Transcriptome analysis to reveal immune cell landscape of inflammatory infiltrates in giant cell arteritis

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20th International Vasculitis and ANCA Workshop 2022

Background

Giant Cell Arteritis (GCA):

- the most common form of vasculitis in people over 50 years of age
- characterized by inflammation in medium- and large-sized vessels
- serious clinical emergency (can lead to permanent visual loss)

Study motivations:

- **incompletely understood pathogenesis**
  - predisposing factors (genetics, gender, age, environment)[1]
- **limited treatment options**
  - high-dose steroids are the only first line effective therapy[2]
  - steroid therapy leads to significant toxicity > 80% of patients[3]

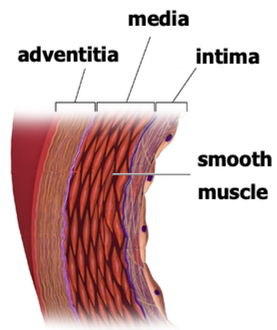


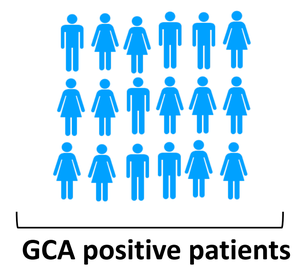
Figure 1. Arterial wall structure showing three layers (tunicae): adventitia, media and intima.

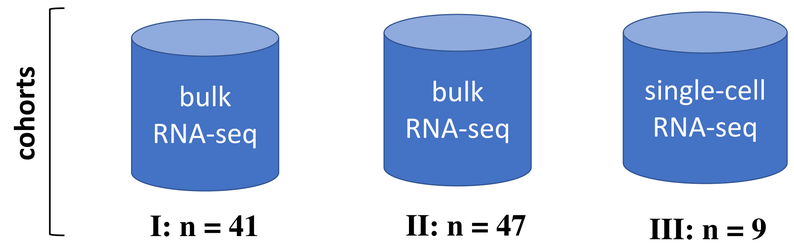
Image credit: Blausen.com (2014)

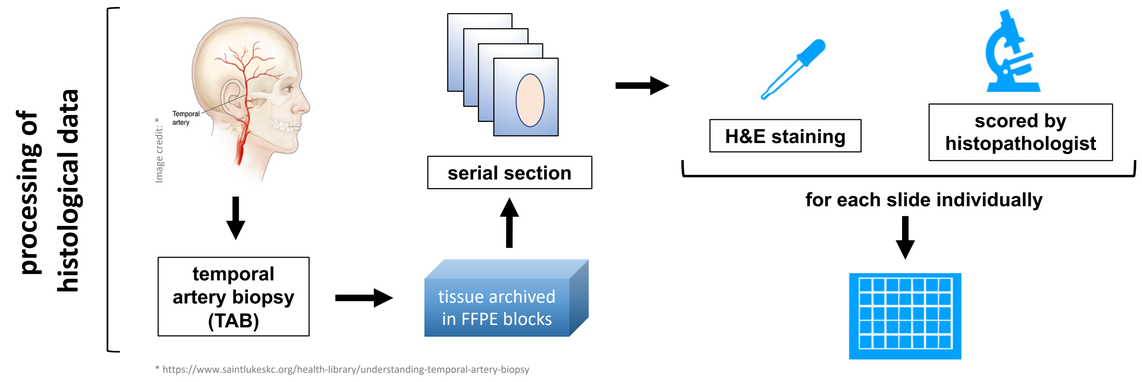
AIMS

- assess confounding influence of clinical variables (sex, gender, steroids duration)
- examine associations with histological phenotypes in GCA
- identify transcripts that relate to GCA subtypes and outcomes

Materials & Methods







Results

Table 1. Number of statistically significant genes detected in Mann-Whitney testing. FDR – False Discovery Rate

Features	Comparison groups	$P < .05$ (.01) TOTAL	FDR-corrected $P < .05$ (.01) down-regulated	up-regulated	TOTAL
clinical					
age	18 — 21	3 189 (980)	3	0	3 (2)
gender	15 — 24	1 447 (334)	3	19	22 (20)
steroids treatment	19 — 20	1 030 (157)	0	0	0
visual loss	24 — 15	1 527 (297)	0	1	1 (0)
histological					
severity of inflammation:					
∘ in adventitia	8 — 31	8 106 (4 911)	2 380	2 979	5 359 (2 455)
∘ in intima	16 — 23	6 409 (3 568)	1 665	1 636	3 301 (1 247)
∘ in media	21 — 18	6 257 (3 255)	987	1 650	2 637 (405)
giant cells presence	29 — 10	5 369 (2 587)	879	692	1 571 (0)
infiltrate around vasa vasorum	22 — 17	5 273 (2 242)	340	466	806 (9)
media destruction	19 — 20	5 387 (2 621)	872	948	1 820 (349)
neoangiogenesis	28 — 11	3 257 (1 190)	29	18	47 (0)
degree of arterial occlusion	17 — 22	4 067 (1 796)	363	267	630 (11)

