Genotype-phenotype correlation in Bulgarian patients with c.293-13A/C>G splice mutation of *21CYPA2* picked up by Neonatal Screening (NS)

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Background

80-95% of Congenital Adrenal Hyperplasia (CAH) cases are due to mutations in *CYP21A2* gene encoding adrenal steroidogenic enzyme 21-hydroxylase [(1,2)]. The residual activity of 21-hydroxylase defines the clinical form of CAH patients. c.293-13A/C>G (I2G) splice mutation is associated with ≤1% enzyme activity, *in vitro,* and could be manifested either by salt wasting (SW) or simple virilizing (SV) forms [(3,4)] (Fig. 1).

Objective

To study genotype-phenotype correlations in Bulgarian patients with homozygous I2G *CYP21A2* mutation.

Methods

Newborns and siblings with elevated 17-OHP picked up by NS; 17-OHP (Delfia®), clinical evaluation, electrolytes; MLPA (multiplex ligation-dependent probe amplification), direct sequencing of *CYP21A2*.

Results

222 827 newborns were screened (2010 to April 2014-coverage 82.5%); 25 patients with *CYP21A2* mutations were characterized (Fig. 2).

70% of the I2G homozygous patients are males and from Roma ethnical group (Fig. 3, 4), found mainly in the Northeast part of the country (Fig. 5).

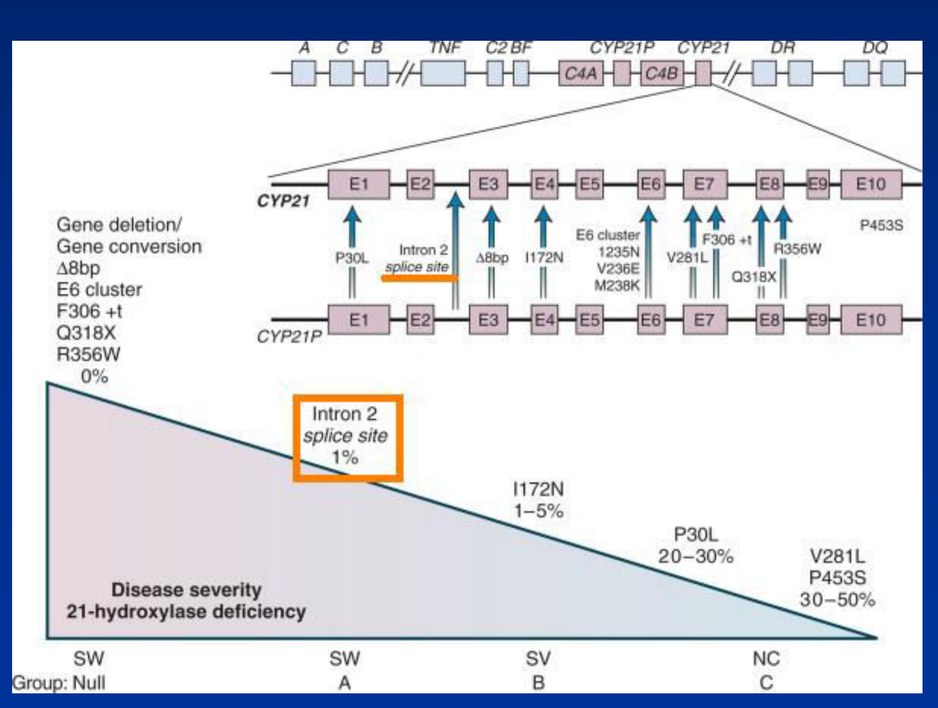

Fig.1 Common CYP21A2 mutations and phenotype groups. Kronenberg et al. (2008) Williams Textbook of Endocrinology, el ed. Sounders

