

FAST-TRACK ARTICLE

Semen quality of fertile US males in relation to their mothers' beef consumption during pregnancy

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BACKGROUND: To look at possible long-term risks from anabolic steroids and other xenobiotics in beef, we examined mens' semen quality in relation to their mother's self-reported beef consumption during pregnancy. **METHODS:** The study was carried out in five US cities between 1999 and 2005. We used regression analyses to examine semen parameters in 387 partners of pregnant women in relation to the amount of beef their mothers reported eating while pregnant. Mothers' beef consumption was also analysed in relation to the son's history of previous subfertility. **RESULTS:** Sperm concentration was inversely related to mothers' beef meals per week ($P = 0.041$). In sons of 'high beef consumers' (>7 beef meals/week), sperm concentration was 24.3% lower ($P = 0.014$) and the proportion of men with sperm concentration below 20×10^6 /ml was three times higher (17.7 versus 5.7%, $P = 0.002$) than in men whose mothers ate less beef. A history of previous subfertility was also more frequent among sons of 'high beef consumers' ($P = 0.015$). Sperm concentration was not significantly related to mother's consumption of other meat or to the man's consumption of any meat. **CONCLUSIONS:** These data suggest that maternal beef consumption, and possibly xenobiotics in beef, may alter a man's testicular development *in utero* and adversely affect his reproductive capacity.

Keywords: beef; fertility; meat; semen quality; sperm

Introduction

Diethylstilbestrol (DES), the first synthetic hormone, was formulated in 1933 and was rapidly marketed for a wide range of medical indications (Swan, 2000). In 1947, the year that DES was approved for use by pregnant women, investigators at Purdue University demonstrated the ability of this synthetic hormone to stimulate growth in cattle (Raun and Preston, 2002). In 1954, the US Food and Drug Administration (FDA) approved DES for use in cattle, and by 1956 more than two-thirds of the nation's feeder cattle were receiving DES (Marcus, 1994). Simultaneously, other hormonal additives were being tested for use in cattle; between 1956 and 1958, estradiol benzoate and progesterone implants were approved, first for use in steers and then heifers. Approval for use of other anabolic hormones in cattle followed rapidly. While the US FDA withdrew approval of DES for use in cattle in 1979, other anabolic hormones continue to be used legally in the USA and elsewhere as growth promoters in meat production. Six hormones are now in common use in Canada and USA: the three natural steroids, estradiol, testosterone and

progesterone, and the three synthetic hormones, zeranol (an estrogen), trenbolone acetate (a steroid with androgen and glucocorticoid action) and melengestrol acetate (a potent progestin) (Meyer, 2001). The anabolic steroids are most often used in combination; often an androgen and estrogen are used simultaneously, e.g. estradiol and trenbolone acetate. 'Good veterinary practice' prescribes that melengestrol acetate be given in the feed and the other hormones given as capsules implanted subcutaneously in the calf's ear from where the hormones continuously are released into circulation. All six hormones can induce increased growth and development of the animal by mechanisms similar to that of the 'peripheral' form of human precocious puberty, which is also associated with a growth spurt (Parent *et al.*, 2003). At slaughter, not all steroids have been metabolized or excreted; measurable levels are, in fact, present in muscle, fat, liver, kidney and other organs present in meat products (Henricks *et al.*, 2001). Therefore, it has been necessary to regulate the use of these growth promoters to avoid unintended adverse effects in humans eating these meat products. FDA has defined an

'acceptable daily intake' (ADI) for each of these veterinary drugs (Henricks *et al.*, 2001), and since 1988 the use of these hormones has been banned in Europe (Stephany, 2001). The International Joint Food and Agricultural Organization's World Health Organization Expert Committee on Food Additives (JECFA) has also published ADIs for all hormones in current use on the basis of animal testing.

These ADIs are based on traditional toxicological testing, and the possible effects on human populations exposed to residues of anabolic sex hormones through meat consumption have never, to our knowledge, been studied. Theoretically, the fetus and the prepubertal child are particularly sensitive to exposure to sex steroids (Andersson and Skakkebaek, 1999; Bay *et al.*, 2004). Therefore, the consumption of residues of steroids in meat by pregnant women and young children is of particular concern. Recent animal and human studies have suggested that perinatal exposures, including exposure to sex steroids, can induce a testicular dysgenesis syndrome (Skakkebaek *et al.*, 2001), and result in a number of testicular disorders in adulthood, including poor spermatogenesis.

