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ORIGINAL PAPER

Radiological evaluation of the normal patella position using the Insall-Salvati ratio

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ABSTRACT

Introduction and aim. Normal patellofemoral relationship relies on the patella's location which is vital in the knee's stability and biomechanics. This research aimed to determine the normal Insall Salvati Ratio (ISR) and the cut-off values that will be useful for diagnosing patella alta and baja in Delta State in Nigeria.

Material and methods. This retrospective cross-sectional study assessed the Insall-Salvati Ratio by dividing the patella tendon length by the patella length. These lengths were measured on lateral knee radiographs of 300 patients (149 males, 151 females) aged 20 years and older using digital calipers calibrated in cm.

Results. With exception to the ISR, the measured variables showed sexual dimorphism and a significant weak negative association with age ($p<0.05$). Based on the international ISR cut-off values (<0.8 and >1.2), the prevalence of patella baja and alta was 15 (5%) and 64 (21.3%) respectively using the calculated cut-off values (<0.73 and >1.41). A lower prevalence of 6 (2%) and 9 (3%) were recorded correspondingly.

Conclusion. The normal ISR cut-offs provided by this study will aid radiologists and orthopedic specialists in Delta State, Nigeria to precisely diagnose patella alta and baja and ensure proper restoration of the knee's stability and biomechanics as well as minimizing complications.

Keywords. alta, baja, height, insall-salvati ratio, length, patella

Introduction

Among all joints, the knee is the most prone to injuries with a high tendency of natural degeneration.[1] The chief orthopedic complaint among physically active adults and adolescents is anterior knee pain.[2,3] The complex knee joint contains the patellofemoral joint which is the articulation between the femoral trochlea and a sesamoid bone called the patella.[4] This bone is an essential part of the knee's extensor mechanism, together with the patella retinaculum, quadriceps muscle and tendon, tibial tubercle and patella ligament.[5,6] The patellofemoral joint's stability depends on the congruence of articulating surfaces, patella tendon (PT)/quadriceps tendon and joint capsule. Patellofemoral pain occurs in 25% of all knee problems.[4,7] It has a multifactorial pathogenesis such as malalignment and maltracking which pre-

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dispose to osteoarthritis.[3] Patellofemoral malalignment can be managed by strengthening exercises, taping and orthoses.[7]

Normal patellofemoral relationship relies on the patella's location which is vital in the knee's stability and biomechanics.[8,9] Abnormal patella height is linked with patella instability, extensor mechanism disorders, patella arthritis, meniscal tears, patella chondromalacia, and anterior cruciate ligament (ACL) injuries.[1,10] Patella baja and patella alta are the most prevalent patella pathologies physicians and radiologist's encounter.[3]

Patella alta, typically described as an abnormally high riding patella is likely seen in patients with Osgood-Schlatter disease, patella chondromalacia, Sinding-Larsen-Johansson disease, patella ligament rupture, recurrent lateral patellar dislocation or subluxation, and patella tendonitis.[2,10] Patella alta with no instability may be regarded as a normal variant.[11] Patella baja is a low riding patella which can be congenital or acquired.[12] Congenital causes could be due to shortened PT, distal patella location within the femoral trochlear, and reduced distance from inferior patella pole to proximal tibial articular surface.[6] It may also signify neuromuscular conditions and achondroplasia.[2] Acquired patella baja can be due to patella fracture, and quadriceps tendon rupture subsequent to trauma or surgeries like high tibial osteotomy, retrograde femoral nailing, and anterior cruciate ligament (ACL) reconstruction.[6,8] Patella baja, further predisposes to lateral meniscal tears.[1]

Computed tomography (CT), conventional radiography, and magnetic resonance imaging (MRI) can be utilized in characterizing patella maltracking, and malalignment.[1] Conventional radiography is routinely used due to its availability and affordability, although, it has inherent magnification and limited visualization of the joint cartilage.[11] Three-dimensional visualization of the patella-trochlear cartilage overlap is clear on MRI hence, useful in assessing the patella ratios with no risk of exposure to radiation.[4,9] Nonetheless, MRI is expensive, not widely available, and also challenging to standardize the flexion angles of knee in a non- weight bearing position.[11]

