CARDIOVASCULAR RISKS AND BENEFITS OF TESTOSTERONE THERAPY

SANDEEP DHINDSA, M.D.
Associate Professor of Medicine
Chief, Division of Endocrinology and Metabolism
Texas Tech University Health Sciences Center, Odessa, TX

Components of the talk

- Relation of endogenous testosterone with mortality.
- Effect of testosterone replacement therapy on cardiovascular events and mortality—observational and retrospective studies
- Effect of testosterone replacement therapy on cardiovascular risk factors in randomized controlled trials

TABLE 5. HRs of low total testosterone and low bioavailable testosterone for cause-specific mortality by years of follow-up

Cause of death	0–20 yr follow-up n	0–20 yr follow-up HR (95% CI)	5–20 yr follow-up n	5–20 yr follow-up HR (95% CI)
Low total testosterone[a]				
All-cause	529	1.38 (1.12, 1.69)	409	1.60 (1.27, 2.02)
CVD	264	1.38 (1.02, 1.85)	199	1.73 (1.23, 2.45)
Cancer	127	1.34 (0.89, 2.00)	90	1.22 (0.75, 1.99)
Respiratory disease	54	2.29 (1.25, 4.20)	46	2.67 (1.37, 5.20)
Other	96	1.13 (0.68, 1.88)	83	1.51 (0.89, 2.56)
Low biotestosterone[b]				
All-cause	529	1.44 (1.19, 1.74)	409	1.44 (1.16, 1.80)
CVD	264	1.36 (1.04, 1.79)	199	1.39 (1.01, 1.92)
Cancer	127	1.50 (0.99, 2.26)	90	1.38 (0.82, 2.31)
Respiratory disease	54	1.84 (1.03, 3.28)	46	1.65 (0.86, 3.14)
Other	96	1.43 (0.91, 2.24)	83	1.56 (0.96, 2.53)

[a] Adjusted for age, BMI, waist to hip ratio, alcohol use, current smoking, and exercise.

[b] Reference is total testosterone 241 ng/dl or greater.

[c] Reference is bioavailable testosterone 78 ng/dl or greater.

Low serum testosterone and mortality in older men. *JCEM* Jan 2008. Laughlin GA, Barrett-Connor

Prospective, population-based study of 794 men, aged 50-91 (median 73.6) yr who had serum testosterone measurements at baseline (1984-1987) and were followed for mortality through July 2004; average follow-up 11.8yrs

Low serum testosterone and increased mortality in men with coronary heart disease

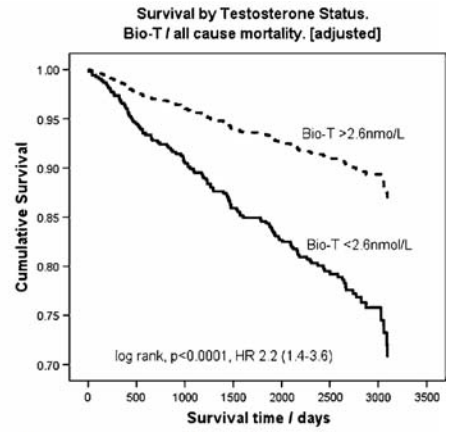
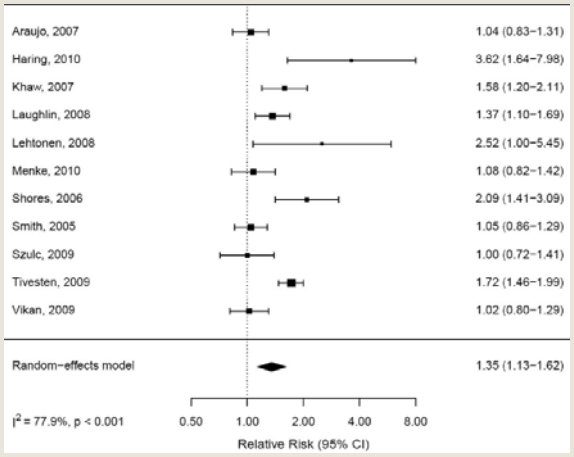


Figure 1 Shows a survival curve of all-cause mortality based on baseline bio-available testosterone (bio-T). The solid line represents patients with baseline bio-T less than 2.6 nmol/l, the broken line represents patients with bio-T greater than 2.6 nmol/l. HR, hazard ratio.

- 930 men with CAD
- mean follow was 7 years
- 21% of men had bio-available T < 2.6 nmol/l

Malkin, Jones, Channer et al; *Heart* 2010;96

Endogenous T and Mortality
Meta-analysis of 11 studies



Not known if low T is a marker or mediator?

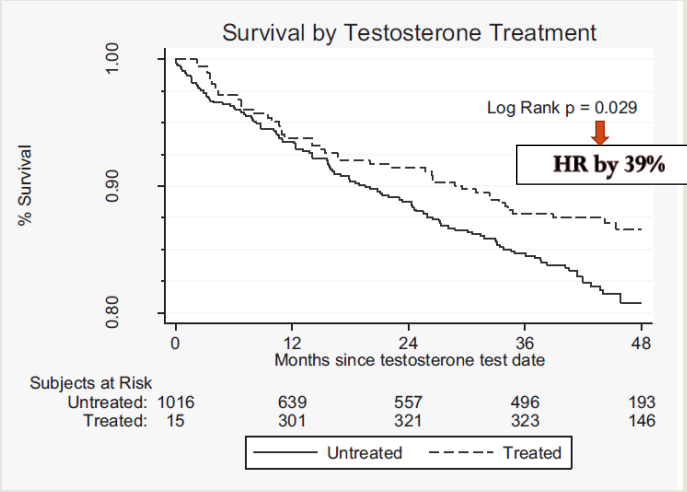
Each *square* shows the study-specific RR estimate comparing the bottom tertile with the top tertile of the testosterone distribution. Araujo; *J Clin Endocrinol Metab*, 2011

Testosterone Treatment and Mortality
Retrospective chart review of VA clinical database

- 1031 men with serum T <250 ng/dl and age >40 years
- 39% of the men (398) initiated testosterone treatment. 89% of men received intramuscular testosterone.
- The mean duration of treatment was 20 months.
- The average follow-up time in the study was 41 months.

