Impact of *TP53* (rs1042522) and *MDM2* (rs2279744) polymorphisms on cervical intraepithelial lesions and cervical cancer in North Macedonian women

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INTRADUCTION

- Persistent human papillomavirus (HPV) infection is a major factor in the onset of cervical intraepithelial lesion (CIN), but additional factors are needed for their further progression to carcinoma (CCa).
- Genetic variant in host cell cycle regulation genes such are single nucleotide polymorphisms (SNPs) rs1042522 within the codon 72 of *TP53* and rs2279744 within *MDM2* promoter genes are plausible factors that could influence cervical carcinogenesis conferring increased attenuation of p53 pathway.
- The p53 tumor suppressor is gate keeper of cell cycle that regulates cellular pathways such as DNA repair, apoptosis, angiogenesis and is important defense mechanism against cancer onset and progression. The C to G base substitution in codon 72 replacing amino acid Prolin with Arginine is considered to produce a more vulnerable variant of p53

MDM2 309 G>T (rs2279744)

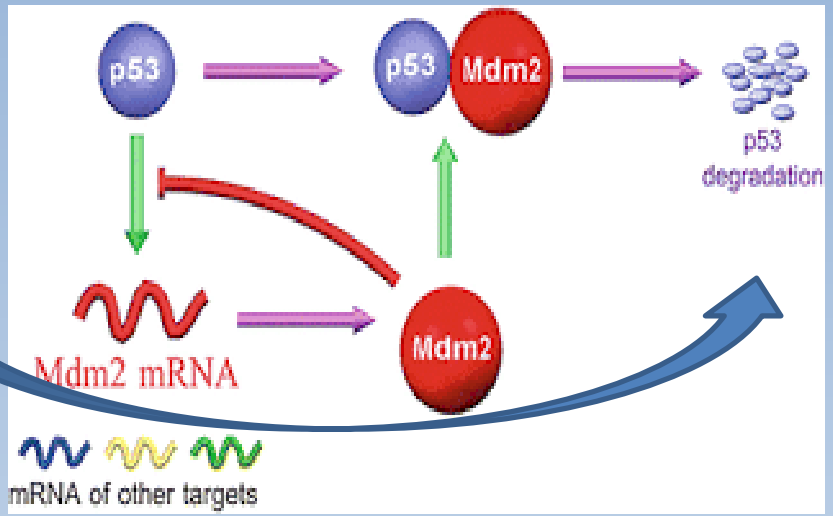
High risk HPV E6 oncoprotein

Higher expression

MDM2

E6/p53 complex

Inhibition of p53 induces apoptosis, HPV persistence, accumulation of mutations



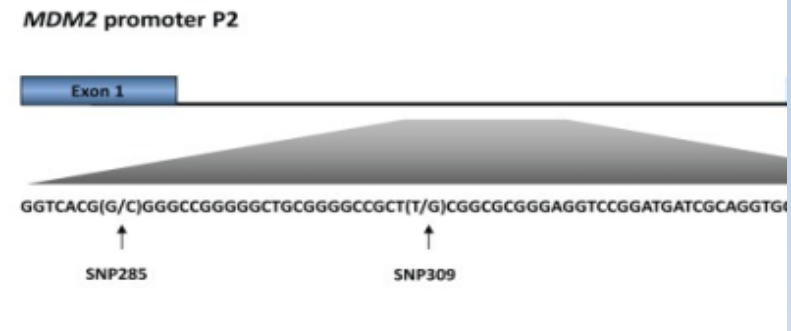
Adapted from Levav-Cohen Y. et al. (2014)

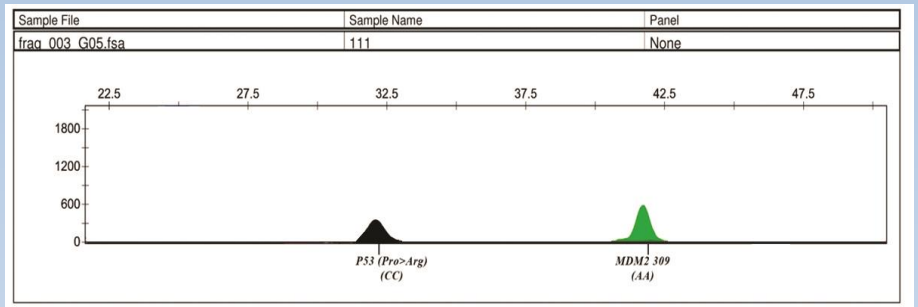
- high risk HPV E6 oncoprotein binds to p53 and this interaction with rs1042522 variant is even stroger that additionally abrogates p53 function leading to its degradation through ubiqutin dependent pathway

