

Association of *pre-miR-146a* rs2910164 GG genotype with papillary thyroid cancer (PTC). A new case-control study on two adjacent genes on chromosome 5, *pre-miR-146a* and *PTTG1*.

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INTRODUCTION

Papillary thyroid carcinoma (PTC) is the most common endocrine malignancy, with a steadily increasing incidence in the last few decades worldwide. Studies revealed the predisposition to PTC by the heterozygous state of rs2910164 within the precursor of microRNA146a. Interestingly, on the same chromosome, 40Kb separate the *pre-miR-146a* from the pituitary tumour transforming gene (*PTTG1*), a proto-oncogene involved in thyroid carcinomas. In this study, we analyzed, with a case-control design, the genetic association between PTC and *pre-miR-146a* rs2910164 as well as *PTTG1* (rs1862391 A/C and rs2910201 C/T).

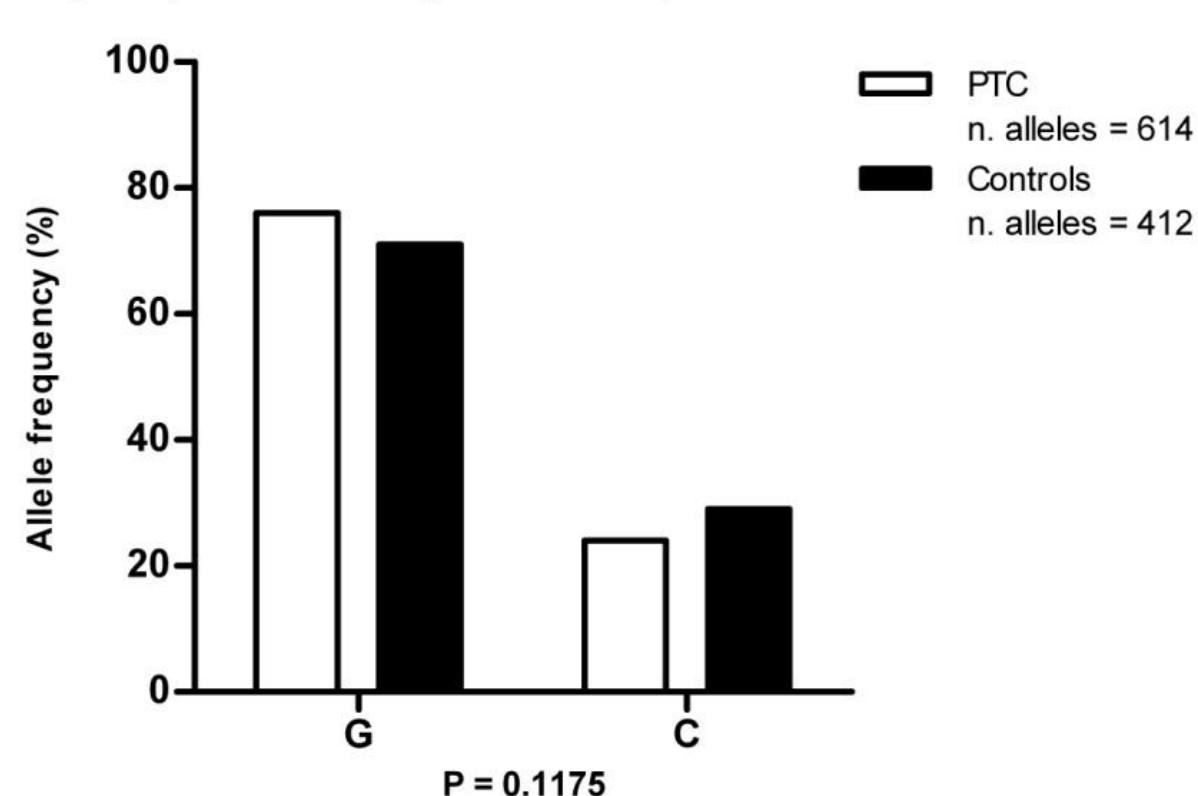
METHODS

206 healthy controls (30 to 78 of age) and 307 PTC patients (30 to 74 of age) were enrolled. The diagnosis of PTC was histological at surgery. Thyroid sonography was performed in controls to exclude nodules. SNP genotyping of *pre-miR-146a* and *PTTG1* was performed by Sanger sequencing and High Resolution Melting (HRM). Linkage Disequilibrium (LD) analysis and statistics were performed with Haplowiew 4.2, GraphPad Prism 5 and Review Manager 5.2 software.

