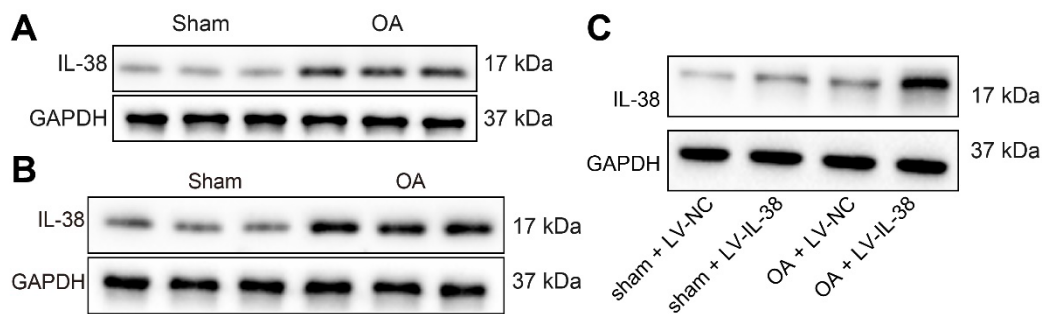# Supplementary Material

Fig. a. Representative western blot images of interleukin (IL)-38 protein. a) Representative western blot images of protein expressions of IL-38 in cartilage tissues from osteoarthritis (OA) mice. b) Representative western blot images of protein expression of IL-38 in knee joint cartilage tissues from OA mice and sham-operated mice in response to lentivirus vector (LV)-IL-38 or LV-NC. c) Representative western blot images of protein expression of IL-38 in chondrocytes after IL-1β treatment. GAPDH, glyceraldehyde 3-phosphate dehydrogenase.
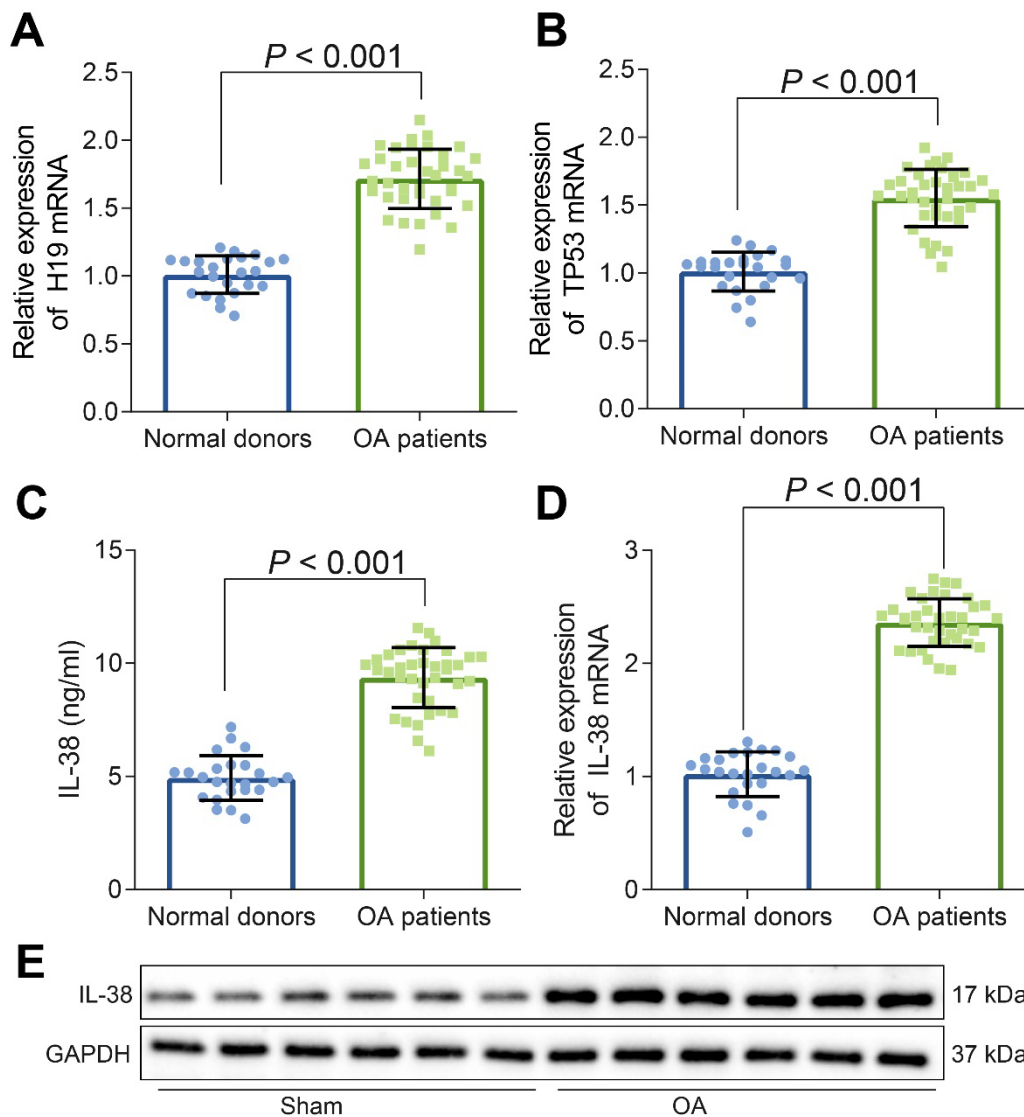
Fig. b. The expression of long noncoding RNA (lncRNA) H19, tumour protein p53 (TP53), and interleukin (IL)-38 in 37 osteoarthritis (OA) patients and 24 healthy subjects. a) The expression level of H19 in cartilage samples of 37 OA patients and 24 healthy subjects determined by quantitative reverse transcription polymerase chain reaction (RT-qPCR). b) The expression level of TP53 in cartilage samples of 37 OA patients and 24 healthy subjects determined by RT-qPCR. c) IL-38 level in synovial fluid from 37 OA patients and 24 healthy subjects (without previous history of arthritis) detected by enzyme-linked immunosorbent assay (ELISA). d) Messenger RNA (mRNA) expression of IL-38 in the cartilage tissues from 37 OA patients and 24 healthy subjects (without previous history of arthritis) determined by RT-qPCR. e) Western blot for IL-38 protein expression. GAPDH,

