# Could a Glucocorticoid Receptor Polymorphism be Protective against Hypothalamic-Pituitary-Adrenal Axis Suppression in Asthmatic Children on Corticosteroids?

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## Background

- 16 % of asthmatic children on corticosteroids were found to have hypothalamic-pituitary-adrenal axis suppression (HPAS)
- A higher BMI was found to be protective
- Single nucleotide polymorphisms (SNPs) rs242941 & rs1876828 of the corticotrophin-releasing hormone receptor 1 (CRHR1) and rs41423247 of the glucocorticoid receptor (NR3C1) gene were found to have lower & higher basal cortisol levels respectively

## Objectives

To determine whether intronic SNPs rs242941 & rs1876828 of CHRH1 & rs41423247 of NR3C1 are associated with hypothalamic-pituitary-adrenal axis suppression (HPAS) in asthmatic school children on CS

## Subjects & Methods

**Subjects:** 96 asthmatic children of mixed ancestry. 5.2-15.6 yrs. on inhaled & nasal corticosteroids. who previously underwent morning serum cortisol (C) and metyrapone (MTP) testing. BMI Z-score (Centre of Disease Control) was computed.

**Samples:** Salivary DNA

**Diagnostic criteria for HPAS:**

- C <83nmol/l
- Post-MTP adrenocorticotropic hormone (PMTP ACTH) <106 pg/ml & 11 deoxycortisol (11DOC) <208 nmol/l & 11DOC+C < 400nmol/l

**ACTH response categories:**

Low = HPAS (36)

Middle (Mid) = 106-319 pg/ml (29)

High (Hi) = > 319 pg/ml (31)

**Genotyping:** Taqman PCR assays

**Statistical analysis:** Fisher's exact test. ANOVA. linear. binomial & multinomial logistic regression

**Functional analysis:** Linkage disequilibrium plots

## Results

**Table 1: Association of HPAS by SNP***

| | rs242941 (A/C) | rs1876828 (C/T) | rs41423247 (G/C) |
|---|---|---|---|
| **OR (95%CI)** | 0.99 (0.43-2.27) | 0.54 (0.05-3.48) | **0.27 (0.06-0.90)** |

* Fisher's exact test

**Fig. 1: Homozygote-Minor-Allele-Heterozygote Scatterplot of rs41423247, rs242941 & rs1876828**

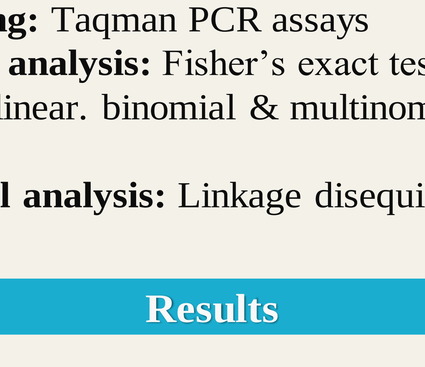



**Fig 2:Genotype Distribution rs41423247**

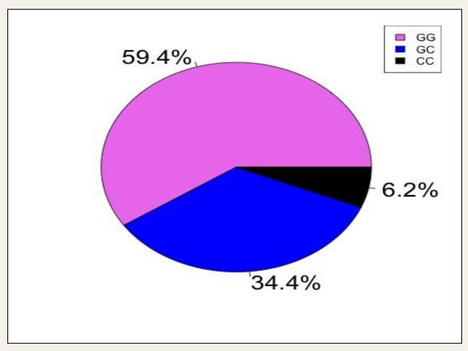
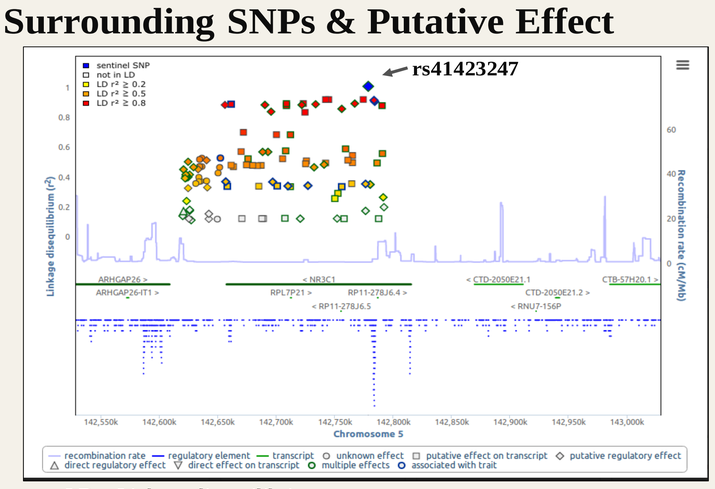



**Table 2: Minor Allele Frequency rs41423247**

| Country | C Allele Frequency (%) |
|---|---|
| Europe (Finnish) | 41 |
| Europe (Non-Finnish) | 37 |
| Africa | 21 |
| South Africa (current study) | **23** |
| Global | 32 |

