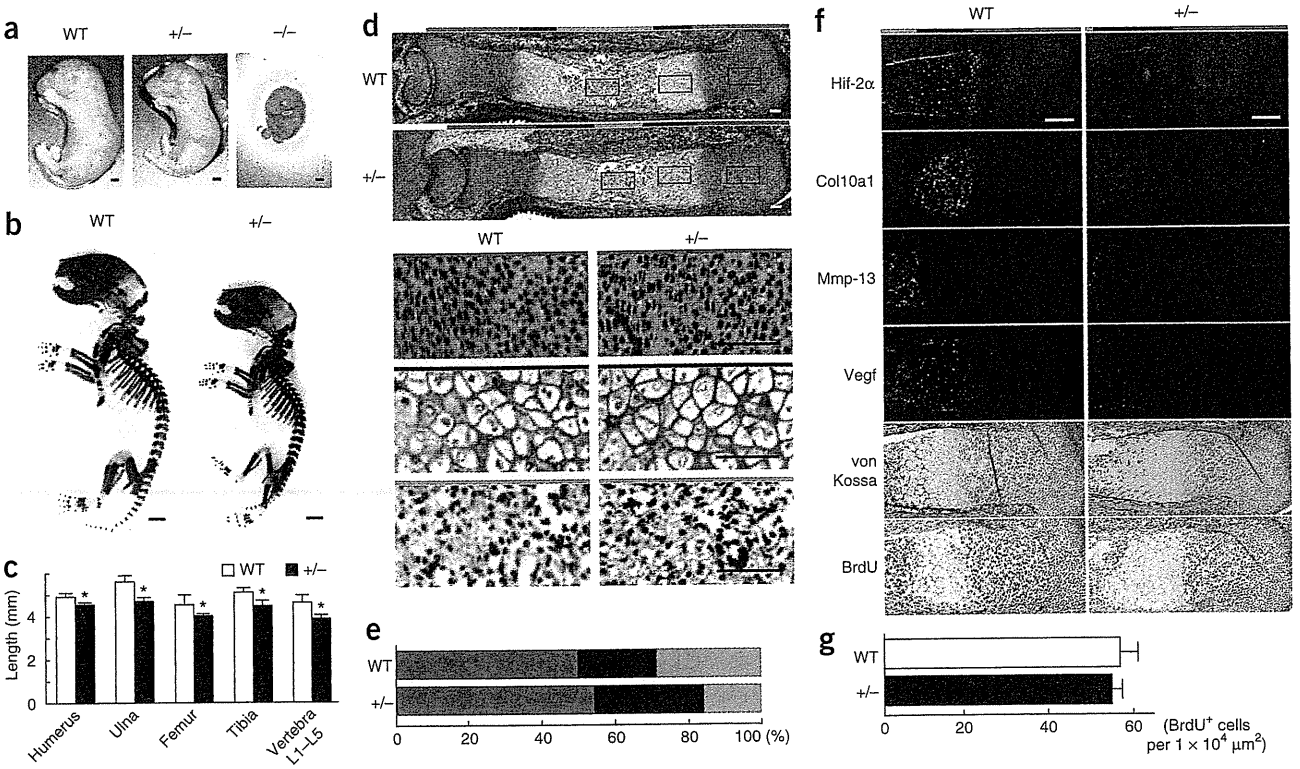

Figure 3 Skeletal abnormality in *Epas1*-deficient mice. (**a**) Wild-type (WT), heterozygous-deficient (*Epas1*$^{+/-}$) and homozygous-deficient (*Epas1*$^{-/-}$) littermate embryos (E17.5). All *Epas1*$^{-/-}$ embryos died at mid-gestation. Scale bars, 1 mm. (**b**) Double staining with Alizarin red and Alcian blue of the whole skeleton of WT and *Epas1*$^{+/-}$ littermate embryos (E17.5). Scale bars, 1 mm. (**c**) Length of long bones and vertebra (first to fifth lumbar spines) of WT and *Epas1*$^{+/-}$ littermate embryos. Data are expressed as means ± s.d. *$P < 0.05$ versus WT. (**d**) H&E staining of whole tibias of the WT and *Epas1*$^{+/-}$ littermate embryos. Inset boxes indicate the regions of the bottom three rows representing proliferative zone, hypertrophic zone and bone area, shown by red, blue and green bars, respectively. Scale bars, 100 μm. (**e**) Percentage of the length of proliferative zone (red), hypertrophic zone (blue) and bone area (green) over the total tibial length of the WT and *Epas1*$^{+/-}$ littermate embryos. (**f**) Immunofluorescence with antibodies to Hif-2α, Col10a1, Mmp-13 and Vegf, as well as bromodeoxyuridine (BrdU) labeling and von Kossa staining of the proximal tibias of WT and *Epas1*$^{+/-}$ littermate embryos (E17.5). Color bars indicate layers as indicated in **d**. Scale bars, 200 μm. (**g**) The number of BrdU-positive cells in 1 × 10^4 μm^2 of the proximal tibia of WT and *Epas1*$^{+/-}$ littermate embryos. Data are expressed as means ± s.d.

Contribution of HIF-2α to osteoarthritis in mice and humans

We next compared osteoarthritis development between adult littermates of wild-type and *Epas1*$^{+/-}$ mice that had undergone comparable skeletal growth after birth (**Supplementary Fig. 1b**) by creating a surgical osteoarthritis model through induction of instability to the knee joints[4,5]. The expression of Hif-2α, as well as of Col10a1, Mmp-13 and Vegf, increased in the joint cartilage with osteoarthritis development for 8 weeks after surgery in the wild-type mice; however, in the *Epas1*$^{+/-}$ littermates, the cartilage degradation and the expression of the three factors were notably suppressed (**Fig. 5a**). Quantification by grading systems[4,26] confirmed that the Hif-2α insufficiency caused significant resistance to cartilage degradation and osteophyte formation (**Fig. 5b**). There was no difference in the subchondral bones between the two genotypes under the sham operation, suggesting that the *Epas1* deficiency does not affect physiological bone homeostasis. However, after surgical induction, subchondral bone sclerosis, an osteoarthritic disorder secondary to cartilage destruction, was apparent in the wild-type joints, whereas it was suppressed in the *Epas1*$^{+/-}$ joints (**Supplementary Table 1**).

In human knee joint samples, as well, the HIF-2α expression increased with osteoarthritis development, reached a maximum at the initial and progressive stages and decreased thereafter at the terminal stage, although it was hardly detected in subchondral bone or synovium (**Fig. 5c**). To further investigate a possible association of the human *EPAS1* gene with knee osteoarthritis of humans, we searched a Japanese population–based cohort of the ROAD study[27] for sequence variations in exons and the 5′-end flanking region up to −1,000 bp from the transcription start site (TSS) of the human *EPAS1* gene and identified only one common SNP with a minor allele frequency >0.1, rs17039192 (+18C and +18T for major and minor alleles, respectively, relative to the TSS; minor allele frequency = 0.132) (**Fig. 5d**). A comparison of allelic frequencies between 397 individuals with knee osteoarthritis and 437 controls showed significant association of the rs17039192 SNP with knee osteoarthritis ($P = 0.013$, odds ratio = 1.44) (**Fig. 5d**). Because this SNP was located close to the TSS, we further examined the effects of the allelic difference (+18C/T) on *EPAS1* promoter activity in chondrogenic and nonchondrogenic cells transfected with a luciferase reporter gene and the *EPAS1* promoter fragment (−1,000 bp to 488 bp) containing +18C or +18T. The susceptibility allele (18C) showed higher promoter activity in chondrogenic cells, but not in nonchondrogenic cells (**Fig. 5e**), confirming that enhanced transactivation of *EPAS1* in chondrocytes is associated with osteoarthritis in humans.

Molecular network around HIF-2α in endochondral ossification

Regarding downstream molecules of HIF-2α, we have focused on COL10A1, MMP-13 and VEGF as representative factors for the

