squares solution was taken as described by McCane et al [22] which gives the Matlab script used to execute this calculation. The positions at which each of these least squares solutions meet was taken as the FCR for those two vertebrae between those two images. The axis of rotation was then displayed relative to the inferior vertebra in a pair as a function of the four-corner template on the inferior vertebra. The superior-posterior corner of the inferior vertebra was taken as the origin for this reference field where the X-axis is along the template on the superior vertebral border and the Y-axis perpendicular to the X-axis passing though the origin. The unit of distance used was the proportion of the average vertebral body depth of superior vertebra (due to the non-uniform shape of the sacral template) where the origin of this co-ordinate system is the anterior-superior corner of the inferior vertebra.

FCR positional data were calculated at the maximum rotation angle between any two template positions where the inter-vertebral angle was greater than 5 degrees as a cut-off - as when intervertebral rotation interval decreases, the variation in FCR position increases. This is a systematic error due to the way in which the FCR positions are calculated. FCR was measured continuously between the first frame of the image sequence and the image frame where angular rotation was at its maximum +/- 0.5°. The limit of +/- 0.5° was selected as this was the increment through which the tracking templates rotated when calculating vertebral body position within each image. The results were taken as the average position of the FCR in X and Y co-ordinates over the 5 trackings.

**Statistical analysis**

For the accuracy study, 10 sets of markings were performed for each specimen. Measured translation was compared with zero translation reference data in the fixed centre specimen (end plate depth 28.77mm) and with translation across 50% of the inferior end plate (depth 34.66mm) in the moveable centre specimen. Disagreement was expressed as the root-
mean-square (RMS) differences between measured and reference values for both translation and FCR. 95% limits of agreement (LoA) were calculated and expressed in VBU [23].

For the repeatability studies, 4 intervertebral levels (L2-S1) were analysed for both flexion and extension translation for each of the 20 participants. For FCR location, data were removed from FCR analysis when rotation did not reach 5°. This range has been suggested as the lowest over which intervertebral FCRs should be calculated from radiographs without unacceptable error [24]. Therefore, in anticipation that not all levels would reach the necessary 5°, the levels were pooled to give a maximum possible 80 observations for each of flexion and extension. Intra and inter-observer reliability were expressed as intraclass correlation coefficients (ICC_{consistency,3,1}) using adjectives proposed by Shrout and Fleiss and revised from the original scale of Landis and Koch [25, 26]. In the Shrout and Fleiss scale, reliability as denoted by an ICC of 0.00-0.01 is considered as “virtually none”, 0.11-0.40 “slight”, 0.41-0.60 “fair”, 0.61-0.80 “moderate” and 0.81-1.00 “substantial”.

Results

Accuracy

The proportion of vertebral body depth that was translated in the moveable centre specimen as measured by the actuator motor was 0.52 VBU (17.95mm). Table 2 shows the RMS differences and 95% LoAs between the reference and measured translation and FCR locations.

For the fixed centre of rotation specimen, the average discrepancy (RMS) in translation range between reference and image data was 0.004 VBU (0.10mm) (LoA 0.01mm). For the translating specimen, the discrepancy when the superior vertebra was translated across 50% of the end-plate of the lower one was 0.062 VBU (2.16mm) (LoA 0.52mm). For FCR, the RMS x and y co-ordinate location differences between the reference and measured locations in the fixed centre specimen were 0.009 VBU(x) or 0.25mm (LoA 1.30mm) and for 0.014 VBU(y) or 0.40mm (LoA 1.20mm). (Table 2). Bland-Altman plots for these are shown in Fig 7 (A-D).
**Repeatability**

The participant sample was made up of 9 females and 11 males aged 26 to 46 (mean age 35.7, SD 7.20). Their mean body mass index was 24.71 (SD 2.22).

Between 6 and 14 observations for each level in the 20 subjects were visible and tracked successfully for translation. Not all levels and directions were visible or trackable in all subjects. Artefacts due to the movement of bowel gas across images and tall patients whose upper vertebral levels did not fit the image field) were the main causes of this. Intra and inter-observer repeatability for each intervertebral level are shown in Table 3. All levels and directions showed at least fair agreement and reliability. The best agreement was between observers at L2-3 in extension (SEM=0.17mm) and the worst within observers at L5-S1 in extension (SEM=1.14mm). The best reliability was within observers at L2-3 in flexion ((ICC=0.998 (0.958-0.997)) and the worst within observers at L3-4 in flexion ((ICC=0.533 (0.406-0.849)).

Table 3 about here

Repeatability results for FCR are shown in Table 4. Five degrees of rotation was reached by 30 intervertebral pairs. For both translation and FCR location, within observer disagreement did not exceed 2mm for either flexion or extension. Inter-observer disagreement was high for FCRY in extension (5.67mm). All directions otherwise showed moderate to substantial reliability, the smallest ICC being 0.621 (0.429-0.813) for FCRX flexion between observers.

