THE ROLE OF APOPTOSIS POLYMORPHISMS IN INDIVIDUAL SUSCEPTIBILITY TO PHILADELPHIA-NEGATIVE MYELOPROLIFERATIVE NEOPLASMS

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1. Background

Although somatic mutations in the Janus kinase 2 gene (*JAK2*) occur in many Philadelphia-chromosome negative chronic myeloproliferative neoplasms (PN-MPNs), disease evolution, distinct phenotypes and the continuous clinical evidence of an increasing number of cases, with younger patients affected, have been pointing to a growing involvement of environmental factors in the pathogenesis of these diseases. Although this association is well established in some solid tumors, like breast and thyroid, this aspect is now being considered for some solid tumors, like breast and thyroid, this aspect is now being considered for hematological malignancies.

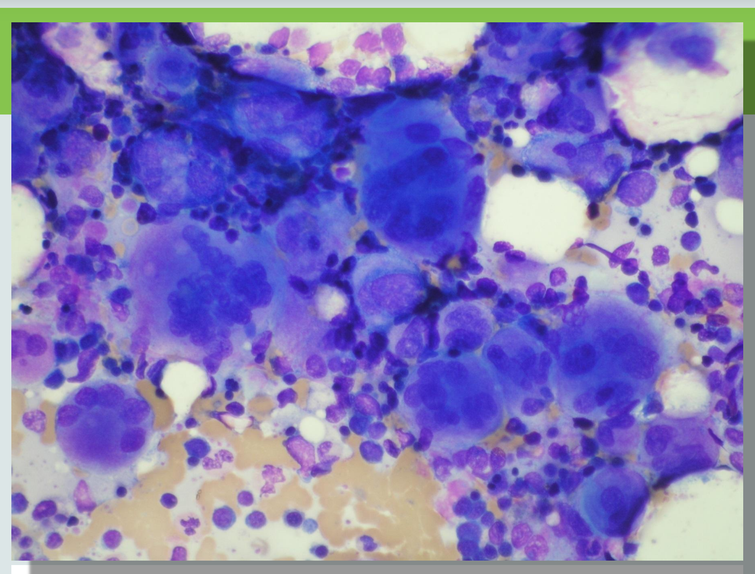

Fig. 1 – Megakaryocytes in primary myelofibrosis, bone marrow aspirate smear (x100).

Exposure to hazardous agents in the environment on a continual basis, can lead to changes at the genome level, alterations in cell cycle regulation and consequently to cancer. Several single nucleotide polymorphisms (SNPs) have been identified, that may influence the DNA repair capacity and apoptotic status, that, in turn, confer genetic predisposition to disease and determine therapeutic response.

Expression deregulation of pro and anti-apoptotic genes promotes cell resistance to apoptosis, culminating with the accumulation of myeloid cells and establishing neoplasms (Fig.1).

2. Objectives

We intend to evaluate the role of apoptosis SNPs in PN-MPNs susceptibility.

3. Methods

We performed a case-control study in 121 Caucasian Portuguese PN-MPNs patients (73 with Essential thrombocythaemia (ET), 35 with Polycythaemia vera (PV) and 13 with Primary Myelofibrosis (PMF)) and 280 matched controls. Most of the patients were diagnosed and are followed by some of the elements of this working group. rs2227309 and rs2227310 (*CASP7*), rs1045485 and rs1035142 (*CASP8*), rs2308950, rs1820204 and rs1052571 (*CASP9*) and rs13006529 (*CASP10*) were genotyped using real-time PCR (RT-PCR 7300 Applied Biosystem), through TaqMan® SNP genotyping assays (Life Technology), according to manufacturer instructions. Differences in genotype frequency, smoking status, age class, gender, therapeutic and pathology distributions between patients and controls were evaluated using SPSS 22.0 (SPSS Inc.).

4. Results

None of the apoptosis polymorphisms studied, individually considered, is associated with PN-MPNs risk.

No significant difference was found between the case and control groups concerning age distribution, gender, smoking habits or genotype frequencies (Table 1). No significant change in crude or adjusted OR was observed for any of the genotypes considered (Table 2).

Studies related with therapeutic response are still ongoing.

5. Conclusions

Our results do not reveal a significant involvement of apoptosis polymorphisms on the individually susceptibility towards PN-MPNs.

However, larger studies are required to confirm these results and to provide conclusive evidence of non association between these and other apoptosis variants and PN-MPNs and therapeutic response.

On the other hand, there are studies that show modifications in the expression of molecules that participate in the regulation of apoptosis, indicating that this mechanism is involved in the pathophysiology of these diseases.

Identification of the main molecules that are altered in MPNs allows the development of drugs more directly targeted to the pathophysiology of the disease, with high efficacy, fewer adverse effects, contributing to compliance of the patients with treatments.

6. References

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Table 1 – General characteristics for the PN-MPNs cases (*n*=121) and control population (*n*=280).

