The Environment and Male Fertility: Recent Research on Emerging Chemicals and Semen Quality

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ABSTRACT

Scientific and public concern about the potential risk of environmental chemicals to male reproductive health has been heightened by reports of downward trends in semen quality, as well as increased rates of developmental urogenital tract anomalies and testicular cancer. Of particular concern is whether some contemporary-use environmental chemicals alter semen quality. Specific toxicants of interest include phthalates and pesticides, as well as polychlorinated biphenyls (PCBs). The human data on the relationship of semen quality with phthalate and pesticide exposure are limited and do not currently allow for a definitive conclusion on whether adult exposure, at background environmental levels, alters semen quality. However, the epidemiologic data support an inverse association of PCBs with reduced semen quality, specifically reduced sperm motility. The associations found were generally consistent across studies despite a range of PCB levels. In addition to the chemicals discussed, there are additional classes of chemicals that require further study on their relation with human semen quality.

KEYWORDS: Environment, pesticides, phthalates, polychlorinated biphenyls, sperm

Scientific and public concern about the potential risk of environmental chemicals to male reproductive health has been heightened by reports of temporal downward trends in semen quality,[1,2] increased rates of development urogenital tract anomalies (specifically, hypospadias and cryptorchidism),[3] and increased rates of testicular cancer.[4–6] These observations, along with documentation of ubiquitous human exposure to environmental chemicals, raise the possibility that environmental chemicals may be partially responsible for these adverse reproductive and developmental outcomes.

Although semen quality is measured in the adult male, it may be affected by exposures during various life stages, such as during gestation, puberty, or as an adult. In addition, as recently shown by an elegant study by Anway et al,[7] there are transgenerational effects of chemicals, whereby exposure of the maternal or paternal (or even of the grandparents') gametes to chemicals may confer an increased risk of altered semen quality in the offspring. Although early life exposure may impair spermatogenesis, as supported by evidence from studies in laboratory animals, the human data are generally limited to the assessment of both semen quality and environmental exposures during adult life.

In the present article, the focus is on epidemiologic studies that explored the relationship of emerging

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contemporary-use environmental chemicals with semen quality. Toxicants discussed in this article include the following: phthalates, pesticides (primarily the contemporary-use nonpersistent insecticides and herbicides), and polychlorinated biphenyls (PCBs). Although PCBs are not an emerging contaminant, they are included in the article because there are many recent publications worthy of summary.

This article is not an exhaustive review of the epidemiologic literature on environmental toxicants and semen quality. For this, the reader is directed toward previously published reviews and book chapters. These earlier publications discuss known human male reproductive toxicants, primarily occupational agents, such as 1,2-dibromo-3-chloropropane, diethylstilbestrol, inorganic lead, alkylating neoplastic agents, ethylene glycol monomethyl and monoethyl ethers, carbon disulfide, ethylene dibromide, and ionizing radiation.[8–10]

PHTHALATES

The diesters of 1,2-benzenedicarboxylic acid (phthalic acid), commonly known as phthalates, are a group of man-made chemicals with a wide spectrum of industrial applications. High molecular weight phthalates (e.g., di(2-ethylhexyl) phthalate [DEHP], di-isononyl phthalate, di-n-octyl phthalate [DnOP]), are primarily used as plasticizers in the manufacture of flexible vinyl, which in turn is used in consumer products, flooring and wall coverings, food contact applications, and medical devices.[11–13] Manufacturers use low molecular weight phthalates (e.g., diethyl phthalate [DEP] and dibutyl phthalate [DBP]) in personal-care products (e.g., perfumes, lotions, cosmetics), as solvents and plasticizers for cellulose acetate, and in making lacquers, varnishes, and coatings, including those used to provide timed releases in some pharmaceuticals.[14,15]

The ubiquitous use of phthalates results in human exposure via dietary ingestion, dermal absorption of low molecular weight phthalates (e.g., DEP and DBP), inhalation of the more volatile phthalates, and parenteral exposure from medical devices containing phthalates, primarily DEHP.[12] Because humans and other mammals rapidly metabolize phthalate diesters to their respective monoesters, phthalates do not bioaccumulate.[12,14,15] Biological half-lives of phthalates are on the order of 1 day or less, and hence urinary biomarkers represent exposure for no more than the 1 or 2 days that precede collection of the urine specimen. Urinary phthalate monoester metabolites are the preferred biomarkers of exposure. By measuring the phthalate metabolites, contamination from the ubiquitous parent compounds (i.e., the diesters) is minimized, thus allowing for the study of environmentally exposed populations. Another advantage of using the phthalate monoesters as biomarkers is that the monoesters are generally considered the biologically active molecules.