The lack of human data on the safety of anabolic steroids in meat production prompted us to analyse mother's beef consumption while pregnant in relation to her son's semen parameters in a large multicenter pregnancy cohort study, the Study for Future Families (SFF).

Materials and Methods

Study population

All subjects were participants in the SFF, a multicenter study of pregnant women and their partners, conducted at prenatal clinics affiliated with university hospitals in five US cities (Harbor-UCLA and Cedars-Sinai Medical Center in Los Angeles, CA; University of Minnesota Health Center in Minneapolis, MN; University Physicians in Columbia, MO; Mt. Sinai School of Medicine, New York City, NY and University of Iowa, Iowa City, IA) between 1999 and 2005. Human subject approvals were obtained from Institutional Review Boards at all participating institutions.

Methods for clinical examination, data collection and semen analysis, which were similar across centres, have been described previously (Swan *et al.*, 2003a). Women were recruited at the prenatal clinic and only those whose pregnancy was conceived without medical assistance were eligible. Both partners completed a questionnaire and most men provided a semen sample. Questions for the men included demographics, recent fever, history of sexually transmitted diseases (STDs), as well as lifestyle factors (smoking, alcohol and caffeine consumption) and diet (including number of servings of beef and other red meat). The men were also asked, 'Have you ever seen a doctor because you thought you might be having trouble fathering a child? Men who responded positively are referred to here as 'self-reporting previous subfertility'.

The man was requested to ask his mother to complete a brief questionnaire, which could, if necessary, be completed by the son in consultation with his mother, or possibly by another proxy respondent. The mother's questionnaire asked, 'In a typical week while you were pregnant with your son —, did any of your meals contain the following foods?' Separate questions on beef, lamb or pork, veal, etc. followed. If the mother responded positively, she was asked to specify the number of meals per week that included that food item (with partial servings rounded up to the next whole serving). There were similar dietary questions in the man's questionnaire referring to the week that preceded

semen collection (or a typical week in the last 3 months, if the last week was atypical). The mother was also asked where she lived at the time her son was born. No data were obtained from the man's father.

Semen collection and analysis

Men collected semen samples by masturbation at the clinic and almost all samples were analysed within 45 min of collection. Sperm concentration was determined by hemocytometer. The percent motile sperm refers to the percentage of sperm with any flagellar movement, whether twitching or progressive. A single technician assessed sperm morphology using the method recommended by the World Health Organization (WHO) in 1987 (WHO 1987). These methods are described in detail elsewhere (Brazil *et al.*, 2004). Although men were requested to observe a 2–5-day abstinence period, the importance of accurately reporting the actual abstinence period was stressed. Abstinence times greater than 240 h were set equal to 240 h because very long abstinence times are not significantly related to sperm concentration (Carlsen *et al.*, 2005).

Statistical methods

We examined mother's beef consumption in relation to semen quality in several ways. A variable denoting the number of beef meals per week consumed by the mother (BEEF) was examined in relation to sperm concentration, as well as sperm motility and morphology, in generalized linear models (GLM) (SAS Institute, 2001). We also dichotomized BEEF at seven meals per week (or one meal a day), a cutpoint chosen prior to data analysis. Mothers who reported consuming >7 meals per week are referred to as 'high beef consumers'. In addition to these analyses of mother's beef meals per week, we also examined consumption of 'other red meat' and 'all red meat' (the sum of beef and other red meat).

We used a logarithmic (base 10) transformation of sperm concentration because the distribution of this parameter is markedly skewed. Regression coefficients were then back-transformed for ease of interpretation. We also examined the proportion of men whose sperm concentration fell below 20×10^6 /ml, a widely used clinical threshold for subfertility (WHO, 1992). The man's response to the question, 'Have you ever seen a doctor because you thought you might be having trouble fathering a child?', was examined in relation to BEEF and the binary variable 'high beef' or 'not high beef'.

Man's characteristics initially examined in these analyses include: age, smoking, alcohol, body mass index (BMI), history of STD and abstinence time, as well as his own meat consumption. The mother's age, whether or not she smoked during pregnancy, and whether she nursed her son were also initially examined in the model. Selection of covariates for the final model was based on their importance in the literature, biological plausibility, sufficient numbers within strata and evidence of an effect on the strength of the association with beef consumption.