Several indices from radiological measurements may be employed in patella height evaluation namely: Insall-Salvati, Blackburne-Pell, modified Insall-Salvati, Caton-Deschamps, and patellotrochlear index.[1,11] Insall-Salvati ratio (ISR) is regarded as a simple, precise, reproducible and reliable index applicable in all flexion degrees of the knee and uses identifiable patella and tibial landmarks.[10,13] It represents the proportion of the patella tendon's length (PTL) to patella's length, introduced by Insall and Salvati[14] from lateral radiographs with knees flexed at 30 degrees.[2] The maximum distance from the distal patella's pole to the tibial tuberosity defines the PTL while the patella's supero-inferior diagonal dimension is the patella length.[1] An ISR of <0.8 suggests patella baja while >1.2 implies patella alta.[14] There is an acceptable correlation between MRI and radiographic Insall –Salvati ratios.[1] The limitations of the ISR encompass the variant shape of the patella's inferior pole, inability to assess the tibial tuberosity, and the fact that measurements don't change after the tibial tubercle distalization.[11,15]

The ISR and the metric parameters of the patella vary in different study cohorts based on factors that impact knee morphology such as genetics, ethnicity, environmental and geographical factors.[2,3,5,8,16]

The patella's position is influenced by lifestyle and socio-cultural practices such as sitting on the floor, kneeling, yoga and squatting.[2,3] The ISR thresholds have previously been established based on previous investigations among Western populations that have distinct lifestyle and cultural patterns like chair sitting. However, these thresholds may not be applicable in other different genetic, racial, cultural and environmental settings.[3,4,8] This highlights the need for population specific cut-offs for diagnosing abnormal patella position.[2,3] Common Nigerian activities involve squatting, kneeling and floor-sitting which affect the patella height and largely the morphology of the knee. There is potential for patient misdiagnosis in Delta State Nigeria owing to scarcity of research to establish the thresholds for normal patella height in the region.

Aim

This study aimed at establishing the ISR thresholds for diagnosing patella baja and alta in Delta State, Nigeria. By developing population-specific diagnostic criteria, the research aimed at improving the accuracy of radiological diagnosis and classification of abnormal patella position. The cut-off values will also aid orthopedic surgeons in monitoring post-operative changes following procedures such as ACL reconstruction and high tibial osteotomy, where patella position is critical to long-term knee function.

Material and methods

This investigation was retrospectively conducted in a single radiological unit and the observational cross-sectional design was used. Lateral radiographic views of knees from 300 patients (149 males and 151 females) seen at the Radiology Department of a Tertiary Hospital in Delta State, Nigeria, from 2nd January, 2017 to 30th December, 2022 were assessed. The convenience sampling technique allowed the inclusion of all available radiographs taken within the specified time frame which fulfilled the selection criteria. The adopted sampling procedure provided a sufficient sample size although the findings can't be generalized to the whole Nigerian population.

The study therefore included patients of both sex groups aged 20 years and older. The exclusion of children was based on the presence of cartilaginous growth plates around joints due to incomplete ossification of bones.[2,17] The study excluded radiographs with evidence of previous knee surgery or knee pathologies namely: fractures, osteoarthritis, inflammatory arthritis, congenital anomalies, tumors, and dislocation. As this was a retrospective investigation, patients with clinical knee symptoms such as pain and abnormal gait could not be excluded. Technically inadequate lateral views with knee rotation or flexion less than 30 degrees were excluded. We did not calculate an estimated sample size. However, from the 794 lateral knee radiographs identified in the database, 300 were utilized, exceeding the sample size used in previous similar studies.[4,8] This was considered sufficient to generate reliable data for this study.

The protocol used was authorized by the Hospital's ethical board (DELSUTH/HREC/2024/004/0742). This board did not deem it necessary to obtain patient consent owing to the retrospective design of the study involving no contact with the patients. The study adhered to the ethical guidelines outlined in the Declaration of Helsinki. According to the center's radiography techniques, only the knee with suspected pathology was imaged to minimize exposure to radioactive rays. Therefore, bilateral measurements could not be obtained. To tense the PT, visualize the PT's tibial insertion and ensure imaging consistency, the lateral knee radiographs were acquired at a 30 degrees semiflexed knee position. The images were acquired 100 cm from the film and rays focused perpendicular to the joint.