**Fig. 3: Correlation of rs41423247 with Surrounding SNPs & Putative Effect**




LD = Linkage disequilibrium

**Table 3:Linear Regression Model of BMI Z-Score vs Genotype**

| Variable | Effect size | SE | P |
|---|---|---|---|
| Intercept | -0.83 | 0.52 | 0.110 |
| Rs41423247 (G/C) | 0.28 | 0.25 | 0.272 |
| Rs41423247 (C/C) | **1.04** | 0.52 | **0.049** |
| Age | 0.03 | 0.04 | 0.490 |
| Sex | 0.35 | 0.24 | 0.159 |

**Table 4: Binomial Regression Model of HPAS vs Genotype**

| Variable | Effect size | SE | P |
|---|---|---|---|
| Intercept | 6.21 | 3.41 | 0.068 |
| Rs41423247 (G/C) | **-1.28** | 0.55 | **0.021** |
| Rs41423247 (C/C) | **-18.12** | 164.08 | 0.991 |
| Age | 0.06 | 0.17 | 0.700 |
| Sex | -0.50 | 0.49 | 0.310 |
| Height | -0.05 | 0.04 | 0.203 |
| Weight | -0.01 | 0.04 | 0.771 |

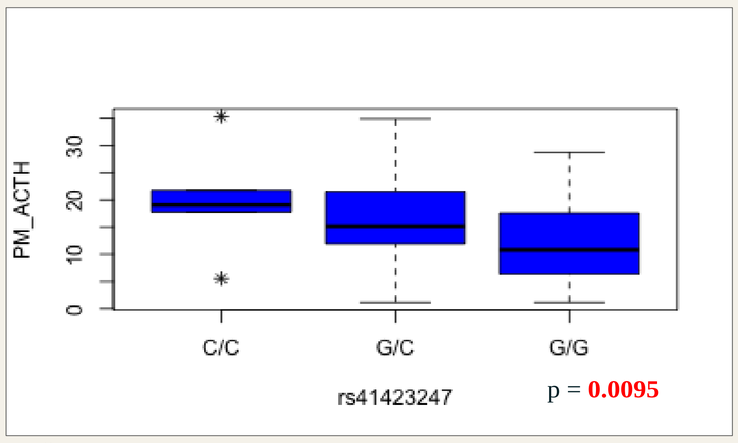
**Table 5:Linear Regression Model of √PMTP ACTH vs Genotype**

| Variable | Effect size | SE | P |
|---|---|---|---|
| Intercept | -5.30 | 10.65 | 0.620 |
| Rs41423247 (G/C) | **4.06** | 1.75 | **0.023** |
| Rs41423247 (C/C) | **10.85** | 3.80 | **0.005** |
| Age | -0.40 | 0.55 | 0.468 |
| Sex | -0.85 | 1.64 | 0.602 |
| Height | 0.14 | 0.11 | 0.232 |
| Weight | 0.06 | 0.10 | 0.591 |

**Table 6: Autosomal Dominance Analysis of PMTP √ACTH vs SNP**

| Variable | Effect size | SE | P |
|---|---|---|---|
| Intercept | -2.44 | 10.32 | 0.812 |
| Rs41423247 | **4.79** | 1.66 | **0.005** |
| Age | -0.55 | 0.54 | 0.311 |
| Sex | -1.61 | 1.58 | 0.311 |
| Height | 0.14 | 0.111 | 0.222 |
| Weight | 0.07 | 0.103 | 0.497 |

**Fig. 4: Difference in PMTP ACTH across Genotypes**[1)]




[1)] ANOVA

**Table 7: Multinomial Regression Model of ACTH Response vs Genotype**

| Variable | Effect size | SE | P |
|---|---|---|---|
| Intercept | -5.46 | 4.07 | 0.180 |
| Hi rs41423247 (G/C) | **1.41** | 0.62 | **0.023** |
| Hi rs41423247 (C/C) | **15.46** | 0.66 | **$< 2x10^{-16}$** |
| Hi age | -0.01 | 0.19 | 0.960 |
| Hi sex | -0.08 | 0.58 | 0.894 |
| Hi height | 0.03 | 0.04 | 0.535 |
| Hi weight | 0.03 | 0.05 | 0.559 |
| Intercept | -8.72 | 4.14 | 0.035 |
| Mid rs41423247 (G/C) | 1.13 | 0.63 | 0.073 |
| Mid rs41423247 (C/C) | **14.48** | 0.66 | **$< 2x10^{-16}$** |
| Mid age | -0.10 | 0.19 | 0.621 |
| Mid sex | 1.07 | 0.59 | 0.069 |
| Mid height | 0.07 | 0.04 | 0.123 |
| Mid weight | -0.02 | 0.05 | 0.712 |

## Conclusions

- The rs41423247 SNP of the NR3C1was found to be protective against HPAS & associated with BMI.
- The protective effect is independent of BMI.
- The homozygous CC genotype is associated with a higher PMTP ACTH response.
- This genetic effect is inherited in a dominant fashion.
- Rs41423247 has a putative regulatory effect.

## Acknowledgements

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## References

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2. Tsartsali L. Papadopoulos M. Lagona E et al. Association of hypothalamic-pituitary-adrenal axis-related polymorphisms with stress in asthmatic children on inhaled corticosteroids. Neuroimmunomodulation 2012; 19: 88-95