We compared participant characteristics between those for whom information on mother's beef meals was available and those for whom it was not, in order to assess possible participation bias. Associations were examined restricted to men born after 1954 when the use of DES was first approved for use in beef. We also compared associations in those born before and after 1970, to assess any effects of changes in contaminants in beef over the study period. Finally, we examined associations by men's place of birth.

Results

Of the 773 men who provided a semen sample, mothers' questionnaires were available for 582 (75.3%), and 66.5% of these

Table 1: Characteristics of participants by availability of mother's beef consumption

	Mean (median) or %		P-value (unadjusted)
	Beef per week available	Beef per week not available	
Number, <i>n</i>	387	386	–
Men's age (years)	31.6 (31.4)	31.5 (30.9)	0.669
Mother's age (years) ^a	25.0 (25.0)	26.2 (26.0)	0.022
Men's alcohol (drinks/week) ^b	4.5 (2.0)	5.1 (1.0)	0.426
Abstinence time (days)	3.3 (3.0)	3.5 (3.1)	0.066
Sperm concentration (10 ⁶ /ml)	82.8 (68.2)	78.2 (67.6)	0.283
Motile sperm (%)	51.6	50.2	0.108
Normal morphology (%)	58.0	57.1	0.243
Reported previous subfertility (%)	6.2	5.0	0.470

^aAt time of pregnancy with son; ^bCurrent.

included information on the number of beef meals consumed during pregnancy. Table 1 compares sperm parameters and other characteristics between participants for whom the weekly amount of beef consumption was available (*n* = 387) and those for whom it was not (*n* = 386). Mean sperm concentration, percent motile sperm and percent normal morphology did not differ significantly between these two groups of men (*P*-values comparing means were 0.283, 0.108 and 0.243, respectively), nor did the proportion of men who had reported previous subfertility differ. Mothers for whom number of beef meals was available were, on average, one year younger than those for whom it was not.

Mothers consumed, on average, 4.3 beef meals per week (range 0–21) and only 15 (4%) reported eating no beef during pregnancy. Table 2 includes characteristics of study participants for both high beef consumers (*n* = 51) and those eating less beef (*n* = 336). High beef consumers ate significantly more other red meat and were more likely to have lived in North America at the time their son was born than women who reported eating less beef.

Among this population of 387 fertile men, born between 1949 and 1983, the mean (median) unadjusted sperm concentration was 82.8 × 10⁶/ml (68.2 × 10⁶/ml). About one-fifth (21%) of these participants were non-Caucasian, and this latter group had a somewhat (but not significantly) lower (unadjusted) sperm concentration (mean 79.3, median 66.9 × 10⁶/ml) than Caucasians (mean 83.8, median 68.3 × 10⁶/ml).

The number of beef meals consumed by the mother was significantly and inversely related to her son's sperm concentration. Results of regression analyses of log sperm concentration using two measures of beef consumed by the mother are shown in Table 3. The adjusted mean sperm concentration (back-transformed from the logarithm of hemocytometer

Table 2: Characteristics of participants by mothers' beef consumption

	Mother's beef meals (per week)		P-value (unadjusted)
	BEEF ≤7 (<i>n</i> = 336)	BEEF > 7 (<i>n</i> = 51)	
Mothers (during pregnancy with son)			
Age (years)	25.1	24.4	0.363
Smoked (%)	19.1	21.6	0.589
Meals of pork, lamb or veal (mean/week)	1.7	4.1	<0.001
Country of son's birth			
North America (%)	79.8	96.1	0.018
Other or missing (%)	20.2	3.9	
Sons (at study entry)			
Age (years)	31.7	31.4	0.732
Caucasian (%)	76.8	90.2	0.107
BMI (kg/m ²)	28.4	29.0	0.400
History of STDs (%)	8.9	7.8	0.797
Alcohol (drinks/week)	4.7	3.1	0.053
Reported previous subfertility (%)	5.7	9.8	0.252
Abstinence time (h)	79.6	77.8	0.637
Sperm concentration (10 ⁶ /ml)	85.0	68.3	0.072
Motile sperm (%)	50.9	51.7	0.627
Normal morphology (%)	57.9	58.8	0.511

count used in regression models) among sons of 'high beef' consumers was 43.1 × 10⁶/ml. For those whose mothers were not high beef consumers it was 24.3% higher (56.9 × 10⁶/ml), a difference that was statistically significant (*P* = 0.014). Among sons of mothers whose beef consumption was not high, only 5.7% had sperm concentration below the WHO threshold for subfertility of 20 × 10⁶/ml. This was significantly less than the percent of men whose mothers were 'high beef' consumers who fell below this threshold (17.7%) (*P* = 0.002).

