Supplementary Materials

Analysis of Phosphodiesterase-5 (PDE5) Inhibitors in Modulating Inflammatory Markers in Humans: A Systematic Review and Meta-Analysis

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Supplementary Materials

Table S1. Description of outcome biomarkers used in included studies.

Abbreviation	Name	Effect	Immune effect
PDE5	Phosphodiesterase 5 inhibitor	Block the action of the enzyme PDE5. [1]	N/A
CRP	C-reactive protein	Produced in the liver from stimulation from IL-6, binds to cells to activate complement system. [2]	Pro-inflammatory
TNF-α	Tumour necrosis factor alpha	Produced by macrophages (primarily), induces inflammation via TNF receptors. [3]	Pro-inflammatory
IL-6	Interleukin 6	Secreted by macrophages in response to specific microbial molecules, referred to as pathogen-associated molecular patterns (PAMPs). [4]	Pro-inflammatory
IL-8	Interleukin 8	Chemokine involved in attracting neutrophils to sites of inflammation and other immune responses. [5]	Pro-inflammatory
ICAM	Intercellular adhesion molecule	Cell surface glycoprotein involved in cell adhesion and immune responses, particularly during inflammation. [6]	Pro-inflammatory
VCAM	Vascular cell adhesion protein	Mediates the adhesion of lymphocytes, monocytes, eosinophils and basophils to vascular endothelium. [7]	Pro-inflammatory
P-Selectin	P-selectin	Cell adhesion molecule belonging to the selectin family, expressed on activated platelets and endothelial cells. [8]	Pro-inflammatory
IL-10	Interleukin 10	Produced by various immune cells, such as macrophages and lymphocytes. Helps to prevent excessive inflammation and autoimmunity. [9]	Anti-inflammatory
NO	Nitric oxide	Produced by nitric oxide synthases, activates cGMP production causing vasodilation. Free radical which causes DNA damage in targeted cells. Reduces immune cell function. [10]	Anti-inflammatory
cGMP	Cyclic Guanosine Monophosphate	Intracellular secondary messenger, upon activation by NO catalyses. [11]	Anti-inflammatory

Table S2. Characteristics of included studies.

No	Study	Clinical population and sample size	Stimulant	Intervention duration	Comparison groups	Outcome measure o□interest	Effect size Std. mean difference, IV, Random, 95% CI
1	Aversa *et al.* (2008)	Men with type 2 diabetes (n=20)	Sildenafil	3 days 100mg/day, 4 days 25mg/day, 3 weeks 25mg/day	Placebo & Intervention	Nitric oxide IL-6 CRP ICAM VCAM	(Negative favours intervention) IL-6: -6.22 (-8.55, -3.90) (Negative favours intervention) CRP: -3.43 (-4.91, -1.96)
2	Burnett *et al.* (2009)	Men with type 2 diabetes and erectile dysfunction (n=292)	Sildenafil	Phase 1: 1-week 50mg/day, 3 weeks 100mg/day Phase 2: 2 weeks 50mg/day, 10 weeks either 25mg, 50mg or 100mg/day (self-managed)	Placebo & Intervention	cGMP IL-6 IL-8	(Positive favours intervention) cGMP: 0.59 (0.08, 1.11) (Negative favours intervention) IL-6: -0.51 (-1.10, 0.09) (Positive favours control) IL-8: 0.36 (-0.23, 0.94)
3	Ceci *et al.* (2015)	Healthy males undertaking exhaustive exercise (n=13)	Tadalafil	48 hours 20mg/day	Placebo & Intervention	IL-6	(Positive favours control) IL-6: 2.33 (1.3, 3.36)
4	Ceci *et al.* (2017)	Healthy males (n=12)	Tadalafil	24 hours 20mg/day	Placebo & Intervention	NO cGMP IL-6 IL-8 IL-10 TNF-α	(Positive favours intervention) cGMP: 0.16 (-0.64, 0.96) (Positive favours control) IL-6: 0.26 (-0.54, 1.06) (Positive favours control) TNF-α: 0.6 (-0.22, 1.42)
5	Corinaldesi *et al.* (2021)	Men and women with systemic sclerosis (n=116)	Sildenafil	Part of treatment regimen for serious cases	Placebo & Intervention	CXCL10	Not estimable.

