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Could androgens be relevant to partly explain why men have lower life expectancy than women?

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Life expectancy is about 5 years shorter for men than for women.[1] At any given age, men are more vulnerable than women to death from most major causes, including infections, cancer and cardiovascular disease.[1] Lifestyle and stress undoubtedly play the same role in this disparity as in other health disparities, particularly given historically higher smoking rates for men than for women. Whether these factors provide a comprehensive explanation and actionable targets of intervention is less clear, particularly as these factors do not fully explain men's cardiovascular disadvantage.[2] Here, to provide another perspective, the sexual disparity in life expectancy is considered in the context of an existing well-established theory from evolutionary biology. Life history theory suggests that animals, including humans, employ environmentally driven strategies to maximise Darwinian fitness, that is, reproductive success.[3] Optimal strategies for reproductive success likely differ by sex, because men may have far more children than women[4] but reproductive costs are far higher for women. As such, not only does a strategy aimed at fitness differ from the public health goal of long and healthy life but may also have sex-specific implications, and so be relevant to the shorter lifespan in men than in women.

Specifically, life history strategies may involve trading off growth or reproduction against longevity,[3] with the role of less growth well known but suppressing the reproductive axis may also increase lifespan;[5] and promoting the reproductive axis may reduce lifespan. For example, a high-protein diet in mice results in bigger gonads and shorter life, while a high-carbohydrate diet results in smaller gonads and longer life;[6] interestingly mice prefer a higher protein diet.[6] In humans, the reproductive axis in women is suppressed at menopause, and artificial supplementation with reproductive hormones in postmenopausal women is not beneficial for lifespan.[7] In contrast, men continue to be fertile throughout adult life; little consideration has been given to whether lifelong fertility in men relates to or trades off against lifespan. Given the shorter lifespan for men than for women, consideration is given here to how male reproductive hormones, namely, androgens, affect health in men beyond reproduction, and whether androgens could have any relevance to the sexual disparity in life expectancy, or any implications for the prevention and treatment of the leading causes of mortality.

NON-REPRODUCTIVE EFFECTS OF ANDROGENS IN MEN

Currently, the non-reproductive effects of androgens on men's health is little researched.[8] The published literature also suffers from citation bias towards studies putting testosterone in a positive light.[9]

Observational evidence concerning endogenous testosterone is difficult to interpret because the most commonly measured androgen biomarker, serum testosterone, falls in men with obesity and ill-health.[10] Observed inverse associations of serum testosterone with major causes of mortality, such as cardiovascular disease, may be confounded in men by health status and so be difficult to interpret, as the authors of the relevant systematic reviews and meta-analyses have been careful to note.[11–13] Other androgen biomarkers that give observational associations more similar to results from randomised controlled trials (RCTs) for ischaemic heart disease or its risk factors are rarely reported.[14] Similarly, observationally, androgens do not predict prostate cancer,[15] but genetic studies have removed doubts about a role for androgens in prostate cancer.[16] Observational evidence concerning exogenous testosterone prescription in men is mixed,[17–20] and difficult to interpret because of the possibility of bias by indication and/or immortal time,[21] which may generate findings at variance with meta-analysis of RCTs.[20 22–24] The only observational study of testosterone prescription that used a self-comparison and a control exposure is probably the most convincing: it found, specifically, that testosterone prescription was associated with a higher risk of non-fatal myocardial infarction.[25]

Evidence about the effects of androgens from RCTs is limited because the US Institute of Medicine advised, in 2004, that no large scale trials of testosterone should be undertaken until benefits over existing treatments had been established in small trials.[26] Health comparisons between men with genetically higher or lower androgens using Mendelian randomisation are limited because androgens are rarely measured, so few genetic variants reliably determining androgens have been identified.[27 28]

Nevertheless, information about the role of androgens in men's health is slowly emerging. Below, the effects of androgens on major contributors to global mortality are considered, including immune function, cancer, cardiovascular disease and diabetes. Given the focus on the effects of androgens, greatest emphasis is placed on evidence about likely causal effects from RCTs, Mendelian randomisation, experiments, natural experiments and knockout animal models, rather than observed associations.