glyceraldehyde 3-phosphate dehydrogenase. In Figures ba to bd, comparison between two groups was conducted by independent-samples *t*-test.
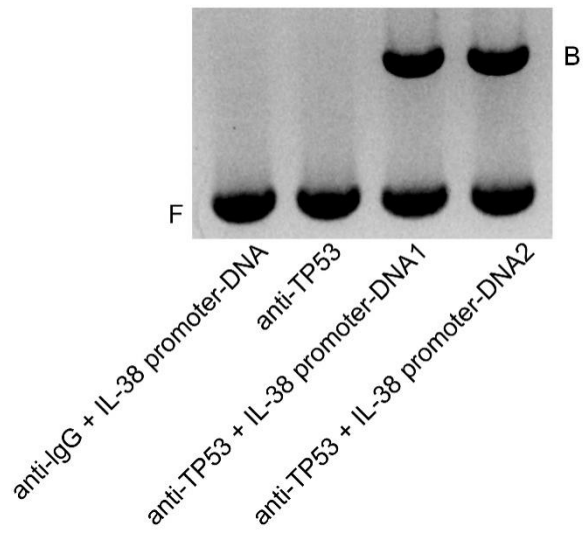
Fig. c. Interleukin (IL)-38 promoter-DNA with mouse tumour protein p53 (TP53) determined by gel electrophoresis mobility shift assay (EMSA). F, free DNA; B, protein-DNA complex; IgG, immunoglobulin G.

Table i. Primer sequences for quantitative reverse transcription polymerase chain reaction.

