BMJ Open

Habitual alcohol consumption associated with reduced semen quality and changes in reproductive hormones; a cross-sectional study among 1221 young Danish men

Tina Kold Jensen,[1,2] Mads Gottschau,[1] Jens Otto Broby Madsen,[1] Anne-Maria Andersson,[2] Tina Harmer Lassen,[2] Niels E Skakkebæk,[2] Shanna H Swan,[3] Lærke Priskorn,[1] Anders Juul,[2] Niels Jørgensen[2]

To cite: Jensen TK, Gottschau M, Madsen JOB, *et al*. Habitual alcohol consumption associated with reduced semen quality and changes in reproductive hormones; a cross-sectional study among 1221 young Danish men. *BMJ Open* 2014;**4**:e005462. doi:10.1136/bmjopen-2014-005462

▸ Prepublication history and additional material is available. To view please visit the journal (http://dx.doi.org/10.1136/bmjopen-2014-005462).

Received 11 April 2014
Revised 24 June 2014
Accepted 26 June 2014

For numbered affiliations see end of article.

Correspondence to
Professor Tina Kold Jensen; tkjensen@health.sdu.dk

ABSTRACT

Objective: Study associations between three measures of alcohol consumption (recent, typical/habitual, binging), semen quality and serum reproductive hormones.

Design: Cross-sectional population based study.

Setting and participants: 1221 young Danish men, aged 18–28 years were recruited when they attended a compulsory medical examination to determine their fitness for military service from 2008 to 2012. Total alcohol consumption: (1) in the week preceding (habitual/typical) the visit (recent alcohol intake),
(2) in a typical week and (3) frequency of 'binge drinking' (consuming more than 5 units/day)) in the past 30 days was estimated.

Main outcome measures: Semen quality (volume, sperm concentration, total sperm count, and percentages of motile and morphologically normal spermatozoa) and serum concentration of reproductive hormones (follicle-stimulating hormone, luteinising hormone, testosterone, sex hormone binding globulin, oestradiol, free testosterone and inhibin B).

Results: Sperm concentration, total sperm count and percentage of spermatozoa with normal morphology were negatively associated with increasing habitual alcohol intake. This association was observed in men reporting at least 5 units in a typical week but was most pronounced for men with a typical intake of more than 25 units/week. Men with a typical weekly intake above 40 units had a 33% (95% CI 11% to 59%) reduction in sperm concentration compared to men with an intake of 1–5 units/week. A significant increase in serum free testosterone with increasing alcohol consumption the week preceding the visit was found. Binging was not independently associated with semen quality.

Conclusions: Our study suggests that even modest habitual alcohol consumption of more than 5 units per week had adverse effects on semen quality although most pronounced associations were seen in men who consumed more than 25 units per week. Alcohol consumption was also linked to changes in testosterone and SHBG levels. Young men should be advised to avoid habitual alcohol intake.

Strengths and limitations of this study

- Our study was large and consisted of young healthy men, of whom the majority had no knowledge of their fertility. It is therefore unlikely to have affected their motivation to participate.
- Our study was cross-sectional and reverse causation cannot be excluded, whereby men with poor semen quality have an unhealthier lifestyle and health behaviour and drink more alcohol even though we adjusted for these factors.
- The men in our study reported daily alcohol consumption the week preceding the visit, as we assumed that to be more accurate to recall than an average intake. This consumption may differ from the typical weekly intake, which can lead to misclassification of exposure.

INTRODUCTION

Alcohol consumption is widespread in the Western world, especially in Europe.[1] Drinking patterns have changed over time and binging (defined here as 5 units or more in a single day) is widespread among young Europeans.[2] Moderate alcohol consumption has been associated with reduced morbidity and mortality although not confirmed in all studies.[3] However, excessive alcohol intake has a negative impact on health (eg, coronary heart disease, stroke and liver disease.[4 5])

Some studies found an association between alcohol intake and semen quality,[6–9] although others did not confirm these findings.[10–18] However, it is difficult to compare across studies, since populations as well as alcohol intake vary considerably between them. In addition, most studies only addressed average alcohol intake by use of only few questions, and within response categories consumption may vary considerably and is likely to be under-reported. Only one study addressed the dose–response relationship between recent alcohol intake (during the past 5 days) and semen quality among 347 young Danish men. Poorer semen quality was found at higher levels of alcohol intake, although not statistically significant.[16] In an earlier multicenter study of over 8000 American and European men, we found no adverse effects of alcohol intake in the week preceding the visit on semen quality. However, in that study most men reported only moderate intake of alcohol.[19] While some men in that study were similar to the men in this study, much less detailed information about drinking habits was collected prior to 2008. To the best of our knowledge no studies have examined the effect of binging on male reproductive parameters nor have the effects of recent versus habitual alcohol intake been studied in healthy populations. We therefore investigated the association between semen quality and serum reproductive hormones and, alcohol consumption during the week preceding the visit in a typical week, and binging in a cross-sectional study of 1221 young Danish men recruited between 2008 and 2012.