A similar association was seen when BEEF was modeled as an ordinal variable in a GLM that controlled for the same covariates. This model predicts that if a mother had eaten seven beef meals per week, her son's sperm concentration would have been 15% lower, on average, than the son of a mother who ate no beef during her pregnancy. Sperm motility and morphology were not significantly related to mother's beef consumption (Table 3).

In this population, the relationship between sperm concentration and man's age is non-linear (increasing slightly until about age 30, leveling off, and then slowly decreasing). This pattern is similar in men whose mothers were high beef consumers, with sperm concentration being reduced a similar amount at each age relative to sons of mothers who ate less beef (data not shown). Sperm concentration was also non-linearly related to the amount of alcohol the man consumed (total drinks of all kinds of alcohol per week). Sperm concentration was linearly related to the period of abstinence (up to the truncation point of 240 h) and significantly lower among men with a history of STDs.

Table 3: Regression analyses of semen parameters in relation to two measures of mother's beef consumption

	Log ₁₀ sperm concentration		% motile sperm		% normal morphology	
	Coefficient	<i>P</i> -value	Coefficient	<i>P</i> -value	Coefficient	<i>P</i> -value
Mothers' beef servings per week						
Number	-0.0102	0.041	-0.1317	0.437	0.0152	0.919
>7 versus ≤7	-0.1208	0.014	-1.0059	0.552	0.8022	0.587
Sons' characteristics ^a						
STD history	-0.1495	0.012	-0.8837	0.659	-0.6452	0.465
Age	0.0563	0.019	0.3221	0.694	0.3552	0.321
Age-squared	-0.0008	0.024	-0.0081	0.522	-0.0056	0.312
Alcohol	0.0178	<0.0001	0.2429	0.107	0.1073	0.105
Alcohol-squared	-0.0003	0.0008	-0.0054	0.109	-0.0014	0.352
Sample characteristics ^a						
Abstinence time	0.0023	<0.0001	NA		NA	
Time to analysis		NA	-0.0752	0.042	NA	NA

^aRegression coefficients and *P*-values for sons' and sample characteristics are from the model in which beef consumption was dichotomized. Differences between regression coefficients and *P*-values from model with mother's beef consumption dichotomized and integer valued are minimal (Data available on request).

Only 75 mothers (19.4%) reported smoking while pregnant, and mother's smoking was unrelated to her beef consumption (Table 2). The son's sperm concentration was only weakly related to mother's smoking. In the regression model, mother's smoking was associated with a slightly (but not significantly) higher sperm concentration (regression coefficient for mother's smoking 0.0570, *P* = 0.1852) and including mother's smoking in the model had a negligible effect on the association between BEEF and any sperm parameter.

Table 4 provides the distribution of mother's meals of beef, other red meat and all red meat. Mothers reported eating less than half as many meals containing red meat other than beef (mean 2.0 meals per week) than beef meals, and ~25% of mothers reported eating >7 meals per week of any red meat, most of which was beef. Consumption of other meat meals was correlated with beef meals (correlation coefficient 0.44, *P* < 0.0001), and high beef consumers consumed more other red meat (mean 4.1 meals per week) than women eating less beef. The relationship between number of meals of other red meat and sperm concentration was somewhat weaker than that for BEEF and non-significant (correlation coefficient -0.0098, *P* = 0.160). The magnitude of the association between total red meat and sperm concentration was also weaker than that for BEEF but significant; regression coefficients for total red meat and beef were -0.0073 (*P* = 0.030) and -0.0102 (*P* = 0.041), respectively.

Mother's consumption of fish, chicken, soy products and vegetables were asked in the same format as the question on beef and other red meat, and analysed using the same regression models. None of these foods were related to the son's sperm concentration (all *P*-values 0.225–0.655).

The son's own beef consumption was correlated with his mother's, but not strongly (correlation coefficient 0.271, *P* < 0.001), and was unrelated to his sperm concentration (regression coefficient -0.005, *P* = 0.319) or other sperm parameters.

