SUPPLEMENTARY CLINICAL AND GENETIC FINDINGS OF SELECTED PATIENTS

Patient P3/FIII and P4/FIII

Patient 3/FIII (Fig. 1C) is the second child born to healthy consanguineous first cousin Pakistani parents. He was born at 33 weeks gestation by Caesarean section after spontaneous rupture of membranes. He was noted to have marked hypotonia in the neonatal period and went on to have motor delay. He first sat at the age of 18 months and walked independently at the age of 4 years. He had failure to thrive requiring percutaneous endoscopic gastrostomy (PEG) in the newborn period and had recurrent hospitalizations during infancy for respiratory tract infections. Severe kyphoscoliosis was noted at 3 months of age with subsequent orthopedic surgery at the age of 4 and reoperation at 9.5 years (growth rods fused). Examination at the age of 6 showed bilateral pes planus secondary to laxity of the subtalar joint, long feet, a broad-based stance and bluish sclerae. His skin was soft and doughy but not particularly hyperextensible or thin. His PEG was reversed at age 7 and he has fed well orally since. He has a bifid uvula and submucous cleft palate. Grommets were inserted at a young age for conductive hearing loss and he wears a hearing aid since the age of 12. For his bilateral severe planovalgus feet he underwent surgery at the age of 11. At the latest clinical follow-up he was 12 years old and able to walk short distances independently. However, he complains of recurrent falls; for longer distances he needs crutches due to partial flexion at knees.

P4/FIII is the younger brother of P3/FIII. He developed cyanotic episodes soon after birth and required a nasopharyngeal airway. U-shaped cleft palate, mild laryngomalacia and micrognathia were noted. Hypotonia, joint laxity and a scoliosis were identified in the newborn period. Growth parameters in the newborn period included an OFC on the 2nd centile and height and weight on 50th centile. He was found to have soft skin with redundancy of skin over abdomen, bluish sclerae, epicanthic folds and valgus deformity of both ankles. He underwent spinal surgery for scoliosis at the age of 3.5 years. He started walking at the age of 4.5 years. He suffers from recurrent otitis media with effusion.

Sanger sequencing of *FKBP14* in both patients revealed homozygosity for the mutation c.197+5_197+8 del in intron 1. Both parents were confirmed to be heterozygous carriers of the mutation. This mutation leads to the insertion of 17 nucleotides in the transcript of *FKBP14* in

between exons 1 and 2 and to a new open reading frame from c.198 on and a p. His67* premature termination codon (CAC -> TAA), as seen by transcript analysis of P5/FIV.

Patient P7/FV

Patient 7/FV (Figure 1F-G) of Croatian origin was born at term after a pregnancy characterized by poor fetal growth and reduced fetal movements. Clinical examination at the age of 2 months showed severe muscular hypotonia, generalized weakness and joint hypermobility along with craniofacial dysmorphisms including micrognathia, a high arched palate, and an asymmetric chest. At the age of one year kyphoscoliosis of the thoracic spine was noted (Figure 3SA). She was able to sit independently by the age of 21 months, and to walk independently by the age of 30 months. Her cognitive and intellectual development was normal. At age 3 years prominent valgus position of the foot and hallux varus were present. At age 4 years despite continuous physical therapy the kyphoscoliosis had worsened. After the use of halo-traction she underwent spinal surgery with insertion of spinal rods. Gastro-esophageal reflux and repeated episodes of vomiting resulted in poor weight gain. Follow-up at age 8 years revealed that motor development and muscular hypotonia improved over time, whereas the kyphoscoliosis was still progressive despite intensive orthopedic management. She presented mild sensorineural hearing loss.

Sanger sequencing of *FKBP14* revealed homozygosity for the common mutation c.362dupC p.(Glu122Argfs*7).