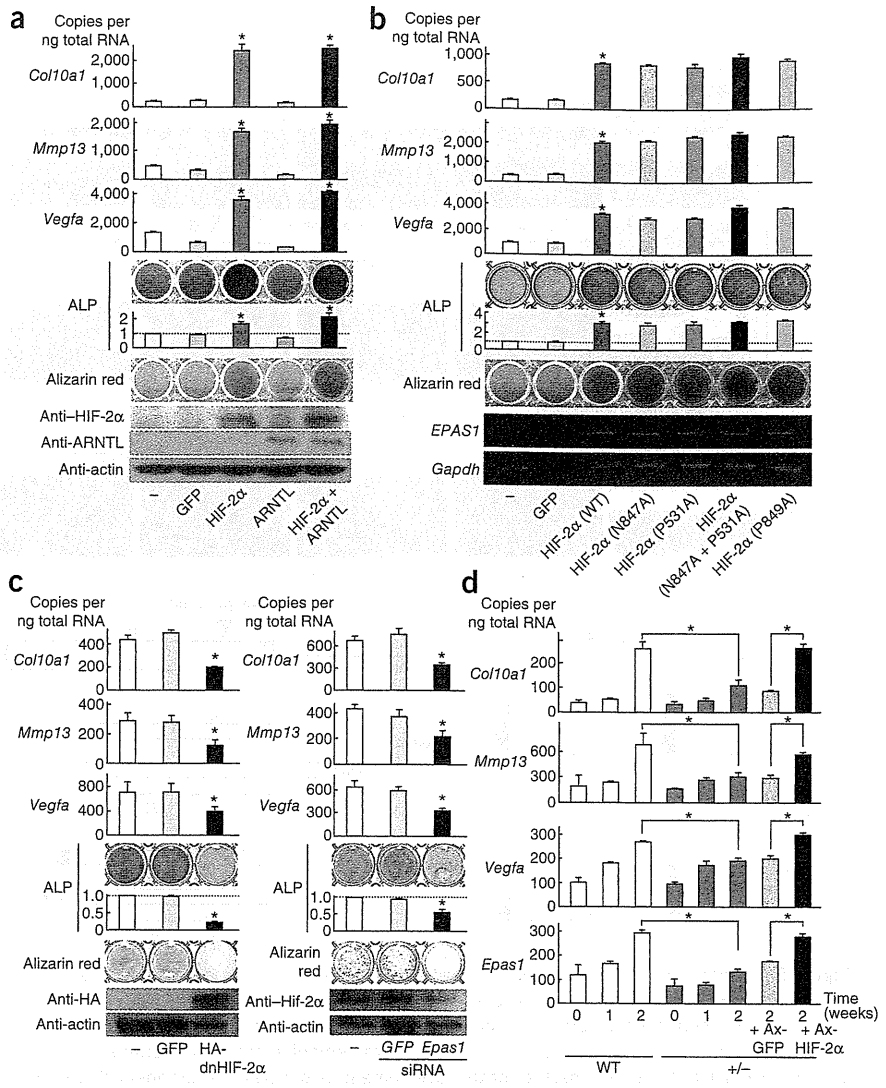

Figure 4 Effects of gain and loss of function of HIF-2α on endochondral ossification parameters in cultures of chondrogenic cells. (**a**) mRNA levels of *Col10a1*, *Mmp13* and *Vegfa*, alkaline phosphatase (ALP) activity (relative to control) and Alizarin red staining in stable lines of ATDC5 cells retrovirally transfected with GFP, HIF-2α, ARNTL or both HIF-2α and ARNTL and in nontransfected parental cells (–) after culture for 3 weeks with ITS and 2 d with Pi. HIF-2α and ARNTL levels were confirmed by western blotting, with the actin level as the internal control. (**b**) Analyses of the parameters in **a** in stable ATDC5 lines transfected with GFP or HIF-2α mutants at the oxygen-dependent hydroxylation residues causing enhancement (N847A and P531A) and abrogation (P849A) of HIF-2α transactivation activity under the culture conditions used in **a**. Gene expression was confirmed by RT-PCR with the *EPAS1* primer set inside the coding sequence, with the *Gapdh* level as the internal control. (**c**) Analyses of the same read-outs in **a** in stable ATDC5 lines transfected with GFP or HA-tagged dominant-negative HIF-2α (HA–dnHIF-2α) (left) or siRNA specific for *GFP* or *Epas1* mRNA (right) under the cultures conditions used in **a**. HA–dnHIF-2α and Hif-2α amounts were confirmed by western blotting. (**d**) mRNA levels of *Col10a1*, *Mmp13* and *Vegfa* and *Epas1* in the pellet cultures of primary chondrocytes derived from wild-type (WT) and *Epas1*$^{+/-}$ littermate embryos for 2 weeks. For the rescue experiment, adenoviral transfection with HIF-2α (Ax–HIF-2α) or the control GFP (Ax-GFP) was performed before the pellet formation. All data are expressed as means ± s.d. $^*P < 0.05$ versus GFP unless otherwise indicated.

central three steps of endochondral ossification (chondrocyte hypertrophy, cartilage degradation and vascularization) and found that all were the direct transcriptional targets. However, there are other factors related to endochondral ossification, including in the earlier cartilage formation step and in the later osteogenesis step, which might also be targets of HIF-2α. We therefore examined the expression patterns of the following factors: type II collagen (COL2A1) and aggrecan (AGC1) as cartilage matrix proteins; RUNX2, Indian hedgehog (IHH), and type 1 PTH/PTHrP receptor (PTH1R) as chondrocyte hypertrophy markers; MMP-3, MMP-9, a disintegrin and metalloproteinase with thrombospondin type 1 motif-4 (ADAMTS4) and ADAMTS5 as cartilage-degradable proteinases; and type I collagen (COL1A1), bone sialoprotein (BSP), osteocalcin, alkaline phosphatase, bone morphogenetic protein-2 (BMP-2), BMP-4 and BMP-7 as osteogenic markers. In cultured ATDC5 cells, expression of chondrocyte hypertrophy markers, cartilage degradable proteinases and osteogenic markers increased in accordance with the cell differentiation and *Epas1* expression (**Supplementary Fig. 3a**). The chondrocyte hypertrophy markers and cartilage degradable proteinases were localized mainly in the hypertrophic zone of the mouse limb cartilage, similarly to Hif-2α (**Supplementary Fig. 3b**). The mRNA levels of cartilage matrix proteins, chondrocyte hypertrophy markers and most of the osteogenic markers were increased in ATDC5 cells overexpressing HIF-2α or HIF-2α–ARNTL (**Supplementary Fig. 4a**). Among cartilage-degradable proteinases, expression of *Mmp3* and *Mmp9* was increased, whereas neither *Adamts4* nor *Adamts5* was affected (**Supplementary Fig. 4a**). In contrast, mRNA levels of the chondrocyte hypertrophy markers *Mmp3* and *Mmp9* were decreased after overexpression of a dominant-negative mutant form of HIF-2α and siRNA specific for *Epas1* mRNA in ATDC5 cells (**Supplementary Fig. 4b,c**); however, cartilage matrix proteins, *Adamts4*, *Adamts5* and most of osteogenic markers were little affected. Primary chondrocytes derived from *Epas1*$^{+/-}$ mice reproducibly showed suppression of the chondrocyte hypertrophy markers, *Mmp3* and *Mmp9*, but not cartilage matrix proteins, *Adamts4*, *Adamts5* or osteogenic factors, and the suppression was restored to wild-type levels by HIF-2α overexpression (**Supplementary Fig. 5**). Further *in vivo* analyses of embryonic limbs (**Supplementary Fig. 6a**) and osteoarthritic knee joints (**Supplementary Fig. 6b**) confirmed the decreases in expression of the chondrocyte hypertrophy markers, Mmp-3 and Mmp-9, but not cartilage matrix proteins Adamts4 or Adamts5, by Hif-2α insufficiency. HIF-2α enhanced the promoter activities of the chondrocyte hypertrophy markers *MMP3* and *MMP9*, as well as *Col10a1*, *Mmp13*, and *Vegfa* mRNA levels, much more strongly than HIF-1α, and the stimulation of the mRNA levels by HIF-2α was not altered by cotransfection of