MATERIALS AND METHODS

Population

Because of the military draft in Denmark, all 18-year-old men, except those suffering from severe chronic disease, are required to undergo a compulsory physical examination to determine their fitness for military service. Since 1996, trained staffs from the Department of Growth and Reproduction at Copenhagen University Hospital (Rigshospitalet, Copenhagen, Denmark) have approached the draftees when they have appeared for their compulsory physical examination and have invited them to participate in a study of semen quality taking place at Rigshospitalet. Only men recruited from January 2008 to April 2012 were included in the present study, since the questionnaire they completed included detailed information about alcohol intake. All participants completed a questionnaire, delivered a semen sample, had a blood sample drawn and underwent a physical examination. They received compensation for their time (DKK 500, equal to approximately US$85). Participants did not differ from non-participants with regard to age, but they were generally better educated than non-participants (data not shown). A detailed description,[20] and other aspects of the study have previously been published.[21–24]

Semen analysis

All men provided a semen sample by masturbation in a room close to the semen laboratory. The period of ejaculation abstinence (time since last ejaculation) was recorded, and the semen sample was analysed for volume, sperm concentration, total sperm count, per cent motile spermatozoa and per cent morphologically normal spermatozoa as described by Jørgensen *et al*,[20] which is in accordance with the most recent guideline from the WHO.[25] Since 1996, our laboratory has led a quality control programme for assessment of sperm concentration; the laboratory has kept the interlaboratory difference unchanged,[26] and the variation between technicians was less than 10%. The same two experienced technicians assessed the sperm morphology according to strict criteria for the first 904 men.[27]

Serum samples

Serum levels of follicle-stimulating hormone (FSH), luteinising hormone (LH) and sex hormone-binding globulin (SHBG) were determined using a time-resolved immunofluorometric assay (Delfia; Wallac Oy, Turku, Finland). Testosterone and oestradiol levels were determined using time-resolved fluoroimmunoassays (Delfia; Wallac Oy). Inhibin B level was determined by means of a specific two-sided enzyme immunometric assay (Inhibin B Gen II; Beckman Coulter Ltd, High Wycombe, UK). The hormones were all measured within the same time period and in the same assay batches. Free testosterone was calculated on the basis of the measured serum concentrations of total testosterone and SHBG using the method of Vermeulen *et al*[28] and a fixed albumin concentration of 43.8 g/L.[28]

Physical examination

Physicians assessed genital development, the possible presence of a varicocele (grades 1–3) or hydrocele, and the location of the testes in the scrotum, and the consistency of the testis and epididymis were recorded. Weight and height was measured, and body mass index (BMI) was calculated as weight in kilograms divided by squared height in metres.

Questionnaire

Prior to the examination, all participants completed a questionnaire that collected information on previous and/or current diseases and genital diseases. Self-reported diseases of the reproductive organs affecting semen quality (torsion of testes, epididymitis or inguinal hernia) were summarised in two variables: 'self-reported genital conditions' and 'sexually transmitted diseases' (gonorrhea or chlamydia).

The mothers of the young men responded to questions about education, which was coded as: less than 9, 9–10 and more than 10 years of schooling. Data on physical activity were converted to watts per week using the method of Craig *et al*.[29] Men were asked about current smoking habits and whether they were exposed to

smoking in utero. Daily caffeine intake was estimated based on their reported intake of caffeine-containing beverages the week prior to the visit. Men completed a diary reporting their daily intake of red and white wine, beer, strong alcoholic drinks, alcopops and others during the week prior to participation and delivery of the semen and blood samples (recent intake). Men were told that one beer, one glass of wine or 40 mL of spirits contained 1 unit of alcohol (≈12 g of ethanol), whereas one strong beer or one alcopop contained 1.5 units of alcohol and one bottle of wine contained 6 units of alcohol and were asked to convert their intake to units. Alcohol intake was calculated as the sum of daily reported unit intakes within that week. In addition, the men were asked whether their alcohol intake in the week preceding the visit represented a typical week (typical/habitual intake). They were also asked how many times during the past 30 days they had been drunk or had consumed more than 5 units of alcohol on one occasion, which we defined as binging.

Statistics

Exposure variables were total number of alcohol units in the week preceding the visit (recent intake) and in a typical week (typical/habitual intake). Alcohol units were divided into 5 unit intervals. Because abstainers may differ from light-moderate drinkers we selected 1–5 drinks/week as the reference category. In addition, number of binge episodes and number of times being drunk during the past 30 days were categorised as; 0 (reference), 1–2, 3–5, 6–9 and more than 9.

Sperm parameters and reproductive hormone 0 levels were compared in relation to alcohol intake and binging the distributions of the relevant covariates from the questionnaires and physical examinations among men with different alcohol intake were compared by χ^2 test in order to identify potential confounders. Finally, data were analysed using multivariable linear regression models. Because of the non-normal (skewed) distributions of semen quality and serum reproductive hormones, semen parameters were transformed by cubic root and reproductive hormones by natural logarithmic scale and the latter back-transformed to obtain the expected percent change per unit increase in exposure. Covariates were then excluded stepwise if their exclusion did not change effect estimate by more than 10%. In final models, the same set of covariates was used for all semen parameters: period of abstinence, current smoking and BMI, except that period of abstinence was not included for sperm morphology and motility models and duration between the time of ejaculation and analysis of the sample was included only for models predicting sperm motility. Models predicting reproductive hormones were adjusted for time of blood sampling, current smoking and BMI. We initially adjusted alcohol intake for binge episodes, but as estimates were unchanged, binging was not included. Tests for linear trend were performed after excluding men with no alcohol intake. Finally, analyses were performed separately for beer adjusting for total alcohol intake, since beer was consumed by most men. We evaluated the fit of the regression models by testing the residuals for normality and by inspecting the residual plots. SPSS statistics V.19 was used and the results are presented with 95% CIs.

RESULTS

A total of 1221 men were included with a mean age of 19.1 years. The median alcohol intake the week preceding the visit was 11 units (25 and 75 centiles 1–21 units) and 64% and 59% of men had binged or had been drunk more than twice during the past 30 days, respectively. Beer was the favourite alcoholic beverage and the median beer intake the week preceding the visit was 5 units (alcohol intake 0–13 units). A total of 553 men (45%) reported that the week preceding the visit represented a typical week and these were used in the analyses of typical/habitual alcohol intake. These men did not differ from the total population (N=1221) in semen or hormone parameters.