Gene	Primer sequence
lncRNA H19 mouse	F: 5'-CATGCTCCAGAGGGAATCGT-3'
	R: 5'-GCTTCAACTGATTCCGTGGC-3'
TP53 mouse	F: 5'-CCTCACCATCATCACACTGG-3'
	R: 5'-CCTCATTCAGCTCTCGGAAC-3'
IL-38 mouse	F: 5'-CCCTACAGCTGGAGGATGTGA-3'
	R: 5'-GAAGCGTGTGGCCTCTTCA-3'
IL-38 human	F: 5'-AGAAGAGGGGCCTTCCCTAC-3'
	R: 5'-GAAGAAGGTGAAGCGTGTGG-3'
H19 human	F: 5'- GCCTCGCGGGCGGCGACG -3'
	R: 5'- GCACGTCCACCCCAGCTG -3'
TP53 human	F:5'- ACTCATGTTCAAGACAGAAG -3'
	R:5'- CTAACCCTTAACTGCAAGAAC -3'
GAPDH human	F:5'- AATGGAAATCCCATCACCA -3'
	R:5'- CCACTTGATTTTGGAGGGA -3'
GAPDH mouse	F: 5'-TCAGCAATGCCTCCTGCAC-3'
	R: 5'-TCTGGGTGGCAGTGATGGC-3'

lncRNA, long noncoding RNA; TP53, tumour protein 53; IL-38, interleukin-38; GAPDH, glyceraldehyde

3-phosphate dehydrogenase; F, forward; R, reverse.

Table ii. Original data of quantitative reverse transcription polymerase chain reaction.

H19	Target unknown (H19)	Reference unknown (GAPDH)	2^△(Ct(A-B))
Normal			
N1	25.57573342	16.73450419	0.871
N2	25.74209238	17.00095596	0.934
N3	25.21140512	16.65724887	1.063
N4	25.84962319	17.41882689	1.158
N5	25.99833744	17.2462418	0.927
N6	25.03355614	16.43487731	1.031
N7	26.00467408	17.50298524	1.103
N8	25.31170349	16.84137883	1.127
N9	25.65952295	16.78617615	0.852
N10	25.47864676	16.642424	0.874
N11	25.88188619	17.16395715	0.949
N12	25.68470019	17.07893889	1.026
N13	25.85277516	16.82628526	0.766
N14	25.34721799	16.70113924	0.998
N15	25.55103648	17.15011586	1.182
N16	25.78467084	17.28694002	1.106
N17	25.98940665	16.84617179	0.707
N18	25.26308953	16.78888143	1.124
N19	25.87911459	17.39710812	1.118
N20	25.28926371	16.92002517	1.209
N21	25.52674505	16.60649134	0.825
N22	24.62237551	16.16620134	1.138
N23	25.41754722	16.92376358	1.109
N24	26.21116454	17.62093902	1.037
OA	**Target unknown**	**Reference unknown**	**2^△(Ct(A-B))**
OA-1	24.52012485	16.83798193	1.946
OA-2	24.21643211	16.45191424	1.838
OA-3	24.39052904	16.54944698	1.743
OA-4	24.62059754	16.58201423	1.520
OA-5	25.08389	17.26661397	1.772
OA-6	25.44012702	17.59573032	1.739
OA-7	24.58188164	16.6262877	1.610
OA-8	24.07961941	16.09688934	1.580
OA-9	24.804374	16.86393362	1.627
OA-10	24.32795108	16.78896193	2.149
OA-11	24.64978602	17.01360895	2.009
OA-12	24.93780548	17.26968016	1.965
OA-13	24.66131393	17.04510528	2.037
OA-14	25.3775512	17.65099099	1.887
OA-15	25.65417394	17.26852992	1.195
OA-16	24.81535213	16.92544609	1.685
OA-17	24.68815858	17.01193441	1.954
OA-18	24.23723502	16.09232047	1.412