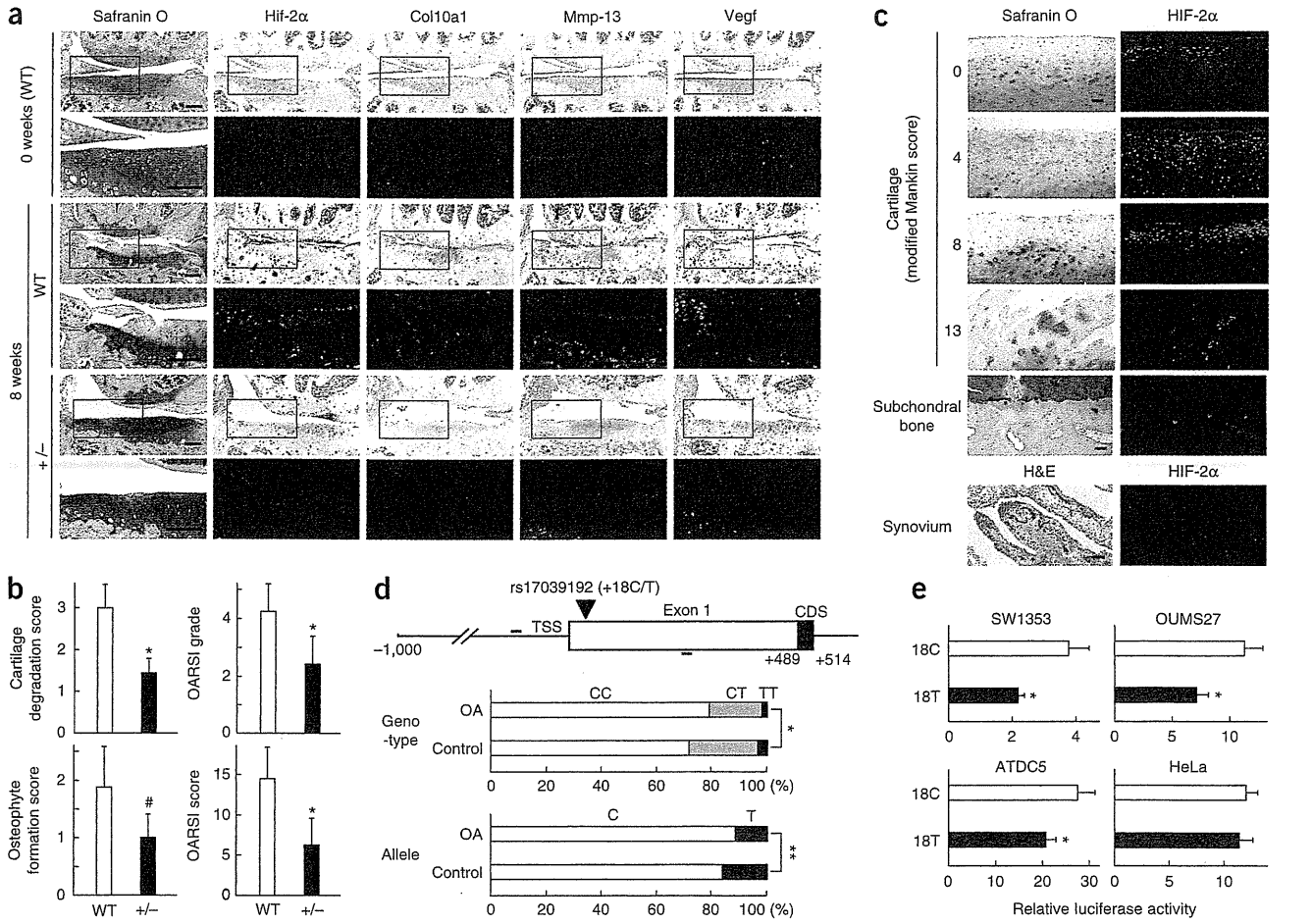

Figure 5 Contribution of HIF-2α to osteoarthritis development in mice and humans. (**a**) Cartilage degradation assessed by safranin O staining and expression of Hif-2α, Col10a1, Mmp-13 and Vegf by immunostaining (brown) and immunofluorescence (green) in mouse knee joints 0 and 8 weeks after creating a surgical osteoarthritis model in 8-week-old wild-type (WT) and *Epas1*$^{+/-}$ littermates. Boxed areas in each safranin O–stained or each immunostained image indicate the regions shown in the enlarged safranin O–stained or immunofluorescent image immediately below. Scale bars, 100 μm. (**b**) Quantification of osteoarthritis development by our (two left graphs) and OARSI (two right graphs) grading systems. Data are expressed as means ± s.d. #$P < 0.05$, *$P < 0.01$ versus WT. (**c**) Safranin O staining, H&E staining and immunofluorescence with an antibody to HIF-2α in human tibial cartilages of various degradation stages, subchondral bone (beneath the cartilage with Mankin score = 8) and synovium (around the cartilage with Mankin score = 8), obtained as surgical specimens of total knee arthroplasty. Scale bars, 100 μm. (**d**) Top, the identified SNP, rs17039192, and primers used for genotyping (red lines) in the human *EPAS1* gene. CDS, coding sequence. Bottom, association of the rs17039192 (+18C/T) SNP with knee osteoarthritis (OA) diagnosed on radiographs using the Kellgren/Lawrence grade in a Japanese population. The odds ratio of the susceptibility allele was 1.44 (95% confidence interval: 1.08–1.92). *$P = 0.05$, **$P = 0.013$. (**e**) Luciferase activities in chondrogenic SW1353, OUMS27 and ATDC5 cells and nonchondrogenic HeLa cells transfected with a luciferase reporter gene construct ligated to a fragment (−1,000 bp to +488 bp) containing +18C or +18T. Data are shown as means ± s.d. *$P < 0.05$ versus 18C.

HIF-1α (**Supplementary Fig. 7a,b**). Furthermore, the endochondral ossification parameters stimulated by HIF-2α overexpression in ATDC5 cells were not inhibited by suppression of RUNX2 through overexpression of a dominant-negative mutant RUNX2 (**Supplementary Fig. 7c**).

Finally, to identify the upstream mechanism that regulates HIF-2α, we performed a screen of transcription factors with the *EPAS1* promoter fragment including the +18C SNP described above. Among candidate molecules that are known to regulate chondrocyte differentiation, such as sex-determining region Y box (SOX), RUNX, CCAAT/enhancer binding protein (C/EBP), MEF2, SP/Kruppel-like factor (KLF), activating transcription factor (ATF), cAMP responsive element–binding protein (CREB), Notch and NF-κB family members, we found that v-rel reticuloendotheliosis viral oncogene homolog A (RELA or NF-κB p65), an essential molecule of the NF-κB signal, showed the strongest activation in all cells (**Fig. 6a**). In the *EPAS1* promoter we identified an NF-κB motif and found that site-directed mutagenesis in the motif caused suppression of transactivation by RELA (**Fig. 6b**). The allelic difference (+18C/T) of the rs17039192 SNP described above also altered the activation of *EPAS1* promoter by RELA but did not affect it when the NF-κB motif was mutated, suggesting the involvement of this SNP in the *EPAS1* transactivation and osteoarthritis development caused by the NF-κB signal. We further confirmed that the proinflammatory cytokines tumor necrosis factor-α (TNF-α) and interleukin-1β (IL-1β), putative inducers of the NF-κB signal[28], increased *EPAS1* expression in cultured chondrogenic cells (**Fig. 6c**). In mouse knee joint cartilage, the expression of Rela was increased during osteoarthritis development, similarly to Hif-2α expression (**Fig. 6d**).