Patient P8/FVI

Patient 8/FVI (Figure1M-O) is the second child of Austrian parents originating from a small village. The mother reported feeble fetal movements during pregnancy. The girl was born at 38 weeks gestation and showed significant hypotonia. Neonatal sepsis was suspected and antibiotic treatment started. The general condition of the newborn rapidly stabilized, but muscular hypotonia persisted. No thoracic asymmetry or kyphosis were reported in the newborn period. Congenital flexion contractures of the elbows and the wrists improved, and initial feeding problems resolved. At the age of 3 months, the first signs of scoliosis developed. Muscle tone and strength increased during the first year of life, but motor development was delayed. Head control was achieved in the second year

of life, and she walked without assistance at 3.5 years. Eventually at the age of 9 years mild to moderate sensorineural high frequency hearing loss was diagnosed. Ophthalmological examinations showed strabismus and mild hyperopia. Scoliosis progressed despite management with a thoracolumbar orthosis (Figure 1M; Figure S3C) and at the age of 6.5 years spinal surgery was performed. In the pressure areas under the thoracolumbar orthosis follicular hyperkeratosis respectively hyperkeratotic skin eruptions evolved (Figure 1O). Cognitive development was normal. At the age of 8 years she had contractures of the elbows (Figure 1N) and distally pronounced hypermobility of the remaining joints. Muscular strength and endurance were reduced.

Homozygosity for the common c.362dupC; p.(Glu122Argfs*7) variant in FKBP14 was found by Sanger sequencing, whereas both parents were heterozygous carriers.

P12/FX

The girl (Figure 1I), an only child of young, healthy, non-consanguineous parents, was born after an uneventful pregnancy at 35 weeks of gestation after premature rupture of the membranes. She had mild perinatal asphyxia and the neonatal period was complicated by neonatal sepsis and severe muscular hypotonia. Unilateral hip dysplasia and valgus deformity of feet were noted. Neonatal hearing test was normal. During the first months of life a patent foramen ovale and flaccid leaflets of the mitral valve were diagnosed. Motor milestones were delayed and physiotherapy was started. Physical examination at the age of 9 months showed a chest deformity, excessive sweating, generalized hypotonia and diminished deep tendon reflexes. Improvement of motor function was observed in subsequent follow up visits. At the age of 2 years the foramen ovale had closed, spontaneously. At the same time, sensorineural hearing impairment was diagnosed and she was provided with a hearing aid. She was found to have myopia, pes planus, and generalized hypermobility with recurrent dislocations of the patella, skin hyperextensibility, and easy bruising. Since the age of 6 years she complains of severe myalgia with normal CK level. Thoraco-lumbar scoliosis, diastasis recti and lumbar hyperlordosis started at the age of 7 years. Based on these findings FKBP14-deficient EDS was suspected and confirmed by sequence analysis revealing homozygosity for the common mutation c.362dupC p.(Glu122Argfs*7), while the parents were carriers.

SUPPLEMENTARY TABLES AND FIGURES

Table S1. Biochemical and neuromuscular investigations

Patient	**P1/FI**	**P2/FII**	**P3/FIII**	**P4/FIII**	**P5/FIV**	**P6/FIV**	**P7/FV**	**P8/FVI**	**P9/FVII**
Age / gender	9 y / F	9 y / F	13 y / M	6 y / M	8 y / M	4 y / M	11 y / F	9 y / F	2 y / F
LABORATORY									
Creatine kinase	ND	Normal	Normal	ND	Normal	Normal	Normal	Normal	ND
Urinary LP/HP ratio	Decreased	Decreased	Normal	ND	Normal	ND	Normal	ND	Normal
NEUROMUSCULAR									
Nerve conduction (age)	Normal	Normal	Normal	ND	Normal (9 m)	ND	Normal (3 m)	ND	Normal (2 m)
Electromyography (age)	Myopathic (4 m)	Normal	Normal (8 y)	ND	Normal (9 m)	ND	Myopathic (3 m) Myopathic (1 y) Normal (8 y)	ND	Normal (2 m)
Muscle biopsy (age)	ND	ND	Normal (8 y)	ND	ND	ND	Myopathic with increased variability of fiber size (3 y)	Normal (1 y)	ND
Muscle MRI (age)	ND	ND	ND	ND	ND	ND	Mild atrophy of rectus femoris, mild hypertrophy of vastus med. (9 y)	ND	Normal (2 m)
Muscle ultrasound (age)	ND	ND	Increased echogenicity (11 y)	ND	ND	ND	ND	ND	ND