Immune function

Androgens are generally understood to suppress the immune system;[29–32] whereas oestrogen promotes immune response.[33] For example, testosterone may inhibit CD4 T-cell function,[30] while androgen deprivation is associated with enlargement of the thymus, which promotes immune function.[29] Experiments in rodents suggest androgens are pro-inflammatory and delay wound healing.[34 35]

Men tend to be more vulnerable to infections than women.[33 36 37] Lifestyle undoubtedly plays a part. However, the male vulnerability is most evident at ages when sexual dimorphism in sex hormones is greatest, those being, early infancy, puberty and young adult life, and includes periods, such as early infancy, when lifestyle choices are unlikely to differ by sex.[37] Male disadvantage is less evident for infections where a pathological immune response occurs.[37]

On the other hand, androgens may have the benefit of suppressing autoimmune responses,[29–31] consistent with men being less vulnerable to autoimmune diseases than women,[29 31] with the difference

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emerging at puberty. Androgen administration has shown some promise in trials as a treatment for rheumatoid arthritis.[38 39] Whether biological treatments for autoimmune diseases, such as tumour necrosis factor-α inhibitors, operate via a similar mechanism, is unknown.

Cancer

Given androgens' suppressive effects on immune function,[29–32] androgens are thought to play a role not only in prostate and testis cancer, but also in cancer more generally,[29] although evidence in men is scarce. Based on androgen knockout models, androgens could play a permissive role in several cancers with a male predondrance,[40] including cancer of the bladder,[41] kidney,[42] liver[43] and lung,[44] as well as, perhaps, of the esophagus[45] and colorectum.[46] Castration protects male rats against colonic adenomas.[46] Whether androgens contribute to men's vulnerability to cancer, and whether cancers with male predominance share features, has not been comprehensively investigated. Androgens' role in immune function also raises questions about other major chronic diseases with an inflammatory component[47 48] to which men are more vulnerable, such as cardiovascular disease.

Cardiovascular disease

Men have higher cardiovascular disease mortality rates than women,[49] particularly at the period in life, early adulthood, when male hormones are highest.[49 50] However, haemorrhagic stroke mortality rates differ little by sex,[51] suggesting that the relevant causative factor underlying men's cardiovascular disadvantage is specific to processes that drive non-haemorrhagic forms of cardiovascular disease, such as ischaemia, thrombosis, embolism and aneurysm. Notably, androgens may raise clotting factors, such as thromboxane.[52 53]

Currently, at the same level of the major risk factors for atherosclerotic cardiovascular disease, specifically, age, smoking, low-density lipoprotien (LDL)-cholesterol, high-density lipoprotein (HDL)-cholesterol, blood pressure and diabetes, validated risk prediction models give substantially higher absolute risk for men than for women.[54] Although menopause precedes an increase in ischaemic heart disease rates for women,[55] RCTs have shown that oestrogen does not protect against cardiovascular disease in either men[56] or women.[7] Recent promotion of highly profitable testosterone to older men as an antidote to ageing has led to warnings about the cardiovascular risk of testosterone from regulators. In June 2014, the US Food and Drug Administration (FDA) added a warning about venous blood clots to the testosterone product label.[57] In July 2014, Health Canada warned about "serious and possible life-threatening heart and blood vessel problems such as heart attack, stroke, blood clot in the lungs or legs; and increased or irregular heart rate with the use of testosterone replacement products".[58] The European Medicines Agency was less clear in November 2104,[59] but advised that testosterone should be restricted to men with pathological hypogonadism.[59] In March 2015, the US FDA warned about "the possible increased risk of heart attacks and strokes associated with testosterone use" and required a label change.[60] Sales of testosterone are already falling in the USA[61] and litigation against pharmaceutical companies is underway.[62]

These warnings from regulators about the cardiovascular risk of testosterone[57 58 60] suggest that substantially higher androgens in men than women could underlie higher cardiovascular disease mortality rates in men than in women, although any adverse effects of exogenous androgens in unhealthy older men[57 58 60] might not translate to endogenous androgens in healthy men. In addition, few androgen-related genetic variants have been found in genome-wide association studies of cardiovascular disease. However, few such studies are sex-specific, few androgen related genetic variants have been identified[27 28] and many genetic variants are of unknown function, so this may be absence of evidence rather than evidence of absence.