RESULTS

Pre-miR-146a rs2910164 allelic frequencies were not statistically different in patients and controls and the SNP was not in LD with the investigated *PTTG1* SNPs. We did not confirm a previously described association of the CG genotype with PTC and our meta-analysis suggested that there was no association between rs2910164 and PTC. However, a moderate non significant association between the GG genotype and PTC (GG vs. GC+CC Odds Ratio = 1.38, 95% CI 0.8-2.4) was found. The *PTTG1* SNPs (rs1862391A/C and rs2910201C/T), in perfect LD, had the same allelic frequency in patients and controls and were not associated with PTC.

Allele frequency of rs2910164 (pre-miR-146a) between PTC and controls



Allelic frequencies of pre-miR-146a rs2910164 in our PTC cases and controls (Modena + Milan)

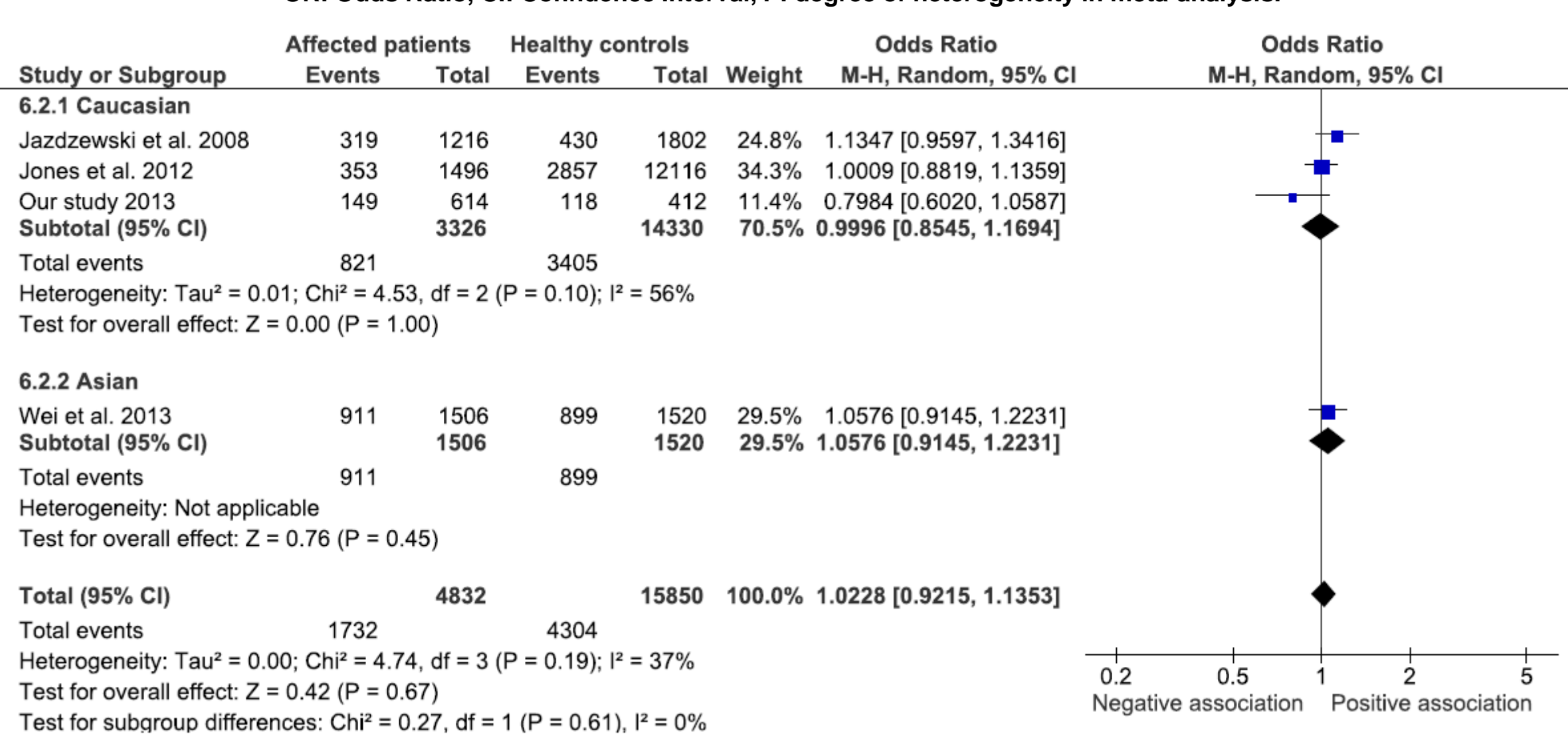
| | PTC | Controls |
|------|-------------|-------------|
| C | 149 (24,3%) | 118 (28,6%) |
| G | 465 (75,7%) | 294 (71,4%) |
| Tot. | 614 | 412 |

Distribution of pre-miR-146a rs2910164 genotypes in cases and controls, and their association with risk of PTC.