Using a desktop computer, the selected images in the picture archiving communications systems were examined and the indicated age and gender of the patients were recorded. Using a digital ruler calibrated in centimeters (cm), a single investigator measured each metric variable three times, and the averages were documented to minimize inter- and intra-observer variability. To ensure reliability, repeatability of measurements was assessed through consistency checks during initial trials. The maximum supero-inferior distance from the proximal to distal patella poles diagonally was the patella length while the patella's tendon length was the maximum distance from the patella's inferior pole to tuberosity of tibia (Fig. 1).[1] The ratio of these two lengths; PTL to the patella length was calculated as the Insall-Salvati ratio.[14]

Based on the international ISR thresholds, patella height was classified as patella baja (ISR <0.8), normal (ISR 0.8-1.2) and patella alta (ISR >1.2).[14] Furthermore, we established cut-off values at two standard deviations above and below the mean ISR as described by Di et al.[3] Based on these, the prevalence of patella baja and alta were determined.

Fig. 1. Right lateral knee radiograph showing the patella tendon length (4.24 cm) and patella length (4.51cm)

Data gathered was explored utilizing Version 27.0 of the statistical package of social sciences (SPSS) software (Inc. Chicago, IL, USA). Descriptive statistics were presented in tables. The independent t-test compared the quantitative variables in the two sex groups while the analysis of variance (ANOVA) assessed how these variables differed among the age groups. The Pearson's correlation test was used to investigate the inter-variable associations. Significance level was fixed at $p<0.05$.

Results

The study assessed the ISR using 300 radiographs comprising 149 (49.7%) male patients and 151 (50.3%) female patients. The mean age and age-range of the patients was 43.82±15.81 years and 20-80 years respectively. The average age of males and females was 48.02±14.79 years and 39.57±15.72 years respectively. The age-group with the highest frequency of patients was the 21–30 years while oldest age group (71–80 years) had the least frequency (10, 3.3%) (Table 1).

Table 1. Patient distribution based on age-groups

Age group (years)	N (%)		
	Males	Females	Total Population
<20	9 (6)	7 (4.6)	16 (5.3)
21–30	47 (31.5)	17 (11.3)	64 (21.3)
31–40	35 (23.5)	24 (15.9)	59 (19.7)
41–50	18 (12.1)	32 (21.2)	50 (16.7)
51–60	19 (12.8)	36 (23.8)	55 (18.3)
61–70	15 (10.1)	31 (20.5)	46 (15.3)
71–80	6 (4)	4 (2.6)	10 (3.3)
Total	149 (100)	151 (100)	300 (100)

The descriptive statistics following the analysis of the lengths and ISR are depicted in Table 2. The patella and its tendon were significantly longer in males than

females and varied across age-groups (p<0.05). However, the ISR lacked sexual dimorphism and age-related variances (p=0.583, 0.690) (Tables 2 and Table 3).

Table 2. Descriptive statistics of the metric variables and their gender based comparison*

		PTL (cm)	PL (cm)	ISR
Male	Min	3.07	3.22	0.6
	Max	7.87	6.18	1.9
	Mean±SD	5.00±0.77	4.75±0.47	1.06±0.19
Female	Min	3.20	3.53	0.7
	Max	5.88	5.81	1.5
	Mean±SD	4.58±0.55	4.29±0.39 Eo	1.07±0.15
Total Population	Min	3.20	3.22	0.59
	Max	7.87	6.18	1.86
	Mean±SD	4.79 ±0.70	4.52 ±0.49	1.07 ± 0.17
p		0.001	0.001	0.583

* PTL – patella tendon length, PL – patella length, ISR – Insall-Salvati ratio, SD – standard deviation

Table 3. Differences in the variables based on age group*

Variables	Age (years)							p
	<20	21–30	31–40	41–50	51–60	61–70	71–80	
PTL	5.10±0.81	5.00±0.76	4.83±0.64	4.63±0.77	4.74±0.67	4.60±0.48	4.55±0.70	0.010
PL	4.56±0.38	4.68±0.56	4.58±0.43	4.40±0.51	4.45±0.41	4.42±0.50	4.55±0.52	0.025
ISR	1.13±0.21	1.08±0.20	1.06±0.17	1.06±0.17	1.07±0.15	1.06±0.16	1.01±0.14	0.690

* PTL – patella tendon length, PL – patella length, ISR – Insall-Salvati ratio

The PTL and the patella length showed a significant weak negative correlation with age and a significant weak positive association with one another (p<0.05). The ISR had a significant strong positive relationship with PTL and weak negative association with patella length respectively (p<0.05). The ISR lacked a significant association with age (p=0.117) (Table 4).

