#### <u>Materials Design Analysis Reporting (MDAR)</u> Checklist for Authors

The MDAR framework establishes a minimum set of requirements in transparent reporting applicable to studies in the life sciences (see Statement of Task: doi:10.31222/osf.io/9sm4x.). The MDAR checklist is a tool for authors, editors and others seeking to adopt the MDAR framework for transparent reporting in manuscripts and other outputs. Please refer to the MDAR Elaboration Document for additional context for the MDAR framework.

# **Materials**

Antibodies	Yes (indicate where provided: section/paragraph)	n/a
For commercial reagents, provide supplier		x
name, catalogue number and RRID, if available.		
Cell materials	Yes (indicate where provided: section/paragraph)	n/a
<b>Cell lines:</b> Provide species information, strain.		х
Provide accession number in repository <b>OR</b>		
supplier name, catalog number, clone number,		
Primary cultures: Provide species, strain, sex of		х
origin, genetic modification status.		
Everymental animals	Vac (indicate where provided, castion (nerograph)	-
Experimental animals	res (indicate where provided: section/paragraph)	n/a
caporatory animals: Provide species, strain, sex, age,		x
pumber in repeation status. Provide accession		
number clone number <b>OR</b> Supplier name, catalog		
Animal observed in or cantured from the		×
field: Provide species, say and age where		X
nossible		
Model organisms: Provide Accession number		v
in repository (where relevant) <b>OR</b> RRID		^
in repository (where relevancy on hind		
Plants and microbes	Yes (indicate where provided: section/paragraph)	n/a
Plants: provide species and strain, unique accession		х
number if available, and source (including location		
for collected wild specimens)		
Microbes: provide species and strain, unique		х
accession number if available, and source		
Human research participants	Yes (indicate where provided: section/paragraph)	n/a
Identify authority granting ethics approval (IRB or	Patients and control subjects were recruited	
equivalent committee(s), provide reference number	prospectively and consented for this study under the	
for approval.	auspices of IRB protocol # PRO15030072 issued through	
	the University of Pittsburgh.	
Provide statement confirming informed consent	Detionts were concented wing a recorded as a second	
-	Patients were consented using a research consent	
obtained from study participants.	separate from procedural consent.	
obtained from study participants. Report on age and sex for all study participants.	Patients were consented using a research consent   separate from procedural consent.   Participants >18y were recruited. Both sexes were	
obtained from study participants. Report on age and sex for all study participants.	Patients were consented using a research consent separate from procedural consent. Participants >18y were recruited. Both sexes were recruited	
obtained from study participants. Report on age and sex for all study participants.	Participants >18y were recruited. Both sexes were recruited	

# <u>Design</u>

Study protocol	Yes (indicate where provided: section/paragraph)	n/a
For clinical trials, provide the trial registration number <b>OR</b> cite DOI in manuscript.		x
Laboratory protocol	Yes (indicate where provided: section/paragraph)	n/a
Provide DOI or other citation details if detailed step-		х
by-step protocols are available.		
Experimental study design (statistics details)	Yes (indicate where provided: section/paragraph)	n/a
State whether and how the following have been		
done, or if they were not carried out.	Samples from natients were collected sequentially	
	(when possible). Based on differences between	
	historical controls demonstrating large differences	
	between certain mediators between the groups, we	
	calculated that we would a minimum of 6 controls and	
	6 patients. This would give an alpha of 0.05 and power	
	differences. We limited our N to the number of	
	controls.	
Randomisation	No randomisation	
Blinding	Blinding occurred with Multiplex measurements and	
	analysis	
Inclusion/exclusion criteria	Inclusion (patients): Age >18yrs, meeting clinical and	
	anatomic criteria for endovenous ablation or	
	Scieroinerapy Exclusion (natients): cancer active infection active	
	ulcer, peripheral arterial disease, kidney failure, CHF,	
	active heart disease	
	Inclusion (controls): Age >18	
	Exclusion (controls): no swelling, no varicose veins, no	
	telanglectasias, no evidence of vehous reliux.	
Sample definition and in-laboratory replication	Yes (indicate where provided: section/paragraph)	n/a
State number of times the experiment was		х
Define whether data describe technical or biological		x
replicates		^
Ethics	Voc (indicate where provided, section (paragraph)	n/2
Studies involving human participants: State details of	Patients and control subjects were recruited	11/ 4
authority granting ethics approval (IRB or equivalent	prospectively and consented for this study under the	
committee(s), provide reference number for	auspices of IRB protocol # PRO15030072 issued through	
approval.	the University of Pittsburgh.	
of authority granting ethics approval (IRB or		х
equivalent committee(s), provide reference number		
for approval.		
Studies involving specimen and field samples: State if		х
relevant permits obtained, provide details of		
authority approving study; if none were required,		
	l	l
Dual Use Research of Concern (DURC)	Yes (indicate where provided: section/paragraph)	n/a
II SLUDY IS SUDJECT TO DUAL USE RESEARCH OF CONCERN,		x
number for the regulatory approval		