Semen quality decreased with increasing recent alcohol intake (data not shown) and binging (table 1). Testosterone and calculated free testosterone (cFT) increased and SHBG decreased with increasing recent alcohol intake (table 2) and binging already from an intake above 5 units/week. Men with an intake of 30 units in a typical week or binging were more often smokers, had a higher caffeine intake, more often reported having had STDs or fever, were younger and their mothers had a higher education (see online supplementary table S1).

No clear association between recent alcohol intake (the week preceding the visit; data not shown), binging (table 3) and semen quality was found after controlling for confounders. A dose–response association with recent alcohol intake from 1 unit/week (abstainers excluded) and higher testosterone (p trend=0.01) and cFT (p trend<0.01) and lower SHBG (p trend<0.01) was found (table 3, figure 1) after control for confounder. Similar associations were found with number of binge episodes and being drunk during the past 30 days (table 3). Men with a weekly alcohol intake above 40 units the week preceding the visit had 20% (95% CI 9% to 31%) higher cFT after control for confounders. No association with LH, FSH, inhibin B and oestradiol was found (data not shown).

Among the 553 men with a habitual alcohol intake (alcohol intake the week preceding the visit represented a typical week) we found an inverse dose–response association between alcohol intake and sperm concentration (p trend=0.02), total sperm count (p trend=0.01) and percentage morphologically normal sperms (p trend=0.01) (table 3, figure 2) after adjustment. The trend was more pronounced among men with a typical weekly alcohol intake above 25 units. Cubic root

Table 1 Semen quality according to typical (last week represented a typical week) alcohol intake and binging during the past 30 days among respectively 553 and 1221 healthy, young Danish men

		Semen volume (mL)		Sperm concentration (million/mL)		Total sperm count (million)		Motility (%)		Morphology* (%)	
Alcohol intake	**N**	**M**	**25–75**	**M**	**25–75**	**M**	**25–75**	**M**	**25–75**	**M**	**25–75**
Units in a typical week, N=553											
0	122	3.3	2.6; 4.4	39	19; 68	132	62; 249	55	46; 67	7.8	3.9; 12.0
1–5	93	3.3	2.3; 4.2	50	28; 87	175	82; 291	58	48; 68	7.8	4.5; 12.0
6–10	72	3.2	2.3; 4.1	50	23; 89	187	64; 302	58	47; 67	5.8	3.0; 11.1
11–15	82	3.2	2.6; 4.1	41	21; 73	152	70; 260	55	46; 69	7.0	4.1; 11.6
16–20	64	3.0	2.2; 4.1	51	32; 83	163	81; 254	58	48; 69	7.5	5.0; 11.0
21–25	47	2.9	2.1; 4.0	50	30; 97	141	98; 262	60	50; 71	7.0	5.9; 8.8
26–30	27	3.4	2.2; 4.8	34	13; 81	147	18; 285	53	39; 66	8.0	4.5; 10.5
31–35	14	3.0	1.7; 4.7	43	18; 59	96	41; 176	55	46; 61	6.3	4.0; 8.8
36–40	11	3.6	2.2; 4.0	41	13; 77	108	57; 158	67	52; 79	8.0	2.3; 11.0
>40	21	3.6	2.6; 4.7	33	12; 70	86	48; 245	62	50; 71	6.5	2.5; 11.0
Number of binge episodes during the past 30 days† N=1221											
0	176	3.2	2.3; 4.2	49	23; 81	145	67; 257	57	46; 68	7.5	3.5; 11.1
1–2	255	3.3	2.4; 4.5	47	24; 80	153	76; 282	59	47; 68	7.5	4.0; 11.5
3–5	425	3.3	2.4; 4.4	48	22; 88	164	69; 300	58	47; 68	7.3	4.0; 11.5
6–9	258	3.3	2.3; 4.2	43	22; 77	137	69; 259	57	47; 69	6.5	4.0; 9.5
>9	92	3.2	2.3; 4.2	41	20; 73	130	61; 239	56	44; 70	6.0	3.5; 9.1

Presented as median (M) and 25 and 75 centiles (25–75).
*Counted for 904 men of whom 397 stated that last weeks alcohol intake represented a typical week.
†Binging defined as alcohol intake of more than 5 units on one occasion.

transformed sperm concentration and percentage morphologically normal spermatozoa were, respectively, 0.39 (95% CI −0.92 to 0.14) and 0.51 (95% CI 1.03 to 0.01) lower among men with a typical alcohol intake of more than 40 units compared to men with an intake of 1–5 units in a typical week. No alcohol intake was also associated with reduced semen quality. Percentages of motile spermatozoa and semen volume were not

Table 2 Reproductive hormones according to recent (the week preceding the visit) alcohol intake and binging during the past 30 days among 1194 healthy, young Danish men

