Genetic screening in adolescents with steroid-resistant nephrotic syndrome

Beata S. Lipska[1,2], Paraskevas Iatropoulos[3], Ramona Maranta[3], Gianluca Caridi[4], Fatih Ozaltin[5,6], Ali Anarat[7], Ayse Balat[8], Jutta Gellermann[9], Agnes Trautmann[1], Ozlem Erdogan[10], Bassam Saeed[11], Sevinc Emre[12], Radovan Bogdanovic[13], Marta Azocar[14], Irena Balasz-Chmielewska[15], Elisa Benetti[16], Salim Caliskan[17], Sevgi Mir[18], Anette Melk[19], Pelin Ertan[20], Esra Baskin[21], Helena Jardim[22], Tinatin Davitaia[23], Anna Wasilewska[24], Dorota Drozdz[25], Maria Szczepanska[26], Augustina Jankauskiene[27], Lina Maria Serna Higuita[28], Gianluigi Ardissino[29], Ozan Ozkaya[30], Elzbieta Kuzma-Mroczkowska[31], Oguz Soylemezoglu[32], Bruno Ranchin[33], Anna Medynska[34], Marcin Tkaczyk[35], Amira Peco-Antic[36], Ipek Akil[20], Tomasz Jarmolinski[37], Agnieszka Firszt-Adamczyk[38], Jiri Dusek[39], Giacomo D. Simonetti[40], Faysal Gok[41], Alaleh Gheissari[42], Francesco Emma[43], Rafael T. Krmar[44], Michel Fischbach[45], Nikoleta Printza[46], Eva Simkova[47], Caterina Mele[3], Gian Marco Ghiggeri[4], Franz Schaefer[1] and the PodoNet Consortium