Characteristics	Cases, *n* (%)	Controls, *n* (%)	*P* value
Gender			
Male	55 (45.8)	132 (47.1)	0.8
Female	65 (54.2)	148 (52.9)	
Age [a, b]			0.9
30-49	16 (13.2)	43 (15.4)	
50-69	47 (38.8)	106 (37.9)	
≥70	58 (47.9)	131 (46.8)	
Smoking habits			0.9
Never	93 (76.9)	212 (76.0)	
Current	28 (23.1)	67 (24.0)	
CASP7 (Arg249Lys)			0.9
Arg/Arg	70 (57.9)	153 (55.0)	
Lys/Arg	45 (37.2)	109 (39.2)	
Lys/Lys	6 (5.0)	16 (5.8)	
CASP7 (Asp255Glu)			1.0
Asp/Asp	7 (5.8)	16 (5,8)	
Asp/Glu	47 (39.2)	106 (38.5)	
Glu/Glu	66 (55.0)	153 (55.6)	
CASP8 (Asp302His)			0.7
Asp/Asp	91 (75.8)	219 (78.8)	
Asp/His	24 (20.0)	51 (18.3)	
His/His	5 (4.2)	8 (2.9)	
CASP8 (Tyr12STOP)			0.6
Tyr/Tyr	20 (16.5)	45 (16.2)	
Tyr/Stop	53 (43.8)	136 (48.9)	
Stop/Stop	48 (39.7)	97 (34.9)	
CASP9 (Phe136Leu)			0.2
Phe/Phe	28 (23.1)	87 (31.3)	
Phe/Leu	66 (54.5)	128 (46.0)	
Leu/Leu	27 (22.3)	63 (22.7)	
CASP9 (Arg173His			0.2
Arg/Arg	117 (96.7)	275 (98.9)	
Arg/His	3 (2.5)	3 (1.1)	
Arg/His	1 (0.8)	0 (0.0)	
CASP9 (*V*al762Ala)			0.3
Val/Val	24 (19.8)	69 (24.8)	
Val/Ala	66 (54.5)	129 (46.4)	
Ala/Ala	31 (25.6)	80 (28.8)	
CASP10 (Ile522Leu)			0.6
Ile/Ile	27 (22.3)	74 (26.6)	
Ile/Leu	58 (47.9)	122 (43.9)	
Leu/Leu	36 (29.8)	82 (29.5)	

[a] Age of diagnosis for cases
[b] Age of control population at the time of diagnosis for the matched case

Table 2 – ORs (95% CI) for apoptosis polymorphisms and PN-MPNs association.

	n	OR crude (95% CI)	OR adjusted (95% CI)[a]
CASP7 (Arg249Lys)	121		
Arg/Arg		1 (Reference)	1 (Reference)
Arg/Lys		0.9 (0.6-1.4)	0.9 (0.6-1.4)
Lys/Lys		0.8 (0.3-2.2)	0.9 (0.3-2.6)
CASP7 (Asp255Glu)	120		
Asp/Asp		1 (Reference)	1 (Reference)
Asp/Glu		1.0 (0.7-1.6)	1.0 (0.6-1.6)
Glu/Glu		1.0 (0.4-2.6)	1.1 (0.4-2.9)
CASP8 (Asp302His)	120		
Asp/Asp[b]		1 (Reference)	1 (Reference)
Asp/His		1.1 (0.7-2.0)	1.1 (0.6-1.9)
His/His		1.5 (0.5-4.7)	1.3 (0.4-4.3)
CASP8 (Tyr12STOP)	121		
Tyr/Tyr		1 (Reference)	1 (Reference)
Tyr/Stop		0.8 (0.5-1.3)	0.8 (0.5-1.3)
Stop/Stop		0.9 (0.5-1.7)	1.0 (0.5-1.9)
CASP9 (Phe136Leu)	121		
Leu/Leu		1 (Reference)	1 (Reference)
Leu/Phe		1.6 (1.0-2.7)	1.6 (1.0-2.8)
Phe/Phe		1.3 (0.7-2.5)	1.4 (0.7-2.6)
CASP9 (Arg173His	117		
Arg/Arg		1 (Reference)	1 (Reference)
Arg/His + His/His		3.1 (0,7-14.2)	2.4 (0.5-11.0)
CASP9 (*V*al762Ala)	121		
Val/Val		1 (Reference)	1 (Reference)
Val/Ala		1.3 (0.8-2.2)	1.3 (0.8-2.2)
Ala/Ala		0.9 (0.5-1.7)	0.9 (0.5-1.7)
CASP10 (Ile522Leu)	121		
Ile/Ile		1 (Reference)	1 (Reference)
Ile/Leu		1.1 (0.7-1.8)	1.1 (0.6-1.8)
Leu/Leu		0.8 (0.5-1.5)	0.8 (0.5-1.6)

[a] ORs were adjusted for age (30-49, 50-69 and ≥70), smoking status (never and current)
[b] Reference class