	Untreated (633)	Treated (398)	p
Age	63±11	61±10	0.007
BMI	31±6	33±7	<0.001
Baseline total T (ng/dl)	193±54	160±62	<0.001
Medical morbidity	7±4	7±4	0.18
Diabetes	40%	36%	0.44
Coronary artery disease	23%	20%	0.26

Shores et al; J Clin Endocrinol Metab, June 2012, 97(6):2050–2058



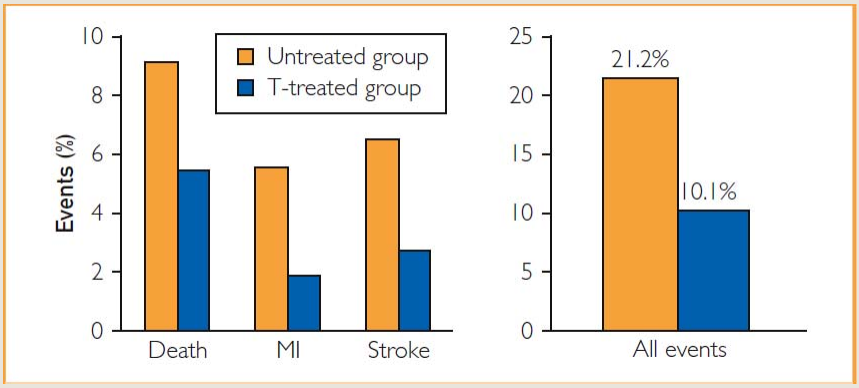
Even after exclusion of men who died in the first year of follow-up, testosterone treatment was associated with decreased mortality (HR 0.47; 95% CI 0.29–0.76, P= 0.003).
Unadjusted mortality rate was 3.4 deaths per 100 person-years in the testosterone-treated men and 5.7 deaths per 100 person-years in the untreated men.

Association of Testosterone Therapy With Mortality, Myocardial Infarction, and Stroke: Retrospective chart review of VA clinical database

- 8709 male veterans who underwent coronary angiography and had a total testosterone < 300 ng/dl.
- Of the 8709 patients, 1223 (14%) initiated testosterone therapy
- The average follow-up was approximately 28 months

	Untreated (7486)	Treated (1223)	p
Age	64±9	61±8	<0.001
Obesity	54%	58%	0.02
Baseline total T (ng/dl)	207±74	176±62	<0.001
Normal Coronaries	12%	16%	<0.001
Diabetes	56%	53%	0.09
Cerebrovascular disease	16%	11%	<0.001

JAMA. 2013;310(17):1829-1836.

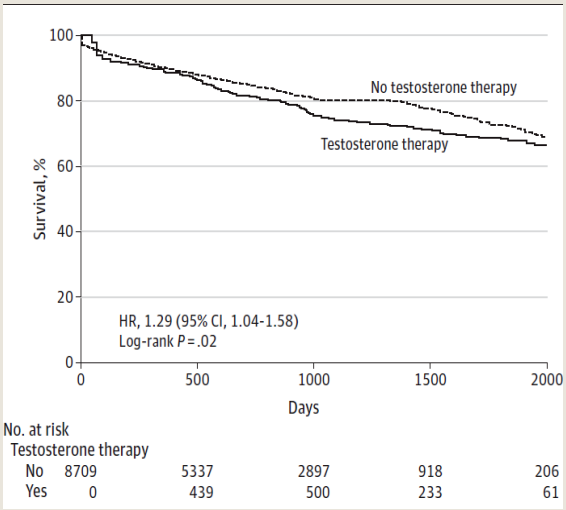


Morgantaler et al; Mayo Clin Proc. February 2015;90(2):224-251

However, the Kaplan-Meier calculation based on statistical adjustment for more than 50 variables converted this into an event rate of 19.9% in the untreated group and 25.7% in the testosterone therapy group at 3 years, thus reversing the results of raw data!

The risk differences were 1.3% (95% CI, −7.1% to 9.7%) at 1 year, 3.1% (95% CI, −4.9% to 11.0%) at 2 years, and 5.8% (95% CI, −1.4% to 13.1%) at 3 years.

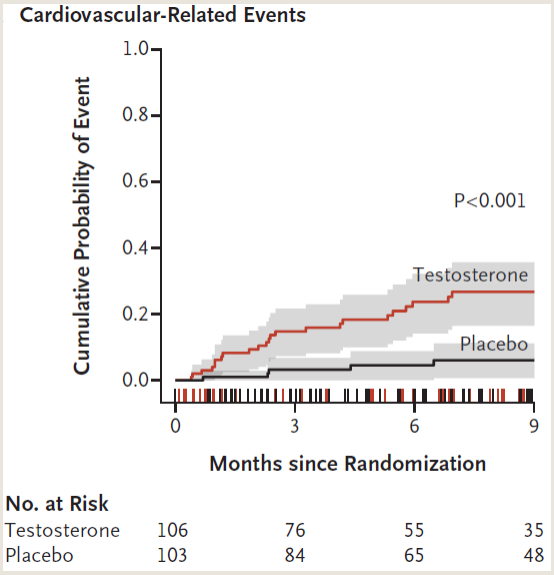
Testosterone use was associated with increased risk of adverse outcomes including all-cause mortality, MI, and ischemic stroke



- The duration of treatment on average was approximately 1 year.
- Only 60% of treated patients had their testosterone concentrations checked while on therapy.
- Mean Testosterone concentration during therapy was 332 ng/dl.
- 36% were on injections and 64% on patches.