Patient	**P9/FVII**	**P10/FVIII**	**P11/FIX**	**P12/FX**	**P13/FXI**	**P14/FXII**	**P15/FXIII**	**P16/FXIV**	**P17/FXV**
Age / gender	2 y / F	15 y / F	6 y / M	9 y / F	5 y / F	16 y / M	36 y / F	24 y / M	11 m / M
LABORATORY									
Creatine kinase	ND	Normal	Normal	Normal	Normal	NR	ND	ND	ND
Urinary crosslinks (LP/HP ratio)	Normal	ND	ND	ND	ND	NR	Normal	Decreased	ND
NEUROMUSCULAR									
Nerve conduction (age)	Normal (2 m)	Normal	ND	Normal (15 m; 8 y)	ND	NR	Normal (7 m)	ND	ND
Electromyography (age)	Normal (2 m)	Non-specific findings	ND	Myopathic (9 m), improvement in follow-up, Normal (8 y)	Normal	NR	Non-specific findings (7 m)	ND	ND
Muscle biopsy (age)	ND	Myopathic with increased variability of fiber size (4 m)	Non-uniform myopathic with mild type I fiber predominance and type I myofiber size disproportion (4 m)	COX-negative fibers EM: no structural abnormality (15 m)	ND	NR	Uniformly small sized muscle fibers (5 m)	ND	ND
Muscle MRI (age)	Normal (2 m)	ND	ND	ND	ND	NR	ND	ND	ND
Muscle ultrasound (age)	ND	ND	Relatively normal appearance with some degree of granularity	ND	ND	NR	ND	ND	ND

The following symbols and abbreviations are used: EM, electron microscopy; F, female; M, male; m, months; ND, not done; NR, not recorded; y, years

Table S2. Follow up and clinical findings of previously published patients (Baumann et al., 2012)

Patient	**P1**	**P2**	**P6**
Age / gender	21 y / M	53 y / F	9 y / F
Origin	Austria	Austria	Germany
FKBP14 mutations	c.362dupC p.(Glu122Argfs*7) homozygous	c.362dupC p.(Glu122Argfs*7) homozygous	c.362dupC p.(Glu122Argfs*7) c.42_60del p.(Thr15*)
SKIN			
Hyperextensible	+	(+)	+
Soft texture	+	+	+
Follicular hyperkeratosis	+	–	+
Easy bruising	–	+	–
Hypertrophic scars	(+)	–	–
Atrophic scars	(+)	–	–
Other skin anomalies	–	–	–
JOINTS AND SKELETON			
Hypermobile large joints	+	+	+
Hypermobile small joints	+	+	+
Beighton score	6/9	6/9	9/9
Recurrent dislocations	–	–	–
Joint contractures	–	–	–
Progressive kyphoscoliosis	+ (17 y op)	+ (11 y op)	Scoliosis
Foot deformities	Pes planus	Pes planus	Congenital unilateral talipes, pes planus
Other skeletal anomalies	–	–	–
Fractures	–	–	–
NEUROMUSCULAR			
Muscle hypotonia at birth	+	+	+
Poor head control in infancy	+	+	+
Weakness improving	+	+	+
Delayed motor development	+	+	+
Walking independently	2.5 y	2.5 y	3.5 y
Muscular atrophy	+	+	(+)
CARDIOVASCULAR			
Cardiac valve abnormalities	–	Mild mitral and pulmonary valve insufficiency at age 50 y	–
Septum defects	–	–	–
Vascular abnormalities	Aortic root and ascending aorta diameters above the upper limit of normal at age 21 y	Internal carotid artery dissection at age 50 y, dilatation of the ascending aorta, small pseudoaneurysm of the right vertebral artery	–
EYES AND EARS			
Bluish sclerae	(+)	–	–
Refraction anomaly	Myopia	Myopia	–
Other eye anomalies	–	–	–
Hearing impairment	Sensorineural	Sensorineural	Sensorineural
MISCELLANEOUS			
Pregnancy and birth		Reduced fetal movements	Polyhydramnios
Cleft palate, bifid uvula	–	–	+
Micrognathia, retrognathia	–	–	+
Herniae	Inguinal	Umbilical	–
Genitourinary system anomalies	Bladder diverticulae	–	–
Speech or language delay	–	–	–
Learning difficulties or Intellectual disability	–	–	–
Brain MRI	Subdural hygroma	Increased tortuosity of the extracranial vessels	Atlantoaxial subluxation with dens dislocation and myelocompression at age 4 y

The following symbols and abbreviations are used: +, present; (+), mildly present; –, absent; F, female; M, male; y, years.