In contrast to the experimental data on the reproductive and developmental toxicity of phthalates, the epidemiologic data are very limited (summarized in Table 1). Human studies are currently limited to cross-sectional studies in which adult exposure levels were measured and their relationships with semen parameters were explored. In one of the early studies, Murature et al[16] recruited 21 university students to explore the relationship between sperm concentration and DBP concentrations in the cellular fractions of ejaculates. They reported an inverse relationship between sperm concentration and DBP. However, the study was small and did not measure or adjust for potential confounders.

In India, Rozati et al[17] studied 21 infertile men with poor semen quality and 32 control men with normal semen parameters. Phthalate esters were measured in seminal plasma and the results were reported as the sum of a mixture of dimethyl phthalate (DMP), DEP, DBP, butylbenzyl phthalate (BBzP), DEHP, and DnOP. The concentration of phthalates was inversely correlated with sperm morphology ($r = -0.77$; $p < 0.001$) but not with ejaculate volume, sperm concentration, or motility. Because diesters were measured, sample contamination is a potential concern in this study.

Duty et al[18,19] have published two articles exploring the relationships between environmental exposure to phthalates and semen characteristics among male partners of subfertile couples that presented to an infertility clinic in Massachusetts. Exposure was assessed by measuring urinary levels of phthalate monoester metabolites. Among 168 men, they found dose–response relationships (after adjusting for age, abstinence time, and smoking status) between monobutyl phthalate (MBP, metabolite of DBP and a minor metabolite of BBzP) and sperm motility (odds ratios [ORs] per tertile: 1.0, 1.8, 3.0; p for trend $= 0.02$) and sperm concentration (OR per tertile: 1.0, 1.4, 5.5; p for trend $= 0.07$). They also found a dose–response relationship between monobenzyl phthalate (MBzP, metabolite of BBzP) and sperm concentration (OR per tertile: 1.0, 1.4, 5.5; p for trend $= 0.02$). There was weak evidence of an association between MBP and sperm morphology, MBzP and sperm motility, and monomethyl phthalate (metabolite of DMP) and sperm morphology, respectively. Among 220 men, MBP, MBzP, and mono(2-ethylhexyl) phthalate (MEHP, metabolite of DEHP) had inverse associations, although not significant, with straight-line velocity (VSL), curvilinear velocity (VCL), and linearity (LIN; LIN $=$ VSL/VCL $\times$ 100) measured by computer-assisted semen analysis (CASA). Unexpectedly, positive relationships were found between MEP and both VSL and VCL.

Table 1 Summary of Epidemiologic Studies (in chronological order) on the Relationship between Phthalates and Semen Quality

Author, Country	Study Population	Exposure	Results	Comments
Murature et al.,[16] 1987; United States	21 young men	DBP in ejaculate cellular fraction	Inverse relationship between sperm concentration and DBP ($r = -0.4$)	Small sample size, no adjustment for confounders
Rozati et al.,[17] 2002; India	53 men (21 infertile and 32 controls)	Phthalate diesters in seminal plasma (DBP, BBzP, DEHP, DnOP)	Sum of phthalates was inversely correlated with sperm morphology ($r = -0.77$; $p < 0.001$), but not with semen volume, sperm concentration, or motility	Measured total phthalate diesters, thus concern with sample contamination
Duty et al.,[18,19] 2003, 2004; United States	168 men from an infertility clinic	Urinary levels of phthalate metabolites (MBP, MBzP, MEP, MEHP)	Adjusted dose–response relationships between MBP and sperm motility (OR per tertile: 1.0, 1.8, 3.0; p for trend = 0.02) and sperm concentration (OR per tertile: 1.0, 1.4, 5.5; p for trend = 0.07). Adjusted dose–response relationship between MBzP and sperm concentration (OR per tertile: 1.0, 1.4, 5.5; p for trend = 0.02). MBP, MBzP, and MEHP inverse nonsignificant associations with straight line velocity (VSL), curvilinear velocity (VCL), and linearity (LIN; LIN = VSL/VCL x 100)	Confounders considered: age, BMI, abstinence time, smoking status, race
Jonsson et al.,[20] 2005; Sweden	234 young men	Urinary levels of phthalate metabolites (MEP, MEHP, MBzP, MBP, and phthalic acid)	No relationships of MBP, MBzP, or MEHP with any of the semen parameters. The highest quartile for MEP had fewer motile sperm (mean difference, 8.8%; 95% CI, 0.8–17) and more immotile sperm (8.9%; 95% CI, 0.3–18). Phthalic acid was associated with improved function as measured by more motile sperm and fewer immotile sperm	Confounders considered: abstinence time, smoking status