[1]Division of Pediatric Nephrology, Center for Pediatric and Adolescent Medicine, University of Heidelberg, Heidelberg, Germany; [2]Department of Biology and Genetics, Medical University of Gdansk, Gdansk, Poland; [3]Clinical Research Centre for Rare Diseases 'ALDO E CELE DACCO', Mario Negri Institute for Pharmacological Research, Ranica (Bergamo), Italy; [4]Laboratorio di Fisiopatologia dell'Uremia e UOC di Nefrologia Dialisi e Trapianto, Istituto G. Gaslini, Genova, Italy; [5]Departments of Pediatric Nephrology and Rheumatology, Hacettepe University Faculty of Medicine, Ankara, Turkey; [6]Nephrogenetics Laboratory, Hacettepe University Faculty of Medicine, Sihhiye, Ankara, Turkey; [7]Pediatric Nephrology Department, Cukurova University Medical Faculty, Adana, Turkey; [8]Department of Pediatric Nephrology, Gaziantep University Medical Faculty, Gaziantep, Turkey; [9]Klinik für Pädiatrie/Nephrologie, Charité Campus Virchow-Klinikum, Berlin, Germany; [10]Sami Ulus Childreńs Hospital, Ankara, Turkey; [11]Department of Pediatric Nephrology, Kidney Hospital of Damascus, Damascus, Syria; [12]Department of Pediatric Nephrology, Istanbul Medical Faculty, University of Istanbul, Istanbul, Turkey; [13]Department of Nephrology, Institute of Mother and Child Healthcare of Serbia, Belgrade, Serbia; [14]Unidad de Nefrología Infantil, Hospital Luis Calvo Mackenna-Facultad de Chile, Santiago de Chile, Chile; [15]Department of Pediatric and Adolescent Nephrology, Medical University of Gdansk, Gdansk, Poland; [16]Department of Pediatrics, Pediatric Nephrology/Dialysis and Transplant Unit, Agenda Ospedaliera University of Padova, Padova, Italy; [17]Department of Pediatric Nephrology, Cerrahpasa Medical Faculty Istanbul University, Istanbul, Turkey; [18]Department of Pediatric Nephrology, Ege University Medical Faculty, Izmir, Turkey; [19]Pediatric Kidney, Liver and Metabolic Disease, MHH Children's Hospital, Hannover, Germany; [20]Department of Pediatric Nephrology, Celal Bayar University, Manisa, Turkey; [21]Department of Pediatric Nephrology, Baskent University Hospital, Ankara, Turkey; [22]Centro Hospitalar do Porto, Porto, Portugal; [23]M. Iashvili Children Central Hospital, Tbilisi, Georgia; [24]Department of Pediatric Nephrology, University of Bialystok, Bialystok, Poland; [25]Department of Pediatric Nephrology, Dialysis Unit, Jagiellonian University Medical College, Krakow, Poland; [26]Dialysis Division for Children, Department and Clinics of Pediatrics, Medical University of Silesia in Katowice, Zabrze, Poland; [27]Vilnius University Children's Hospital, Vilnius, Lithuania; [28]Hospital Pablo Tobon Uribe, Medellin, Antioquia, Colombia; [29]IRCCS Fondazione Ospedale Maggiore Policlinico, UOC Nefrologia e Dialisi Pediatrica, Milano, Italy; [30]Department of Pediatric Nephrology, Faculty of Medicine, Ondokuz Mayis University, Samsun, Turkey; [31]Department of Pediatric Nephrology, University of Warsaw, Warsaw, Poland; [32]Pediatric Nephrology Department, Gazi University Hospital, Ankara, Turkey; [33]Service de Pédiatrie & Inserm U820, Centre de Référence des Maladies Rénales Rares, Hôpital Femme Mère Enfant and Université de Lyon, Lyon, France; [34]Department of Pediatric Nephrology, Medical University Wroclaw, Wroclaw, Poland; [35]Nephrology Division, Department of Pediatrics and Immunology, Polish Mothers Memorial Hospital Research Institute, Lodz, Poland; [36]Division of Pediatric Nephrology, University Children's Hospital, Belgrade, Serbia; [37]Department of Pediatrics, Regional Hospital, Miedzyrzecz, Poland; [38]Wojewodzki Szpital Dzieciecy Oddzial Pediatrii, Nefrologii ze Stacja Dializ, Torun, Poland; [39]1st Department of Pediatrics, University Hospital Motol, Prague, Czech Republic; [40]Department of Pediatric Nephrology, Inselspital Bern, University Children's Hospital, Bern, Switzerland; [41]Gulhane Military Medical Academy, Ankara, Turkey; [42]Pediatric Nephrology Department, Isfahan University of Medical Science, St Al Zahra Hospital, Isfahan, Iran; [43]Nephrology and Dialysis Unit, Ospedale Pediatrico Bambino, Rome, Italy; [44]Division of Pediatrics, Karolinska University Hospital, Stockholm, Sweden; [45]Unité de Dialyse pour Enfants Service de Pédiatrie 1, Hôpital de Hautepierre, Hopitaux Universitaires de Strasbourg, Strasbourg, France; [46]1st Department of Pediatrics, Aristotle University, Hippokration General Hospital, Thessaloniki, Greece and [47]Paediatric Department, Dubai Hospital, Dubai, United Arab Emirates

Correspondence: *Beata S. Lipska, Department of Biology and Genetics, Medical University of Gdansk, Debinki 1, Gdansk, 80211, Poland. E-mail: b.lipska@gumed.edu.pl*

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Genetic screening paradigms for congenital and infantile nephrotic syndrome are well established; however, screening in adolescents has received only minor attention. To help rectify this, we analyzed an unselected adolescent cohort of the international PodoNet registry to develop a rational screening approach based on 227 patients with nonsyndromic steroid-resistant nephrotic syndrome aged 10–20 years. Of these, 21% had a positive family history. Autosomal dominant cases were screened for *WT1*, *TRPC6*, *ACTN4*, and *INF2* mutations. All other patients had the *NPHS2* gene screened, and *WT1* was tested in sporadic cases. In addition, 40 sporadic cases had the entire coding region of *INF2* tested. Of the autosomal recessive and the sporadic cases, 13 and 6%, respectively, were found to have podocin-associated nephrotic syndrome, and 56% of them were compound heterozygous for the nonneutral p.R229Q polymorphism. Four percent of the sporadic and 10% of the autosomal dominant cases had a mutation in *WT1*. Pathogenic *INF2* mutations were found in 20% of the dominant but none of the sporadic cases. In a large cohort of adolescents including both familial and sporadic disease, *NPHS2* mutations explained about 7% and *WT1* 4% of cases, whereas *INF2* proved relevant only in autosomal dominant familial disease. Thus, screening of the entire coding sequence of *NPHS2* and exons 8–9 of *WT1* appears to be the most rational and cost-effective screening approach in sporadic juvenile steroid-resistant nephrotic syndrome.