Table 4 about here

**Discussion**

Where mechanical impairment of intervertebral motion in the spine is at issue, its assessment will depend on the availability of technology with which to perform standardised measurements in patients during motion and to provide reference values and error estimates for the various parameters. This study is the first to assess the accuracy and level by level repeatability of the measurement of sagittal plane translation and FCR location from moving vertebral images using low dose 2-D QF. Its results indicate where the current strengths and weaknesses in the technique lie when reporting results of patient studies to clinicians.

The accuracy of techniques for radiographic measurement of intervertebral kinematics has been determined using calibration models for roentgen stereophotogrammetry, (which although highly invasive, is sometimes considered the gold standard), biplanar radiography
and QF [10, 15, 27, 28]. In this study, idealised conditions were also avoided by degrading the images with animal soft tissue and in the upright position, although it is not uncommon for such studies to be undertaken with no loading or in an animal model with no tissue degradation [16, 29, 30].

In this study, we compensated for radiographic image distortion using distortion-compensated roentgen analysis and used an image intensifier that incorporated automatic distortion correction [21]. Measurement is virtually independent of distortion of the radiographic image resulting from central projection, axial rotation, lateral tilt, and off-centre position with an error for translation of between 0.4 and 0.8mm. Measurement of translation was determined from the vertebral body centres, making it independent of rotation. Previous QF studies have also shown that degrading the alignment by axially rotating it 10° out of plane and inclining the X-ray beam inclined 10° inferiorly results in minimal loss of accuracy in rotational studies [10]. Thus the technique should be sufficiently accurate to give useful information about ranges and motion patterns. However, this technique is not thought to be possible in scoliotic spines due to failure of image tracking.

This study found the current QF method to have fair to substantial repeatability for all levels and directions using the current protocol. It also found acceptable accuracy in vitro for the measurement of FCR location and translation during continuous spinal motion. Reliability was mainly good, but at some levels and directions suggests that training and quality assurance are needed when applying the measurement to comparisons between individuals and reference standards [31].

The inter-observer y-error in determination of FCR in extension (5.67mm) and the intra-observer ICC (0.644) for extension translation at L5-S1 point to a need for caution. Closer inspection of the data revealed that the former was also greatest at L5-S1, where image quality and consequently co-ordinate placement may be rendered problematical by the super-imposition of the ilia and/or lack of perfect orthogonal alignment of the central X-ray beam with the vertical axis of the vertebrae. Previous work found radiographic positioning to be more important than tracking accuracy as a contributor to the variability in measurement of angular position, but that this does not preclude high repeatability and accuracy of measurement of rotation [19, 48]. However, for translation and FCR this may be more critical.

FCR was once thought to be promising as a way of assessing abnormal loading during intervertebral motion in patients [32, 33] but fell out of favour owing to high errors in measurement and the intrinsic computational errors that occur when rotational range is low.
The suggestion that it might be used to measure stability has therefore also not generally been taken up [14]. However, the present study has shown that despite the use of continuous motion data, as is necessary in patient studies, greater accuracy was achieved for determining the FCR (average error 0.3mm$_x$, 0.4mm$_y$) than was found in a previous study with such a specimen that used stepped rotation positions (average error 2mm)[15].

The repeatability study utilised information from participants undergoing passive recumbent and not weight bearing motion. It may be thought that weight bearing information would have been preferable to study the repeatability of translation and FCR measurement. However, this would have meant irradiating additional participants to obtain the same data and differences in motion patterns associated with weight bearing should not affect their measurement. Indeed, Wood concluded that the lateral decubitus position was superior for the detection of instability in patients with spondylolisthesis and Yeager et al used these interchangeably for their repeatability analysis of rotation and translation at pooled levels [37][5].

FCR, at least in the sagittal plane, could therefore be used to inform both patient care and patient-specific mathematical models. However, further studies are needed to establish normative in vivo reference standards at individual levels using QF. It would also be beneficial to explore the effects of spinal geometry and muscle contraction on FCR location, to add coronal plane validation and to confirm whether the FCR locus might be used to assess relationships between structural change and the in vivo biomechanical performance characteristics of discs under load. Finally, rotational cut-offs for accurately locating the FCR should be revisited in the light of the greater standardisation offered by QF protocols.

Diagnostic advances in spine biomechanics have also been made using kinetic MRI [37-41] and SPECT-CT imaging [42, 43]. However, although kinetic MRI locates points of encroachment on neural tissues and SPECT-CT contributes to the identification of potential sites of pain generation, neither can extract end-range or continuous inter-vertebral motion. In addition, the radiation dosage from SPECT-CT is considerably larger than that of QF.

Improvements in repeatability and accuracy are ongoing requirements for any diagnostic test, which means that reference standards will always be imperfect. Validation of QF will therefore require that scientists and practitioners also examine the extent to which test results are meaningful in practice [44]. This may be appreciated from patient register data. In parallel with this, technology development should address any measurement deficiencies.

Limitations
Participants with a BMI over 31 or aged over 51 were excluded from the study and none had osteoporosis, osteoarthritic change, vertebral deformities or curvatures; which may precipitate tracking failures. In the accuracy study, the translation error was considerably higher (2.10mm) in the translating specimen than in the fixed specimen (0.10mm). This may have been due to the resolution of the actuator motor in the latter (0.01mm), or by a small amount of out of plane motion due to imperfections in the mechanical linkage of this specimen. However, this discrepancy is well below the generally accepted cut-off of 4mm for excessive translation [45-48].