OA-19	24.94039477	16.93279797	1.553
OA-20	24.38021052	16.60624265	1.826
OA-21	25.43703555	17.33639127	1.456
OA-22	24.41692172	16.67344271	1.865
OA-23	24.6456797	16.90065275	1.863
OA-24	25.01204762	17.08396822	1.641
OA-25	25.61270973	17.61806028	1.567
OA-26	24.76143203	16.59386228	1.390
OA-27	24.72637883	16.9389166	1.809
OA-28	24.07936364	16.41050393	1.964
OA-29	25.45791252	17.25461507	1.356
OA-30	24.12353214	16.31113283	1.778
OA-31	24.80373751	16.87389873	1.639
OA-32	25.25570174	17.07980472	1.382
OA-33	25.15857475	17.23576067	1.647
OA-34	24.27178668	16.32957176	1.625
OA-35	25.27284267	17.48378454	1.807
OA-36	24.84373105	16.96745956	1.701
OA-37	24.37967178	16.70344761	1.954
IL-38	**Target unknown (IL-38)**	**Reference unknown (GAPDH)**	**2^△(Ct(A-B))**
Normal			
N1	26.88736903	17.87963921	1.208
N2	26.38318026	17.13272013	1.021
N3	25.75571237	16.37890651	0.935
N4	25.62208269	16.2534047	0.941
N5	25.62420273	16.56115103	1.162
N6	27.44485984	17.94385625	0.858
N7	25.60690191	16.71319218	1.307
N8	26.29229537	17.08740555	1.054
N9	26.48339649	17.24186948	1.027
N10	26.17369419	17.12759256	1.176
N11	26.64202193	16.75180683	0.655
N12	26.3646014	17.38189727	1.229
N13	26.70123131	17.4774063	1.040
N14	26.1346167	16.990645	1.099
N15	26.77762232	17.1031359	0.761
N16	26.10978462	17.03090794	1.150
N17	25.93868875	16.74962916	1.065
N18	27.05725887	16.80380869	0.509
N19	25.33411391	16.33393748	1.214
N20	26.09893834	16.83346564	1.010
N21	26.3103272	17.33751305	1.237
N22	25.77554332	16.60497096	1.079
N23	26.60921874	16.9064036	0.746
N24	25.75260875	16.53024915	1.041
OA	**Target unknown (IL-38)**	**Reference unknown (GAPDH)**	**2^△(Ct(A-B))**
OA-1	24.68148523	16.83798193	2.707
OA-2	24.55834536	16.45191424	2.256

OA-3	24.69345378	16.54944698	2.198
OA-4	24.50251679	16.60581555	2.609
OA-5	25.21982349	17.02164657	2.117
OA-6	26.08665328	17.90406589	2.140
OA-7	24.85575266	16.94236625	2.579
OA-8	25.12395066	17.30265938	2.749
OA-9	24.59476403	16.70853112	2.628
OA-10	24.81161142	16.48969913	1.943
OA-11	24.23183994	16.22246967	2.413
OA-12	24.53095871	16.69118124	2.714
OA-13	25.47211594	17.45555316	2.401
OA-14	25.16551193	16.95981918	2.106
OA-15	25.09496762	17.14303146	2.511
OA-16	25.79573334	17.7315243	2.323
OA-17	24.28704847	16.15935823	2.223
OA-18	24.74001289	16.67144996	2.316
OA-19	24.66649634	16.64210099	2.388
OA-20	25.1455971	17.14040596	2.420
OA-21	24.76407171	16.78604632	2.466
OA-22	24.53431395	16.33409113	2.114
OA-23	24.38563052	16.42793738	2.501
OA-24	24.78277184	16.69792125	2.290
OA-25	25.37682033	17.12235971	2.036
OA-26	24.87652219	16.80109076	2.305
OA-27	24.37662931	16.46044318	2.574
OA-28	24.87543304	16.7893219	2.288
OA-29	24.34356645	16.32640266	2.400
OA-30	24.68310758	16.80180696	2.637
OA-31	24.39796327	16.24407722	2.183
OA-32	25.73080161	17.41629537	1.953
OA-33	25.39316876	17.37540373	2.399
OA-34	25.09403388	16.93219542	2.171
OA-35	24.9457068	16.93991938	2.419
OA-36	25.13201137	17.15865868	2.474
OA-37	25.22955592	17.21659388	2.407
TP53	**Target unknown (TP53)**	**Reference unknown (GAPDH)**	**2^△(Ct(A-B))**
Normal			
N1	23.94029384	16.54314099	1.131
N2	25.01981627	17.50589635	1.043
N3	24.44881132	16.98840691	1.083
N4	24.78804688	17.15620124	0.961
N5	23.76495839	16.29509365	1.076
N6	24.02296458	16.24601026	0.869
N7	25.49355725	17.7725411	0.904
N8	25.06138492	17.44011959	0.968
N9	24.22563751	16.91844804	1.204
N10	24.45970033	17.01936154	1.098
N11	24.38377016	16.77300812	0.975
N12	24.55603685	17.0407183	1.042