DEHP, di(2-ethylhexyl) phthalate; MEHP, monoethylhexyl phthalate; DiNP, di-isononyl phthalate; DnOP, di-n-octyl phthalate; BMI, body mass index; DEP, diethyl phthalate; MEP, monoethyl phthalate; BBzP, butylbenzyl phthalate; DBP, dibutyl phthalate; MBP, monobutyl phthalate; CI, confidence interval.

In a recently published study from Sweden, Jonsson et al[20] recruited 234 young Swedish men at the time of their medical conscript examination. Each man provided a single urine sample used to measure concentrations of MEP, MEHP, MBzP, MBP, and phthalic acid. Urinary phthalate levels were divided into quartiles and were used to calculate the mean difference and 95% confidence interval (CI) between the lowest and highest quartiles. Because multivariate adjusted and unadjusted results differed by less than 15%, potential confounders, such as abstinence time and smoking status, were not retained in the models. In contrast to the U.S. study,[18] there were no relationships of MBP or MBzP with any of the semen parameters. MEHP was also not associated with any of the semen parameters. Men in the highest quartile for MEP had fewer motile sperm (mean difference, 8.8%; 95% CI, 0.8 to 17) and more immotile sperm (mean difference, 8.9%; 95% CI, 0.3 to 18) than men in the lowest MEP quartile. Contrary to their hypothesis, phthalic acid was associated with more motile sperm and fewer immotile sperm. Phthalic acid is a nonspecific marker of phthalate exposure, formed as the result of the hydrolysis of any of the phthalates measured. Interactions between urinary phthalate levels and PCB 153 (measured previously in serum samples from these men) were assessed by including an interaction term in the models. There was no evidence of multiplicative interactions between PCB 153 and any of the phthalates with the semen parameters. This is in contrast to a previous study by Hauser et al,[21] in which they found interactions of MBP and MBzP with PCB 153 in relation to sperm motility among the same men from the infertility clinic described in the study by Duty et al.[18]

Although the Swedish study had some similarities in design and execution to the U.S. study, there were many differences. The population in the Swedish study consisted of young men (median age, 18 years; range, 18 to 21 years) who were undergoing a medical examination before military service. Given that ~95% of young men in Sweden undergo the conscript examination, these young men reflected the general population of young Swedish males. In contrast, in the U.S. study, the median age of the men recruited from an infertility clinic was 35.5 years and ranged from 22 to 54 years. None of the men from the infertility clinic were 21 years of age or younger. The recruitment of men from an infertility clinic compared with young men from the general population may account for some of the differences in results between studies. For instance, it is unclear whether men presenting to an infertility clinic are more susceptible to reproductive toxicants, including phthalates, than men from the general population. Furthermore, it is also unclear whether middle-aged men, compared with young men, are more susceptible to reproductive toxicants because of an age-related response to the toxicant.

Although only 14% of the young Swedish men, compared with 65% of men in the U.S. study, agreed to participate, it is unlikely that the young Swedish men did so differentially in relation to reproductive function and phthalate levels. Therefore, selection bias as a result of the low participation rate is unlikely in the Swedish study.