Figure S1. Normal appearance of the hands of patients with *FKBP14*-kEDS.

A) Patient P7/FV at age 2 years.

B) Patient P1/F1 at age 8 years.

C) Patient P15/FXIII at age 36 years with slender fingers, but no signs of arachnodactyly.

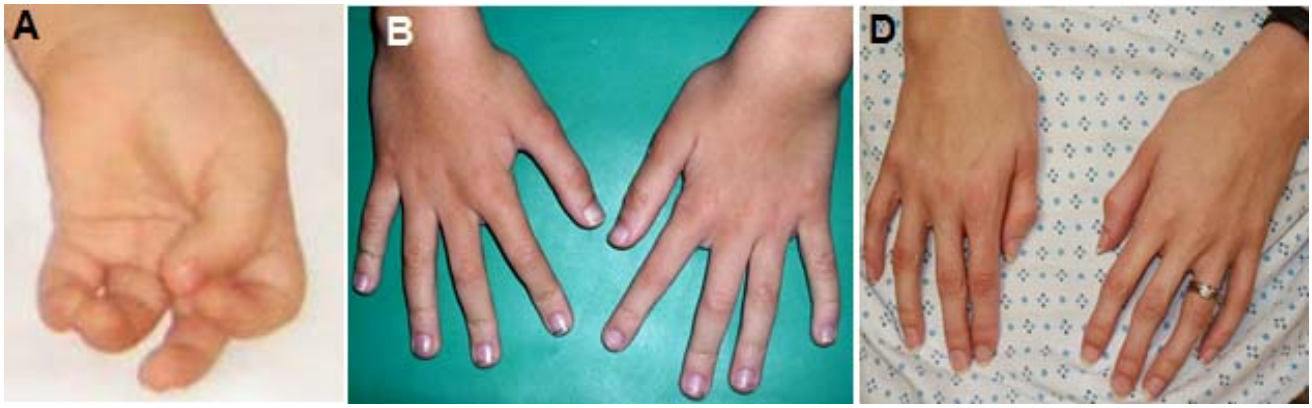

Figure S2. Hypertrophic and atrophic scars in a patient with *FKBP14*-kEDS.

Patient P10/FVIII at the age of 11 years showing hypertrophic and atrophic scars after multiple surgeries for the correction of a severe scoliosis.

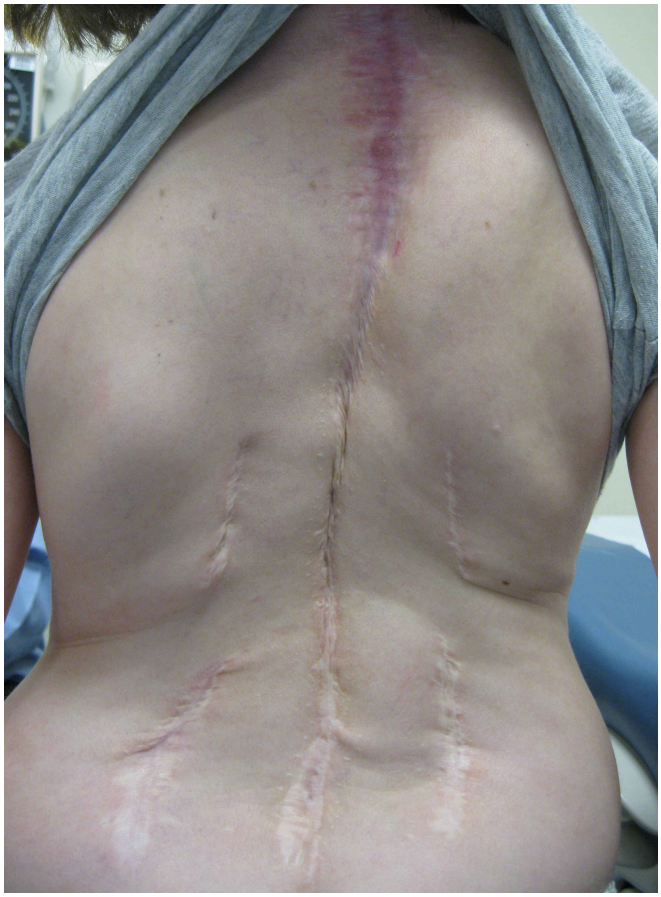

Figure S3. X-rays of the spine showing thoracic kyphoscoliosis in patients with *FKBP14*-kEDS.