Table 4. Correlation between the quantitative variables*

		Age (years)	PTL (cm)	PL (cm)	ISR
Age (years)	r	1	-0.219*	-0.167*	-0.091
	P		0.001	0.004	0.117
PTL (cm)	r	-0.219*	1	0.210*	0.753*
	P	0.001		0.001	0.001
PL (cm)	r	-0.167*	0.210*	1	-.474*
	P	0.004	0.001		.001
ISR	r	-0.091	0.753*	-0.474*	1
	P	0.117	0.001	0.001	

* PTL – patella tendon length, PL – patella length, ISR – Insall-Salvati ratio, P- probability value, r – Pearson's correlation coefficient, *p considered significant at <0.05

Table 5 shows the percentage of patella variants classified based on cutoffs by Install and Salvati 1971 with ISR <0.8 classified as patella baja and ISR >1.2 classified as patella alta. Based on these cut-offs, the frequency of patella baja and alta was 15 (5%) and 64 (21.3%) respectively. From the calculated thresholds herein, the occurrence of patella baja and alta was 6 (2%) and 9 (3%) correspondingly. The prevalence of patella baja was equal in both males (2%) and females (2%) while patella alta had a higher prevalence in males (4%) than in females (2%) (Table 5).

Table 5. Frequency of patella variants

		N (%)		
		Male	Female	Total
Based on Insall and Salvati	Patella baja	10 (6.7)	5 (3.3) .e	15 (5)
	Patella alta	31 (20.8)	33 (21.9)	64 (21.3)
	Normal	108 (72.5)	113 (74.8)	221 (73.7)
	Total	149 (100)	151 (100)	300 (100)
Based on current study	Patella baja	3 (2)	3 (2)	6 (2)
	Patella alta	6 (4)	3 (2)	9 (3)
	Normal	140 (94)	145 (96)	285 (95)
	Total	149 (100)	151 (100)	300 (100)

The midline of the graph in Figure 2 represents the mean ISR (1.07) in this study while the dotted lines show the normal range cut-off values within the 2 SD range (0.73, 1.41). The patella height was classified as patella baja and alta when the ISR was below 0.73 and above 1.41 respectively (Fig. 2).

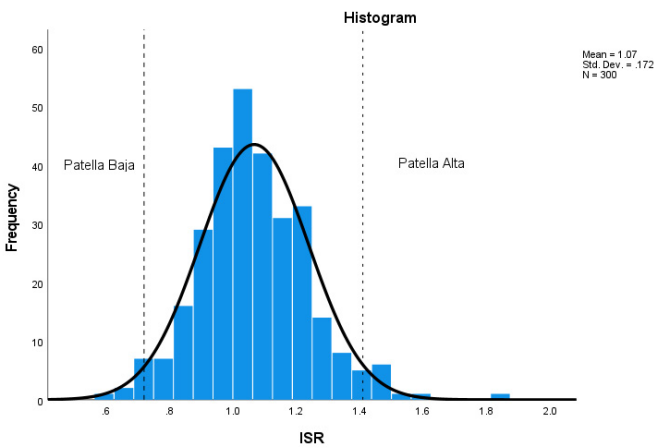

Fig. 2. Distribution of the ISR in the study population

The mean variables, ISR cut-offs and frequency of patella baja and alta in diverse population groups are compared on Table 6.

Discussion

The mean PTL and patella length in this study were higher than the findings by Di et al.[3] The Indians evaluated

Table 6. Comparison of cut-offs and prevalence of patella baja and alta in different populations*

Author	Country	n	Modality	Gender	Patella tendon length (cm)	Patella length (cm)	Insall-Salvati ratio	ISR Cut-offs		Prevalence (%)	
								Baja	Alta	Baja	Alta
Althani et al.[2]	United Arab Emirate (Emirati)	400	Radiographs	Males			1.22	<0.86	>1.54		
				Females			1.18	<0.80	>1.20	4	38
				Total			1.20				
Rhatomy et al.[4]	Indonesia	100	MRI	Males			1.09				
				Females			1.09				
				Total			1.09				
Di et al.[3]	Vietnam	455	MRI	Males	4.21	4.19	1.01	<0.72	>1.32	2.4	0.9
				Females	3.88	3.76	1.03				
				Total	4.06	4.00	1.02				
Kumar and Ram[8]	South India	200	Radiographs	Males			1.41				
				Females			1.28				
				Total			1.34				
Upadhyay et al.[17]	India	400	Radiographs	Males	5.16	4.63	1.12	<0.70	>1.50	1	2.8
				Females	4.14	3.80	1.17	<0.80	>1.20	3	19
				Total	4.80	4.22	1.14				
Present Study	Nigeria	300	Radiographs	Males	5.00	4.75	1.06	<0.73	>1.41	2	3
				Females	4.58	4.29	1.07	<0.80	>1.20	5	21.3
				Total	4.79	4.52	1.07				