		FSH (IU/L)		LH (IU/L)		Testosterone (nmol/L)		SHBG (nmol/L)		Free testosterone pmol/L		Inhibin B (pg/mL)		Oestradiol (nmol/L)	
Alcohol intake	**N**	**M**	**5–95**	**M**	**5–95**	**M**	**5–95**	**M**	**5–95**	**M**	**5–95**	**M**	**5–95**	**M**	**5–95**
Units the week preceding the visit, N=1194															
0	243	2.4	0.9; 5.2	3.4	1.6; 6.8	19.1	10.2; 35.4	29	13; 52	439	249; 749	166	81; 271	81	48; 128
1–5	198	2.4	1.0; 5.8	3.4	1.4; 6.1	19.5	12.1; 32.5	28	15; 51	446	288; 694	169	73; 275	81	48; 134
6–10	154	2.5	0.9; 5.9	3.3	1.7; 7.1	20.8	12.1; 32.3	28	12; 46	486	260; 742	162	98; 283	79	46; 128
11–15	162	2.3	0.9; 6.2	3.3	1.3; 6.4	20.8	12.4; 32.8	29	14; 48	483	283; 770	169	67; 295	80	32; 125
16–20	131	2.3	1.0; 6.0	3.1	1.4; 6.7	21.1	11.8; 34.5	28	14; 50	480	298; 752	163	76; 266	80	54; 120
21–25	92	2.7	0.8; 6.3	3.4	1.6; 7.4	22.1	11.8; 36.2	30	14; 56	473	283; 883	159	72; 290	88	42; 141
26–30	72	2.5	1.0; 6.1	3.4	1.7; 5.8	21.4	12.3; 30.5	27	14; 47	476	285; 779	164	76; 257	81	39; 143
31–35	48	2.4	0.6; 5.6	3.5	1.7; 7.0	21.2	13.0; 36.7	28	12; 55	526	334; 858	185	93; 321	83	44; 131
36–40	28	2.5	0.8; 6.1	3.6	1.7; 7.0	21.1	11.5; 36.6	26	12; 44	497	326; 900	157	86; 226	75	38; 133
>40	66	2.4	0.8; 6.6	3.3	1.8; 7.8	22.9	13.8; 36.1	27	11; 47	541	335; 920	158	82; 290	84	41; 140
Number of binge episodes during the past 30 days*, N=1194															
0	171	2.4	0.9; 5.8	3.6	1.6; 7.4	17.9	10.9; 32.9	27	12; 51	440	260; 740	166	84; 281	83	50; 127
1–2	250	2.6	1.0; 5.7	3.2	1.3; 6.5	19.9	11.4; 32.3	28	13; 54	449	257; 737	164	71; 266	80	43; 131
3–5	405	2.3	1.0; 5.8	3.2	1.4; 6.5	20.8	12.0; 33.1	28	14; 49	476	286; 765	170	86; 277	80	46; 125
6–9	254	2.4	0.9; 6.5	3.4	1.7; 6.6	21.3	12.0; 35.6	28	14; 48	496	300; 842	164	73; 297	80	41; 140
>9	90	2.5	0.8; 6.1	3.3	1.8; 7.0	21.9	12.5; 37.2	27	13; 55	523	310; 858	164	79; 291	85	41; 137

Presented as median (M) and 5 and 95 centiles (5–95).
FSH, follicle-stimulating hormone; LH, luteinizing hormone; SHBG, sex hormone binding globulin.
*Binging defined as alcohol intake of more than 5 units on one occasion.

Table 3 Results from linear regression analyses of semen quality (adjusted β-coefficients) and serum reproductive hormones (percent change) among young, Danish men according to habitual alcohol intake (last week represented a typical week) or recent (the week preceding the visit) or binging during the past 30 days

		Sperm concentration*† (million/mL)		Total sperm count*† (million)		Morphology†‡ (%)			Testosterone§¶ (nmol/L)		SHBG§¶ (nmol/L)		Free testosterone§¶ (pmol/L)	
Alcohol intake	**N**	**B**	**95% CI**	**β**	**95% CI**	**β**	**95% CI**	**N**	**%**	**95% CI**	**%**	**95% CI**	**%**	**95% CI**
Units in a typical week, N=553								Units the week preceding the visit, N=1194						
0	121	−0.32	−0.62 to −0.03	−0.42	−0.86 to 0.01	−0.21	−0.54 to 0.12	242	−2.7	−8.5 to 3.5	−0.6	−8.6 to 6.9	−3.3	−9.1 to 2.6
1–5	92	Reference		Reference		Reference		193	Reference		Reference		Reference	
6–10	71	−0.04	−0.38 to 0.3	−0.06	−0.56 to 0.45	−0.12	−0.49 to 0.26	154	3.3	−3.6 to 10.6	−3.0	−10.6 to 5.2	5.5	−1.4 to 12.9
11–15	80	−0.21	−0.54 to 0.12	−0.29	−0.77 to 0.20	−0.19	−0.56 to 0.18	160	2.0	−4.9 to 9.2	−3.1	−10.6 to 5.1	3.6	−3.1 to 10.7
16–20	62	−0.03	−0.39 to 0.33	−0.18	−0.70 to 0.34	−0.09	−0.46 to 0.29	130	3.0	−4.2 to 10.8	−2.1	−10.1 to 6.7	4.3	−3.0 to 12.0
21–25	45	0.25	−0.15 to 0.65	0.07	−0.52 to 0.65	−0.13	−0.55 to 0.29	92	7.3	−1.1 to 16.4	−0.2	−9.4 to 9.9	7.9	−0.3 to 16.9
26–30	25	−0.35	−0.83 to 0.14	−0.65	−1.37 to 0.08	−0.19	−0.71 to 0.34	71	1.0	−7.7 to 10.4	−9.0	−18.1 to 1.1	5.9	−3.1 to 15.5
31–35	14	−0.29	−0.92 to 0.33	−0.60	−1.51 to 0.31	−0.56	−1.19 to 0.06	47	6.1	−4.5 to 17.8	−9.8	−20.3 to 2.1	11.9	0.8 to 23.9
36–40	11	−0.33	−1.02 to 0.35	−0.73	−1.73 to 0.28	−0.54	−1.20 to 0.13	28	9.1	−4.2 to 17.8	−9.3	−22.2 to 5.7	16.0	2.1 to 31.7
>40	21	−0.39	−0.92 to 0.14	−0.54	−1.32 to 0.23	−0.46	−0.99 to 0.08	66	10.6	0.8 to 21.3	−12.0	−21.1 to −1.9	19.5	9.2 to 30.9
p trend**		0.02		0.01		0.02			0.01		<0.01		<0.01	
Number of binge episodes during the past 30 days††														
0	174	Reference		Reference		Reference		171	Reference		Reference		Reference	
1–2	248	−0.01	−0.23 to 0.22	0.15	−0.18 to 0.48	0.07	−0.16 to 0.31	246	4.0	−2.5 to 10.8	7.9	0.1 to 16.4	1.0	−5.2 to 7.6
3–5	407	0.08	−0.13 to 0.29	0.26	−0.05 to 0.56	0.10	−0.12 to 0.31	401	6.2	0.1 to 12.6	4.1	−2.9 to 11.6	6.1	0.1 to 12.4
6–9	253	0.00	−0.23 to 0.23	0.08	−0.26 to 0.41	−0.04	−0.28 to 0.19	252	8.4	1.6 to 15.6	1.1	−6.7 to 9.2	10.2	3.4 to 17.5
>9	88	−0.02	−0.32 to 0.29	0.03	−0.41 to 0.47	−0.22	−0.52 to 0.08	89	13.0	3.8 to 23.1	−2.9	−12.1 to 7.5	17.2	7.8 to 27.6
p trend**		0.93		0.87		0.16			<0.01		0.31		0.01	