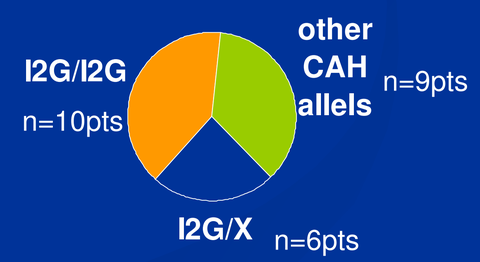

Fig. 2 I2G alleles in CAH patients. 59% I2G allele frequency

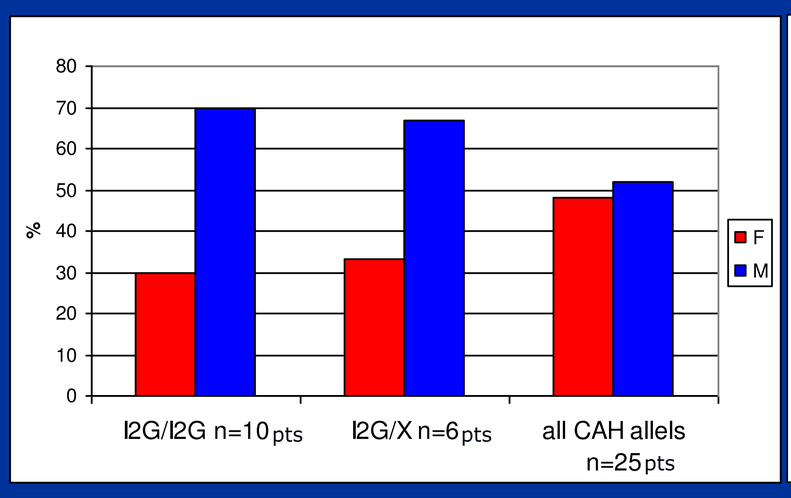

Fig. 3 Sex distribution of I2G allele in Bulgaria

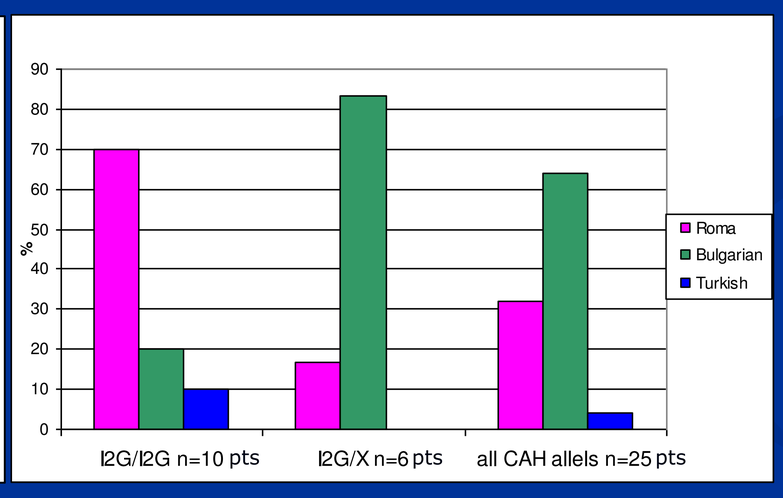

Fig. 4 I2G allele in Bulgarian ethnical groups

Fig. 5 Geographical I2G allele distribution in Bulgaria

												17-OHP (nmol/l)				
N	Pthenotypic group	Genotype	Ethnic group	civil gender	genetic gender	GAH clinical form	phenotype determines by the genotype	virilisation	weight (g)	G.A (Weeks)	age (days)	1FPC first drop	1FPC second drop	norm ISNS	2FPC	day of treatment
P1	A	I2G/I2G	T	F	F	SW	SW	III	2400	35	2	703	570	44		9
P2	A	I2G/I2G	R	M	M	SW	SW		3000	39	3	348	447,2	23	392	15
P3	A	I2G/I2G	R	M	M	?	SW		-	-	-	-	-	-		
P4	A	I2G/I2G	R	M	M	SW	SW		3620	40	3	227,5	171,6	23	344,5	13
P5	A	I2G/I2G	R	M	F	SW	SW	III-IV	3000	40	3	560,75		23	392	41
P6	A	I2G/I2G	B	M	M	SW	SW		2900	38	5	564,8	490,4	20	197,2	14
P7	A	I2G/I2G	R	M	M	SW	SW		3300	39	3	96	94,5	23	116	7
P8	A	I2G/I2G	R	M	M	SW	SW		1300	32	3	534		70		
P9	A	I2G/I2G	R	M	M	SW	SW		3100	38	5	480	362,5	20	890	12
P10	A	I2G/I2G	B	F	F	SW	SW	III	4300	39	4	69,9	73,4	23	91,2	12

P2 Diagnosed by NS, Hospital admission at 13d, vomiting, SW form (Na-138; K-7,3, Cl-99), therapy from 15d

P3 Difficult to classify form: reported vomiting in early childhood, hospital admissions, pseudoprecocious puberty at 6 yrs. No regular corticosteroid treatment. At 13y-short stature and masculine constitution, no salt crises.

Table 1. 9 patients with clear SW form; Virilisation (Prader 2-4) in all of the diagnosed girls. The 17-OHP screening levels were elevated: average 398.2 nmol/l (SD± 224), but varied widely (median 480, range 69.9-703);

Conclusions

Homozygous I2G patients showed variable phenotype, even within a family; the I2G splice mutation is the most common in Bulgaria; Our mutational screening strategy is currently adapted to the results.

References:

1. New MI. (1987) Basic and clinical aspects of congenital adrenal hyperplasia. J Steroid Biochem. 27:1-7 **2.** Wajnrajch MP, New MI (2010) Defects of adrenal steroidogenesis. Endocrinology, eds Jameson JL, De Groot LJ (Elsevier, Philadelphia), 6th Ed, Vol 2, pp 1897–1920. **3.** Delague V, Souraty N, Khallouf E, Tardy V, Chouery E, Halaby G, Loiselet J, Morel Y, Mégarbané A. (2000) Mutational analysis in Lebanese patients with congenital adrenal hyperplasia due to a deficit in 21-hydroxylase. Horm Res. 53(2):77-82. **4.** New MI, Abraham M, Gonzalez B, Dumic M, Razzaghy-Azar M, Chitayat D, Sun L, Zaidi M, Wilson RC, Yuen T. (2013) Genotype-phenotype correlation in 1,507 families with congenital adrenal hyperplasia owing to 21-hydroxylase deficiency. Proc Natl Acad Sci U S A. Feb 12;110(7):2611-6.

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