In an unadjusted comparison, sons of high beef consumers appeared to be more likely to be classified as 'self-reporting previous subfertility' (9.8%) compared to those whose mothers ate less beef (5.7%) (Table 2 unadjusted *P*-value = 0.252 non-significant). However, this difference was significant in the logistic analysis (*P* = 0.015) controlling for age and age-squared, the only covariates that influenced the association between BEEF and 'self-reporting previous subfertility'.

Mothers' beef consumption varied by centre. Mothers reported eating 2.2 beef meals per week in NY (*n* = 18), 3.8 in CA (*n* = 79), 4.1 in MN (*n* = 111), 4.2 in IA (*n* = 53) and 5.2 in MO (*n* = 126). Sperm concentration also varied by centre, with the most significant difference being between a mean (median) of 101.3 (88.2) × 10⁶/ml in MN and 60.1 (53.5) × 10⁶/ml in MO (Swan *et al.*, 2003a). Therefore, we examined the relationship between sperm concentration and

Table 4: Servings of meat per week consumed by mothers during pregnancy

Servings/ week	Beef		Other red meat ^b		All red meat ^c	
	Number	%	Number	%	Number	%
0	15	3.9	82	21.8	11	2.8
1–2	110	28.4	203	53.8	46	11.9
3–4	123	31.8	54	14.3	98	25.3
5–7	88	22.7	29	7.7	132	34.1
>7	51	13.2	9	2.4	100	25.9
Total ^a	387	100	377	100	387	100

^aTotals differ because some women who reported on amount of beef servings/week did not report on amount of other meat servings/week; ^bPork + lamb + veal; ^cBeef + pork + lamb + veal.

BEEF within MO and MN, where the difference in semen quality was greatest, but the populations were otherwise very similar. Regression coefficients for BEEF differed slightly, with the association being somewhat stronger in MN (-0.0098 and -0.0138 for MO and MN, respectively). We also examined a possible interaction between center and BEEF, but found none (for all centre \times BEEF interaction terms P -values >0.792).

In some cases ($n = 50$), the mother's questionnaire had been completed by the son (presumably in collaboration with his mother), the father, or someone else other than the mother. After excluding men for whom the questionnaire was completed by someone other than the mother, the regression coefficient for high compared to not high beef consumption changed only slightly, from -0.1208 ($P = 0.014$) for all participants to -0.1201 ($P = 0.021$). We also added a variable indicating who completed the questionnaire to the model containing all 387 women, but this was not significant ($P = 0.980$), and its addition did not alter the strength of the association between beef and sperm concentration.

We saw no evidence of modification of the association between BEEF and sperm concentration with year of birth. After excluding the few men ($n = 4$) born before 1954 (when DES was first approved for use in meat production), the regression coefficient (and P -value) for BEEF was unchanged from that for all 387 men (-0.0101 ($P = 0.044$) and -0.0102 ($P = 0.041$) for men born after 1954 and all men, respectively). When we divided men into those born before and after 1970 (the median year of birth for our subjects), to look for possible changes in the association over the study period, associations were similar in the two groups.

Most men (83.2%) were born in North America, 8.4% in South and Central America, 4.5% in Europe and 3.9% in Asia or elsewhere, and mothers of sons born in North America consumed an average of 1.3 more beef meals a week than other mothers ($P < 0.0001$). In a model that included only men born in North America, the association with beef appeared slightly weaker than that including all men. However, only two mothers living outside North America (one living in Mexico and one in Iran) ate >7 beef meals per week (3.1%), compared to 15.5% of those living in the North America at the time of their son's birth. Therefore, this study can provide little information about the association between semen quality and high beef consumption outside North America.

Discussion

In this study of fertile US men, we found a significant association between a mother's reported beef consumption while pregnant and her son's sperm concentration. In addition, the proportion of men with a sperm concentration below the WHO threshold for subfertility ($20 \times 10^6/\text{ml}$) and of men with a history of possible subfertility increased with higher beef consumption. These findings suggest that maternal beef consumption is associated with lower sperm concentration and possible subfertility, associations that may be related to the presence of anabolic steroids and other xenobiotics in beef. It must, however, be noted that most mothers in this study were living in North America, and our findings may

not apply to other regions of the world where beef is produced by other methods. Additionally, our participants were fertile men who conceived without medical assistance, and the associations reported here might differ among those less fertile.