*Adjusted for period of abstinence, smoking and body mass index (BMI) categorised according to table 2 (sperm morphology not adjusted for period of abstinence).
†Transformed by cubic root.
‡Counted for 904 men of whom 397 stated that last weeks alcohol intake represented a typical week.
§Adjusted to a time at 8.00, BMI and smoking.
¶Transformed by the use of natural logarithm and back transformed giving the percentages change.
**Test for trend was performed by inserting the categorical alcohol variable into the model assuming the association to be linear with 1–5 units weekly as reference and 0 units excluded.
††Binging defined as alcohol intake for more than 5 units on one occasion.

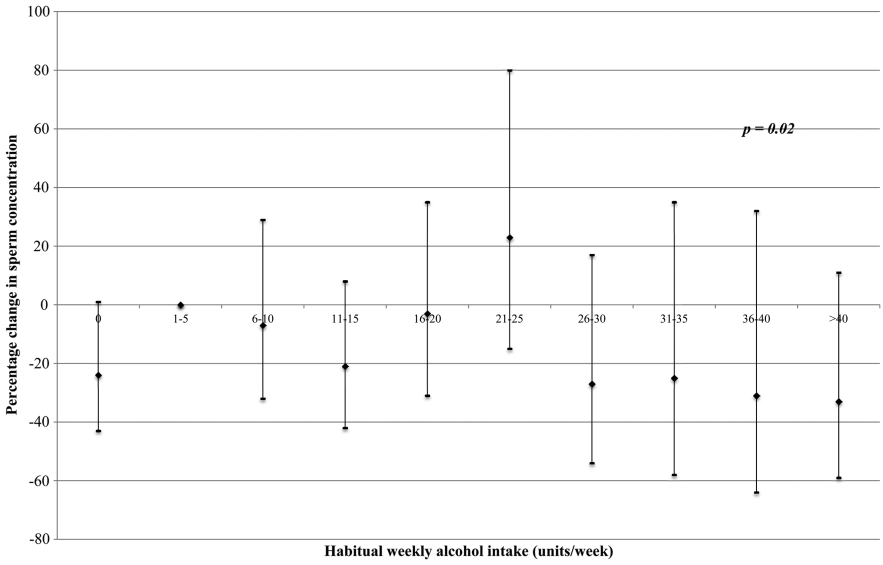

Figure 1 Adjusted (for period of abstinence, body mass index and smoking) changes in sperm concentration (%) according to habitual alcohol intake (reference 1–5 units in a typical week) among 553 young, Danish men. The p-value refers to the linear trend from the reference alcohol intake to the highest intake (abstainers excluded).

associated with habitual alcohol intake (data not shown). Habitual alcohol intake was also associated with serum reproductive hormones although not as strongly as the recent intake (data not shown). The associations between recent alcohol intake from beer were similar to that of total alcohol.

DISCUSSION

Findings

In this cohort of more than 1200 young healthy men with detailed questionnaire information on alcohol intake we found that a habitual alcohol intake was associated with a reduction in semen quality already from more than 5 units/week in a typical week although the decreasing trend was most apparent for men with a typical weekly intake above 25 units. In addition, recent alcohol intake (for the week preceding the visit) was associated with an increase in serum testosterone and reduction in SHBG. No independent adverse effect of binging was found. The negative association between alcohol intake and semen quality may be attributed to a direct adverse effect of alcohol on spermatogenesis or it may be a result of differences in lifestyle, health behaviour and diet found among high alcohol consumers, despite adjustment for these factors.

This is, to our knowledge, the first study to separate the effects of recent versus habitual alcohol exposure, and as the duration of spermatogenesis is approximately 72 days[30] the typical intake is probably a more appropriate exposure measure than the intake during the week preceding the delivery of the semen sample. Contrary to this, serum reproductive hormone levels fluctuate[31] and are theoretically more susceptible towards recent changes (within days) induced by recent alcohol exposure (the week preceding the blood sampling).