Despite similarities in urinary concentrations of the phthalate monoesters across studies, the analytical methods differed between the Swedish and U.S. study. The detection limits for MEP, MBP, MBzP, and MEHP in the Swedish study were 30, 15, 7, 15 ng/mL, respectively, many-fold higher than the detection limits (~1 ng/mL) in the U.S. study. The higher limits of detection in the Swedish study may contribute to measurement error of urinary phthalate levels and may result in bias to the null hypothesis. However, by categorizing the phthalate levels into quartiles for the statistical analysis, potential measurement error resulting from high detection limits may be minimized. Another difference between studies is that the Swedish study used urinary creatinine to adjust for urine dilution compared with specific gravity (SG) in the US study. In the Swedish study, the median creatinine-adjusted phthalate metabolite concentrations were quite different from the unadjusted values. In contrast, in the U.S. study, medians between SG-adjusted and unadjusted values were not markedly different. The role, if any, that the methods used for urine dilution adjustment may play in explaining differences across studies is unclear.

The statistical methods used for the data analysis also differed between studies. In the U.S. study, multivariate logistic regression with categorized semen parameters was used as the primary outcome. Men with all three semen parameters above the reference range were used as comparison subjects in these analyses. In contrast, in the Swedish study, for the primary analysis, semen parameters were used as a continuous measurement and mean differences between men in the highest and lowest phthalate quartiles were calculated. In addition, logistic regression analyses were performed, although the results of these analyses, reported to be consistent with their primary analyses, were not shown. However, it is unclear whether the comparison group in the logistic regression analyses included only men with all three semen parameters above the reference range. If not, dilution of associations between phthalates and semen parameters may occur because the comparison group does not consist of a homogenous group of men with normal semen parameters. For instance, dilution of the associations between sperm concentration and phthalate monoesters may occur if phthalates alter both sperm concentration and motility.

In conclusion, the epidemiologic data on the relationship between semen quality and phthalate exposure remain limited and inconsistent. Although the

two recent studies by Duty et al[18,19] and Jonsson et al[20] had similarities, important differences existed. The US study recruited older men from an infertility clinic while the Swedish study recruited young men from the general population. It is currently unclear whether these differences in age and recruitment source may partially account for the inconsistent results across studies, especially for MBP and MBzP. Additional studies are critically needed to help elucidate possible explanations for differences across studies, and whether phthalate exposure alters semen quality.

POLYCHLORINATED BIPHENYLS

PCBs are a class of synthetic, persistent, lipophilic, halogenated aromatic compounds that were widely used in industrial and consumer products for decades before their production was banned in the United States in the late 1970s. PCBs were used in cutting oils, lubricants, and as electrical insulators. As a result of their extensive use and persistence, PCBs remain ubiquitous environmental contaminants. They are distributed worldwide and have been measured in air, water, aquatic and marine sediments, fish, and wildlife.[22] Furthermore, they are biologically concentrated and stored in human adipose tissue. The general population is exposed primarily through ingestion of contaminated foods (e.g., fish, meat, and dairy products); PCBs can bioaccumulate up the food chain. However, exposure may also occur through dermal contact (soil and house dust) and inhalation (indoor air in residential buildings and workplaces, as well as outdoor air). As a result of their persistence and ubiquity, measurable levels of serum PCBs are found in the majority of the U.S. general population.[23] Serum levels of PCBs are an integrated measure of internal dose, reflecting exposure from all sources over the previous years; depending on the congener, the half-life of PCBs in the blood ranges from 1 to 10 or more years.[24]

Studies on Background Environmental Exposure

The epidemiologic data on the relationship between PCBs and semen quality is summarized in Table 2. In an early study on PCBs and semen quality, Bush et al[25] studied fertile men (n = 33), men with oligozoospermia (n = 50) or azoospermia (n = 50), and men after vasectomy (n = 25). The average (standard deviation [SD]) age of the men from these groups were 33 (7), 32 (4), 33 (5), 38 (7) years, respectively. The seminal concentrations of PCBs 153, 138, and 118 were inversely related to sperm motility only among semen samples with a sperm count less than 20 million/mL. The authors cautioned against overinterpreting these associations because they were found only among a subset of subjects.