6	Fryk *et al*. (2023)	Men and women with type 2 diabetes (n=36)	Tadalafil	6 weeks 20mg/day	Placebo & Intervention	CRP TNF-α IL-6	(Positive favours control) CRP: 0.26 (-0.40, 0.91) (No favour) IL-6: 0.00 (-0.65, 0.65)
7	Giannattasio *et al*. (2018)	Men with diabetic cardiomyopathy (n=61)	Sildenafil	3 months 100mg/day	Placebo & Intervention	IL-8	(Negative favours intervention) IL-8: -0.61 (-1.20, -0.02)
8	Goudie *et al*. (2014)	Men and women with chronic pulmonary disease (COPD) (n=120)	Tadalafil	12 weeks 10mg/day	Placebo & Intervention	CRP	(Positive favours control) CRP: 0.10 (-0.27, 0.47)
9	Jamaluddin *et al*. (2019)	Men with erectile dysfunction (n=220)	Tadalafil	6 weeks 10mg/day	No placebo, Before and after intervention	CRP	(Negative favours intervention) CRP: -0.22 (-0.41, -0.04)
10	Kilic *et al*. (2023)	Men with erectile dysfunction (n=185)	Either of Sildenafil or Tadalafil	4 days of either 100mg/day sildenafil or 20mg/day tadalafil	No placebo, Before and after intervention	NLR (neutrophil/lymphocyte ratio) LMR (lymphocyte/monocyte ratio) PLR (platelet/lymphocyte ratio) MHR (monocyte/high-density lipoprotein cholesterol ratio) Neutrophils Lymphocytes Monocytes	Not estimable.

11	Kumar *et al.* (2020)	Men and women with acute kidney injury undergoing cardiac surgery (n=125)	Sildenafil	12.5mg/150 minutes (given during surgery)	Placebo & Intervention	IL-10 IL-6 IL-8 NO	Not estimable.
12	Li *et al.* (2018)	Overweight men (n=16)	Sildenafil	1-week 100mg/day	Placebo & Intervention	cGMP TNF-α IL-6 CRP	(Positive favours intervention) cGMP: 3.47 (1.78, 5.17) (Positive favours control) TNF-α: 0.02 (-0.96, 1.0) (No favour) IL-6: 0.00 (-0.98, 0.98) (Negative favours intervention) CRP: -0.29 (-1.28, 0.70)
13	Mandosi *et al.* (2015)	Men with type 2 diabetes (n=28)	Sildenafil	3 months 100mg/day	No placebo, Before and after intervention	P-selectin ICAM	(Negative favours intervention) P-selectin: -0.59 (-1.13, -0.06) (Negative favours intervention) ICAM: -0.19 (-0.72, 0.33)
14	Morano *et al.* (2007)	Men with type 2 diabetes and erectile dysfunction (n=32)	Sildenafil	12 weeks 50mg/twice weekly	Placebo & Intervention	P-selectin ICAM	(Negative favours intervention) P-selectin: -0.51 (-1.51, 0.49) (Positive favours control) ICAM: 3.11 (1.53, 4.69)
15	Pofi *et al.* (2022)	Men and women with long duration diabetes (n=122)	Tadalafil	20 weeks 20mg/day	Placebo & Intervention	cGMP	(Positive favours intervention) cGMP: 1.07 (0.69, 1.45)
16	Santi *et al.* (2016)	Men with type 2 diabetes and erectile dysfunction (n=54)	Vardenafil	24 weeks 10mg/day	Placebo & Intervention	CRP ICAM VCAM IL-6	(Positive favours control) CRP: 0.11 (-0.42, 0.65) (Positive favours control) ICAM: 3.01 (2.22, 3.81) (Negative favours intervention) IL-6: -0.75 (-1.30, -0.20)