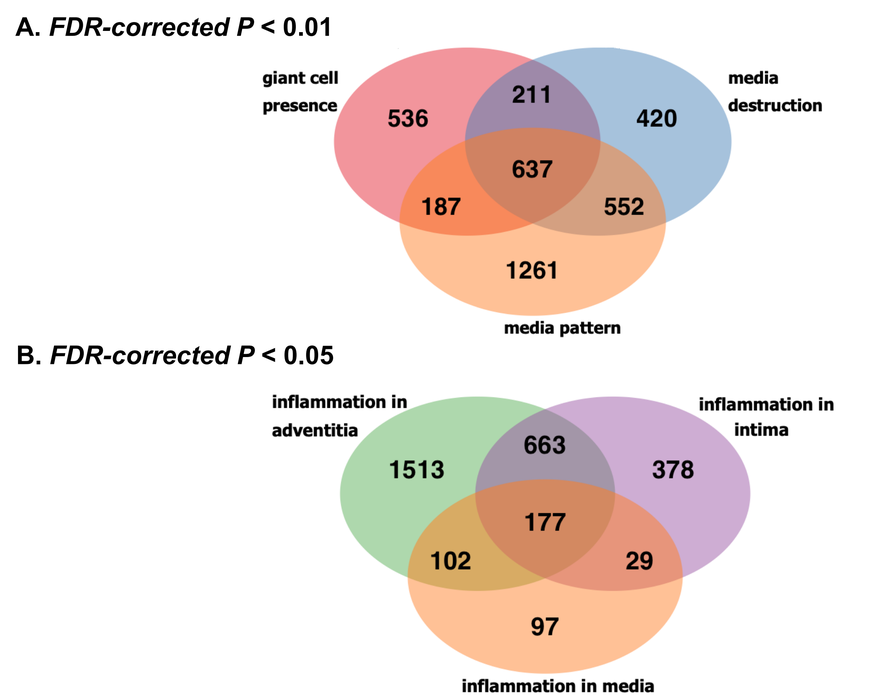


Figure 1. Venn diagrams showing number of statistically significant genes overlapping between different histological features.

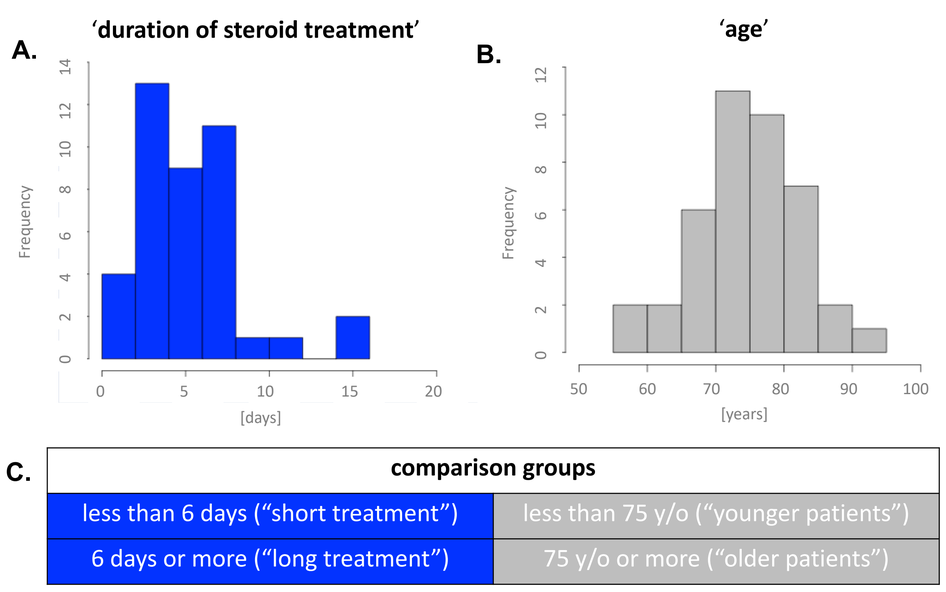


Figure 2. Frequency plots showing variable distribution of (A) duration of steroid treatment and (B) age, and (C) comparison groups that were used in Mann-Whitney testing to assess the influence of these variables on gene expression profiles. Results of these analyses are included in Table 1.