* MRI – magnetic resonance imaging, ISR – Insall-Salvati ratio

by Upadhyay et al.[17] had longer PTL and shorter patella length than the Nigerians herein. The ISR was higher than the reports by Uduoka and Bienonwu,[5] Capkin et al.[13] and Di et al.[3] and lower than the observations by several other authors (Table 6).[2,4,8,17] Variations in the PTL have been linked to individual differences in height.[18] Taller people have longer patella tendons than shorter people. Long PTs predispose to high riding patella (patella alta).[11] The variant PTL is influenced by its migration rate. As bones grow, tendons and ligaments migrate at varying rates depending on the different insertions. The migration is greater when the insertion to the bone is nearer. Migration is caused by periosteal growth and stretching caused by inherent epiphyseal plate activity. As the PT migrates, its deeper layer lengthens at its insertion point while the superficial fibers slide over the tibial tuberosity's surface.[17] Awareness of the patella's morphometry is necessary in the design of implants and minimizing surgical complications.[18]

The variability in morphometry could have resulted from discrepant imaging modalities, landmarks used and knee positioning, thus highlighting the importance of standardized imaging protocol.[3,11] With the improved soft tissue contrast offered by MRI, the surface of the tendon measured affects the length recorded since it is significantly shorter posteriorly than anteriorly.[11] The tendon's central fascicles inserting on the patella's most inferior pole are shorter than the lateral and medial tendon fascicles.[3,11]

The experience of the researchers to recognize the tendon's insertion at the tibial tuberosity may explain the dissimilarities in measurements.[10] Additionally, diversity could stem from variant knee positioning whereby in MRI, the knee is imaged in passive extension while in lateral radiograph, different protocols with variable flexion degrees are used.[13,18] Although some studies report consistency of the ISR in the all the flexion degrees.[1,2] Shape variations of the patella's inferior pole such as blunt, intermediate or pointed also affects the measurements.[11,15] This can be avoided by calculating the modified Insall-Salvati ratio involving the measurement from the lower limit of the patella's articular surface to the PT's insertion and the extent of the patella's articular surface.[8,9] Sampling biasness of normal subjects versus symptomatic knees and age differences contributes to the diversities observed.[3,18] In China, there is rare over-exercising among the young population who also have a low hospital visit rate due to knee pain or discomfort.[16]

The ISR discrepancies have been ascribed to diverse ethnic and cultural practices.[3,10] Compared to western cultures, the patella position is higher in Asians and does not exhibit any differences from Africans.[2,5] Unlike the Western and African cultures, Asians have unique lifestyle activities involving knee flexion namely; praying, squatting, and sitting cross-legged. These continuously stretch the PT and lengthen the patella hence causing a high ISR.[2,8,17] Clinicians and radiologists need to apply the population-specific ISR for accurate diagnosis of patella instability, malalignment and PT pathologies.

The patella and PT were significantly longer in males than females. This corresponded with Upadhyay et al.[17] Sex bias in sampling could explain the variant degree of sexual dimorphism of patella variables in different studies. Males are involved in many sporting activities and are more likely to have knee complaints or injuries earlier than females whose knee pain is experienced later, mainly caused by degenerative arthri-

tis.[3,18] Intense physical activities in males are responsible for their larger skeletal morphology and muscle mass.[3] The larger patella height in males could be ascribed to a higher circulating testosterone in men than women which is responsible for more periosteal bone thickening.[19] Appreciating the sexual dimorphism of patella variables improves the understanding of sex related patellofemoral problems and their surgical management.

Consistent with several researchers, the ISR lacked sexual dimorphism.[3-5,8] Conversely, Upadhyay et al.[17] documented significantly higher ISR in females while Althani et al.[2] documented significantly larger ISR in males. The sex differences in the distribution of stresses within the PT and in the prevalence of tendinopathy may explain the ISR's sexual dimorphism.[15] The ISR lacked significant association with age. Therefore, clinicians in Delta State can estimate the patella position by applying the normative ISR reference values obtained regardless of the gender or age.