In the Netherlands, Dallinga et al[26] studied the relationship between PCBs and semen quality among men who were partners in couples visiting an infertility treatment center. They identified two groups of men, those men with good semen quality (n = 31) and men with very poor semen quality (n = 34) based on three semen samples. Progressive motile sperm concentration was used to make the classification. Blood and semen were analyzed for PCBs 118, 138, 153, and 180 and their hydroxylated metabolites. The mean (SD) non–lipid-adjusted levels of PCB 153 were 0.41 (0.22) ng/g blood. Assuming that serum percent lipid is ~0.5%, the estimated lipid adjusted concentration would be 82 ng/g lipids. Contrary to expectations, the sum of PCBs in seminal plasma of men with good semen quality were higher than among men with poor semen quality (0.071 and 0.022 ng/mL seminal plasma, respectively; $p = 0.06$). However, within the group of men with good semen quality, there were inverse associations between serum levels of sum of PCB metabolites and sperm count ($p = 0.04$) and progressive motile sperm concentration ($p = 0.02$). There were also inverse nonsignificant corresponding associations in the men with poor semen quality. Because associations with semen quality were found for PCB metabolites and not the parent PCBs, these results suggested that the PCB metabolites were the biologically active compounds.

Richthoff et al[27] conducted a study on the relationship between PCB 153 and semen parameters among 305 young men undergoing a conscript examination for military service. The men ranged in age from 18 to 21 years (median age, 18.1 years). PCB 153 levels were considered representative of background environmental levels for men from Southern Sweden; the median was 65 ng/g lipid (range, 23 to 250 ng/g lipid). The following confounders were considered for inclusion in the models: body mass index (BMI), abstinence period, and smoking status. There were significant inverse associations between PCB 153 and percent motile sperm; a 10 ng/g lipid increase in PCB 153 was associated with a 1.0% decline in percent CASA motile sperm (95% CI, −2.0 to −0.13). The association between PCB 153 and conventional sperm motility was slightly weaker. There were no associations between PCB 153 and sperm concentration or total sperm count. The study was relatively large and well conducted. Although the participation rate was very low (only 13.5% of eligible subjects agreed to participate), it is unlikely that this would introduce bias because young men are likely to be unaware of their fertility or exposure levels.

Rozati et al[17] measured PCBs in seminal plasma and explored relationships with semen parameters among men in India. Details of the study are provided in the Phthalates section. PCBs were detected in the

seminal plasma of infertile men but not controls. They reported a negative correlation between seminal plasma PCB levels and total progressive motility ($r = -0.5$). No correlations were found among PCBs and sperm count, rapid progressive motility, or normal morphology. The authors reported results for total PCBs and not for individual congeners. Potential confounders were considered in the methods section but no statistical adjustments were made.

Hauser et al[28] conducted a study on 212 male partners of subfertile couples visiting an infertility clinic in Massachusetts. The mean (SD) age was 36.0 (5.4) years. Fifty-seven PCB congeners were measured, and included PCB 118, 138, 153, and 180. The median level for PCB 153 was 42 ng/g lipid (range, 9.3 to 361 ng/g lipid). Multivariate logistic regression analyses were used in which semen parameters were dichotomized based on World Health Organization reference values (1999).[29] The comparison groups for each analysis were defined as men with all three semen parameters above reference values. There were significant dose–response relationships (OR per tertile adjusted for age, abstinence time, and smoking status) between PCB 138 and below-reference sperm motility (1.00, 1.68, 2.35, respectively; p for trend $= 0.04$) and sperm morphology (1.00, 1.36, 2.53; $p = 0.04$). Associations between semen parameters and PCB 153 were not consistent.

Rignell-Hydbom et al[30] reported on the associations between PCBs with semen parameters among Swedish fishermen from the east and west coasts; 195 Swedish fishermen (median age, 50.6 years; range, 24 to 65 years) participated. The median serum level of PCB 153 was 193 ng/g lipid (range, 39 to 1460 ng/g lipid). When PCB 153 was categorized into quintiles, the highest quintile had decreased sperm motility compared with men in the lowest quintile. The age-adjusted mean difference was 9.9% (95% CI, −1.0 to 21%; $p = 0.08$). There were no consistent associations of PCB 153 with sperm concentration.