Testosterone in Older Men with Mobility Limitations trial; NEJM, 2010

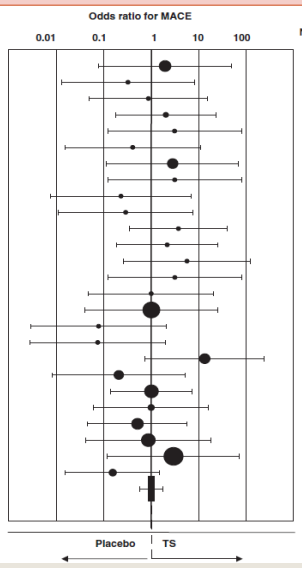
- 209 elderly frail men (mean age was 74 years), with limitations in mobility and
- low total serum testosterone levels
- Randomly assigned a placebo gel (n=103) or testosterone gel (n=106) for 6 months.
- A total of 23 men in the testosterone group and 5 in the placebo group had cardiovascular-related events



- The majority of "events" would not be classified as MACE, such as peripheral edema, hypertension, and tachycardia, as well as non-specific EKG changes.
- Men with testosterone levels in the highest quartile during the intervention period were at elevated risk for cardiovascular-related events (hazard ratio, 2.4; P =0.05).
- 14 subjects achieved T >1000 ng/dl

Meta-analysis of 26 randomized trials that reported major adverse cardiovascular events (MACE)

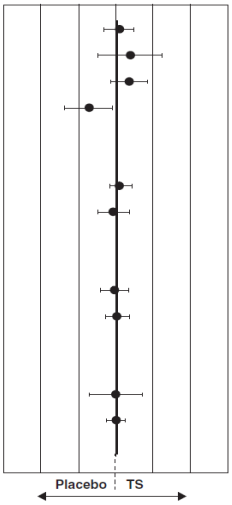
Source	MH-OR	LL	UL	P	TS No. events	TS No. patients	Placebo No. events	Placebo No. patients
Copenhagen SG 1986 [31]	1.97	0.08	48.82	0.68	1	134	0	87
Hall *et al.*, 1996 [34]	0.32	0.01	8.23	0.49	0	35	1	35
Sih *et al.*, 1997 [36]	0.88	0.05	15.33	0.93	1	17	1	15
Snyder *et al.*, 1999 [40]	2.04	0.18	23.17	0.57	2	54	1	54
English *et al.*, 2000 [42]	3.12	0.12	80.39	0.49	1	25	0	25
Seidman *et al.*, 2001 [47]	0.41	0.02	10.83	0.59	0	13	1	17
Steidle *et al.*, 2003 [52]	2.83	0.11	70.27	0.53	1	106	0	99
Amory *et al.*, 2004 [54]	3.13	0.12	80.68	0.49	1	24	0	24
Kenny *et al.*, 2004 [56]	0.23	0.01	7.05	0.40	0	6	1	5
Svartberg *et al.*, 2004 [60]	0.29	0.01	7.74	0.46	0	15	1	14
Brockenbrough *et al.*, 2006 [63]	3.75	0.36	39.59	0.27	3	19	1	21
Malkin *et al.*, 2006 [69]	2.17	0.19	25.01	0.53	2	37	1	39
Nair *et al.*, 2006 [72]	5.70	0.26	123.78	0.27	2	30	0	32
Svartberg *et al.*, 2008 [81]	3.16	0.12	82.64	0.49	1	19	0	19
Chapman *et al.*, 2009 [84]	1.00	0.05	20.83	1.00	1	6	1	6
Legros *et al.*, 2009 [85]	1.01	0.04	25.01	1.00	1	237	0	79
Aversa *et al.*, 2010 [89]	0.08	0.00	2.07	0.13	0	40	1	10
Aversa *et al.*, 2010 [90]	0.07	0.00	1.97	0.12	0	42	1	10
Basaria *et al.*, 2010 [11]	13.39	0.74	240.78	0.08	6	106	0	103
Kalinchenko *et al.*, 2010 [92]	0.21	0.01	5.15	0.34	0	113	1	71
Srinivas-Shankar *et al.*, 2010 [93]	1.01	0.14	7.31	0.99	2	136	2	138
Ho *et al.*, 2011 [95]	1.00	0.06	16.37	1.00	1	60	1	60
Jones *et al.*, 2011 [96]	0.51	0.05	5.57	0.59	1	108	2	112
Kaufman *et al.*, 2011 [97]	0.87	0.04	18.48	0.93	2	234	0	40
Behre *et al.*, 2012 [99]	2.95	0.12	72.91	0.51	1	183	0	179
Hildreth *et al.*, 2013 [100]	0.15	0.02	1.53	0.11	1	96	3	47
Overall	**1.01**	**0.57**	**1.77**	**0.98**	**31**	**1895**	**20**	**1341**



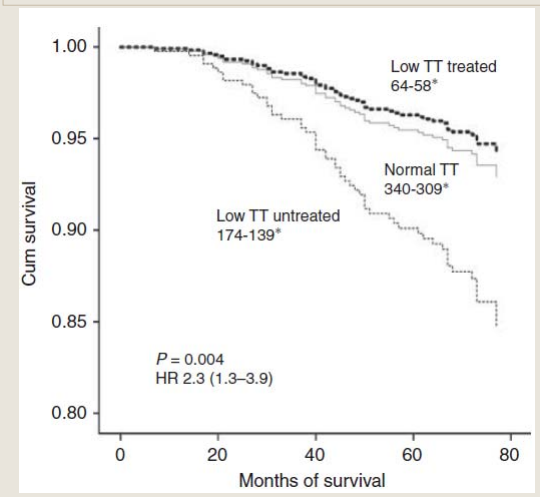
Odds ratio for MACE in subjects treated with testosterone substitution (TS) or placebo. Corona, *Expert Opin. Drug Saf.*,2014

Odds ratio for MACE according to baseline characteristics in subjects treated with testosterone substitution (TS) or placebo. Corona, *Expert Opin. Drug Saf.*,2014

Source	No. Trials	MH-OR	LL	UL	P	TS No. Events	TS No. Patients	Placebo No. Events	Placebo No. Patients
Associated diseases									
Elderly men	10	1.22	0.49	3.03	0.67	13	954	6	549
Men with CVD	2	2.48	0.35	17.45	0.36	3	62	1	64
Frail men	5	2.25	0.72	7.08	0.17	13	401	4	355
Men with metabolic diseases	4	0.19	0.04	0.85	0.03	1	303	5	203
Hypogonadism status									
Mixed population	14	1.26	0.58	2.73	0.56	15	1066	11	865
TT < 12 nM	12	0.84	0.32	2.23	0.73	16	829	9	476
Type of support									
Drug company not supported	12	0.94	0.39	2.24	0.88	10	437	8	332
Drug company supported	14	1.07	0.51	2.24	0.86	21	1458	12	1009
Trial duration									
≤ 12 weeks	4	1.02	0.20	5.29	0.98	2	147	2	145
> 12 weeks	22	1.01	0.55	1.84	0.98	29	1746	18	1196