The known effects of testosterone administration in men from meta-analyses of RCTs, include lower HDL-cholesterol,[63] higher haemoglobin,[63] lower adiponectin[64] and higher risk of venous thrombosis.[22] Some of these changes also take place in boys at puberty under the influence of endogenous testosterone,[65] suggesting similar effects of exogenous and endogenous testosterone. Mendelian randomisation studies also suggest endogenous testosterone lowers HDL-cholesterol[66] as well as possibly raising heart rate,[67] consistent with the warning from Health Canada, and again suggesting similar effects of endogenous and exogenous testosterone. Most systematic reviews and meta-analyses of RCTs have shown non-significantly higher risk of cardiovascular events on testosterone,[68–71] but are underpowered, partly because of incomplete event reporting in the underlying RCTs,[70] and, in one meta-analysis, a numerical error (reporting on 308 instead of 485 men).[69] Notably, the most recent trial of testosterone found 12 major adverse cardiovascular events among 155 men in the treatment arm and 4 among 151 men in the placebo arm, OR 3.08, 95% CI 0.97 to 9.79.[72] Men with lower endogenous testosterone due to legal castration have relatively lower rates of specifically myocardial infarction.[73] Similarly, men with lifelong low testosterone due to Klinefelter's syndrome have low rates of ischaemic heart disease,[74] despite well-known vulnerability to diabetes.[75]

Diabetes

Diabetes is a strong, well-established risk factor for cardiovascular disease, although sex differences in diabetes rates are minor and inconsistent.[76] RCTs suggest androgens improve glucose metabolism,[77] perhaps through building muscle mass, which is a sink for glucose disposal. Whether treatments for diabetes operate by raising androgens has rarely been assessed. However, insulin may raise androgens,[78] which could be relevant to the difference in magnitude between observed and causal estimates for the association of diabetes with cardiovascular disease.[79]

ENVIRONMENTAL DRIVERS OF ANDROGENS

Despite the potential benefits of androgens for some autoimmune diseases and diabetes, consideration of the possibility that androgens could have a role in chronic diseases with a male preponderance would be consistent with evolutionary biology and the newly emerging evidence, as well as potentially providing aetiological insight concerning some diseases acknowledged to be poorly understood.[2] Many may feel that such a perspective would not be useful in clinical or public health practice because antiandrogens would be unacceptable to men. However, understanding alone could be valuable. For example, androgens lowering HDL-cholesterol and raising cardiovascular disease risk could underlie the observed inverse association of HDL-cholesterol with cardiovascular disease and thereby explain the failure of HDL-cholesterol raising therapies in cardiovascular disease.[80] Similarly, androgens generating systemic inflammation and raising cardiovascular risk could explain the failure of several anti-inflammatories.[81] Pharmaceutical companies are investing less in cardiovascular therapies[82] following expensive failures with lipid modulators and anti-inflammatories, highlighting the need for

new approaches. Moreover, antiandrogens are quite widespread and in therapeutic use. Obesity in men reduces testosterone.[83] Antiandrogens are used in prostate cancer. Several therapies for cardiovascular disease have anti-androgen properties, such as spironolactone, statins[84] and perhaps digoxin.[85] Whether any other therapies for cardiovascular disease currently in use or being trialled, such as diuretics or methotrexate,[81] reduce androgens is unknown. Diuretics are thought to affect sexual function adversely.

Whether androgenic effects could potentially provide a new classifying principle for common modifiable environmental exposures as healthy or otherwise has rarely been considered. Environmental drivers of androgens are usually conceptualised as xenobiotic endocrine disruptors disrupting androgens, with harmful effects.[86] However, evolutionary biology would predict that androgens in men should be sensitive to environmental indicators of breeding conditions, so as to focus reproductive effort at suitable times while minimising the risk attached to extreme masculinity, which may have been selected against in human history.[87] Few common modifiable exposures are known to affect testosterone, apart from obesity in men, driven by the conversion of testosterone to oestrogen in fat cells.[83] As might be expected, chronic undernutrition reduces testosterone.[88] However, the physiological processes that regulate testosterone in response to environmental conditions have not been systematically investigated. Androgens are sensitive to environmental influences, because they share metabolism and catabolism pathways with major drugs, foods and environmental exposures.[89] These catabolism pathways have been studied in the context of drug–drug interactions,[90] are polymorphic,[89] and are susceptible to epigenetic programming during early life.[91] Notably, diclofenac,[92] rofecoxib,[93] ibuprofen[92] and nicotine,[94] all inhibit these pathways, thereby most likely raising androgens, and all have unexpected adverse cardiovascular side effects.[95 96] Testing common modifiable exposures, selected according to their role in androgen metabolism or catabolism, for their effects on androgens in vitro, might provide a preliminary classification of exposures as potentially adverse or protective for further investigation in vivo.