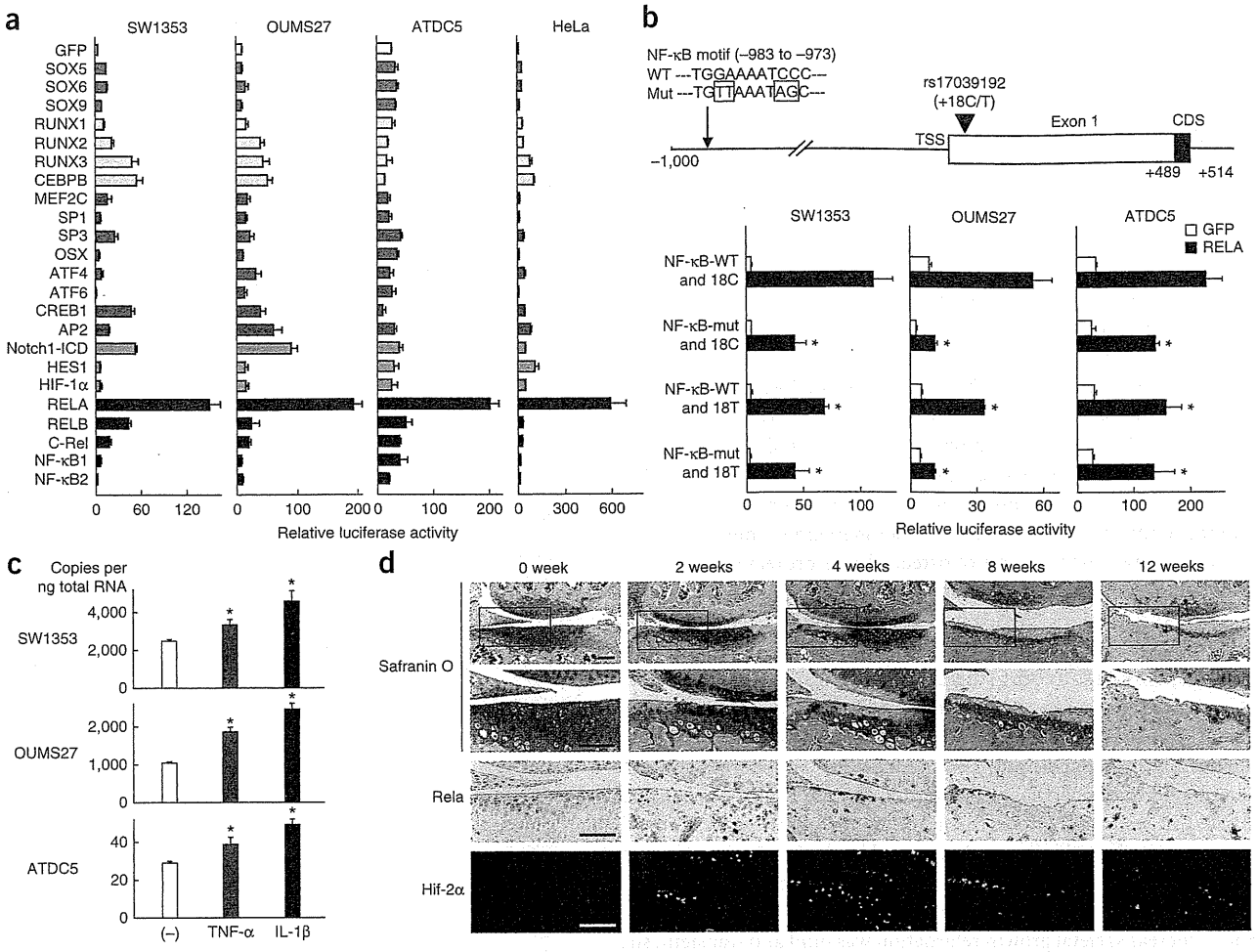

Figure 6 Upstream mechanism that regulates HIF-2α. (**a**) Luciferase activities after transfections of putative chondrocyte-related transcription factors into chondrogenic SW1353, OUMS27 and ATDC5 cells and nonchondrogenic HeLa cells with a reporter construct containing a fragment (–1,000 bp to +488 bp) of the *EPAS1* gene. OSX, osterix; AP2, transcription factor AP-2α; Notch1-ICD, intercellular domain of Notch1; HES1, hairy and enhancer of split 1. Data are shown as means ± s.d. (**b**) Top, depiction of the NF-κB motif (–983 to –973) in the human *EPAS1* gene. Bottom, site-directed mutagenesis analyses of the luciferase assay in the three chondrogenic cell lines transfected with GFP or RELA. Luciferase activities were compared with or without mutation in the NF-κB motif and with +18C or +18T of the rs17039192 SNP. Data are shown as means ± s.d. $^*P < 0.05$ versus wild-type NF-κB and 18C with RELA. (**c**) mRNA levels of *EPAS1* in the three chondrogenic cells cultured with or without TNF-α or IL-1β (each 1 ng ml^{-1}) for 2 d. Data are expressed as means ± s.d. $^*P < 0.05$ versus control. (**d**) Time course of degradation in mouse knee joint cartilage, as shown by Safranin O staining and expression of Rela and Hif-2α by immunostaining and immunofluorescence, respectively, in a surgical osteoarthritis model in 8-week-old mice. Boxed areas in each of the top images are enlarged in the bottom images directly beneath. Scale bar, 100 μm.

DISCUSSION

Among the sequential steps of endochondral ossification—cartilage formation, chondrocyte hypertrophy, cartilage degradation, vascularization and osteogenesis—this study reveals that HIF-2α functions as an extensive transcriptional regulator of the central three steps. HIF-2α shares about 50% amino acid homology with HIF-1α (ref. 20), a potent regulator of cartilage homeostasis[14–16]; however, accumulating evidence has shown distinct expression patterns and functions between the two HIF proteins[17–21]. HIF-1α is expressed mainly in hypovascular and hypoxic tissues[16,19,29], whereas HIF-2α is expressed even in vascularized tissues[11,29]. In cartilage as well, previous studies and our current study show that HIF-1α is expressed from the early stage of cartilage formation, and its activity is enhanced by hypoxia[13–16,30]. In contrast, HIF-2α is expressed mainly in highly differentiated chondrocytes, and its function is independent of oxygen-dependent hydroxylation. Likewise, although cartilage-specific knockout of HIF-1α leads to defects in the earlier stage of cartilage formation and the later stage of chondrocyte survival and osteogenesis[14–16], *Epas1*$^{+/-}$ mice show growth retardation with defects in the central steps of endochondral ossification. Hence, HIF-1α and HIF-2α may have distinct roles via different mechanisms: hypoxia-dependent cartilage formation and maintenance by HIF-1α and less hypoxia-dependent endochondral ossification by HIF-2α. We have also confirmed that neither gain nor loss of function of HIF-2α alters HIF-1α expression during chondrocyte differentiation, nor does HIF-1α transfection affect *EPAS1* promoter activity. Furthermore, HIF-1α hardly stimulates the expression of markers of the central steps of endochondral ossification in the presence or absence of HIF-2α, indicating independent functions of HIF-1α and HIF-2α at least in the central steps. However, we do not deny the possibility of interactions between HIF-1α and HIF-2α in earlier and later stages of endochondral ossification. The HIF-2α function may possibly be compensated by HIF-1α in this earlier stage, as cartilage matrix proteins are not altered by HIF-2α suppression. In the later or severe stage of osteoarthritic

cartilage, expression of HIF-2α decreases after reaching a maximum at the initiation of cartilage degradation in mice and humans, as was reported in a previous study[31]. In this terminal stage, the decreased HIF-2α expression may enhance autophagy in mature chondrocytes of osteoarthritic cartilage, as HIF-2α is known to antagonize the autophagy-accelerator function of HIF-1α[32].

Our study reveals that *COL10A1*, *MMP13* and *VEGFA* are the direct transcriptional targets of HIF-2α. The functional relationships *in vivo* are supported by previous reports that a deficiency of *Mmp13* or *Vegfa* in mice causes a skeletal phenotype similar to that in *Epas1*$^{+/-}$ mice, with elongation of the hypertrophic zone and delay of ossification in the limb cartilage[9,10], although the skeletal phenotypes of *Col10a1*-knockout mice differs among the reports[33–35]. Our further studies identify *RUNX2*, *IHH*, *PTH1R*, *MMP3* and *MMP9* as possible transcriptional targets of HIF-2α. We have recently reported that HIF-2α enhances *Runx2* promoter activity[36] and that *Runx2*$^{+/-}$ mice show resistance to osteoarthritis development under mechanical instability, similarly to *Epas1*$^{+/-}$ mice[5]. However, HIF-2α and RUNX2 may promote endochondral ossification via independent mechanisms.

There are two mechanisms of osteoarthritis protection: induction of anabolism or inhibition of catabolism in joint cartilage. The protection in *Epas1*$^{+/-}$ mice is not likely to be due to induction of anabolism, as the anabolic markers COL2A1 and AGC1 are unaffected in joint cartilage. Although recent studies have identified ADAMTS5 and related molecules as key catabolic regulators of osteoarthritis development[37–40], neither ADAMTS4 nor ADAMTS5 is regulated by HIF-2α, implicating another pathway. Because *Mmp13*$^{-/-}$ mice are reported to be protected from cartilage degradation despite considerable aggrecan loss after surgical osteoarthritis induction[41], similarly to *Epas1*$^{+/-}$ mice, the osteoarthritis protection caused by the Hif-2α insufficiency might occur principally through regulation of Mmp-13.

Suppression of osteoarthritis development was obvious in *Epas1*$^{+/-}$ mice, whereas skeletal growth retardation was mild and transient, suggesting that pathological endochondral ossification is more dependent on HIF-2α than is physiological endochondral ossification. As a trigger of osteoarthritis, mechanical stress may induce the upstream NF-κB signal and HIF-2α expression in joint cartilage, which causes endochondral ossification by transactivation of *COL10A1*, *MMP13*, *VEGFA* and other factors. Recent comprehensive profiling analyses of not only genes and proteins, but also microRNAs, is unraveling the molecular network underlying osteoarthtitis development[42]; however, we hereby propose that signals in the HIF-2α axis from NF-κB signaling to endochondral ossification–related molecules may represent a rational therapeutic target for osteoarthritis with minimal effects on physiological skeletal homeostasis.

METHODS

Methods and any associated references are available in the online version of the paper at http://www.nature.com/naturemedicine/.

Note: Supplementary information is available on the Nature Medicine website.

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AUTHOR CONTRIBUTIONS

T.S., T.I. and H.K. performed project planning; T.S., A.F., A.M., F.Y. and S.O. performed the experiments; T.S., A.M., N.N., T.A., N.Y., T.N., K.N., K.T., U.-i.C. and H.K. conducted data analysis; T.S. and H.K. wrote the manuscript.

COMPETING FINANCIAL INTERESTS

The authors declare no competing financial interests.

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POSITION PAPER

IOF position statement: vitamin D recommendations for older adults

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Abstract This position paper of the International Osteoporosis Foundation makes recommendations for vitamin D nutrition in elderly men and women from an evidence-based perspective.

Keywords Musculoskeletal health · Requirement · Vitamin D

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This statement has been endorsed by the IOF Committee of Scientific Advisors

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Introduction

Vitamin D is important for bone and muscle development, function, and preservation. The serum 25OHD concentration is the best available clinical indicator of vitamin D status. Until recently, optimal serum 25OHD concentration was considered to be that level associated with maximal parathyroid hormone (PTH) suppression. Estimates of that threshold level have clustered around 32–50 nmol/L (12.8–20 ng/ml) and 68–75 nmol/L (27.2–30 ng/ml), depending upon analytical approach used [1]. In the last decade, however, the evidence base for older men and women has grown to include many randomized, controlled clinical trials (RCTs) with falls and fracture endpoints. Because the RCTs have for the most part been conducted in men and women over the age of 60 to 65 years, our recommendations are directed at this large and growing older segment of the adult population. Our objective is to use available evidence to support recommendations for optimal vitamin D status. We approach this by examining

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the efficacy of different vitamin D doses administered and levels of 25OHD achieved in reducing risk of falls and fractures. In this process, it is important to consider other factors that influence serum 25OHD levels and responses to oral vitamin D supplementation.