Testosterone replacement improves survival in men with type 2 diabetes



- Chart review at a diabetes clinic
- Mean age: 60 years
- Mean follow up: 6 years
- Mortality rates:-

20% (untreated low T)

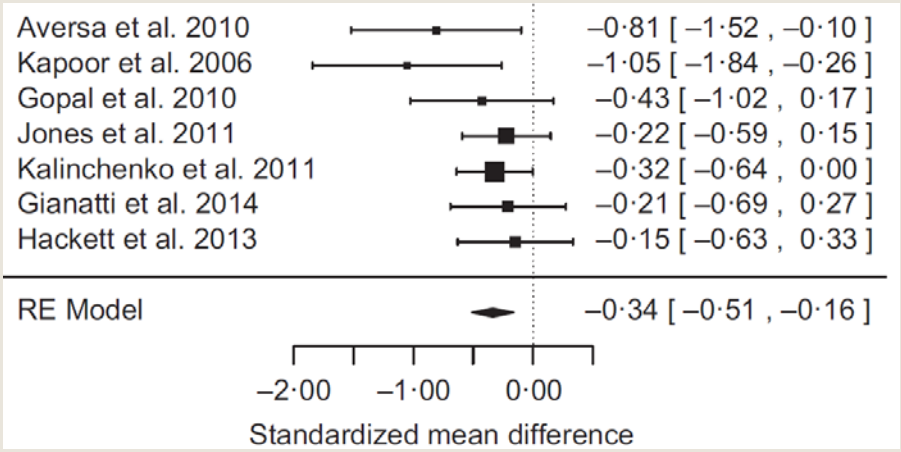
9% (Eugonadal group)

9% (TRT group)

*The number of patients alive at the start of the study and at the end of the study.

Muraleedharan et al; European Journal of Endocrinology (2013)

Meta-analysis of effect of T on HOMA-IR in seven randomized controlled trials in patient with type 2 Diabetes



Grossmann; Clinical Endocrinology (2014)

EFFECT OF TESTOSTERONE REPLACEMENT ON INSULIN SENSITIVITY

- We conducted a randomized placebo controlled trial to evaluate the effect of T replacement on insulin resistance in men with T2D and hypogonadotropic hypogonadism (HH).
- 94 men with T2D were recruited into the study.
- HH was defined as free T concentrations <5 ng/dl with normal or low LH and FSH.
- 44 men had HH and 50 men had normal free T concentrations.
- Men with HH were randomized to receive intramuscular T(250 mg) or placebo(1ml saline) every 2 weeks for 24 weeks.

We hypothesized that…

- T2D men with HH are more insulin resistant than those with normal T;
- T replacement in T2D men with HH reverses insulin resistance.
- T replacement increases lean body mass, decreases fat mass and improves sexual function in T2D men with HH

STUDY HYPOTHESIS

- TESTOSTERONE replacement INCREASES in the expression of mediators of insulin signaling and a REDUCTION in factors that interfere with insulin signaling
- Insulin signaling mediators (IR, IRS-1 and GLUT-4) are decreased in Adipose Tissue in patients with (HH) and Type 2 DM and increase after T replacement
- Since free fatty acids (FFA) and inflammatory mediators impair insulin signaling, we further hypothesized that T replacement decreases 1) serum FFA, CRP and TNF-α concentrations; 2)mRNA expression in mononuclear cells(MNC) of suppressor of cytokine signaling (SOCS)-3, c-Jun N-terminal kinase (JNK)-1, IκB kinase (IKK) β.

STUDY PROCEDURES

- Fasting blood samples, fat biopsies and Hyperinsulinemic euglycemic clamps were done at baseline and at 6 months.
- Insulin sensitivity was calculated from the glucose infusion rate **(GIR)** during the last 30 min of a 4 hour hyperinsulinemic-euglycemic clamp (80 mU/m²/min) and expressed as mg/kg lean body mass/min.
- Body composition: Lean mass and fat mass were measured by **DEXA and MRI**.
- Sexual Function: Subjects were asked to complete a questionnaire daily for 7 consecutive days.
- Total and free testosterone were measured by **liquid chromatography tandem mass spectrometry and equilibrium dialysis**.

Comparison of T2D men with and without HH

	Hypogonadal	Eugonadal	p
N	44	50	
Age (years)	54±8	52±9	0.15
BMI (kg/m^2)	40±8	34±7	<0.001
Total testosterone (ng/dl)	253±84	484±183	<0.001
Free testosterone (ng/dl)	4.3±1.7	7.6±2 .2	<0.001
Calculated Free testosterone (ng/dl)	5.4±1.1	9.5±1.8	<0.001
SHBG (nmol/L)	27±13	36±24	0.02
LH	3.8 [2.3, 5.5]	5.0 [3.6, 6.1]	0.02
FSH	5.3 [3.5, 9.8]	6.9 (4.2, 9.3]	0.38
Testicular size (ml)	15 [14, 20]	20 [15, 23]	0.20

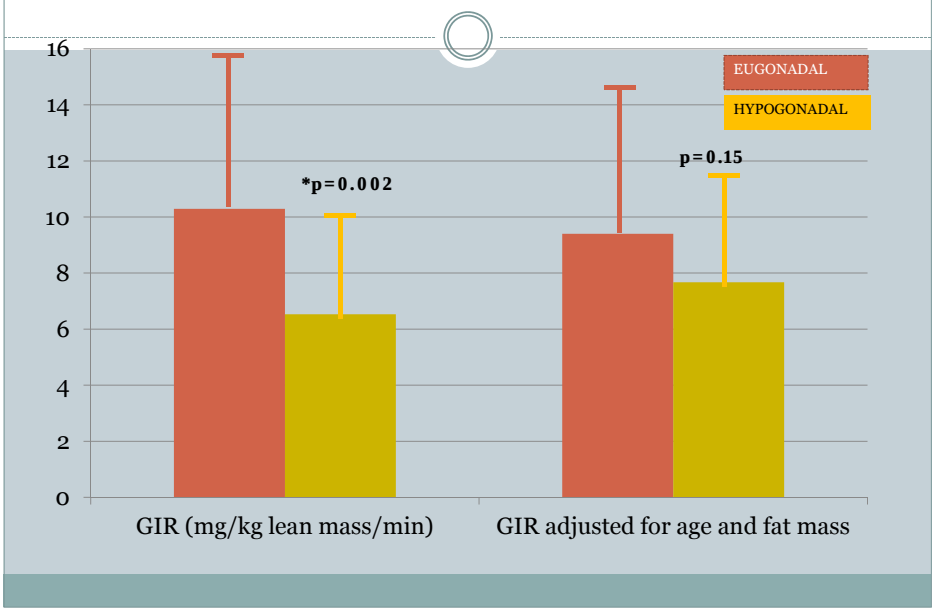
Comparison of T2D men with and without HH