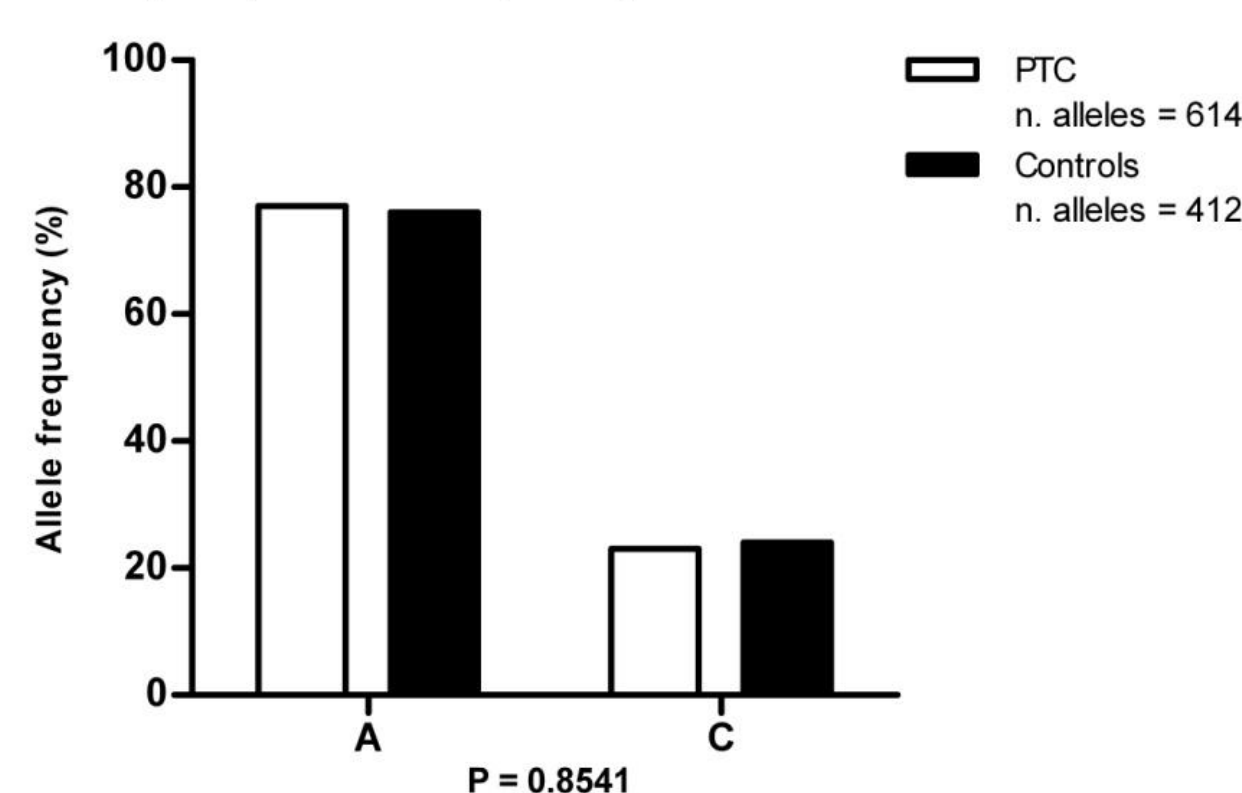
| rs2910164 | PTC | Controls | OR for genotype | CI 95% | p Value |
|-----------|------------|-----------|-----------------|-----------------|---------|
| G/G | 180 (58,6) | 105 (51) | 1,363 | 0,9556 to 1,945 | 0,0869 |
| C/G | 105 (34,2) | 84 (40,8) | 0,755 | 0,5243 to 1,087 | 0,1302 |
| C/C | 22 (7,2) | 17 (8,2) | 0,8582 | 0,4439 to 1,659 | 0,649 |
| TOT | 307 | 206 | | | |

P value = 0,2299 (Chi-square test)

Meta-analysis for the association between PTC and the *pre-miR-146a* rs2910164 polymorphism (C vs. G). OR: Odds Ratio, CI: Confidence Interval, I²: degree of heterogeneity in meta-analysis.



Allele frequency of rs1862391 (PTTG1) between PTC and controls



Allelic frequencies of PTTG1 rs1862391 in our PTC cases and controls (Modena + Milan)

| | PTC | Controls |
|------|-------------|-------------|
| A | 471 (76,7%) | 314 (76,2%) |
| C | 143 (23,3%) | 98 (23,8%) |
| Tot. | 614 | 412 |

CONCLUSIONS

The study showed a moderate association between *pre-miR-146a* rs2910164 GG and PTC but the meta-analysis, among all published studies and our data, demonstrated no association of *pre-miR-146a* rs2910164 with the pathology. *PTTG1* did not seem to be associated to PTC.