

# Inflammatory CD14+CD16++ monocytes are increased in Cushing's Syndrome

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## Introduction

Chronic hypercortisolism is marked by an increased cardiovascular risk pattern. Atherosclerosis is a chronic inflammation that involves both innate and adaptive immunity. Glucocorticoids (GC) are immune-suppressors and adrenocorticotrophic hormone (ACTH) possesses immune-modulatory activities. GC and ACTH may act in atherothrombotic inflammatory pathways.

## Aim

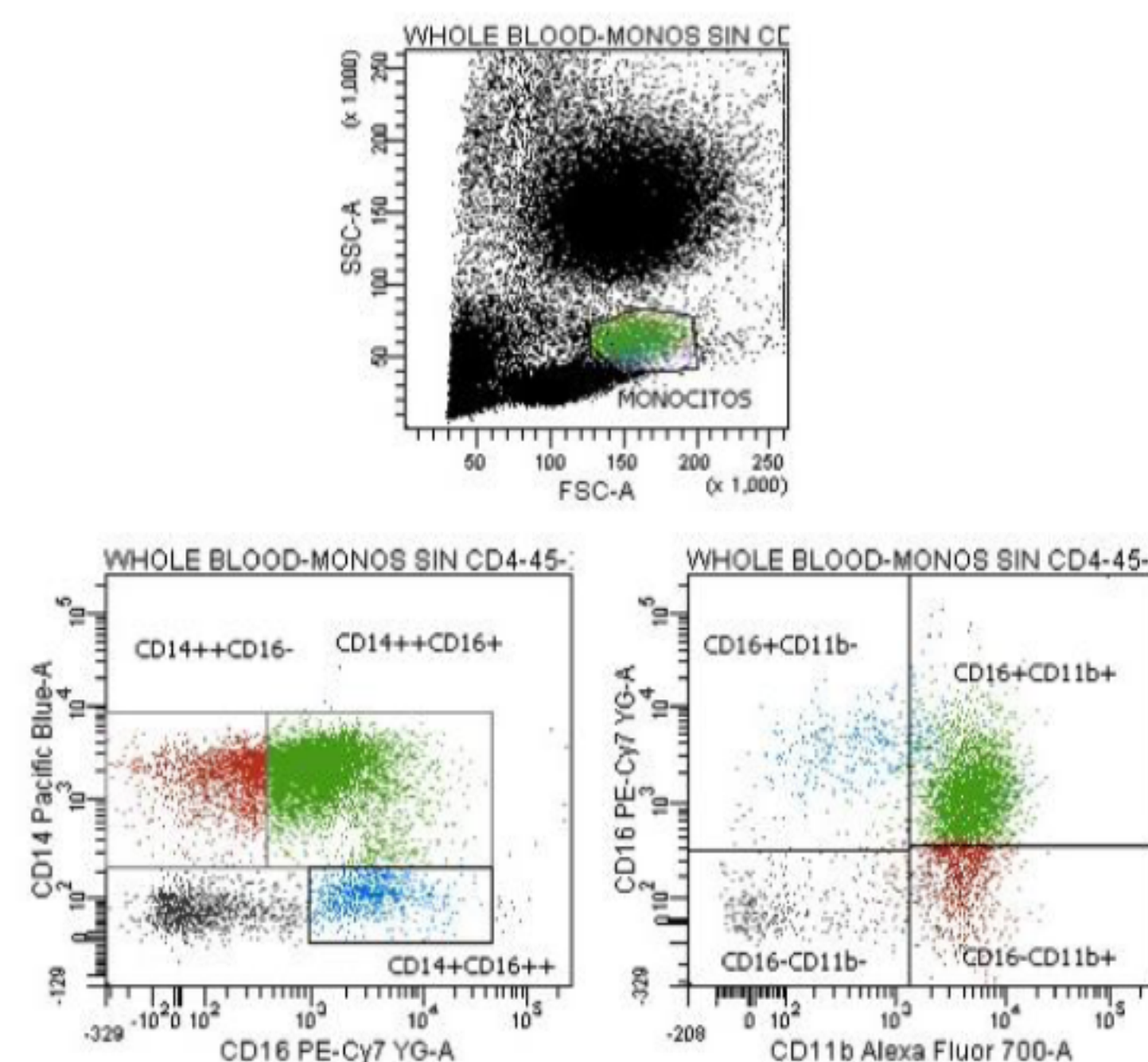
To analyse the immune cells pattern in endogenous Cushing syndrome (CS) in order to investigate their atherosclerotic risk phenotype and to evaluate the immune modulator role of ACTH on this pattern.