	Hypogonadal	Eugonadal	p
N	44	50	
PSA (ng/dl)	0.6 [0.4, 0.8]	0.6 [0.4, 1.0]	0.85
HbA1c%	7.0±1.1	7.2±1.2	0.51
Total Cholesterol (mg/dl)	158±36	173±51	0.16
HDL (mg/dl)	38±9	45±14	0.03
LDL (mg/dl)	87±30	98±35	0.18
Triglycerides (mg/dl)	124 [100, 212]	113 [80, 195]	0.27
Waist/hip ratio	1.06±0.07	1.01±0.10	0.02
Waist circumference (cm)	125±22	115±18	0.04

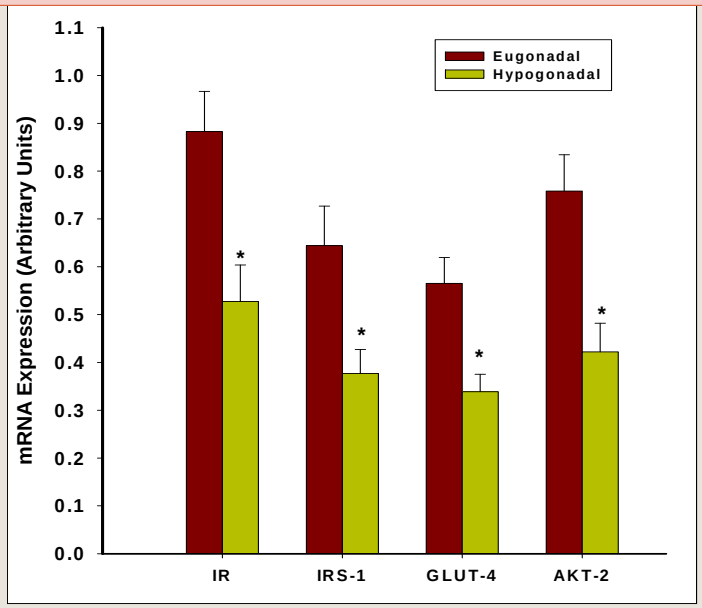
Comparison of T2D men with and without HH

	Hypogonadal	Eugonadal	p
N	44	50	
Total fat mass(kg)	47±14	34±12	<0.001
Trunk fat mass (kg)	28±7	22±8	<0.001
Visceral fat mass (L)	8.6±3.4	5.9±2.5	<0.001
Hepatic fat (%)	5.4±6.2	7.1±7.9	0.29
Total lean mass(kg)	71±11	64±9	0.001
Lean mass % body wt	58±5	62±7	0.004

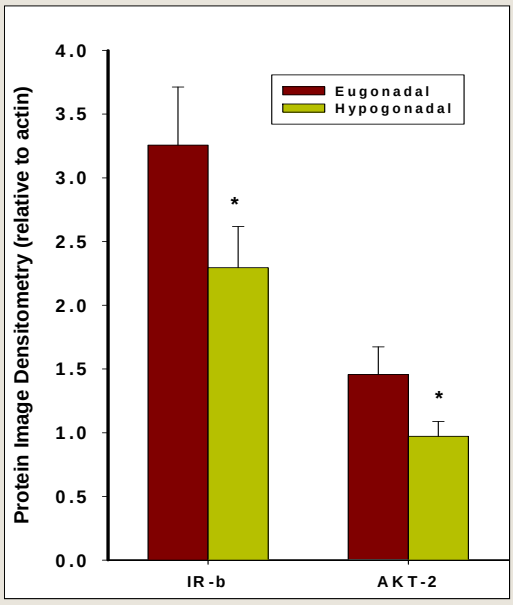
INSULIN RESISTANCE in MEN with HH and T2D

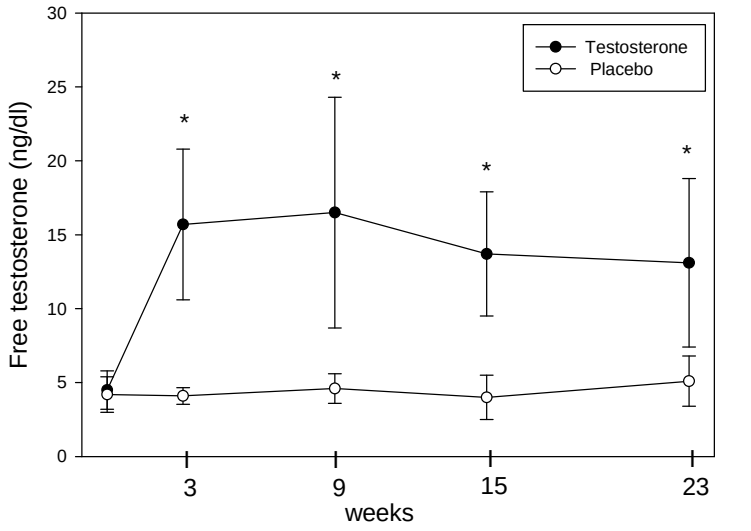


mRNA expression of insulin signaling mediators in adipose tissue



Basal protein levels of IR-b subunit and Akt-2 in adipose tissue





Effect of Testosterone or placebo for 24 weeks in men with T2D and HH

	TESTOSTERONE (n=20)			PLACEBO (n=14)		
	0 week	24 weeks	p	0 week	24 weeks	p
Age (years)	54±7			54.2±8.9		
BMI (kg/m²)	39.0±7.8	38.9±8.3	0.66	40.0±7.8	39.9±8.5	0.91
Total testosterone (ng/dl)	260±87	562±189	<0.001	239±81	294±140	0.10
Free testosterone (ng/dl)	4.5±1.3	13.1±5.7	<0.001	4.2±1.2	4.9±1.8	0.14
Calculated free testosterone (ng/dl)	5.6±1.1	15.5±6.9	<0.001	5.2±1.0	5.7±2.0	0.42
SHBG (nmol/l)	27±14	24±10	0.07	26±13	29±13	1.0
PSA (ng/dl)	0.8±0.7	1.0±0.7	0.02	1.0±1.0	1.1±1.2	0.17
Testicular size (ml)	18±4	14±4	0.01	17±5	18±6	0.71