CONCLUSION

Considering androgens as potential contributors to major diseases represents a major paradigm shift that flies in the face of individual level data from observational studies. However, in addition to experimental evidence and warnings from regulators, relevant physiological pathways exist. Androgens suppressing the immune system could increase vulnerability to infections and cancer. Androgens increasing clotting could increase vulnerability to cardiovascular disease. Despite some benefits of androgens for autoimmune diseases and diabetes, men have shorter lives than women. Current understanding of major chronic diseases is incomplete. Surely, we should capitalise on the insight unexpectedly provided by commercial promotion of androgens to older men to rethink the role of androgens, particularly, in immune function, cancer and cardiovascular disease, as potentially providing an underlying explanatory mechanism that could address the major sexual disparity in life expectancy, help identify new specific targets of intervention, explain unexpected side effects of commonly used drugs and eventually provide targets for precision medicine.

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REFERENCES

1 Mathers CD, Boerma T, Ma FD. Global and regional causes of death. *Br Med Bull* 2009;92:7–32.
2 Ezzati M, Obermeyer Z, Tzoulaki I, *et al*. Contributions of risk factors and medical care to cardiovascular mortality trends. *Nat Rev Cardiol* 2015;12:508–30.
3 Lemaitre JF, Berger V, Bonenfant C, *et al*. Early-late life trade-offs and the evolution of ageing in the wild. *Proc Biol Sci* 2015;282:20150209.
4 Balaresque P, Poulet N, Cussat-Blanc S, *et al*. Y-chromosome descent clusters and male differential reproductive success: young lineage expansions dominate Asian pastoral nomadic populations. *Eur J Hum Genet* 2015;23:1413–22.
5 Mukhopadhyay A, Tissenbaum HA. Reproduction and longevity: secrets revealed by C. elegans. *Trends Cell Biol* 2007;17:65–71.
6 Solon-Biet SM, Walters KA, Simanainen UK, *et al*. Macronutrient balance, reproductive function, and lifespan in aging mice. *Proc Natl Acad Sci U S A* 2015;112:3481–6.
7 Manson JE, Chlebowski RT, Stefanick ML, *et al*. Menopausal hormone therapy and health outcomes during the intervention and extended post-stopping phases of the Women's Health Initiative randomized trials. *JAMA* 2013;310:1353–68.
8 Schooling CM, Xu L, Zhao J, *et al*. Norms hide causes-the example of testosterone. *Int J Epidemiol* 2014;43:1987–8.
9 Zhao J, Schooling CM. Promotion of "Low T" and citation bias in testosterone studies. *Int J Cardiol* 2015;184:510–11.
10 Shi Z, Araujo AB, Martin S, *et al*. Longitudinal changes in testosterone over five years in community-dwelling men. *J Clin Endocrinol Metab* 2013;98:3289–97.
11 Araujo AB, Dixon JM, Suarez EA, *et al*. Clinical review: endogenous testosterone and mortality in men: a systematic review and meta-analysis. *J Clin Endocrinol Metab* 2011;96:3007–19.
12 Ruige JB, Mahmoud AM, De BD, *et al*. Endogenous testosterone and cardiovascular disease in healthy men: a meta-analysis. *Heart* 2011;97:870–5.
13 Holmegard HN, Nordestgaard BG, Jensen GB, *et al*. Sex hormones and ischemic stroke: a prospective cohort study and meta-analyses. *J Clin Endocrinol Metab* 2015:jc20152687.
14 Schooling CM. Androgen activity, ischaemic heart disease and risk factors among men in NHANES III. *Eur J Clin Invest* 2013;43:1273–81.
15 Roddam AW, Allen NE, Appleby P, *et al*. Endogenous sex hormones and prostate cancer: a collaborative analysis of 18 prospective studies. *J Natl Cancer Inst* 2008;100:170–83.
16 Sissung TM, Price DK, Del RM, *et al*. Genetic variation: effect on prostate cancer. *Biochim Biophys Acta* 2014;1846:446–56.
17 Vigen R, O'Donnell CI, Baron AE, *et al*. Association of testosterone therapy with mortality, myocardial infarction, and stroke in men with low testosterone levels. *JAMA* 2013;310:1829–36.
18 Sharma R, Oni OA, Gupta K, *et al*. Normalization of testosterone level is associated with reduced incidence of myocardial infarction and mortality in men. *Eur Heart J* 2015;36:2706–15.
19 Baillargeon J, Urban RJ, Kuo YF, *et al*. Risk of myocardial infarction in older men receiving testosterone therapy. *Ann Pharmacother* 2014;48:1138–44.
20 Baillargeon J, Urban RJ, Morgentaler A, *et al*. Risk of venous thromboembolism in men receiving testosterone therapy. *Mayo Clin Proc* 2015;90:1038–45.
21 Faillie JL, Suissa S. [Immortal time bias in pharmacoepidemiological studies: definition, solutions and examples]. *Therapie* 2015;70:259–63.
22 Xu L, Schooling CM. Differential risks in men and women for first and recurrent venous thrombosis: the role of genes and environment: comment. *J Thromb Haemost* 2015;13:884–6.
23 Bosco C, Bosnyak Z, Malmberg A, *et al*. Quantifying observational evidence for risk of fatal and nonfatal