## Methods

26 CS: 16 ACTH dependent (D), 10 ACTH independent (ID) and 12 healthy controls (CTR) were included.

Peripheral immune cells, respectively monocytes (MN), lymphocytes (L) and neutrophils (N) were isolated from fresh blood and analysed by flow cytometry for the presence of cell surface activation markers previously associated with atherosclerosis.

Flow cytometry analysis of monocyte pattern in whole blood.



## Results

Leucocytes, N, MN, NKL were increased in CS ( $p < 0.05$ ). Atypical CD14+CD16++ MN were higher in ACTH-ID CS (8.9 3.5%) vs. ACTH-D CS (4.2 1.9%). Other tendencies: CD11b+ cells increased in ACTH-ID CS; CD15+, CD15+CD16+ and CD15+CD16+11b+N were higher in CS.

| Immune cell phenotyping       | ACTH dependent CS (n:16) | ACTH independent CS (n:10) | Controls (n:12) | P value      |
|-------------------------------|--------------------------|----------------------------|-----------------|--------------|
| <b>MONOCYTES</b>              |                          |                            |                 |              |
| CD14+CD16- Classical (%)      | 32.7 ± 29.2              | 36.4 ± 33.8                | 42.2 ± 32.5     | 0.805        |
| CD14+CD16-11B+ (%)            | 63.4 ± 37.5              | 86.6 ± 15.5                | 80.8 ± 19.6     | 0.195        |
| CD14+CD16+ Intermediate (%)   | 37.6 ± 25.1              | 34.2 ± 27.0                | 28.7 ± 26.2     | 0.748        |
| CD14+CD16+11B+ (%)            | 47.7 ± 43.7              | 59.8 ± 49.1                | 48.1 ± 48.7     | 0.845        |
| CD14+CD16++ Non Classical (%) | 4.2 ± 1.9                | 8.8 ± 4.5                  | 7.1 ± 2.3       | <b>0.009</b> |
| CD14+CD16++11B+ (%)           | 29.1 ± 38.7              | 52.9 ± 48.0                | 35.4 ± 41.1     | 0.508        |
| <b>NEUTROPHILS</b>            |                          |                            |                 |              |
| CD15+ (%)                     | 65.5 ± 27.5              | 66.3 ± 21.2                | 47.1 ± 13.9     | 0.205        |
| CD15+CD16+ (%)                | 64.4 ± 28.0              | 70.2 ± 21.6                | 44.3 ± 12.7     | 0.103        |
| CD15+CD16+11B (%)             | 74.2 ± 30.7              | 90.5 ± 15.3                | 69.2 ± 29.1     | 0.313        |
| CD16+11B (%)                  | 85.9 ± 28.8              | 96.4 ± 1.5                 | 94.2 ± 4.2      | 0.495        |
| <b>LYMPHOCYTES</b>            |                          |                            |                 |              |
| CD3+CD8+ (%)                  | 27.9 ± 14.1              | 33.2 ± 8.0                 | 26.8 ± 8.1      | 0.496        |
| CD3+CD4+ (%)                  | 43.6 ± 21.6              | 53.4 ± 12.6                | 54.2 ± 6.6      | 0.300        |
| CD3+CD4+CD25+ (%)             | 18.7 ± 16.7              | 23.7 ± 22.7                | 22.7 ± 11.1     | 0.813        |
| NK (%)                        | 30.8 ± 31.8              | 40.3 ± 34.4                | 18.4 ± 18.5     | 0.359        |
| NKCD11B+ (%)                  | 26.4 ± 31.2              | 47.6 ± 40.9                | 37.5 ± 44.4     | 0.541        |

## Conclusion

High chronic exposure to GC in CS increase in the absence of the protective immune-modulator presence of ACTH the non-classical atherosclerotic risk CD14+CD16++ monocytes.

EP733