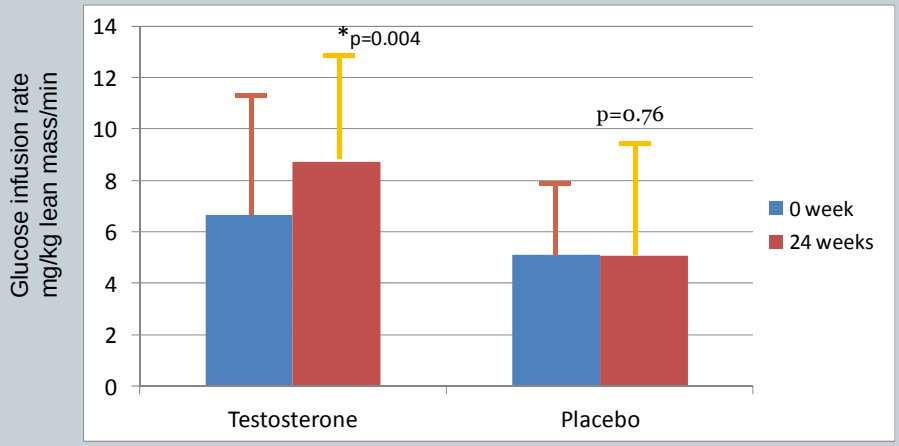
Effect of Testosterone or placebo for 24 weeks in men with T2D and HH

	TESTOSTERONE			PLACEBO		
	0 week	24 weeks	p	0 week	24 weeks	p
Total Cholesterol (mg/dl)	155±38	145±28	0.26	156±38	151±32	0.78
HDL (mg/dl)	35±8	34±9	0.36	39±10	41±12	0.84
LDL (mg/dl)	87±37	78±21	0.20	83±23	77±28	0.84
Triglycerides (mg/dl)	182±122	191±142	0.36	167±96	163±100	0.98
HbA1c%	6.9±0.9	7.2±0.9	0.27	6.7±0.9	7.1±1.6	0.78
Hemoglobin (g/dl)	14.0±0.8	14.8±1.4	0.001	13.7±1.0	14.0±1.1	0.02
Sexual desire (on a scale of 1-7)	2.7±1.8	3.7±1.2	0.03	2.3±1.5	3.0±1.9	1.0
Satisfaction with erection (on a scale of 1-7)	2.5±1.9	3.5±1.7	0.03	2.8±2.4	3.2±1.4	0.23

Effect of Testosterone or placebo for 24 weeks in men with T2D and HH

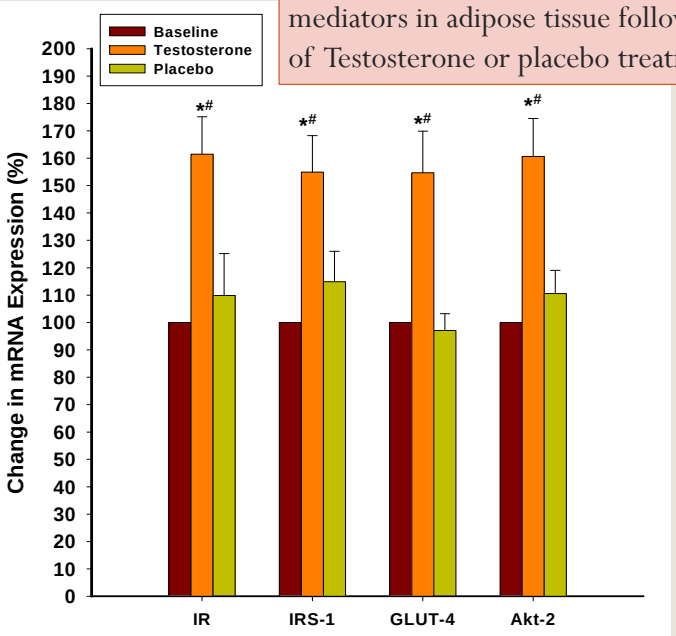
	TESTOSTERONE			PLACEBO		
	0 week	24 weeks	p	0 week	24 weeks	p
Weight (kg)	123±23	123±25	0.63	126±30	126±31	0.70
Waist/hip ratio	1.06±0.08	1.04±0.07	0.21	1.05±0.05	1.00±0.04	0.02
Total lean mass(kg)	70.9±9.0	73.5±10.7	0.001	69.1±13.4	68.3±13.0	0.40
Total fat mass(kg)	45.2±14.0	43.1±12.8	0.02	44.5±15.0	45.4±14.4	0.11
Trunk fat mass (kg)	27.7±8.2	25.9±7.5	0.05	26.7±6.9	27.2±7.9	0.35
Visceral fat mass (L)	7.8±3.3	7.3±2.9	0.40	7.3±2.3	7.4±3.3	0.69
Hepatic fat %	6.2±6.4	4.4±3.8	0.22	3.9±6.1	3.6±4.2	0.56
Spine BMD (g/cm^2)	1.26±0.18	1.28±0.16	0.35	1.19±0.19	1.27±0.23	0.07
Hip BMD (g/cm^2)	1.12±0.18	1.12±0.18	0.61	1.06±0.08	1.06±0.09	0.89

Effect of Testosterone on insulin resistance in men with Type 2 Diabetes and Hypogonadotropic Hypogonadism