Determinants of serum 25OHD levels and of the serum 25OHD response to oral vitamin D

Vitamin D intake and effective sun exposure are the major determinants of the serum 25OHD level. Several factors influence the increment in serum 25OHD in response to a given dose of vitamin D_3, including the starting level of 25OHD. At a dose of 2.5 μg (100 IU/d), the mean increment ranges from 2.75 nmol/l (1.1 ng/ml) at low starting 25OHD levels to 1.75 nmol/l (0.7 ng/ml) at higher (near optimal) starting levels [2]. The increment in 25OHD in response to a given dose of vitamin D also varies with body size. It is smaller in subjects with high BMI than in individuals with normal BMI [3, 4]. Other factors affect 25OHD levels but have no known impact on 25OHD responses to supplemental vitamin D. Estrogen use increases measured serum 25OHD levels by increasing levels of vitamin D binding protein [5] but does not alter the serum 25OHD increment achieved with supplementation. Serum 25OHD levels decline with aging, but the serum 25OHD response to a given dose of supplemental vitamin D_3 is not affected by age [6]. Similarly, the dietary calcium intake, within the range usually consumed, does not affect the serum 25OHD response to vitamin D supplementation [7]. (The latter statement should not be confused with the observations that the *calcium* requirement may be dependent upon vitamin D status and that an adequate calcium intake is important for bone health [8].) Finally, serum 25OHD levels vary widely across commonly used assays. Until this problem is addressed by widespread use of standard reference material such as the NIST standards [9] and participation in the DEQAS quality control program (www.deqas.org), assay variability will continue to complicate the process of determining the desired 25OHD level and the impact of a given dose on serum 25OHD levels.

Falls

Vitamin D is thought to act on myocyte vitamin D receptors to exert its effect on muscle tissue. In prospective studies, lower serum 25OHD levels have been associated with decreased grip strength and appendicular muscle mass in older men and women [10, 11] Supplementation with vitamin D has improved lower extremity muscle performance and reduced risk of falling in several high-quality double blind RCTs [12]. These trials have employed doses up to 25 μg (1,000 IU) of vitamin D per day, with and without calcium. Supplementation in amounts of 17.5 to 25 μg/day (700–1,000 IU/day) lowered risk of falling by 20% in older individuals, independent of their calcium intake level. In contrast, supplementation with doses of <17.5 μg/day (<700 IU/day) had no detectable effect on falls. From this meta-analysis of available data, it appears that a mean serum 25OHD level of at least 60 nmol/L (24 ng/ml) is needed for optimal fall risk reduction. Observational studies suggest that there may be benefit to increasing serum 25OHD levels beyond 60 nmol/L (24 ng/ml), but higher levels (and doses) have not been evaluated in RCTs.

Fractures

Vitamin D affects fracture risk through its effects on bone metabolism and on risk of falling. Randomized controlled trials indicate that supplementation with vitamin D reduces rates of bone loss in older women [13]. The impact of supplemental vitamin D on fracture risk has been examined mainly in men and women age 65 and older. A recent meta-analysis revealed that vitamin D in doses in the range of more than 10 through 20 μg/day (>400–800 IU/day) reduced risk of non-vertebral and hip fracture by approximately 20% whereas doses up through 10 μg/day (400 IU/day) had no evident effect [14]. Doses above 20 μg/day (800 IU/day) have not been studied. The mean serum 25OHD level associated with reduction in non-vertebral fracture risk was 66 nmol/L (26.4 ng/ml). Hip fracture risk reduction was observed at a mean 25OHD level of 74 nmol/L (29.6 ng/ml) and higher. Based on this

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and other evidence, eight of ten IOF Working Group members felt that 75 nmol/L (30 ng/ml) is the appropriate target level of serum 25OHD for older individuals; in contrast, two members felt that the target should be 50 to 75 nmol/l (20 to 30 ng/ml). The estimate of 75 nmol/l (30 ng/ml) is close to the higher cluster of 25OHD levels associated with maximal PTH suppression [1].

Other potential benefits

Vitamin D insufficiency has been implicated as a contributing factor in a growing number of important chronic diseases including type 2 diabetes, cardiovascular disease, selected cancers, and autoimmune diseases as well as infections, and also to increased mortality. RCTs are needed before causal relationships can be determined and optimal 25OHD levels for prevention can be established. The doses and mean serum levels needed to achieve optimal impact on these non-classical outcomes are not clear.

Global vitamin D status

Vitamin D insufficiency, whether defined as 25OHD levels <75 or <50 nmol/L (<30 or <20 ng/ml), is prevalent worldwide [15]. For instance, the prevalence of levels <75 nmol/L (<30 ng/ml) in postmenopausal women has been reported to be approximately 50% in Thailand and Malaysia, 75% in the USA, and 90% in Japan and South Korea. Vitamin D deficiency, defined as a level below 25 nmol/L (10 ng/ml) is very common in the Middle East and South Asia where mean levels range from 10 to 30 nmol/L (4 to 12 ng/ml) [15, 16]. The high prevalence of suboptimal 25OHD levels in older men and women around the world raises the possibility that many falls and fractures can be prevented with vitamin D supplementation.

Vitamin D preparations

Vitamin D is available in two forms, vitamin D_2 (ergocalciferol) and vitamin D_3 (cholecalciferol). Some find that the two forms are equally effective in raising the serum 25OHD level [17] whereas others find that vitamin D_3 gives a larger increment [18]. We generally recommend that vitamin D_3 be used when available. In some parts of the world, active metabolites are available for use in the treatment of osteoporosis. These metabolites are not a substitute for adequate vitamin D intake. Vitamin D is the substrate for 25OHD and the circulating 25OHD level may be important to support the non-renal production of 1,25-dihydroxyvitamin D. Local production of 1,25-dihydroxyvitamin D appears to mediate some of the non-classical effects of vitamin D.

Recommendations

The estimated average vitamin D requirement for older adults to reach a serum 25OHD level of 75 nmol/L (30 ng/ml) is 20 to 25 μg/day (800 to 1,000 IU/day). Considerably higher doses would be needed to ensure that almost all older adults reached 75 nmol/l (30 ng/ml). Efficacy of doses higher than 20 μg/day (800 IU/day) for fractures and 25 μg/day (1,000 IU/day) for falls however have not been evaluated in RCTs. It is therefore premature to recommend higher intakes for all older adults at this time.

The repletion dose will vary among individuals according to their starting level, their BMI, their effective sun exposure, and other unidentified factors. An intake lower than 20 μg/day (800 IU/day) may be adequate for individuals with regular effective sun exposure. Intake may need to be adjusted upward to as much as 50 μg/day (2,000 IU/day) in individuals who are obese, and in those with osteoporosis, limited sun exposure (institutionalized, homebound), and malabsorption, and in non-European populations known to be at high risk for vitamin D deficiency such as those in the Middle East and South Asia, or immigrants from such regions living in Europe. In these and other high-risk individuals, we recommend measuring the serum 25OHD level. The required dose to reach 75 nmol/L can be estimated from the measured level. Each 2.5 μg (100 IU) of added vitamin D will increase the serum 25OHD level by about 2.5 nmol/L (range 1.75–2.75 nmol/L) or 1.0 ng/ml (range 0.7 to 1.1 ng/ml) [2]. Because of the variability in individual 25OHD responses to supplemental vitamin D, however, in high-risk individuals, the serum 25OHD levels should be retested after about 3 months of supplementation to confirm that the target 25OHD level has been reached.

Conflicts of interest None.

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ORIGINAL ARTICLE

Health-related quality of life with vertebral fracture, lumbar spondylosis and knee osteoarthritis in Japanese men: the ROAD study

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Abstract
Summary This study examined associations of VFx, lumbar spondylosis and knee OA with QOL in 767 men over 40 years old from the ROAD study (mean, 69.7 years.). Multiple regression analysis showed VFx and knee OA as significantly associated with lower PCS scores, but lumbar spondylosis was not.

Purpose Little data have been accumulated regarding associations of vertebral fracture (VFx), lumbar spondylosis and knee osteoarthritis (OA) with quality of life (QOL) in men. The purpose of the present study is to clarify the impact of these diseases on QOL parameters such as the Medical Outcomes Study Short Form 8 (SF-8) and the EuroQOL (EQ-5D). In addition, to provide greater insight into the magnitude of QOL loss, a comparison was made with cerebral stroke.
Methods From the 3,040 participants in the ROAD study, this study analyzed 767 men over 40 years who had completed the questionnaires (mean, 69.7 years.). Vertebral fracture was assessed by lateral radiography of the lumbar spine. Lumbar spondylosis and knee OA were defined as Kellgren/Lawrence grade ≥3. Cerebral stroke was assessed by self-report.
Results Multiple regression analysis after adjustments for age, body mass index and presence of the above four diseases showed VFx was significantly associated with lower scores in physical function (PF), role physical (RP), bodily pain (BP) and vitality (VT) domains as well as physical component summary (PCS). Knee OA were significantly associated with lower scores in PF, RP, BP and PCS scores. Lumbar spondylosis was not associated with any domains of the SF-8. Lumbar spondylosis and knee OA were significantly associated with EQ-5D utility scores, but VFx was not. The impact for VFx on BP, VT and PCS scores was larger than cerebral stroke.
Conclusions This study revealed that VFx and knee OA impaired physical QOL in men, rather than lumbar spondylosis.