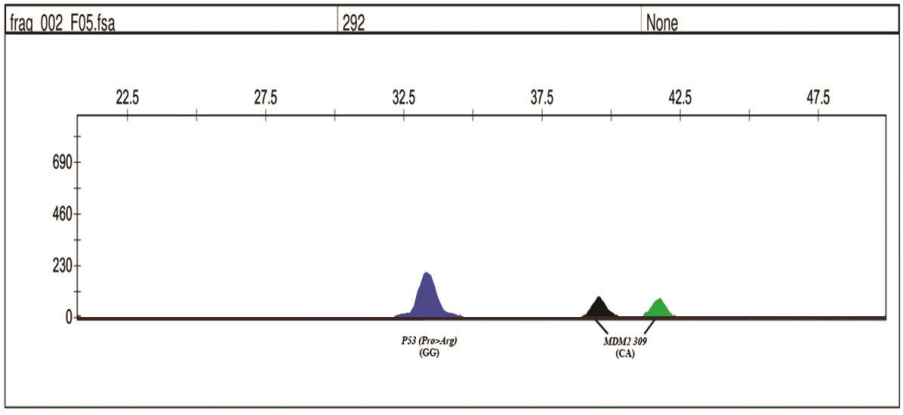
- MDM2 oncoprotein is negative regulator of p53 tumor suppression. The variant rs2279744 in its promoter's region could influence cervical carcinogenesis increasing its affinity of the Sp1 transcription factor

MATERIALS AND METHODS

- Objective: We investigated the association of these SNPs with the CIN and CCa among women from the Republic of North Macedonia.
- Using a multiplex PCR SNaPShot analysis we genotyped rs1042522 and rs2279744 in 131 women with CIN or CCa and 110 cytologicaly and Human papillomavirus negative women.
- The allele and genotype frequencies of the variants were analysed using x^2 – test in SISA statistic software.







• For association analysys the cases were stratified in subgroups

RESULTS

*p53*Pro72Arg	Cases (CIN + CCa) n (%)	CIN2+ n (%)	CIN1 n (%)	controls n (%)	p1*	p 2*	p 3*
Allele C	78(30.2)	45 (24.4)	33(44.6)	63(31.2)	0.8	0.15	**0.001**
G	180 (68.8)	140 (75.6)	41(55.4)	139 (68.8)			
Genotype GG	66 (51.2)	53 (57.6)	13 (35.1)	51 (50.5)	0.7	0.11	**0.001**
GC	48 (37.2)	33 (35.9)	15 (40.6)	37 (36.6)	0.8	0.2	**0.04**
CC	15(11.6)	6 (6.5)	9 (24.3)	13 (12.9)	Ref	Ref	
GG/ GC+ d.CC					0.1	0.36	0.02
r. GC+GG/CC					0.7		**0.004**
n-total	129(100)	92 (100)	37 (100)	101 (100)	-	-	-

For association analysis the cases were stratified in subgroups

Value of chi square test: P1*- cases vs controls; P2*-CIN2 /controls; P3* CIN2+/CIN1

RESULTS

MDM2309 T>G	Cases (CIN + CCa) n (%)	CIN2+ n (%)	CIN1 n (%)	controls n (%)	p1*	p 2*	p 3*
Allele T	183 (62.0)	97 (52.7)	53 (68.0)	124 (56.4)	0.21	0.4	**0.02**
G	112 (48.0)	87 (47.3)	25 (32.0)	96 (43.6)			
Genotype TT	39 (29.8)	21 (22.8)	18 (46.2)	30 (27.3)	Ref	Ref	Ref
GT	72 (55.0)	55 (59.8)	17 (43.6)	64 (58.2)	0.6	0.54	0.14
GG	20 (15.2)	16 (17.4)	4 (10.2)	16 (14.5)	1	0.43	**0.04**
Dominant GT+GG/TT					0.66	0.40	**0.007**
Recessive GG/ TG + TT	-				0.87	0.5	0.29
n-total	131(100)	92 (100)	39 (100)	110 (100)	-	-	-

Value of chi square test: P1*- cases vs controls; P2*-CIN2 /controls; P3* CIN2+/CIN1

RESULTS

- The *TP53* rs1042522 and *MDM2* rs2279744 polymorphic variants showed no association with initiation and development of CIN and CCa. No signicant difference in either genotype or allelic frequencies for rs1042522 and rs2279744 between cases and control was found.
- Stratication of cases group based on grade of the lesion, revelled lower frequency of CC genotype and C allele of **rs1042522** in CIN2+ and CCa compared to CIN1 [GG vs CC; p=0.001, OR=0.4; CG vs CC; p=0.04, OR=0.03 and CG+ GG vs CC; p=0.004, OR=0.2]. Furthermore, GG genotype and G allele of rs2279744 showed signicantly lower frequency in CIN2+ and CCa cases then in CIN1 [G vs T p=0.02, OR=0.52; GG vs TT; p=0.04, OR=0.29; TT vs TG+GG; p=0.007, OR=0.34].

CONCLUSION

- The Arg variant of rs1042522 and T allele/TT genotype of rs2279744 are associated with progression of CIN1 to CIN2+ or CCa and may be used as prediction markers in CCa management. Still, clinical importance of these variants warrants further validation in large and more comprehensive studies.