cardiovascular disease following androgen deprivation therapy for prostate cancer: a meta-analysis. *Eur Urol* 2015;68:386–96.
24 Nguyen PL, Je Y, Schutz FA, *et al*. Association of androgen deprivation therapy with cardiovascular death in patients with prostate cancer: a meta-analysis of randomized trials. *JAMA* 2011;306:2359–66.
25 Finkle WD, Greenland S, Ridgeway GK, *et al*. Increased risk of non-fatal myocardial infarction following testosterone therapy prescription in men. *PLoS ONE* 2014;9:e85805.
26 Liverman CT & Blazer DG (eds), *Testosterone and Aging: Clinical Research Directions.Institute of Medicine (US) Committee on Assessing the Need for Clinical Trials of Testosterone Replacement Therapy*. Washington: National Academies Press, 2004.
27 Chen Z, Tao S, Gao Y, *et al*. Genome-wide association study of sex hormones, gonadotropins and sex hormone-binding protein in Chinese men. *J Med Genet* 2013;50:794–801.
28 Vandenput L, Ohlsson C. Genome-wide association studies on serum sex steroid levels. *Mol Cell Endocrinol* 2014;382:758–66.
29 Trigunaite A, Dimo J, Jorgensen TN. Suppressive effects of androgens on the immune system. *Cell Immunol* 2015;294:87–94.
30 Kissick HT, Sanda MG, Dunn LK, *et al*. Androgens alter T-cell immunity by inhibiting T-helper 1 differentiation. *Proc Natl Acad Sci U S A* 2014;111:9887–92.
31 Sakiani S, Olsen NJ, Kovacs WJ. Gonadal steroids and humoral immunity. *Nat Rev Endocrinol* 2013;9:56–62.
32 Olsen NJ, Kovacs WJ. Evidence that androgens modulate human thymic T cell output. *J Investig Med* 2011;59:32–5.
33 Giefing-Kroll C, Berger P, Lepperdinger G, *et al*. How sex and age affect immune responses, susceptibility to infections, and response to vaccination. *Aging Cell* 2015;14:309–21.
34 Hofer MD, Cheng EY, Bury MI, *et al*. Androgen supplementation in rats increases the inflammatory response and prolongs urethral healing. *Urology* 2015;85:691–7.
35 Lai JJ, Lai KP, Chuang KH, *et al*. Monocyte/macrophage androgen receptor suppresses cutaneous wound healing in mice by enhancing local TNF-alpha expression. *J Clin Invest* 2009;119:3739–51.
36 Bernin H, Lotter H. Sex bias in the outcome of human tropical infectious diseases: influence of steroid hormones. *J Infect Dis* 2014;209(Suppl 3): S107–13.
37 Guerra-Silveira F, Abad-Franch F. Sex bias in infectious disease epidemiology: patterns and processes. *PLoS ONE* 2013;8:e62390.
38 Cutolo M, Balleari E, Giusti M, *et al*. Androgen replacement therapy in male patients with rheumatoid arthritis. *Arthritis Rheum* 1991;34: 1–5.
39 Booji A, Biewenga-Booji CM, Huber-Bruning O, *et al*. Androgens as adjuvant treatment in postmenopausal female patients with rheumatoid arthritis. *Ann Rheum Dis* 1996;55:811–15.
40 Chang C, Lee SO, Yeh S, *et al*. Androgen receptor (AR) differential roles in hormone-related tumors including prostate, bladder, kidney, lung, breast and liver. *Oncogene* 2014;33:3225–34.
41 Zhang Y. Understanding the gender disparity in bladder cancer risk: the impact of sex hormones and liver on bladder susceptibility to carcinogens. *J Environ Sci Health C Environ Carcinog Ecotoxicol Rev* 2013;31:287–304.
42 He D, Li L, Zhu G, *et al*. ASC-J9 suppresses renal cell carcinoma progression by targeting an androgen receptor-dependent HIF2alpha/VEGF signaling pathway. *Cancer Res* 2014;74:4420–30.