17	Semen *et al*. (2016)	Men and women with pulmonary arterial hypertension (n=14)	Sildenafil	12 weeks at 25mg/three times a day	Placebo & Intervention	4-hydroxynonenal (4HNE)	Not estimable.
18	Taylor-Cousar *et al*. (2015)	Men and women with mild to moderate cystic fibrosis lung disease (n=20)	Sildenafil	6 weeks of either 20mg/day or 40mg/day	No placebo, Before and after intervention	Sputum elastase activity	Not estimable.
19	Vignozzi *et al*. (2013)	Men with benign prostatic hyperplasia ± metabolic syndrome (n=43)	Vardenafil	12 weeks 10mg/day	Placebo & Intervention	Anti-pan leukocytes (CD45)	Not estimable.
20	Vlachopoulos *et al*. (2015)	Men with erectile dysfunction (n=20)	Sildenafil	Over either 2, 4 or 8 hours 100mg	Placebo & Intervention	CRP IL-6 VCAM-1 TNF-α	(Negative favours intervention) CRP: -1.65 (-2.37, -0.92) (Negative favours intervention) IL-6: -1.09 (-1.76, -0.42) (Negative favours intervention) TNF-α: -1.27 (-1.95, -0.58)

Table S3. List of excluded articles and reasons.

Reasons for exclusion	**Articles**
In vitro data only	1. Kohzadi, R.; Cheraghi, E.; Mehranjani, M.S.; Shariatzadeh, M. Sildenafil citrate ameliorates the adverse effects of cryopreservation on sperm quality in asthenozoospermic men. Cryobiology 2023, 111, 126-133, doi:10.1016/j.cryobiol.2023.05.005. 2. Di Luigi, L.; Sgrò, P.; Duranti, G.; Sabatini, S.; Caporossi, D.; Del Galdo, F.; Dimauro, I.; Antinozzi, C. Sildenafil Reduces Expression and Release of IL-6 and IL-8 Induced by Reactive Oxygen Species in Systemic Sclerosis Fibroblasts. Int J Mol Sci 2020, 21, doi:10.3390/ijms21093161.

Table S4. Risk of bias of included studies.
Key: 1= Risk of bias arising from the randomization process, 2= Risk of bias due to deviations from the intended interventions (effect of assignment to intervention, 3= Missing outcome data, 4= Risk of bias in measurement of the outcome, 5= Risk of bias in the selection of the reported result. L= Low, H= High, SC= Some concerns.

Articles	**Domain 1**	**Domain 2**	**Domain 3**	**Domain 4**	**Domain 5**	**Overall Risk o bias**
Aversa *et al.* (2008)	L	L	L	L	L	L
Burnett *et al.* (2009)	L	L	L	L	L	L
Ceci *et al.* (2015)	L	L	L	L	L	L
Ceci *et al.* (2017)	L	L	L	L	L	L
Corinaldesi *et al.* (2021)	L	L	SC	L	L	L
Fryk *et al.* (2023)	L	L	L	L	L	L
Giannattasio *et al.* (2018)	L	L	L	L	L	L
Goudie *et al.* (2014)	L	L	L	L	L	L
Jamaluddin *et al.* (2019)	L	L	SC	L	L	L
Kilic *et al.* (2023)	L	SC	L	L	L	L
Kumar *et al.* (2020)	L	L	L	L	L	L
Li *et al.* (2018)	L	L	L	L	L	L
Mandosi *et al.* (2015)	L	L	L	L	L	L
Morano *et al.* (2007)	L	L	L	L	L	L
Pofi *et al.* (2022)	L	L	SC	L	L	L
Santi *et al.* (2016)	L	L	L	L	L	L
Semen *et al.* (2016)	H	SC	SC	SC	L	SC
Taylor-Cousar *et al.* (2015)	SC	SC	SC	L	SC	SC
Vignozzi *et al.* (2013)	L	L	L	L	L	L
Vlachopoulos *et al.* (2015)	L	L	SC	L	L	L

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