N13	24.74831759	17.48526526	1.241
N14	25.035794	16.81670852	0.640
N15	24.29616915	16.78783026	1.047
N16	24.59142057	17.11339731	1.069
N17	25.19128757	17.18970236	0.744
N18	24.70343491	16.80216923	0.798
N19	23.81939709	16.32904888	1.060
N20	24.06254787	16.71107784	1.168
N21	24.12655371	16.64032553	1.063
N22	24.558102	17.11110564	1.093
N23	24.1281355	16.66098005	1.078
N24	24.64169957	16.92229627	0.905
OA	**Target unknown (TP53)**	**Reference unknown (GAPDH)**	**2^△(Ct(A-B))**
OA-1	23.91111835	16.83798193	1.416
OA-2	23.35512566	16.45191424	1.593
OA-3	23.23843991	16.54944698	1.848
OA-4	23.48586225	16.51876746	1.524
OA-5	24.4029151	17.0933943	1.202
OA-6	23.76068122	16.8447347	1.579
OA-7	23.93798015	17.11234218	1.681
OA-8	24.21086602	17.14382979	1.422
OA-9	23.32180397	16.57222489	1.772
OA-10	23.89719934	17.10800143	1.724
OA-11	23.26137194	16.62977301	1.923
OA-12	24.95187818	17.77964267	1.322
OA-13	23.39095759	16.55411919	1.668
OA-14	24.21483604	17.35357417	1.640
OA-15	23.01041725	16.23537576	1.741
OA-16	24.40565891	17.1199455	1.222
OA-17	23.84853047	16.82644166	1.467
OA-18	23.48584541	16.53942804	1.546
OA-19	23.40472913	16.67295133	1.794
OA-20	23.71924481	16.71377822	1.484
OA-21	23.98530452	17.05835201	1.567
OA-22	24.40905921	17.6064095	1.708
OA-23	24.05910769	16.99511192	1.425
OA-24	23.75480337	16.89442092	1.641
OA-25	25.28922734	17.90835671	1.144
OA-26	24.25941659	17.40517514	1.648
OA-27	23.88714483	16.94817365	1.554
OA-28	23.44897869	16.70751834	1.782
OA-29	24.42147081	17.54606476	1.624
OA-30	23.86615954	16.94104721	1.569
OA-31	24.50601099	17.14642128	1.161
OA-32	24.09540576	17.06344895	1.457
OA-33	24.83668313	17.32384715	1.044
OA-34	23.15328957	16.4430628	1.821
OA-35	23.06918344	16.24010844	1.677
OA-36	24.18098781	17.32762152	1.649

OA-37	23.92397755	16.81994712	1.386

H19, long noncoding RNA H19; TP53, tumour protein 53; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; OA, osteoarthritis.

ARRIVE

The ARRIVE guidelines 2.0: author checklist

The ARRIVE Essential 10

These items are the basic minimum to include in a manuscript. Without this information, readers and reviewers cannot assess the reliability of the findings.