Keywords Quality of life · Vertebral fracture · Lumbar spondylosis · Knee osteoarthritis · Men

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Introduction

Vertebral fracture (VFx) is reportedly associated with functional impairment [1], back pain, kyphosis [2, 3], esophageal reflux [4], depressive mood [5], respiratory dysfunctions [6] and mortality [7]. Lumbar spondylosis and knee osteoarthritis (OA), characterized by pathological features including disk or joint space narrowing and osteophytosis, are also major public health issues causing chronic pain and disability among the elderly [8–12]. In fact, prevalences of lumbar spondylosis and knee OA are quite high in the elderly in Japan [13–15], and 37,900,000 and 25,300,000 people ≥40 years old would be affected by radiographic lumbar spondylosis and knee OA, respectively [15]. Furthermore, according to the recent National Livelihood Survey of the Ministry of Health, Labour and Welfare in Japan, OA and fracture represent the fourth and fifth among diseases that cause disabilities, respectively, subsequently requiring support with regard to activities of daily living (ADL) [16].

Gender differences have been observed in these bone and joint diseases. The prevalence of knee OA is higher in women than men [14, 15], while that of lumbar spondylosis is higher in men [13, 15]. For VFx, prevalence is higher in women [17], while mortality is higher in men [18], so the impact of these diseases on quality of life (QOL) may also differ between genders. Although several studies have examined associations of VFx [19–27] and knee OA [28–32] with QOL, men and women were not separated [20, 21, 28, 29, 31, 32] or only women were focused [22, 23], and few large-scale population-based studies have examined bone and joint diseases in men [19, 24, 27, 30]. Furthermore, the association of VFx, lumbar spondylosis and knee OA with ADL and QOL may not be independent, but no studies have examined VFx, lumbar spondylosis and knee OA simultaneously in the same population using the same tools.

The objective of the present study is to clarify the impact of VFx, lumbar spondylosis and knee OA on QOL among 767 men using the cohorts of the ROAD study. In addition, to provide greater insight into the magnitude of QOL loss with VFx, lumbar spondylosis and knee OA, we made a comparison with cerebral stroke. Cerebral stroke is ranked first among diseases causing disabilities according to the recent National Livelihood Survey of the Ministry of Health, Labour and Welfare in Japan [16], and several studies have already reported that QOL is greatly affected after cerebral stroke [33]. Thus, such information can provide the health care physician with insights into the influence of VFx, lumbar spondylosis and knee OA on QOL.

Methods

Subjects

The ROAD study is a nationwide prospective study for bone and joint diseases (with OA and osteoporosis as the representative bone and joint diseases) constituting population-based cohorts established in several communities in Japan. As detailed profile of the ROAD study has already been described elsewhere [13–15, 34], the brief summary is provided here. To date, we have completed creation of a baseline database including clinical and genetic information for 3,040 inhabitants (1,061 men, 1,979 women) in the age range of 23 to 95 years (mean, 70.6 years), recruited from listings of resident registrations in three communities: an urban region in Itabashi, Tokyo, a mountainous region in Hidakagawa, Wakayama, and a seacoast region in Taiji, Wakayama. All participants provided written informed consent, and the study was conducted with the approval of ethical committees of the University of Tokyo and the Tokyo Metropolitan Institute of Gerontology. Participants completed an interviewer-administered questionnaire of 400 items that included lifestyle information such as smoking habits, alcohol consumption, family history, history, physical activity, reproductive variables and health-related QOL. Anthropometric measurements included height, weight, bilateral grip strength and body mass index (BMI) (weight [kg]/height^2 [m^2]). In the present study, to compare the magnitude of QOL loss in VFx, lumbar spondylosis and knee OA with another chronic disease, we assessed medical history of cerebral stroke by self-report. The following question was asked by an interviewer: "Have you ever experienced cerebral stroke?" Furthermore, to assess the impact of these bone and joint diseases according to symptoms, all participants were also interviewed regarding low back pain (LBP) by asking, "Have you experienced pain on most days in the past month, in addition to now?" Subjects who answered "yes" were defined as having LBP.

From the baseline data of the overall participants, the present study analyzed 767 men ≥40 years old who had completed questionnaires for the Medical Outcomes Study Short Form 8 (SF-8) and the EuroQOL (EQ-5D).

Radiographic assessment

All participants underwent radiographic examination of the lumbar spine including intervertebral levels from L1/2 to L5/S with anteroposterior and lateral views and both knees using anteroposterior and lateral views with weight-bearing and foot map positioning. Vertebral fracture, lumbar

spondylosis and knee OA were determined by a single well-experienced orthopaedist blinded to participant clinical status (S.M.). Vertebral fracture was assessed by lateral radiographs of the lumbar spine (L1–L5) in terms of a wedge, biconcave, or crush appearance according to the Japanese Society for Bone and Mineral Research (JSBMR) criteria [35]. The films were marked up, and morphometric measurements of anterior, middle and posterior heights on lateral radiography of the thoracic and lumbar spine were made. Wedge appearance was defined as a site where anterior height of the vertebra was ≤75% than posterior height. Biconcave appearance was where the height of the central part of the vertebra was ≤80% than that of the anterior or posterior parts of the vertebra. Crush appearance was where the height of the anterior, central and posterior parts of an axial vertebra was all reduced to ≤80% of the normal value (Fig. 1). Lumbar spondylosis and knee OA were assessed using the Kellgren/Lawrence (KL) radiographic atlas, and severity by KL grading was determined [36]. We have defined lumbar spondylosis and knee OA as KL ≥3 in at least one knee and in one intervertebral level, respectively. To evaluate intraobserver agreement of the JSBMR criteria for VFx and the KL grade (0–4) for the lumbar spine and knee, 100 randomly selected radiographs were scored by the same observer at >1 month after the first reading. Furthermore, 100 other radiographs were scored by two experienced orthopaedic surgeons (S.M. and H.O.) using the same radiographic atlas for interobserver agreement. Intra- and interobserver agreements were evaluated by kappa analysis. Intra- and interobserver agreements in JSBMR criteria for VFx and KL grade for lumbar spine and knee have been shown to be sufficient for assessment (0.93 and 0.91 for VFx, 0.84 and 0.76 for lumbar spine and 0.86 and 0.80 for knee, respectively).

Instruments

To carry out the QOL assessment, we used the SF-8 Health Survey (SF-8) scale. The SF-8 is an alternate form of the SF-36 Health Survey (SF-36) [37], the most widely used patient-based health status survey. The SF-8 was constructed to provide an even shorter alternative to the SF-36 for use in large population-based surveys of general and specific populations. The SF-8 uses one question to measure each of the eight SF-36 domains. Although none of the SF-8 items are identical to SF-36 items, the item pool including the SF-36 survey, SF-8 single-item scales and summary measures is scored on the same metric as the SF-36 scales and summary measures. The SF-8 and SF-36 measure eight concepts: general health (GH), physical function (PF), role physical (RP), bodily pain (BP), vitality (VT), social function (SF), mental health (MH) and role emotional (RE). Each domain includes questions regarding overall health, limitations to usual physical activities due to physical health problems, difficulties in performing daily work due to physical health, severity of pain, energy levels, limitations to usual social activities due to physical health or emotional problems, severity of emotional problems and difficulties with usual work, school or other daily activities due to personal or emotional problems, respectively. The SF-8 was scored by assigning the mean SF-36 scale score from the 2002 general Japanese population to each response category of the SF-8 measuring the same concept and then weighting each SF-8 item to compute aggregate physical (PCS) and mental (MCS) summary scale measures. The SF-8 may be scored using a published algorithm for Japanese versions of the SF-8, which have been well validated [38]. We also used the EuroQOL (EQ-5D) questionnaire [39] translated into Japanese [40]. This five-dimensional health care classification included questions on the status of morbidity, self-care, usual activities, pain/discomfort and anxiety/depression. Participants were asked to indicate current health status by ticking the most appropriate of three statements about each of five QOL dimensions. Each statement represents an increasing degree of severity. These results were coded and converted to a score of utility using the tables of values [40].

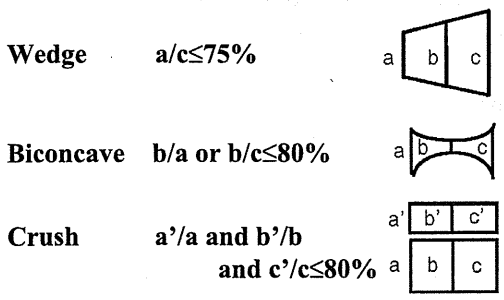

Fig. 1 Diagnostic criteria for VFxs accoding to the JSBMR

Statistical analysis

We performed nonpaired Student's *t* test to compare mean scores of QOL parameters between subjects with and without each chronic disease. Associations of VFx, lumbar spondylosis and knee OA with QOL parameters were determined by multiple regression analysis after adjustment for age and BMI. Next, to determine the independent impact of these bone and joint diseases, multiple regression analysis was used by age, BMI and presence of VFx, lumbar spondylosis and knee OA as independent variables. Furthermore, to compare the magnitude of QOL loss of the

three bone and joint diseases to cerebral stroke, multiple regression analysis was performed by age, BMI and presence of cerebral stroke in addition to VFx, lumbar spondylosis and knee OA as independent variables. Tukey honestly significant difference (HSD) test after adjustment for age and BMI was used to determine the differences of PCS values among VFx with LBP, VFx without LBP and no VFx and the differences among lumbar spondylosis with LBP, lumbar spondylosis without LBP and no lumbar spondylosis. Data analyses were performed using SAS version 9.0 (SAS Institute, Cary, NC).