High PCB Exposure Studies

Guo et al[31] studied the relationship between semen quality and prenatal exposure to PCBs and polychlorinated dibenzofurans (PCDFs) after the poisoning episode in Taiwan in 1979, in which PCB-contaminated rice oil was ingested. In 1998, 12 men pre-natally exposed to contaminated rice oil and 23 healthy unexposed subjects of comparable age provided a semen sample. The unexposed men had no unusual chemical exposure and were recruited from a local high school. The mean (SD) age of the exposed men was 17.3 (1.2) years and 17.6 (1.0) for the unexposed men. The proportion of sperm with abnormal morphology was increased in the exposed men (37.5 versus 25.9% for unexposed men). In the exposed men the percentage of motile sperm (35.1 versus 57.1% in unexposed men) and rapidly motile sperm (25.5 versus 42.4% in unexposed men) were reduced. Several of the CASA parameters were reduced in the exposed men, in particular, including average path velocity, VSL, and VCL. Sperm from exposed men had reduced hamster oocyte penetration compared with unexposed men. This small study provided the opportunity to explore high prenatal exposure to PCBS and PCDFs.

In a second study on men from the Taiwan PCB poisoning, Hsu et al[32] studied the relationship between semen quality and levels of PCBs among men who had consumed contaminated rice oil some 20 years earlier. They identified 40 exposed men and 28 unexposed men who were matched using an address registry. Mean ages of exposed (37.9 years) and unexposed (40.4) men were similar. Exposed men had a higher percentage of sperm with abnormal morphology (27.5 versus 23.3%) and a higher oligozoospermia rate (9 versus 1%). The ability of sperm to penetrate the hamster oocyte was reduced in exposed men. The results of this small study provide evidence of adverse effects of exposure to PCBs and PCDFs among men exposed 20 years earlier to the contaminated rice oil.

In conclusion, the data on the relationship between PCBs and semen quality support an inverse association of PCBs with reduced semen quality, specifically reduced sperm motility. The observed associations generally were consistent across studies performed in different countries (India, Netherlands, Taiwan, Sweden, and United States) that used different methods to measure semen quality and PCB exposure. Furthermore, associations were consistently found despite a range of PCB levels; that is, there did not appear to be a threshold. The PCB levels in these studies ranged from low background levels,[26–28] to high background levels due to consumption of contaminated fish,[30] to even higher exposure levels due to ingestion of accidentally contaminated rice oil.[31,32] Although the data across studies generally support a relationship between PCBs and poor semen quality, there are possible alternative explanations. One potential alternative explanation is that PCBs are a surrogate for exposure to other environmental factors that may predict semen quality. Although this is possible, there currently is no evidence identifying potential alternative exposures. Another explanation is that there may be confounding of the associations by some currently unrecognized or unmeasured confounders. Although possible, this is also unlikely because the more recent studies considered important potential confounders and the results were consistent across studies, suggesting that it is unlikely that there is a strong unmeasured confounder. In conclusion, although PCBs are no longer used, these data, along with ongoing human exposure, albeit at lower levels than several

Table 2 Summary of Epidemiologic Studies (in chronological order) on the Relationship between Polychlorinated Biphenyls (PCBs) and Semen Quality