Distortion that changes during motion is not correctable if the templates that track the images from frame to frame do not change to accommodate it. In the future, this could be provided by adaptations to the tracking codes [8]. The US versions of this technology image the upper and lower lumbar levels separately to minimise out of plane images and ensure inclusion of all lumbar levels. While this increases the X-ray dose, it also makes for better reliability in the measurement of translation than was found here [5].

Future studies of accuracy and repeatability are needed to substantiate the present work. These could use a larger number of examiners, a range of rotational angles for FCR accuracy and a more elaborate calibration set up that combines rotation and translation. A larger number of human participants would overcome the problem of low angles of rotation and enable determination of the level by level repeatability of FCR location at 5° and above. For example, poorer agreement was found at L5-S1 than other levels, possibly owing to lower image quality resulting from superimposition of both ilia on the vertebral images.

**Conclusion**

Quantitative fluoroscopy was found to have a high level of accuracy as well as moderate to substantial observer agreement and reliability for the measurement of FCR and translation. Exceptions were in the reliability of measuring translation at L3-4 and agreement between observers in locating the FCR in extension. The development of reference standards and analysis quality assurance measures will be essential for optimal clinical use [6].
List of Figures

Figure 1. Lumbar intervertebral motion specimens. (A) Fixed centre specimen (B) Movable centre specimen

Figure 2. Diagram of patient positioning for fluoroscopic imaging (Ortho Kinematics Inc., with permission)

Figure 3. Example of translation data for extension at L5-S1 (live participant). Solid line shows filtered average of 25 trackings. Shaded area represents all data.

Figure 4. Graphical representation of two lumbar vertebrae undergoing extension in the sagittal plane with a four-point reference template marked on the corner of each vertebra to calculate the bisectrix. The bisectrix is to be used as a basis of calculation of translation changes.

Figure 5 A and B. Depiction of translation measurement calculation between two adjacent lumbar vertebrae in (A) full extension (B) full flexion

Figure 6 A and B. Examples of computer-generated measurements of: (A) FCR in fixed centre specimen (B) translation in movable centre specimen

Figure 7 A to D. Bland-Altman plots: (A) Translation in fixed centre specimen (B) Translation in movable centre specimen (C) FCRx in fixed centre specimen (D) FCRy in fixed centre specimen
Acknowledgements

This article presents independent research and the views expressed are those of the author(s) alone. The authors wish to acknowledge the contributions of the participants and of Fiona Mellor for sharing some of her participant image sequences and contributing to the participant image registration.

Competing Interests

The authors have performed research for the Ortho Kinematics Company, which is commercialising a version of this technology in the United States.

Sources of funding

No external funding was obtained for this research.

Ethical Approval

Ethical approval was given by the National Research Ethics Service (REC reference 0/H0502/99).
References


Figure legends

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Figure

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\[ M((Reference - Measured)^2) \]
Figure

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mean difference $t$ 95% TMris of agreement

Mean ((Reference /Afo/ - Me wred /Afo/J 12)
### Table 1. Participant inclusion and exclusion criteria for repeatability study

<table>
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<th>Inclusion criteria</th>
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<td>Male and female.</td>
<td>Pregnancy</td>
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<tr>
<td>Age 21–51 years.</td>
<td>Mental illness</td>
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<td>Able to understand written information.</td>
<td>Poor understanding of English</td>
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<td>Willing to participate</td>
<td>Recent abdominal or pelvic surgery.</td>
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<tr>
<td>Able to freely give informed consent.</td>
<td>Previous mid-lumbar spinal surgery</td>
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<td>Menstruation within last 28 days, or evidence of contraceptive use, or sterility (females).</td>
<td>Body mass index (BMI)&gt;31</td>
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<td>Consent to GP being informed of inclusion in study.</td>
<td>Medical radiation exposure in the past 2 years with a dose of greater than 8 mSv</td>
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<td>Able to tolerate 80 degrees of flexion–extension passive trunk motion</td>
<td>(defined as CT scan of chest, abdomen or pelvis or interventional procedures under</td>
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<td>radiological control, i.e. angiography).</td>
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<td>Current involvement in any other research study.</td>
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<td>Hyper-mobility syndrome</td>
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<td>Pathology such as fracture, infection, neoplasm.</td>
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<td>Spinal stenosis.</td>
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<td>Spondyololisthesis.</td>
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<td>Radicular pain.</td>
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<td>Litigation or compensation pending.</td>
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### Table 2. RMS differences between reference and measured translation and FCR locations

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### Table 3. Intra and interobserver repeatability of translation by level and direction

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<th>Interobserver n</th>
<th>SEM (mm)</th>
<th>ICC (95%CI)</th>
<th>Intraobserver n</th>
<th>SEM (mm)</th>
<th>ICC (95%CI)</th>
<th>Interobserver n</th>
<th>SEM (mm)</th>
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<tr>
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Table 4. Intra and interobserver repeatability of FCR location (pooled data)