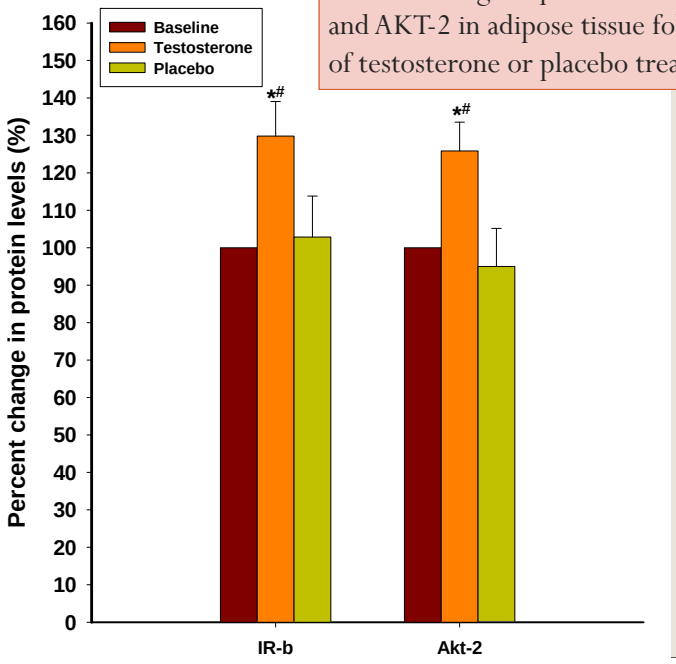


Increase in insulin sensitivity by 30%

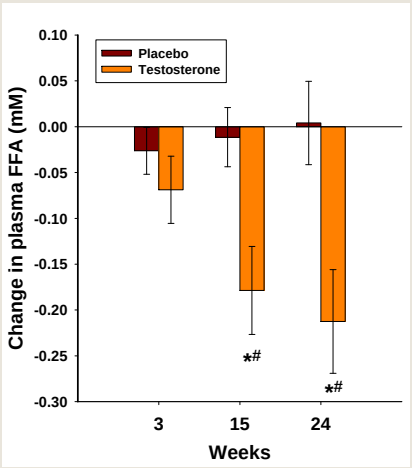
Change in mRNA expression of insulin signaling mediators in adipose tissue following 24 weeks of Testosterone or placebo treatment

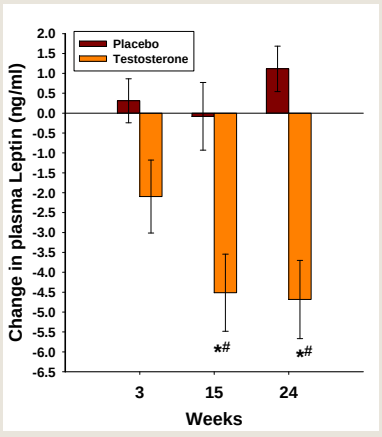


Percent change in protein levels of IR-b subunit and AKT-2 in adipose tissue following 24 weeks of testosterone or placebo treatment.

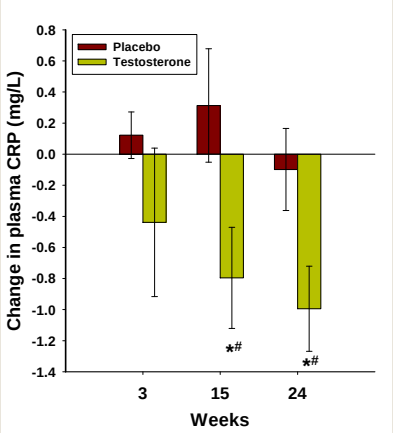


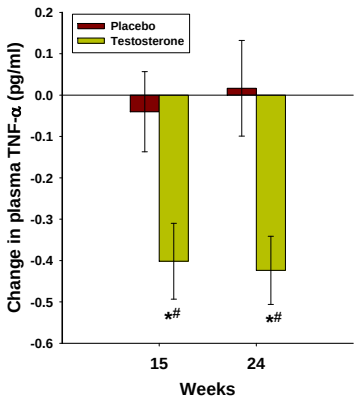
Change in serum FFA and Leptin after 6 months of testosterone or placebo