Comparison with previous studies

Our findings are in accordance with a recent study among 347 young Danish men in which a non-significant dose–response association between recent alcohol intake (5 days preceding the delivery of the

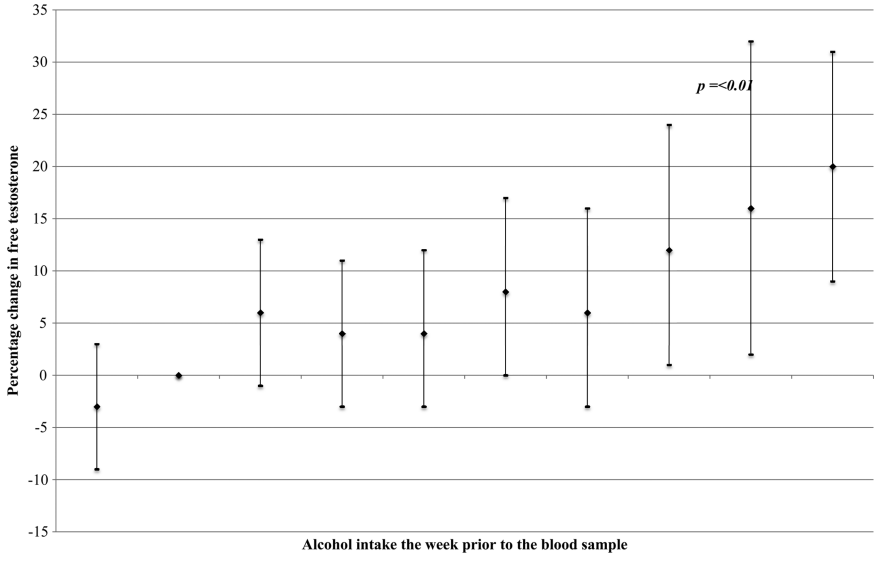

Figure 2 Adjusted (for body mass index, time 8:00 and smoking) changes in free testosterone (%) according to recent alcohol intake (reference 1–5 units the week preceding the visit) among 1194 young Danish men. The p value refers to the linear trend from the reference alcohol intake to the highest intake.

sample) and semen quality was found.[16] The study did not obtain information on typical alcohol exposure nor on binging. However, a Chinese study among 1346 men did not find association between semen quality or alcohol intake even in high doses (more than 120 units/months).[18] Other previous studies of association between alcohol intake and semen quality have shown contradictory results,[6 7 9–15 17] but have been conducted in small selected population and not been able to address dose–response associations and none have been able to separate the effect of recent versus habitual intake. A previous multicenter study including young and fertile men did not find adverse effect of recent alcohol intake (the week preceding the visit) on semen quality, although most men only had a moderate alcohol intake.[19] The young Danish men in that study were also conscripts but included from 1996 to 2007 after which the questionnaire included more detailed information on alcohol intake. The men in this study were included from 2008. No alcohol consumption was also associated with reduced semen quality, which may be attributed to social or health parameters differentiating non-drinkers from drinkers.

We found no independent adverse effect of binging on semen quality, which to our knowledge has not previously been reported. It was, however, difficult to separate binging from typical alcohol intake as most young men who binged also had a high alcohol intake. The percentage of Danes drinking 5 units or more in a typical drinking occasion has been reported to be 23%. Furthermore, young people aged 15–24 years are more likely (25%) to drink 5 units or more on one occasion compared with people above 55 years of age (11%).[1]

Animal studies have suggested that alcohol may affect the hypothalamic-pituitary-gonadal axis, change sperm morphology and directly negatively affect the testis.[32 33] In addition, analysis of histological samples from 195 deceased men showed that high alcohol consumption (>80 g alcohol/>7 units/day) was associated with significantly reduced spermatogenesis, including spermatogenic arrest and sertoli-cell-only syndrome.[34]

Our observed association between alcohol intake, testosterone and cFT is in accordance with previous studies showing increased total testosterone and cFT or increased cFT in combination with decreased SHBG,[16 19 35 36] whereas other studies found no association with cFT.[37–40] If SHBG levels are affected this could explain the observed increase in cFT. Otherwise, it may be explained by alcohol detoxification leading to a changed metabolism of steroids in the liver. In contrast, decreased testosterone levels have been reported in male alcoholics suggesting that habitual alcohol abuse may damage Leydig cells or impair the hypothalamic-pituitary-gonadal axis.[6 41]

Strengths and weaknesses

Our study has several strengths. It was large and consisted of young healthy men and the participation rate was approximately 30%, which is higher than in other population-based semen-quality studies.[42–44] The drinking habits of these men resembled those of Danish men aged 16–20 years in 2008,[45] suggesting that they are not selected. In addition, the majority of our young men had no knowledge of their fertility potential and this is unlikely to have affected their motivation to participate. Our study was, however, cross-sectional and reverse causation cannot be excluded, whereby men with poor semen quality have an unhealthier lifestyle and health behaviour and drink more alcohol even though we adjusted for these factors.

The men in our study reported daily alcohol consumption the week preceding the visit, as we assumed that to be more accurate to recall than an average intake. This consumption may differ from the typical weekly intake, which can lead to misclassification of exposure, and we therefore repeated the analyses among men stating that that week represented a typical week. We used diary information on alcohol consumption, which makes it easier to recall the units consumed, but it may still be underreported. Further, the definition of a unit may vary according to size, method of preparation and brand. We defined binging as an intake of 5 units or more in a single day, which is also the definition used by The Danish National Board of Health.[46] These potential sources of exposure misclassification are likely to be unrelated to semen quality, since the men responded to the questionnaire, before they knew the result of their semen and blood analysis. Such non-differential misclassifications would underestimate the associations between alcohol habits and semen quality and reproductive hormones and cannot explain our findings.