43 Kalra M, Mayes J, Assefa S, *et al*. Role of sex steroid receptors in pathobiology of hepatocellular carcinoma. *World J Gastroenterol* 2008;14: 5945–61.
44 Harlos C, Musto G, Lambert P, *et al*. Androgen pathway manipulation and survival in patients with lung cancer. *Horm Cancer* 2015;6:120–7.
45 Sukocheva OA, Li B, Due SL, *et al*. Androgens and esophageal cancer: What do we know? *World J Gastroenterol* 2015;21:6146–56.
46 Amos-Landgraf JM, Heijmans J, Wielenga MC, *et al*. Sex disparity in colonic adenomagenesis involves promotion by male hormones, not protection by female hormones. *Proc Natl Acad Sci U S A* 2014;111:16514–19.
47 Swirski FK, Nahrendorf M. Leukocyte behavior in atherosclerosis, myocardial infarction, and heart failure. *Science* 2013;339:161–6.
48 Tabas I, Glass CK. Anti-inflammatory therapy in chronic disease: challenges and opportunities. *Science* 2013;339:166–72.
49 Nikiforov SV, Mamaev VB. The development of sex differences in cardiovascular disease mortality: a historical perspective. *Am J Public Health* 1998;88:1348–53.
50 Bhasin S, Pencina M, Jasuja GK, *et al*. Reference ranges for testosterone in men generated using liquid chromatography tandem mass spectrometry in a community-based sample of healthy nonobese young men in the Framingham Heart Study and applied to three geographically distinct cohorts. *J Clin Endocrinol Metab* 2011;96:2430–9.
51 Barker-Collo S, Bennett DA, Krishnamurthi RV, *et al*. Sex differences in stroke incidence, prevalence, mortality and disability-adjusted life years: results from the Global Burden of Disease Study 2013. *Neuroepidemiology* 2015;45:203–14.
52 Ajayi AA, Mathur R, Halushka PV. Testosterone increases human platelet thromboxane A2 receptor density and aggregation responses. *Circulation* 1995;91:2742–7.
53 Ajayi AA, Halushka PV. Castration reduces platelet thromboxane A2 receptor density and aggregability. *QJM* 2005;98:349–56.
54 Stone NJ, Robinson J, Lichtenstein AH, *et al*., American College of Cardiology/American Heart Association Task Force on Practice Guidelines. 2013 ACC/AHA Guideline on the treatment of blood cholesterol to reduce atherosclerotic cardiovascular risk in adults: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. *J Am Coll Cardiol* 2014;63(25 Pt B):2889–934.
55 Tunstall-Pedoe H. Myth and paradox of coronary risk and the menopause. *Lancet* 1998;351:1425–7.
56 [No authors listed]. The Coronary Drug Project. Findings leading to discontinuation of the 2.5-mg day estrogen group. The coronary Drug Project Research Group. *JAMA* 1973;226:652–7.
57 FDA adding general warning to testosterone products about potential for venous blood clots. 2014. http://www.fda.gov/Drugs/DrugSafety/ucm401746.htm
58 Health Canada. Information Update—Possible cardiovascular problems associated with testosterone products. 2014. http://healthycanadians.gc.ca/recall-alert-rappel-avis/hc-sc/2014/40587a-eng.php
59 European Medicines Agency. No consistent evidence of an increased risk of heart problems with testosterone medicines. 2014. http://www.ema.europa.eu/ema/index jsp?curl=pages/news_and_events/news/2014/11/news_detail_002218.jsp&mid=WC0b01ac058004d5c1
60 FDA. FDA Drug Safety Communication: FDA cautions about using testosterone products for low testosterone due to aging; requires labeling change to inform of possible increased risk of heart attack and stroke with use. 2015. http://www.fda.gov/Drugs/DrugSafety/ucm436259.htm