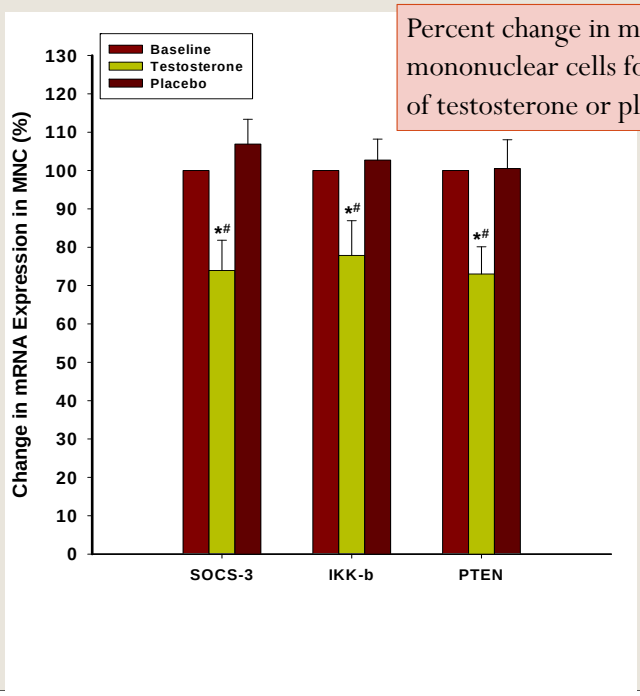




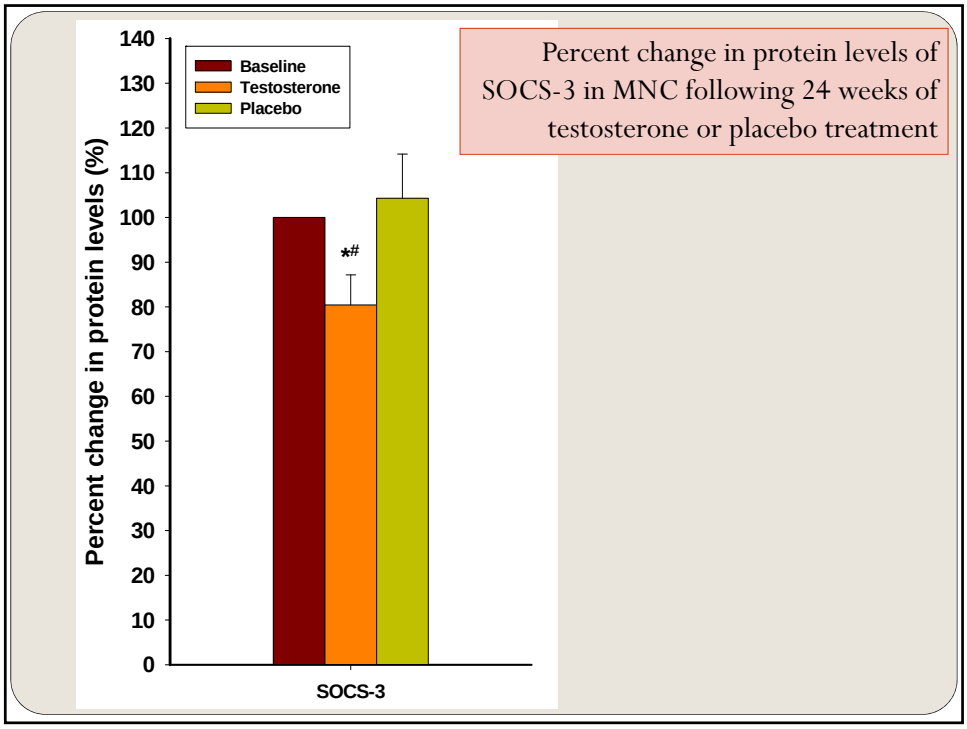
Change in serum CRP and TNF-α after 6 months of testosterone or placebo



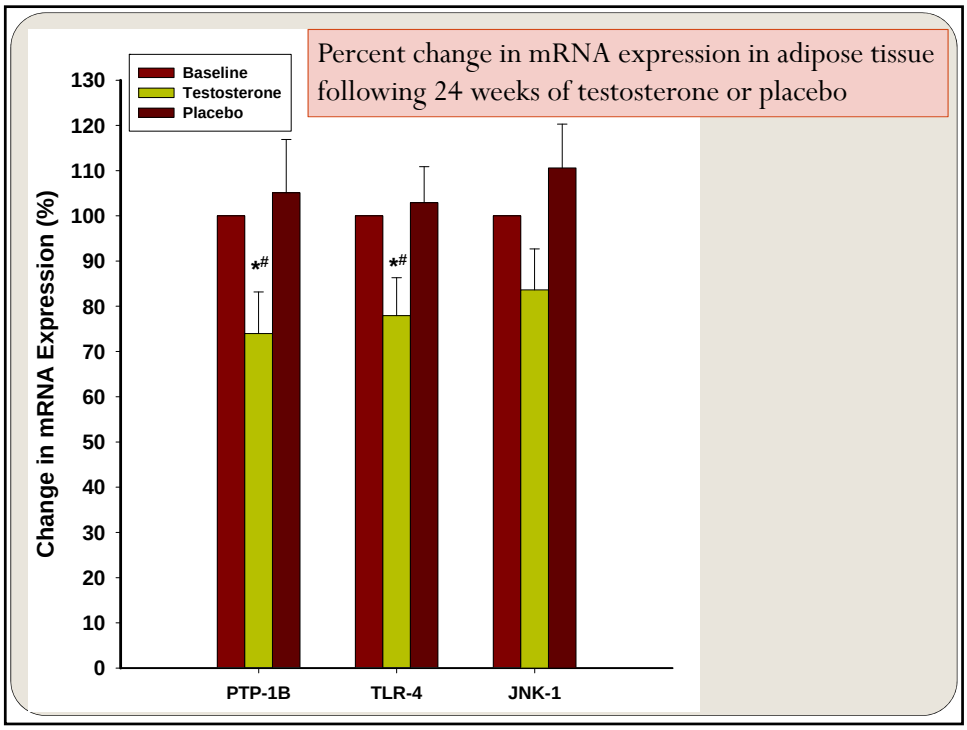




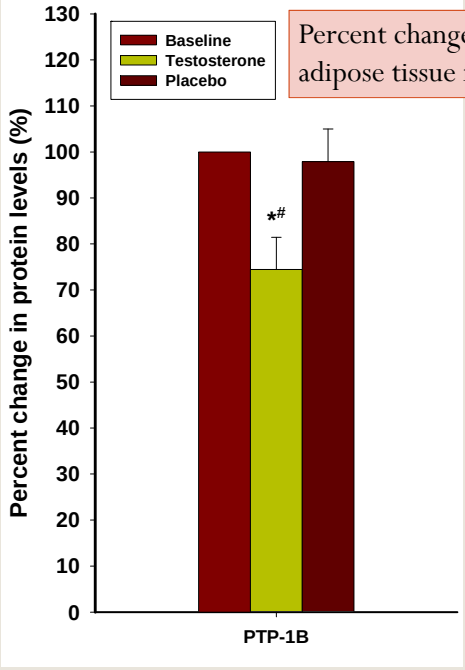
Percent change in mRNA expression in mononuclear cells following 24 weeks of testosterone or placebo treatment.



Percent change in protein levels of SOCS-3 in MNC following 24 weeks of testosterone or placebo treatment



Percent change in mRNA expression in adipose tissue following 24 weeks of testosterone or placebo



Percent change in protein levels of PTP-1-B in adipose tissue following 24 weeks of T or placebo

CONCLUSIONS

Our data show for the first time that

1) men with T2D and HH are more insulin resistant than those with normal T based on HE clamps; Insulin sensitivity is restored following T administration.
2) HH is associated lower expression of mediators of insulin signaling in adipose tissue as compared with eugonadal patients
3) Following Testosterone replacement the expression of mediators of insulin signaling increases and there is reduction in expression of markers interfering with insulin signaling.

- Epidemiological studies suggest that low T may increase all-cause and cardiovascular mortality.
- In fact, no study has suggested that low endogenous testosterone may decrease mortality.
- However, low testosterone concentration could be marker of illness and not causal factor.
- Effect of testosterone replacement therapy on cardiovascular events or mortality has not been studied in randomized controlled trials.
- Retrospective, observational or underpowered randomized trials have shown, increased, decreased or no change in MACE or mortality after testosterone replacement.
- Testosterone therapy decreases cardiovascular risk factors such as fat mass, insulin resistance and inflammatory mediators