A) Patient P7/FV at age 1 year. Thoracic kyphoscoliosis worsened overtime despite intensive orthopedic management.

B) Patient P9/FVII at age 3 years. Kyphoscoliosis was first noticed at the age of 7 months, and subsequently progressed.

C) Patient P8/FVI at age 4 years. First signs of scoliosis developed at the age of 3 months. Scoliosis progressed despite management with a thoracolumbar orthosis, and at the age of 6 years spinal surgery was performed.

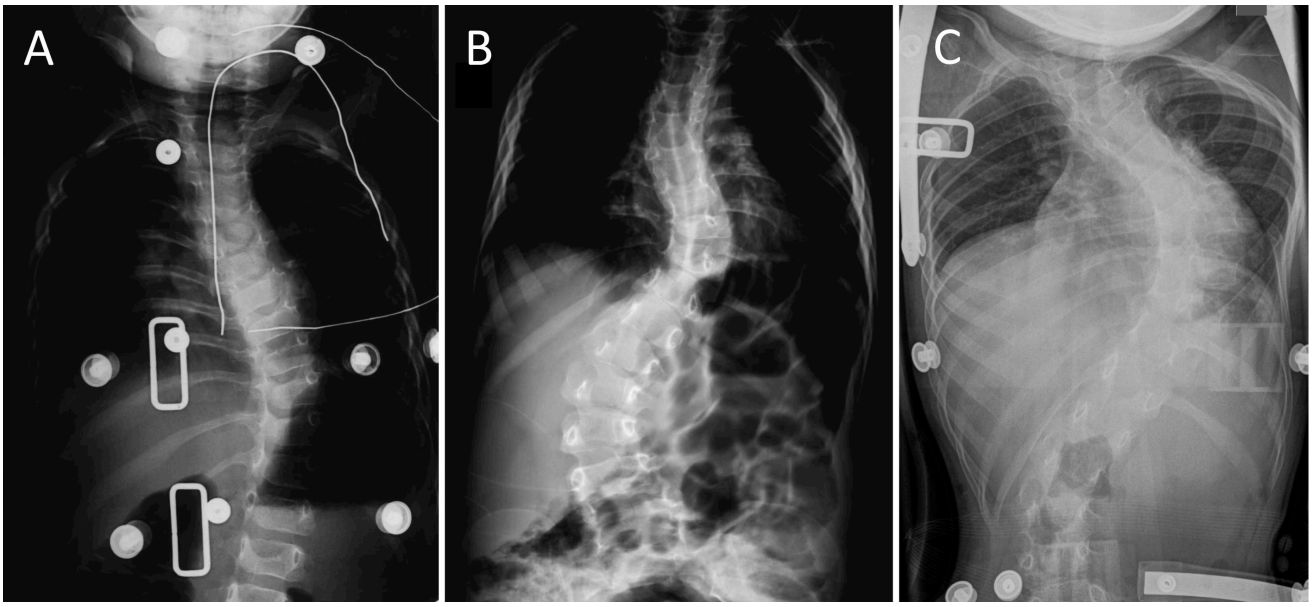

Figure S4. Structure of FKBP14 and cartoon topology diagram of its crystal structure.

A) Structure of FKBP14 with one PPIase FKBP-type domain (blue) and two EF-hand motifs (orange). The ER retention signal at the C-terminus is indicated (green). The disease causing variants identified to date are reported on the protein structure. In purple is the hitherto only missense variant p.Met48Lys.

B) Cartoon diagram showing one monomer colored blue to red from N-term to C-term and the other monomer in grey; in the grey monomer amino acid Met48 is shown in pink; putative PPIase active site amino acids (Y52, I75, W88 and F127) are shown in yellow.