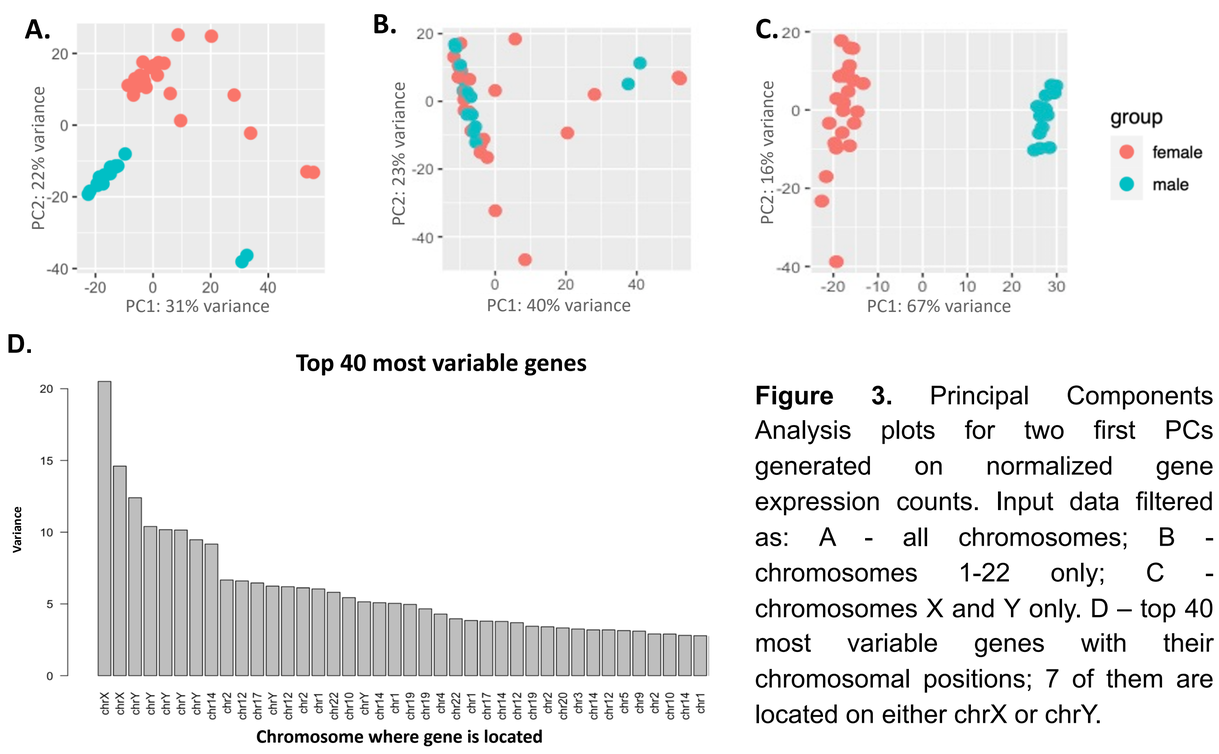


Figure 3. Principal Components Analysis plots for two first PCs generated on normalized gene expression counts. Input data filtered as: A - all chromosomes; B - chromosomes 1-22 only; C - chromosomes X and Y only. D – top 40 most variable genes with their chromosomal positions; 7 of them are located on either chrX or chrY.

Conclusions

- gender was found to likely have an influence on gene expression, while no statistical evidence for influence from duration of steroid treatment or age was found
- four histological phenotypes were found to have the strongest associations: presence of giant cells and extent of inflammation in the intima, media and adventitia

Future work

- pathway analysis and further interpretation of statistical testing results
- deconvolution of bulk data to estimate cell type proportions and to examine cell-type-specific associations with histological phenotypes

(1) Terrades-Garcia, N., & Cid, M.C. (2018); doi:10.1093/rheumatology/kex423; **(2)** Borchers, A.T. & Gershwin, M.E. (2012); doi:10.1016/j.autrev.2012.01.003; **(3)** Ponte, C. et al. (2015); doi:10.12998/wjcc.v3.i6.484

HELICAL · Newcastle University · UNIVERSITY OF LEEDS · MARIE CURIE ACTIONS · Horizon2020 European Union Funding for Research & Innovation · NIHR National Institute for Health Research · LEEDS Institute for Data Analytics · UKRI Medical Research Council

This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 813545, MRC TARGET partnership grant, and NIHR Leeds Biomedical Research Centre and In-vitro diagnostics co-operative grant.