Mother's beef consumption may be related to other lifestyle factors. We examined several maternal variables in relation to her beef consumption: smoking, employment outside the home and parity. We saw little or no association of these factors with her beef consumption (P -values for regression coefficients were 0.889, 0.801 and 0.364 for smoking, working outside the home and parity, respectively), but cannot rule out the role of other factors on which we had no data. Of these factors, mothers' smoking is the only one previously related to the son's semen quality (Jensen *et al.*, 2004). While we did not see an association with maternal smoking, the prevalence of smoking in our population was somewhat lower than in the European population in which this association was reported.

We previously reported reduced semen quality in men from rural MO relative to urban centres, most notably MN, a difference that was associated with higher concentration of some pesticide metabolites in MO men (Swan *et al.*, 2003b). Therefore, it is important to note that the association between BEEF and sperm concentration we are reporting here was slightly higher among MN men than MO men when these groups were examined separately. Thus, the association between BEEF and sperm concentration is not simply the reflection of this previously reported reduced semen quality in MO.

The mothers' recall of her food consumption when pregnant with her son is undoubtedly subject to error. However, the magnitude (or direction) of that error is unlikely to be related to the son's semen quality, which was unknown to the men or their mothers. In fact, since all men had recently conceived a pregnancy, the man's fertility is unlikely to have been of concern to mother or son.

While our *a priori* hypothesis was that beef consumption was associated with semen quality, we also examined mother's consumption of other foods, including other red meat, at a similar level of detail. We did not observe a significant association between sperm concentration and consumption of other foods. However, only nine women (2.4%) reported eating >7 meals per week of other red meat (pork, lamb or veal), and all but three of these were also high consumers of beef, so it was not possible to examine the association between sperm concentration and high consumption of meat other than beef.

A crucial question is whether the anabolic hormone residues in beef can explain our findings. We have no information on the type of beef that the mothers were eating when pregnant (1949–1983), but most American beef during that time was produced by administration of anabolic hormones (growth promoters) (Epstein, 1990), and it is well documented that such use of anabolic steroids generally results in beef products with residues of the administered hormones (Wade, 1972; Henricks *et al.*, 2001). Moreover, it would be difficult for most consumers to avoid hormone-containing meat, since, while some meat is labeled as 'hormone-free', there is no requirement that meat be labeled as to hormone content. Thus, it seems likely that mothers' increasing beef consumption was accompanied by greater exposure to the sex steroids used in beef production.

Several animal studies carried out prior to the ban in 1979 on the use of DES as an anabolic agent in beef production demonstrated that residues of DES present in meat waste could harm reproduction. Thus, studies showed that mink colonies fed with waste products from chicken raised using estrogenic anabolic agents suffered from infertility (Howell and Pickering, 1964; Duby and Travis, 1971; Sundqvist *et al.*, 1989).

Although evidence is accumulating that the fetal organism may be most sensitive to endocrine disrupters (Sharpe, 2006), we cannot exclude the possibility that remnants of anabolic hormones in meat may also affect children and adults. There are, to our knowledge, no previous human studies relating consumption of meat products containing residues of anabolic hormones to fertility of offspring. However, our hypothesis is consistent with results from a recent 12-year follow-up of >90,000 American women that found that higher consumption of red meat was associated with breast cancer incidence, but only with ER+/PR+ cancers (Cho *et al.*, 2006).

Beef, which contains fat in varying concentrations, may also contain pesticides and remnants of other persistent and lipophilic industrial chemicals. These agents can act as endocrine disrupters, which theoretically may have adverse effects on the human fetal testis similar to the effects of anabolic steroids. Recent studies have, in fact, shown that pesticides and persistent chemicals in breast milk were associated with testicular problems, including undescended testes (Damgaard *et al.*, 2006) in newborn offspring. Increased exposure to such agents through mothers' beef consumption may have contributed to these results. Further, heterocyclic amines, produced in cooking and processing of red meat, which are estrogenic, may play a role (Cho *et al.*, 2006).

In conclusion, in our large study of fertile American men, we found a negative association between the number of servings of beef the mother ate per week while pregnant and the sperm concentration and fertility of her son. Several alternative explanations for these findings are possible. We cannot rule out unknown confounders associated with both the mother's beef consumption and her son's testicular development. As discussed, pesticides and other contaminants in animal feed may play a role, as may lifestyle factors correlated with greater beef consumption. Whether prenatal exposure to anabolic steroids is responsible for our findings in whole or in part could be clarified by repeating this study in men born in Europe after 1988 when anabolic steroids were no longer permitted in beef sold or produced there.

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