61 Safety Concerns Slow Sales of Testosterone Therapy. 2014. http://www.bloomberg.com/bw/articles/2014-11-06/safety-concerns-hurt-sales-of-testosterone-replacement-therapy
62 What's Next For The Thousands Of Angry Men Suing Over Testosterone? 2015. http://www.forbes.com/sites/arleneweintraub/2015/04/06/whats-next-for-the-thousands-of-angry-men-suing-over-testosterone/
63 Fernandez-Balsells MM, Murad MH, Lane M, *et al*. Clinical review 1: adverse effects of testosterone therapy in adult men: a systematic review and meta-analysis. *J Clin Endocrinol Metab* 2010;95:2560–75.
64 Schooling CM, Zhao J. Higher adiponectin and lower hemoglobin levels in older men: causal or confounded by androgens? *J Intern Med* 2015;278: 95–6.
65 Morrison JA, Barton BA, Biro FM, *et al*. Sex hormones and the changes in adolescent male lipids: longitudinal studies in a biracial cohort. *J Pediatr* 2003;142:637–42.
66 Zhao J, Jiang C, Lam TH, *et al*. Genetically predicted testosterone and cardiovascular risk factors in men: a Mendelian randomization analysis in the Guangzhou Biobank Cohort Study. *Int J Epidemiol* 2014;43:140–8.
67 Zhao J, Jiang C, Lam TH, *et al*. Genetically predicted testosterone and electrocardiographic QT interval duration in Chinese: a Mendelian randomization analysis in the Guangzhou Biobank Cohort Study. *Int J Epidemiol* 2015;44:613–20.
68 Calof OM, Singh AB, Lee ML, *et al*. Adverse events associated with testosterone replacement in middle-aged and older men: a meta-analysis of randomized, placebo-controlled trials. *J Gerontol A Biol Sci Med Sci* 2005;60:1451–7.
69 Haddad RM, Kennedy CC, Caples SM, *et al*. Testosterone and cardiovascular risk in men: a systematic review and meta-analysis of randomized placebo-controlled trials. *Mayo Clin Proc* 2007;82:29–39.
70 Xu L, Freeman G, Cowling BJ, *et al*. Testosterone and cardiovascular events among men: a systematic review and meta-analysis of placebo-controlled randomized trials. *BMC Med* 2013;11:108.
71 Corona G, Maseroli E, Rastrelli G, *et al*. Cardiovascular risk associated with testosterone-boosting medications: a systematic review and meta-analysis. *Expert Opin Drug Saf* 2014;13: 1327–51.
72 Basaria S, Harman SM, Travison TG, *et al*. Effects of testosterone administration for 3 years on subclinical atherosclerosis progression in older men with low or low-normal testosterone levels: a randomized clinical trial. *JAMA* 2015;314:570–81.
73 Eyben FE, Graugaard C, Vaeth M. All-cause mortality and mortality of myocardial infarction for 989 legally castrated men. *Eur J Epidemiol* 2005;20: 863–9.
74 Swerdlow AJ, Higgins CD, Schoemaker MJ, *et al*. Mortality in patients with Klinefelter syndrome in Britain: a cohort study. *J Clin Endocrinol Metab* 2005;90:6516–22.
75 [No authors listed]. Klinefelter's syndrome and diabetes mellitus. *Lancet* 1966;2:747–8.
76 Gale EA, Gillespie KM. Diabetes and gender. *Diabetologia* 2001;44:3–15.
77 Grossmann M, Hoermann R, Wittert G, *et al*. Effects of testosterone treatment on glucose metabolism and symptoms in men with type 2 diabetes and the metabolic syndrome: a systematic review and meta-analysis of randomized controlled clinical trials. *Clin Endocrinol (Oxf)* 2015;83:344–51.
78 Tong G, Hua X, Zhong Y, *et al*. Intensive insulin therapy increases sex hormone-binding globulin in newly diagnosed type 2 diabetic patients. *Eur J Endocrinol* 2014;170:237–45.