Using the international ISR cut-offs (<0.8 and >1.20) by Insall and Salvati,[14] the prevalence of patella baja and alta (5% and 21.3%) were higher than the prevalence obtained (2% and 3%) with the calculated cut-offs (<0.73 and >1.41). Althani *et al*.,[2] observed higher rates of patella alta using the Insall and Salvati's threshold in the Emirati population of Middle East, therefore recommending an extension of the cutoffs to <0.86 and 1.54. In Vietnam the cut-off documented was <0.72 and >1.32.[3] Among central Indians, the cut-off for patella alta was >1.5; 10% more than the internationally recommended threshold of >1.2, hence, proposing the normal range among Indian squatters to be 0.7 to 1.5.[17] Therefore, ISR should be specific for each ethnic or population group for accurate evaluation of patella position (Table 6).[3] Although this study establishes normative ISR values for the local population, further validation in multi-ethnic cohorts is necessary to generalize findings.

Patella alta is more common than patella baja in most populations but vice versa is true among Asians.[2,3,17] This inconsistency shows ethnic diversity in patella position.[3] Nevertheless, the race bias in the use of the international ISR threshold from western culture in other populations contributes greatly to the disparities in the frequency of patella baja and alta.[16] The occurrence of patella baja was equal in both males and females while patella alta had a slightly higher frequency in males than females. Conversely, using modified Insall-Salvati ratio, a cadaveric study by Wambua *et al*.[15] revealed more patella alta cases among black Kenyan females and concluded that this predisposes them to patella tendinopathy. Females have higher occurrence of patella alta than males due to their weaker hamstrings and higher strength ratio of quadriceps to hamstrings.[18] Females are more prone to patella instability due to ligamentous laxity and larger Q angle caused by the wider pelvis. The Q angle is a known indicator of patellofemoral stability although in individuals of the same height, it shows no sexual dimorphism.[15]

The findings herein are important to radiologists and orthopedic surgeons in evaluating knee conditions, planning of treatment, and monitor the postoperative prognosis.[3,10] In patella alta, performing patella tenodesis and tibial tubercle distalization normalizes the patella height.[11] On the contrary, Patella baja may occur inadvertently after some surgical knee procedures. The prevalence of total knee arthroplasty (TKA) has increased in the recent past and these predispose to postoperative patella infera due to subsequent PT shortening by scarring. Additionally, elevation of femorotibial joint line in TKA may also change the patella height leading to restricted range of motion, pain and crepitation.[6,10] Patella baja may also ensue following progressive PT shortening after surgical harvesting of the PT autograft for arthroscopic ACL reconstruction.[2,6] Moreover improper technique during total or unicondylar knee arthroplasty may lead to patella baja.[13]

The use of readily available knee radiographs and utilizing simple and reproducible morphometric parameters to establish diagnostic ISR cut-off values were the current study's strengths. However, the findings may not represent the broader Nigerian population due to the limited sample size based on the use of a single study center and retrospective study design. The sample entailed symptomatic patients hence, the data does not represent asymptomatic knees. The clinical impact of abnormal ISR values could not be ascertained from the radiographic findings of abnormal patella position.

Recommendations

To increase the sample size, a multi-institutional study is recommended to ascertain the ISR thresholds obtained in this study. Future studies incorporating MRI could provide more accurate ISR measurement owing to high precision in assessing soft tissue and cartilage.

Conclusion

This study established the normal ISR cutoff range of 0.73 and 1.4 which will assist orthopedic specialists and radiologists in accurately diagnosing abnormal patella positions regardless of patient's sex or age. Precise diagnosis will enhance better surgical outcomes through restoration of the knee's stability and biomechanics.

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Declarations

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Author contributions

Conceptualization, B.S.O. and J.E.I.; Methodology, O.G.E.; Software, J.E.I.; Validation, O.F.O, P.S.I., M.T.E. and J.E.I.; Formal Analysis, O.G.E, O.J. and B.S.O.; Investigation, O.F.O., O.G.E. and J.E.I.; Resources, O.G.E. and P.S.I.; Data Curation, O.J.; Writing – Original Draft Preparation, B.S.O. and M.T.E.; Writing – Review & Editing, B.S.O., O.G.E., O.F.O. and P.S.I.; Visualization, M.T.E, P.S.I; Supervision, J.E.I. and P.S.I; Project Administration, J.E.I.

Conflicts of interest

The authors declare no conflict of interest.

Data availability

The data for this study are not publicly available to protect patient information but will be available on reasonable request from the corresponding author.

Ethics approval

This study due to its retrospective nature did not receive consent from subjects however ethical approval was sought from the Hospital's research and ethics committee (DELSUTH/HREC/2024/004/0742). The study was carried out in compliance with the Declaration of Helsinki.

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