Results

Impact of VFx, lumbar spondylosis and knee OA on QOL scores

Characteristics of the 767 men ≥40 years old in the ROAD study are shown in Table 1. Prevalence of lumbar spondylosis and knee OA were approximately 42% and 12%, respectively, compared to 12% for VFx. Six percent of all subjects had a medical history of cerebral stroke.

Table 1 Characteristics of participants

N	767
Age, years	69.7±10.5
Height, cm	162.8±6.7
Weight, kg	61.5±10.8
BMI, kg/m2	23.1±3.4
Medical history (%)	
Cerebral stroke	5.8
Prevalence (%)	
VFx	11.6
LS	41.6
KOA	12.0
LBP	15.4
SF-8	
GH	50.2±5.5
PF	49.9±6.2
RP	50.2±6.7
BP	50.4±9.2
VT	50.4±6.3
SF	52.4±5.5
MH	54.4±5.3
RE	52.0±5.2
PCS	47.4±6.8
MCS	53.4±5.3
EQ-5D utility score	0.91±0.14

Except where indicated otherwise, values represent mean ±SD.

BMI body mass index; *VFx* vertebral fracture; *LS* lumbar spondylosis; *KOA* knee osteoarthritis; *LBP* low back pain; *GH* general health; *PF* physical function; *RP* role physical; *BP* bodily pain; *VT* vitality; *SF* social function; *MH* mental health; *RE* role emotional; *PCS* physical component summary; *MCS* mental component summary.

Table 2 shows scores for all domains in the SF-8 and the EQ-5D utility score according to the presence of chronic diseases. Scores for PF, RP BP, VT and PCS in the SF-8 were significantly lower in subjects with VFx compared to those in subjects without VFx, but GH, SF and RE were not. Physical function, RP, SF and PCS were significantly lower in subjects with lumbar spondylosis compared to subjects without lumbar spondylosis. For knee OA, scores of PF, RP, BP and PCS were significantly lower compared to subjects without knee OA. For MCS, the score was higher in subjects with VFx and knee OA compared to those in subjects without them. EQ-5D utility score was significantly lower in subjects with lumbar spondylosis and knee OA compared to those without them, respectively, while no significant difference was apparent in subjects with or without VFx.

We next performed further multiple regression analyses to examine the independent association of VFx, lumbar spondylosis and knee OA with QOL parameters after adjusting for age, BMI and all other bone and joint diseases. Beta values in each domain of SF-8 and EQ-5D utility score after adjusting for age, BMI and all other bone and joint diseases are shown in Table 3. Vertebral fracture was significantly associated with lower scores in PF, RP, BP, VT and PCS, while not in GH, SF and RE. Knee OA was significantly associated with lower PF, BP and PCS scores. For MCS, VFx and knee OA were associated with higher scores, but lumbar spondylosis was not associated. Lumbar spondylosis and knee OA were significantly associated with EQ-5D utility scores, while VFx was not. The Tukey HSD test after adjustment for age and BMI showed that PCS score was significantly lower in subjects having VFx with LBP than in subjects having VFx without LBP (Fig. 2). Vertebral fracture with or without LBP was significantly associated with lower PCS scores compared with no VFx. However, PCS scores were significantly lower in subjects having lumbar spondylosis with LBP than in subjects having lumbar spondylosis without LBP. There were no significant differences in PCS scores between subjects having lumbar spondylosis without LBP and those having no lumbar spondylosis.

Comparison of the magnitude of QOL loss in VFx, lumbar spondylosis and knee OA with that in cerebral stroke

To compare the magnitude of QOL loss in VFx, lumbar spondylosis and knee OA with another chronic disease, we analyzed the association of medical history of cerebral stroke with QOL (Supplementary Table). Multiple regression analysis showed that cerebral stroke was significantly associated with lower QOL scores measured by PF, RP, BP SF, MH and PCS in the SF-8, along with EQ-5D utility

Table 2 Mean scores of all domains, PCS and MCS in the SF-8 and EQ-5D by VFx, LS and KOA

	VFx		LS		KOA		Japanese general population*
	No	Yes	No	Yes	No	Yes	
SF-8							
GH	50.3 (5.5)	49.4 (5.4)	50.3 (5.4)	50.1 (5.6)	50.2 (5.4)	50.6 (6.2)	50.3 (6.6)
PF	50.2 (5.8)	48.1† (8.2)	50.5 (5.7)	49.2† (6.8)	50.3 (5.7)	47.7† (8.7)	49.8 (5.0)
RP	50.5 (6.2)	47.9† (9.1)	50.7 (6.3)	49.6† (7.2)	50.5 (6.3)	48.4† (8.6)	50.3 (5.0)
BP	50.8 (9.2)	47.0† (9.1)	50.7 (9.2)	49.9 (9.3)	50.7 (9.1)	48.3† (10.0)	50.8 (7.9)
VT	50.6 (6.2)	49.2† (6.4)	50.4 (6.1)	50.4 (6.4)	50.5 (6.2)	49.7 (6.4)	52.1 (5.6)
SF	52.4 (5.5)	52.4 (5.6)	52.8 (5.0)	51.9† (6.0)	52.3 (5.4)	53.0 (5.6)	50.3 (6.3)
MH	54.2 (5.4)	55.9† (3.7)	54.5 (5.2)	54.3 (5.3)	54.2 (5.2)	55.5† (5.4)	52.9 (5.9)
RE	51.9 (5.1)	52.6 (5.4)	52.2 (4.9)	51.7 (5.6)	52.0 (5.0)	52.4 (6.1)	51.3 (4.8)
PCS	47.9 (6.5)	43.9† (8.4)	47.8 (6.7)	46.8† (7.5)	47.8 (6.5)	44.7† (8.3)	48.3 (6.2)
MCS	53.1 (5.3)	55.6† (4.8)	53.4 (5.3)	53.4 (5.2)	53.1 (5.2)	55.3† (5.5)	51.9 (5.8)
EQ-5D	0.91 (0.14)	0.89 (0.16)	0.93 (0.13)	0.89† (0.16)	0.92 (0.14)	0.87† (0.17)	

Unless otherwise indicated, values represent mean (SD).

* Reference data derived from the 2002 general Japanese men at the age of 60 to 69 years [38]

† $p<0.05$ vs. subjects without the corresponding disease by non-paired Student's t test

score (Table 4). Adjusted beta values for PF and RP in VFx were lower than those in cerebral stroke, while these for BP, VT and PCS were higher in VFx. For knee OA, adjusted beta values for PF and RP were lower than those in cerebral stroke, while those for BP and PCS were higher. For EQ-5D utility score, lumbar spondylosis and knee OA was significantly associated with lower scores, but the adjusted beta values were lower than that in cerebral stroke.

Discussion

This is the first population-based study to examine the effects of a variety of bone and joint diseases including VFx, lumbar spondylosis and knee OA on QOL as measured by both SF-8 and EQ-5D in Japanese men. In the present study, we performed multiple regression analysis to determine independent associations of QOL parameters with each bone and joint disease after adjustment for age, BMI and all other bone and joint diseases. Vertebral fracture and knee OA were significantly associated with lower PCS scores, while they were associated with higher MCS. Lumbar spondylosis was associated with the EQ-5D utility scores, while not with any domains in the SF-8. The impact of diseases on PCS was largest in VFx among the three bone and joint diseases in men. Furthermore, to provide greater insight into the magnitude of QOL loss with the bone and joint diseases, we compared the

Table 3 Beta values for VFx, LS and KOA in all domains, PCS and MCS in the SF-8 and EQ-5D

	VFx		LS		KOA	
	Beta[a]	Adjusted beta[b]	Beta[a]	Adjusted beta[b]	Beta[a]	Adjusted beta[b]
SF-8						
GH	−0.054	−0.053	−0.015	−0.009	0.018	0.020
PF	−0.094*	−0.088*	−0.051	−0.032	−0.087*	−0.082*
RP	−0.113*	−0.109*	−0.038	−0.016	−0.073*	−0.069
BP	−0.133*	−0.131*	−0.035	−0.010	−0.081*	−0.078*
VT	−0.074*	−0.074*	−0.003	0.011	−0.056	−0.055
SF	0.011	0.018	−0.051	0.060	0.066	0.071
MH	0.106*	0.109*	−0.013	−0.035	0.076*	0.076*
RE	0.038	0.046	-0.052	−0.062	0.032	0.035
PCS	−0.181*	−0.178*	−0.042	−0.007	−0.118*	−0.113*
MCS	0.149*	0.153*	−0.015	−0.047	0.121*	0.122*
EQ-5D	−0.050	−0.035	−0.107*	−0.096*	−0.081*	−0.073*

[a] Beta values are shown using multiple regression analysis after adjustment for age and BMI

[b] Adjusted beta values are shown using multiple regression analysis after adjustment for age, BMI and all other diseases