Conclusion and implications

In conclusion, we found an adverse dose–response association between semen quality and habitual alcohol intake most pronounced among men with an alcohol intake above 25 units in a typical week. In addition, men with a high alcohol intake the week preceding the visit had increased free testosterone. This is to our knowledge the first study among healthy young men with detailed information on alcohol intake and, given the fact that young men in the western world have a high alcohol intake, this is of public health concern and could be a contributing factor to the low sperm count reported among young men.[20] It remains to be seen whether semen quality is restored if alcohol intake is reduced, but young men should be advised that high habitual alcohol intake may affect not only their general but also their reproductive health.

Author affiliations
[1]Department of Environmental Medicine, University of Southern Denmark, Copenhagen, Denmark
[2]The Faculty of Medical and Health Sciences, University Department of Growth and Reproduction, Rigshospitalet, University of Copenhagen, Copenhagen, Denmark

[3]Department of Preventive Medicine, Icahn School of Medicine at Mount Sinai, New York, New York, USA

Contributors TKJ performed data analysis and interpretation and wrote the manuscript. MG and JOBM provided assistance with data analysis and revised and edited the manuscript. THL, LP and NJ performed data collection, provided assistance with data analysis and interpretation, revised and edited the manuscript. NES, SHS and AJ provided assistance with data analysis and interpretation, and revised and edited the manuscript.

Funding This work was supported by the The Danish Council for Strategic Research, Program Commission on Health, Food and Welfare (project number 2101-08-0058), Rigshospitalet (grants 961506336 and R42-A1326), European Union, DEER (grant agreement no 212844), The Danish Ministry of Health and The Danish Environmental Protection Agency and Kirsten and Freddy Johansens Foundation (grant 95-103-72087.

Competing interests None.

Patient consent Obtained.

Ethics approval Ethical approval was obtained from the local ethical committee; Danish Regional Ethical Committee.

Provenance and peer review Not commissioned; externally peer reviewed.

Data sharing statement Additional data regarding technical details, statistical code, and derivative data is available from the principal investigator at tkjensen@health.sdu.dk. Data access for further analyses is possible through direct collaborative agreement or through locally managed access arranged through the study's principal investigator.