Author	Study Population	Exposure	Results	Comments
Bush et al.,[25] 1986	33 fertile, 50 subfertile, 50 infertile, and 25 postvasectomy men	Seminal plasma levels of PCBs	PCB 153, 138, and 118 were inversely related to sperm motility only among samples with a sperm count less than 20 million/mL	Association found only among a subset of men
Rozati et al.,[17] 2002	53 men from India (21 infertile and 32 controls)	Seminal plasma levels of PCBs	Negative correlation between PCBs and total progressive motility ($r = -0.5$). No correlations of PCBs with sperm count, rapid progressive motility, or normal morphology	No data on individual PCB congeners; no statistical adjustment for potential confounders
Dallinga et al.,[26] 2002	65 Dutch men from an infertility clinic	Serum and semen levels of PCB 118, 138, 153, 180 and their metabolites	In men with good semen quality, there were inverse associations between serum levels of sum of PCB metabolites and sperm count ($p = 0.04$) and progressive motile sperm concentration ($p = 0.02$). There were also negative nonsignificant corresponding associations in men with poor semen quality	Confounders considered: age and smoking status; measured PCB metabolites
Richthoff et al.,[27] 2003	305 Swedish young men	Serum levels of PCB 153	Inverse association between PCB 153 and percent motile sperm (10 ng/g lipid increase in PCB 153 associated with a 1.0% decline in percent motile sperm; (95% CI, −2.0 to −0.13). No association of PCB 153 with sperm concentration	Confounders considered: BMI, abstinence period, smoking status
Hauser et al.,[28] 2003	212 US men from an infertility clinic	Serum levels of PCBs and p,p′-DDE	Dose–response relationships (odds ratio per tertile adjusted for age, abstinence time, and smoking status) between PCB 138 and below reference sperm motility (1.00, 1.68, 2.35, respectively; p for trend = 0.04) and sperm morphology (1.00, 1.36, 2.53; $p = 0.04$). DDE had a nonsignificant association with sperm motility	Confounders considered: BMI, abstinence time, age, smoking status
Rignell-Hydbom et al.,[30] 2004	195 Swedish fishermen	Serum levels of PCB 153 and p,p′-DDE	The highest PCB 153 quintile had decreased sperm motility as compared with men in the lowest quintile. The age adjusted mean difference was 9.9% (95% CI, −1.0 to 21%; $p = 0.08$). No significant associations of p,p′-DDE with semen parameters	Confounders considered: age, smoking status, abstinence time, BMI, reproductive hormones
High Exposure Studies				
Guo et al.,[31] 2000	35 young men from Taiwan (12 prenatally exposed to contaminated rice oil, 23 unexposed men)	Maternal ingestion (yes/no) of rice oil contaminated with PCBs and PCDFs	Increased percent abnormal morphology in exposed men (37.5%) compared with unexposed men (25.9%). Exposed men had decreased percentage of motile sperm (35.1 compared with 57.1% in unexposed men) and rapidly motile sperm (25.5 compared with 42.4% in unexposed men). Reduced hamster oocyte penetration in exposed men	Age and percentage of smokers in exposed and unexposed groups were similar; no statistical adjustment for confounders

Hsu et al.,[32] 2003	68 men from Taiwan (40 exposed to contaminated rice oil and 28 unexposed)	Ingestion (yes/no) of rice oil contaminated with PCBs and PCDFs	Exposed men had higher percentage of sperm with abnormal morphology (27.5%) compared with unexposed men (23.3%), and a higher oligospermia rate (9 compared with 1%, respectively). Ability of sperm to penetrate the hamster oocyte was reduced in exposed men	Age and percentage of smokers in exposed and unexposed groups were similar; no statistical adjustment for confounders

BMI, body mass index; p,p′-DDE, p,p′-dichlorodiphenyl-dichloroethene; PCDFs polychlorinated dibenzofurans.

decades ago, raise concerns regarding altered human fertility due to adverse affects on semen quality.

NONPERSISTENT PESTICIDES

The term contemporary-use pesticides refers to chemical mixtures that are currently available to control insects (insecticides), weeds (herbicides), fungi (fungicides), or other pests (e.g., rodenticides), as opposed to pesticides that have been banned from use in most countries (e.g., many of the formerly popular organochlorine pesticides such as dichlorodiphenyltrichloroethane [DDT]). Although environmentally nonpersistent, because of the extensive use of pest control in these various settings, a majority of the U.S. general population is exposed to some of the more widely used pesticides at low levels.

There are several epidemiologic studies on men exposed to contemporary-use pesticides during agricultural work. A cross-sectional study measured sperm concentration, motility, and morphology in 122 greenhouse workers categorized into groups with low, medium, or high exposure to more than a dozen pesticides.[33] After adjustment for abstinence time and other potential confounders, a higher proportion of abnormal sperm was found in the group with high exposure compared with the group with low exposure. Lower median sperm concentration was also observed in workers with more than 10 years of work in the greenhouse compared with men with less than 5 years of experience. In a cross-sectional study on traditional and organic farmers, Juhler et al[34] investigated the relationship between dietary exposure to pesticides and semen quality. Estimating exposure through food frequency questionnaires and data from pesticide monitoring programs, the authors found that men with a lower intake of organic food had a lower proportion of normal shaped sperm according to the strict criteria (2.5 versus 3.7%, $p = 0.003$). However, organic food intake was not associated with the other 14 semen parameters measured in the study. Results in the study were adjusted for age, urogenital disease, spillage, abstinence time, smoking, and alcohol intake. Oliva et al[35] recently investigated the impact of environmental factors on infertility among 177 men in Argentina. Adjusting for age, BMI, abstinence time, income, health center, and smoking, a dose-related response was observed in (primary) infertile men occupationally exposed to pesticides. Significantly elevated odds ratios (ORs) were reported for sperm concentration (less than 1×10^6/mL; OR = 3.4; 95% CI, 1.2 to 7.4), motility (< 50% motile; OR = 3.6, 95% CI, 1.1 to 11.4), and morphology (< 30% normal; OR = 4.1, 95% CI, 1.4 to 12.0) for men exposed to pesticides compared with occupationally nonexposed men. Conversely, in models adjusting for many of the same variables, Larsen et al[36] found only marginal differences among 15 semen quality parameters from Danish farmers who sprayed pesticides compared with farmers who did not spray pesticides. Although these studies reported associations between pesticide exposure and human semen quality, the nonspecific assessment of pesticide exposures makes it impossible to determine which pesticides, if any, were responsible for the observed associations.