Item		Recommendation	Section/line number, or reason for not reporting
Study design	1	For each experiment, provide brief details of study design including: a. The groups being compared, including control groups. If no control group has been used, the rationale should be stated. b. The experimental unit (e.g. a single animal, litter, or cage of animals).	
Sample size	2	a. Specify the exact number of experimental units allocated to each group, and the total number in each experiment. Also indicate the total number of animals used. b. Explain how the sample size was decided. Provide details of any *a priori* sample size calculation, if done.	
Inclusion and exclusion criteria	3	a. Describe any criteria used for including and excluding animals (or experimental units) during the experiment, and data points during the analysis. Specify if these criteria were established *a priori.* If no criteria were set, state this explicitly. b. For each experimental group, report any animals, experimental units or data points not included in the analysis and explain why. If there were no exclusions, state so. c. For each analysis, report the exact value of *n* in each experimental group.	
Randomisation	4	a. State whether randomisation was used to allocate experimental units to control and treatment groups. If done, provide the method used to generate the randomisation sequence. b. Describe the strategy used to minimise potential confounders such as the order of treatments and measurements, or animal/cage location. If confounders were not controlled, state this explicitly.	
Blinding	5	Describe who was aware of the group allocation at the different stages of the experiment (during the allocation, the conduct of the experiment, the outcome assessment, and the data analysis).	
Outcome measures	6	a. Clearly define all outcome measures assessed (e.g. cell death, molecular markers, or behavioural changes). b. For hypothesis-testing studies, specify the primary outcome measure, i.e. the outcome measure that was used to determine the sample size.	
Statistical methods	7	a. Provide details of the statistical methods used for each analysis, including software used. b. Describe any methods used to assess whether the data met the assumptions of the statistical approach, and what was done if the assumptions were not met.	
Experimental animals	8	a. Provide species-appropriate details of the animals used, including species, strain and substrain, sex, age or developmental stage, and, if relevant, weight. b. Provide further relevant information on the provenance of animals, health/immune status, genetic modification status, genotype, and any previous procedures.	
Experimental procedures	9	For each experimental group, including controls, describe the procedures in enough detail to allow others to replicate them, including: a. What was done, how it was done and what was used. b. When and how often. c. Where (including detail of any acclimatisation periods). d. Why (provide rationale for procedures).	
Results	10	For each experiment conducted, including independent replications, report: a. Summary/descriptive statistics for each experimental group, with a measure of variability where applicable (e.g. mean and SD, or median and range). b. If applicable, the effect size with a confidence interval.	

The Recommended Set

These items complement the Essential 10 and add important context to the study. Reporting the items in both sets represents best practice.

Item			Recommendation	Section/line number, or reason for not reporting
Abstract	11		Provide an accurate summary of the research objectives, animal species, strain and sex, key methods, principal findings, and study conclusions.	
Background	12	a.	Include sufficient scientific background to understand the rationale and context for the study, and explain the experimental approach.	
		b.	Explain how the animal species and model used address the scientific objectives and, where appropriate, the relevance to human biology.	
Objectives	13		Clearly describe the research question, research objectives and, where appropriate, specific hypotheses being tested.	
Ethical statement	14		Provide the name of the ethical review committee or equivalent that has approved the use of animals in this study, and any relevant licence or protocol numbers (if applicable). If ethical approval was not sought or granted, provide a justification.	
Housing and husbandry	15		Provide details of housing and husbandry conditions, including any environmental enrichment.	
Animal care and monitoring	16	a.	Describe any interventions or steps taken in the experimental protocols to reduce pain, suffering and distress.	
		b.	Report any expected or unexpected adverse events.	
		c.	Describe the humane endpoints established for the study, the signs that were monitored and the frequency of monitoring. If the study did not have humane endpoints, state this.	
Interpretation/ scientific implications	17	a.	Interpret the results, taking into account the study objectives and hypotheses, current theory and other relevant studies in the literature.	
		b.	Comment on the study limitations including potential sources of bias, limitations of the animal model, and imprecision associated with the results.	
Generalisability/ translation	18		Comment on whether, and how, the findings of this study are likely to generalise to other species or experimental conditions, including any relevance to human biology (where appropriate).	
Protocol registration	19		Provide a statement indicating whether a protocol (including the research question, key design features, and analysis plan) was prepared before the study, and if and where this protocol was registered.	
Data access	20		Provide a statement describing if and where study data are available.	
Declaration of interests	21	a.	Declare any potential conflicts of interest, including financial and non-financial. If none exist, this should be stated.	
		b.	List all funding sources (including grant identifier) and the role of the funder(s) in the design, analysis and reporting of the study.	