* $p<0.05$

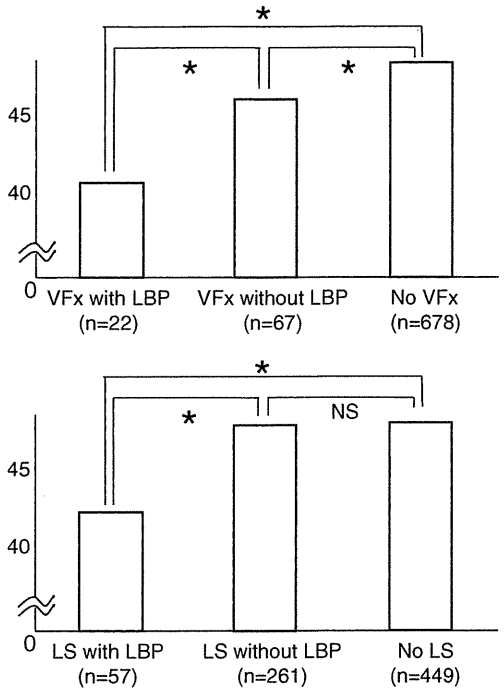

Fig. 2 Physical component summary values among subjects having VFx with LBP, VFx without LBP and no VFx, and those having lumbar spondylosis (LS) with LBP, LS without LBP and no LS. The number of subjects in each group is shown in parentheses. *$p<0.05$ by Tukey HSD test adjustment for age and BMI

impact of these bone and joint diseases on QOL loss with that of cerebral stroke. The impact of VFx and knee OA on BP and PCS loss was larger than that of cerebral stroke.

Few population-based studies have examined relationships between radiographic VFx and QOL [19–21, 24, 27], and genders were only adjusted, not separated in almost all these studies, although the impact of vertebral deformities on QOL may differ between genders. In the Canadian Multicentre Osteoporosis Study [27], no strong fracture-related associations of subclinical vertebral deformity with QOL were found in men as measured by the Health Utilities Index (HUI) Mark II and III Systems [41]. In the present study, radiographic VFx showed a significant association with lower PCS scores in men, with the largest impact among the three bone and joint diseases. Furthermore, multiple regression analysis showed that the magnitude of PCS loss was stronger than that of cerebral stroke, which is ranked first among the diseases causing disabilities according to the recent National Livelihood Survey of the Ministry of Health, Labour and Welfare in Japan [16]. Furthermore, VFx with LBP had a strong effect on PCS, while VFx without LBP had a moderate but significant effect. Reasons for the discrepancy between the present study and the Camos study can be partly attributed to differences in QOL measurements. In HUI scores, multi-attribute utility score reflects global health, but PCS reflects only physical QOL. In fact, for pain domains, subclinical vertebral deformities tend to be associated with lower HUI scores as seen for the SF-8 in the present study. Another reason may be racial differences. Racial variations exist in the prevalence of vertebral deformities [42, 43], so differences may also exist in the impact of QOL. For GH, SF and RE in the SF-8, VFx was not associated with the scores by multiple regression analysis. In this study, VFx was diagnosed according to lumbar radiographs, so many fractures in men may have occurred years previously and may have been related to trauma, which must be one reason why some domains of QOL were not impaired.

The significant effect of knee OA on QOL is shown by poorer scores in PF, BP and PCS domains in the SF-8 in multiple regression analysis. A previous survey in Chinese using a GH-related QOL measure also showed that OA has comparable impact compared with stroke, asthma and chronic obstructive pulmonary disease [32], but men and women were not separated in that analysis. The present study is the first population-based study to clarify that knee OA is significantly associated with lower PCS scores in men. Although multiple regression analysis showed that the magnitude of the impact on PCS is lower than VFx, prevalence is much higher for knee OA than for VFx, so the total burdens of these diseases might be similar.

Likewise, for lumbar spondylosis, few population-based studies have examined QOL [20]. Unlike VFx or knee OA, multiple regression analysis in the present study showed that lumbar spondylosis was not associated with PCS in men, supporting previous findings [20], although gender was only adjusted in that analysis. These results may be

Table 4 Comparison of adjusted beta values for the three bone and joint diseases such as VFx, LS and KOA with that for cerebral stroke in the SF-8 and EQ-5D

	VFx	LS	KOA	Stroke
SF-8				
GH	−0.054	0.007	0.004	−0.070
PF	−0.092*	−0.031	−0.097*	−0.107*
RP	−0.115*	−0.015	−0.083*	−0.125*
BP	−0.136*	−0.008	−0.086*	−0.079*
VT	−0.078*	0.004	−0.063	−0.029
SF	0.013	−0.061	0.061	−0.100*
MH	0.106*	−0.033	0.066	−0.096*
RE	0.042	−0.065	0.028	−0.053
PCS	−0.183*	0.007	−0.128*	−0.107*
MCS	0.151*	−0.049	0.118*	−0.036
EQ-5D	−0.039	−0.090*	−0.081*	−0.122*

Adjusted beta values are shown using multiple regression analysis after adjustment for age, BMI and all other diseases

*$p<0.05$

explained by the fact that associations between lumbar spondylosis and LBP are not so strong [13, 44, 45]. In fact, the domain of BP score in SF-8 was not associated with lumbar spondylosis in this study.

In the present study, VFx and knee OA were significantly associated with lower PCS, while they were associated with higher MCS. Past literatures also showed the dissociation between PCS and MCS in VFx and knee OA [20, 46]. Several factors may contribute to the dissociation between MCS and PCS for VFx and knee OA. First, MCS questions within the SF-8 include generic questions about energy levels, feelings of being "downhearted and blue," and interference in daily activities as a result of emotional problems. These questions are less sensitive to the presence of MH issues than disease-specific scales such as the Kessler psychological distress scale [47]. In fact, Hill et al. [48] showed that psychological distress has been shown to be significantly more frequent in those with arthritis than those without, although scores on the MCS were not significantly different between these two groups. Second, the dissociation may be due to a disability paradox [49], which suggests that people with chronic disabilities report serious limitations in ADL, problems in performing social roles, yet state that they have excellent or good QOL. Many subjects with VFx and knee OA had LBP or knee pain, which leads to functional impairment. This may be associated with lower scores of PCS, but the individual may not feel that the impairment of social activity or ADL was due to mental factors. Particularly in elderly individuals, pain or functional impairment may be considered a natural consequence of being elderly. Vertebral fracture and knee OA were thus not associated with lower scores for SF or RE domains in the SF-8 and, thus, showed no associations with MCS. Conversely, elderly individuals may think that having cerebral stroke is not a natural consequence of being elderly, potentially contributing to the differences between VFx, knee OA and cerebral stroke.

The present study showed that the association of chronic diseases with QOL differed between the SF-8 and EQ-5D. For VFx, PCS of the SF-8 was reduced, while EQ-5D utility score was not, while for lumbar spondylosis, both PCS and MCS of the SF-8 was not associated, but the EQ-5D utility scores were significantly reduced. The reason may be explained by the fact that in the EQ-5D, all five domains are combined together to analyze the association with chronic diseases, while PCS and MCS are analyzed separately in the SF-8. In fact, associations of VFx differed between PCS and MCS of the SF-8, so when all domains were combined together, the results may differ. Lumbar spondylosis reduced both the PCS and MCS scores, although they were not significant, so when combined, the association may be significant. For VFx and knee OA, the SF-8 may be more useful to examine associations with QOL than the EQ-5D.

There are several limitations in the present study. First, this was a large-scale, population-based study, but a cross-sectional study of baseline data. Causal relationships could therefore not be determined. The ROAD study is a longitudinal survey, so further progress may help to elucidate any causal relationships. Second, among the 1,047 men ≥40 years old in the ROAD study, 767 men had completed questionnaires for both the SF-8 and the EQ-5D, so the response rate was 73.7%. Subjects who completed questionnaires may have had better QOL than those who did not, so our results regarding QOL may have represented overestimations. Third, we only used semi-quantitative methods to assess VFx. Furthermore, we used the KL system for lumbar spondylosis and knee OA. Since the KL system emphasizes osteophytosis, it is unclear how to handle lumbar spondylosis or knee OA with disc or joint space narrowing but no osteophytosis. We are currently developing a computer-aided diagnostic program to enable fully automatic measurement of the major features of VFx, lumbar spondylosis and knee OA, including joint and disc space narrowing and osteophytosis on plain radiography [50]. Fourth, cerebral stroke was assessed by self-report, so severity could not be examined. Furthermore, cerebral stroke is a serious disease, so participants are considered highly likely to know if they have been diagnosed with cerebral stroke, although some participants may not; thus, strict comparison with bone and joint diseases was limited.

Conclusions

The present cross-sectional study using a large-scale population from the ROAD study revealed that VFx and knee OA were significantly associated with lower PCS scores of the SF-8 in men, while lumbar spondylosis was not. The impact of diseases on PCS was the largest for VFx in men. Further progress, along with continued longitudinal survey in the ROAD study, will elucidate the environmental and genetic backgrounds of VFx, lumbar spondylosis and knee OA and the relationship with QOL.

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Conflict of Interest All authors have no conflicts of interest.

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