79 Ahmad OS, Morris JA, Mujammami M, *et al*. A Mendelian randomization study of the effect of type-2 diabetes on coronary heart disease. *Nat Commun* 2015;6:7060.
80 Keene D, Price C, Shun-Shin MJ, *et al*. Effect on cardiovascular risk of high density lipoprotein targeted drug treatments niacin, fibrates, and CETP inhibitors: meta-analysis of randomised controlled trials including 117,411 patients. *BMJ* 2014;349:g4379.
81 Ridker PM, Luscher TF. Anti-inflammatory therapies for cardiovascular disease. *Eur Heart J* 2014;35:1782–91.
82 Fordyce CB, Roe MT, Ahmad T, *et al*. Cardiovascular drug development: is it dead or just hibernating? *J Am Coll Cardiol* 2015;65:1567–82.
83 Schulte DM, Hahn M, Oberhauser F, *et al*. Caloric restriction increases serum testosterone concentrations in obese male subjects by two distinct mechanisms. *Horm Metab Res* 2014;46:283–6.
84 Schooling CM, Au Yeung SL, Freeman G, *et al*. The effect of statins on testosterone in men and women, a systematic review and meta-analysis of randomized controlled trials. *BMC Med* 2013;11:57.
85 Stoffer SS, Hynes KM, Jiang NS, *et al*. Digoxin and abnormal serum hormone levels. *JAMA* 1973;225:1643–4.
86 Chevalier N, Fenichel P. Endocrine disruptors: new players in the pathophysiology of type 2 diabetes? *Diabetes Metab* 2015;41:107–15.
87 Cieri RL, Churchill SE, Franciscus RG, *et al*. Craniofacial feminization, social tolerance, and the origins of behavioral modernity. *Curr Anthropol* 2014;55:419–43.
88 Sewani-Rusike CR, Mudambo KS, Tendaupenyu G, *et al*. Effects of the Zimbabwe Defence Forces training programme on body composition and reproductive hormones in male army recruits. *Cent Afr J Med* 2000;46:27–31.
89 Liu W, Ramirez J, Gamazon ER, *et al*. Genetic factors affecting gene transcription and catalytic activity of UDP-glucuronosyltransferases in human liver. *Hum Mol Genet* 2014;23:5558–69.
90 Rowland A, Miners JO, Mackenzie PI. The UDP-glucuronosyltransferases: their role in drug metabolism and detoxification. *Int J Biochem Cell Biol* 2013;45:1121–32.
91 Strassburg CP, Strassburg A, Kneip S, *et al*. Developmental aspects of human hepatic drug glucuronidation in young children and adults. *Gut* 2002;50:259–65.
92 Sten T, Finel M, Ask B, *et al*. Non-steroidal anti-inflammatory drugs interact with testosterone glucuronidation. *Steroids* 2009;74:971–7.
93 Zhang JY, Zhan J, Cook CS, *et al*. Involvement of human UGT2B7 and 2B15 in rofecoxib metabolism. *Drug Metab Dispos* 2003;31:652–8.
94 Yamanaka H, Nakajima M, Katoh M, *et al*. Trans-3′-hydroxycotinine O- and N-glucuronidations in human liver microsomes. *Drug Metab Dispos* 2005;33:23–30.
95 Trelle S, Reichenbach S, Wandel S, *et al*. Cardiovascular safety of non-steroidal anti-inflammatory drugs: network meta-analysis. *BMJ* 2011;342:c7086.
96 Mills EJ, Thorlund K, Eapen S, *et al*. Cardiovascular events associated with smoking cessation pharmacotherapies: a network meta-analysis. *Circulation* 2014;129:28–41.