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Steroid-resistant nephrotic syndrome (SRNS) is a heterogeneous disorder caused either by dysregulation of the immune system or by genetic abnormalities affecting podocyte-specific proteins. Knowledge of the underlying pathology has major impact on the treatment and prognosis of the disorder. The genetic heterogeneity and phenotypic variability of SRNS mandates a rational, adapted approach to genetic screening.

The age of disease onset is an important predictor of the odds of finding an abnormality in a particular gene linked to SRNS. In recent years, several proposals for genetic screening paradigms have been put forward, which preferentially addressed congenital and infantile onset cases.[1–4] SRNS manifesting at adolescent age was addressed as a subject of minor interest in two recent reports only.[3,4] The current literature suggests that at least five genes should be taken into consideration in adolescent-onset SRNS: *NPHS2* in autosomal recessive (AR) and sporadic cases,[5,6] *WT1*[7] in autosomal dominant (AD) and sporadic cases, and *TRCP6*,[8] *ACTN4*,[9] and the recently identified *INF2*[10] in AD cases. In contrast, the occurrence of mutations in the genes *NPHS1*, *PLCE1*, *MYOE1*, and *PTPRO* in this age group is rather anecdotal.[11] Furthermore, *APOL1* variants are considered risk factors for focal segmental glomerulosclerosis (FSGS) in young adults, at least in the African-American population.[12]

Although most previous studies in SRNS focused on familial cases, the vast majority of adolescent SRNS cases are in fact sporadic. The PodoNet registry study collects clinical and genetic information as well as biomaterials from patients with SRNS across the pediatric age range. With almost 1500 cases from 66 pediatric nephrology centers in 21 countries compiled to date, PodoNet is the largest registry worldwide devoted to this rare disorder (Supplementary Material S1 online). Here, we utilized the PodoNet registry to perform comprehensive screening for genetic causes in an unselected population of 297 SRNS patients with disease onset in the second decade of life, including both sporadic and familial cases.

RESULTS

AR cases accounted for ~25% of Polish, Turkish, and Syrian patients enrolled in the study, whereas family history was positive in no more than 10% of patients from Western Europe and Latin America. A total of 38 (17%) patients (including members of 12 AR families) descended from consanguineous marriages; all of these were from Turkey or the Middle East.

The distribution of age at onset and degree of proteinuria was similar in the familial and sporadic forms. On biopsy, patients with familial SRNS showed less frequently minimal change histology (3% vs. 16%, $P=0.05$) and more commonly mesangioproliferative glomerulonephritis (24% vs. 9%, $P<0.05$) than patients with sporadic disease, whereas the proportion of cases with FSGS was similar (63 and 73%, not significant). The fraction of sclerosed glomeruli did not differ significantly in patients with familial (median 36, interquartile range 15–50%) and sporadic FSGS (median 18, interquartile range 10–50%).

Autosomal dominant SRNS

None of the AD patients was found to have a mutation in *TRCP6* or *ACTN4*. One patient was found to have an intronic mutation in *WT1* and two patients were positive for an *INF2* mutation located in its *hot spot* region (exon 4). Details regarding clinical presentation are given in Tables 1 and 2.

Sporadic and autosomal recessive SRNS

NPHS2 screening. In all, 5/38 (13%) AR patients and 11/179 (6%) sporadic cases were found to have podocin-associated SRNS. *NPHS2*-positive patients did not differ from the other adolescents with respect to age at first manifestation, time to end-stage renal disease, histopathology, and degree of proteinuria. FSGS was present in 10 cases, mesangioproliferative glomerulonephritis GN in 2 cases, and minimal change nephropathy and global glomerulosclerosis in 1 case each.

Homozygous mutations in *NPHS2* were found in 5 (all sporadic) patients and compound heterozygous mutations in 11 patients (Table 3). The most common mutations were