#### **Analysis**

Attrition	Yes (indicate where provided: section/paragraph)	n/a
State if sample or data point from the analysis is		х
excluded, and whether the criteria for exclusion were		
determined and specified in advance.		
Chatistics	Mar (in discharged and an and it also a discussion (a supervised a)	
Statistics	Yes (indicate where provided: section/paragraph)	n/a
Describe statistical tests used and justify choice of tests.	Given very prominent differences seen between some mediators in prior work, our power calculation determined that we would need only 6 individuals per group to generate a potentially significant difference if seen. However, we aimed to recruit a minimum of 10 each. Given difficulties associated with recruiting controls we were able to recruit 8 and could not recruit more due to COVID 19. Mean values of inflammatory mediators, age and VCSS scores and their standard deviations were calculated. Individual inflammatory mediator data was compared between controls and subjects using Mann-Whitney U tests. Network analysis was carried out to define the central inflammatory network nodes in the control and CVI groups Connections were created if a correlation between two mediators was ≥ to a specific threshold Pearson correlation, either 0.7 or 0.95 (equivalent to p = 0.05), as indicated. These cross-correlations were visualized as networks created with MATLAB ® software (The MathWorks, Inc., Natick, MA). A robustness index was also calculated to measure network "strength", by comparing the number of connections created with a Pearson correlation of 0.7 to one of 0.95.(15) Spearman's correlation between IL-17A and GM-CSF and between IL-17A and IL-10 (Luminex <sup>TM</sup> data) using a modified version of a MATLAB®-based toolbox (26) as described previously (16). A p-value of less than 0.05 was considered significant.	
Data Availability	Yes (indicate where provided: section/paragraph)	n/a
State whether newly created datasets are available.	Data will be made available upon reasonable request	Π/a
including protocols for access or restriction on		
access.		v
number in repository or DOI or URL.		X
If publicly available data are reused, provide accession number in repository or DOI or URL, where		х

Code Availability	Yes (indicate where provided: section/paragraph)	n/a
For all newly generated code and software essential		x
for replicating the main findings of the study:		
State whether the code or software is available.		х
If code is publicly available, provide accession		х
number in repository, or DOI or URL.		

### **Reporting**

possible.

Adherence to community standards	Yes (indicate where provided: section/paragraph)	n/a
MDAR framework recommends adoption of		
discipline-specific guidelines, established and		

endorsed through community initiatives. Journals have their own policy about requiring specific guidelines and recommendations to complement MDAR.		
State if relevant guidelines (eg., ICMJE, MIBBI, ARRIVE) have been followed, and whether a checklist (eg., CONSORT, PRISMA, ARRIVE) is provided with the manuscript.	ICMJE guidelines were followed, as the journal follows ICMJE recommendations for publication.	

Article information: https://dx.doi.org/10.21037/atm-21-688