REFERENCES

1. Eurobarometer. EU citizens' attitudes towards alcohol. Secondary EU citizens' attitudes towards alcohol 2010. http://ec.europa.eu/health/alcohol/docs/ebs_331_en.pdf
2. Simons-Morton B, Pickett W, Boyce W, *et al*. Cross-national comparison of adolescent drinking and cannabis use in the United States, Canada, and the Netherlands. *Int J Drug Policy* 2010;21:64–9.
3. Saleem TS, Basha SD. Red wine: a drink to your heart. *J Cardiovasc Dis Res* 2010;1:171–6.
4. Farke W, Anderson P. Binge drinking in Europe. *Adicciones* 2007;19:333–9.
5. Dawson DA, Li TK, Grant BF. A prospective study of risk drinking: at risk for what? *Drug Alcohol Depend* 2008;95:62–72.
6. Muthusami KR, Chinnaswamy P. Effect of chronic alcoholism on male fertility hormones and semen quality. *Fertil Steril* 2005;84:919–24.
7. Martini AC, Molina RI, Estofan D, *et al*. Effects of alcohol and cigarette consumption on human seminal quality. *Fertil Steril* 2004;82:374–7.
8. Gaur DS, Talekar MS, Pathak VP. Alcohol intake and cigarette smoking: impact of two major lifestyle factors on male fertility. *Indian J Pathol Microbiol* 2010;53:35–40.
9. Stutz G, Zamudio J, Santillan ME, *et al*. The effect of alcohol, tobacco, and aspirin consumption on seminal quality among healthy young men. *Arch Environ Health* 2004;59:548–52.
10. Chia SE, Tay SK, Lim ST. What constitutes a normal seminal analysis? Semen parameters of 243 fertile men. *Hum Reprod* 1998;13:3394–8.
11. Dunphy BC, Barratt CL, Cooke ID. Male alcohol consumption and fecundity in couples attending an infertility clinic. *Andrologia* 1991;23:219–21.
12. Kunzle R, Mueller MD, Hanggi W, *et al*. Semen quality of male smokers and nonsmokers in infertile couples. *Fertil Steril* 2003;79:287–91.
13. Lopez Teijon M, Garcia F, Serra O, *et al*. Semen quality in a population of volunteers from the province of Barcelona. *Reprod Biomed Online* 2007;15:434–44.
14. Marshburn PB, Sloan CS, Hammond MG. Semen quality and association with coffee drinking, cigarette smoking, and ethanol consumption. *Fertil Steril* 1989;52:162–5.
15. Oldereid NB, Rui H, Purvis K. Life styles of men in barren couples and their relationship to sperm quality. *Int J Fertil* 1992;37:343–9.
16. Hansen ML, Thulstrup AM, Bonde JP, *et al*. Does last week's alcohol intake affect semen quality or reproductive hormones? A cross-sectional study among healthy young Danish men. *Reprod Toxicol* 2012;34:457–62.
17. Povey AC, Clyma JA, McNamee R, *et al*. Modifiable and non-modifiable risk factors for poor semen quality: a case-referent study. *Hum Reprod* 2012;27:2799–806.
18. Li Y, Lin H, Ma M, *et al*. Semen quality of 1346 healthy men, results from the Chongqing area of southwest China. *Hum Reprod* 2009;24:459–69.
19. Jensen TK, Swan S, Jørgensen N, *et al*. Alcohol and male reproductive health; a cross-sectional study of 8,344 healthy men from Europe and USA. *Hum Reprod* 2014;29:1801–9.
20. Jørgensen N, Joensen UN, Jensen TK, *et al*. Human semen quality in the new millennium: a prospective cross-sectional population-based study of 4867 men. *BMJ Open* 2012;2:pii:e000990.
21. Jensen TK, Andersson AM, Jorgensen N, *et al*. Body mass index in relation to semen quality and reproductive hormones among 1,558 Danish men. *Fertil Steril* 2004;82:863–70.
22. Jensen TK, Swan SH, Skakkebaek NE, *et al*. Caffeine intake and semen quality in a population of 2,554 young Danish men. *Am J Epidemiol* 2010;171:883–91.
23. Jensen TK, Andersson AM, Skakkebaek NE, *et al*. Association of sleep disturbances with reduced semen quality: a cross-sectional study among 953 healthy young Danish men. *Am J Epidemiol* 2013;177:1027–37.
24. Jensen TK, Heitmann BL, Jensen MB, *et al*. High dietary intake of saturated fat is associated with reduced semen quality among 701 young Danish men from the general population. *Am J Clin Nutr* 2013;97:411–18.
25. Organisation WH. *WHO laboratory manual for the examination of human semen and sperm-cervical mucus interaction*. 4th edn. Cambridge: Cambridge University Press, 1999.
26. Jorgensen N, Auger J, Giwercman A, *et al*. Semen analysis performed by different laboratory teams: an intervariation study. *Int J Androl* 1997;20:201–8.
27. Menkveld R, Stander FS, Kotze TJ, *et al*. The evaluation of morphological characteristics of human spermatozoa according to stricter criteria. *Hum Reprod* 1990;5:586–92.
28. Vermeulen A, Verdonck L, Kaufman JM. A critical evaluation of simple methods for the estimation of free testosterone in serum. *J Clin Endocrinol Metab* 1999;84:3666–72.
29. Craig CL, Marshall AL, Sjostrom M, *et al*. International physical activity questionnaire: 12-country reliability and validity. *Med Sci Sports Exerc* 2003;35:1381–95.
30. Adler ID. Comparison of the duration of spermatogenesis between male rodents and humans. *Mutat Res* 1996;352:169–72.
31. Jensen RB, Vielwerth S, Larsen T, *et al*. Pituitary-gonadal function in adolescent males born appropriate or small for gestational age with or without intrauterine growth restriction. *J Clin Endocrinol Metab* 2007;92:1353–7.
32. Anderson RA Jr, Willis BR, Oswald C, *et al*. Hormonal imbalance and alterations in testicular morphology induced by chronic ingestion of ethanol. *Biochem Pharmacol* 1980;29:1409–19.
33. Anderson RA Jr, Willis BR, Oswald C, *et al*. Ethanol-induced male infertility: impairment of spermatozoa. *J Pharmacol Exp Ther* 1983;225:479–86.
34. Pajarinen J, Karhunen PJ, Savolainen V, *et al*. Moderate alcohol consumption and disorders of human spermatogenesis. *Alcohol Clin Exp Res* 1996;20:332–7.
35. Muller M, den Tonkelaar I, Thijssen JH, *et al*. Endogenous sex hormones in men aged 40–80 years. *Eur J Endocrinol* 2003;149:583–9.
36. Shiels MS, Rohrmann S, Menke A, *et al*. Association of cigarette smoking, alcohol consumption, and physical activity with sex steroid hormone levels in US men. *Cancer Causes Control* 2009;20:877–86.
37. Svartberg J, Midtby M, Bonaa KH, *et al*. The associations of age, lifestyle factors and chronic disease with testosterone in men: the Tromso Study. *Eur J Endocrinol* 2003;149:145–52.
38. Tamimi R, Mucci LA, Spanos E, *et al*. Testosterone and oestradiol in relation to tobacco smoking, body mass index, energy consumption and nutrient intake among adult men. *Eur J Cancer Prev* 2001;10:275–80.
39. Allen NE, Appleby PN, Davey GK, *et al*. Lifestyle and nutritional determinants of bioavailable androgens and related hormones in British men. *Cancer Causes Control* 2002;13:353–63.

40. Handa K, Ishii H, Kono S, *et al*. Behavioral correlates of plasma sex hormones and their relationships with plasma lipids and lipoproteins in Japanese men. *Atherosclerosis* 1997;130:37–44.
41. Maneesh M, Dutta S, Chakrabarti A, *et al*. Alcohol abuse-duration dependent decrease in plasma testosterone and antioxidants in males. *Indian J Physiol Pharmacol* 2006;50:291–6.
42. Jorgensen N, Carlsen E, Nermoen I, *et al*. East-West gradient in semen quality in the Nordic-Baltic area: a study of men from the general population in Denmark, Norway, Estonia and Finland. *Hum Reprod* 2002;17:2199–208.
43. Jorgensen N, Andersen AG, Eustache F, *et al*. Regional differences in semen quality in Europe. *Hum Reprod* 2001;16:1012–19.
44. Swan SH, Brazil C, Drobnis EZ, *et al*. Geographic differences in semen quality of fertile U.S. males. *Environ Health Perspect* 2003;111:414–20.
45. Brink A-L, Kræftens Bekæmpelse og Sundhedsstyrelsen Unges livsstil og dagligdag 2008. MULD-rapport nr. 7. 2008.
46. Health DNBo. Secondary. http://www.sundhedsstyrelsen.dk/Sundhedogforebyggelse/Alkohol.aspx