Few studies have been conducted that provide information on specific chemicals or classes of contemporary-use pesticides and altered testicular function (Table 3). Padungtod et al[37] studied the relationship between occupational exposure to organophosphates (parathion and methamidophos) and testicular function among Chinese pesticide factory workers. They found a significant reduction in adjusted mean sperm concentration (28.5 versus 49.4 million sperm/mL; $p = 0.01$), and percentage of motile sperm (64 versus 74%; $p = 0.03$) in the 20 exposed workers compared with the 23 unexposed workers. In a recent Japanese study, pesticide sprayers exposed primarily to organophosphates and pyrethroids showed spraying season-dependant reductions in motile sperm velocity measures compared with unexposed controls.[38]

Two publications reported the results from a study on a small cohort of men exposed to carbaryl (1-naphthyl methyl carbamate [commonly known as Sevin]) during the production and packaging of the insecticide.[39,40] Although analyses using sperm counts as a continuous measure failed to find significant differences based on carbaryl exposure, the authors found a greater proportion of oligozoospermic men among the carbaryl workers compared with the chemical workers.[39] In a subsequent publication on the same cohort of carbaryl production workers, Wyrobek et al[40] studied the relationship between sperm shape abnormalities and carbaryl. Morphological analyses showed an elevated percent of abnormal sperm in carbaryl workers compared with comparison subjects, which remained after stratifying on potential confounders such as smoking, medical history, or previous exposure to hazardous agents. The proportion of men defined as teratozoospermics (greater than 60% abnormal sperm) was higher among the carbaryl workers than in the comparison group (28.6 and 11.8%, respectively).

More recently, researchers have used biomarkers of exposure to pesticides in blood or urine to explore associations with reduced semen quality. In a U.S. study on the male partners of pregnant women, Swan et al[41] compared urinary levels of pesticide biomarkers in 34 men with sperm concentration, motility, and morphology below the median (cases) versus 52 men with above-median semen parameters (controls). They found elevated ORs for alachlor mercapturate (OR = 30.0; 95% CI, 4.3 to 210), 2-isopropoxy-4-methyl-pyrimidinol (diazinon metabolite; OR = 16.7; 95% CI, 2.8 to 98)], atrazine mercapturate (OR = 11.3; 95% CI, 1.3 to 99), 1-naphthol (carbaryl and naphthalene metabolite;

Table 3 Summary of Epidemiologic Studies (in chronological order) on the Relationship between Contemporary-Use Pesticides and Semen Quality

Author, Country	Study Population	Exposure	Results	Comments
Whorton et al.,[39] 1979; United States	47 carbaryl production workers; 90 unexposed chemical workers	Subjective exposure classification based on job tasks	Greater proportion of oligozoospermic men among the carbaryl workers (15%) as compared with the chemical workers (5.5%; $p = 0.07$)	No adjustment for potential confounders; sperm motility not measured
Wyrobek et al.,[40] 1981; United States	50 carbaryl production workers, 34 unexposed comparison men	Exposure ranks/groups based on job for previous year	Elevated percent of abnormal sperm in carbaryl workers (52%) as compared with comparison subjects (42%; $p < 0.005$). The proportion of men defined as teratozoospermics (greater than 60% abnormal sperm) was higher among the carbaryl workers (28.6%) than in the comparison group (11.8%; $p = 0.06$)	Confounders considered: smoking, medical history, previous exposure to hazardous agents
Padungtod et al.,[37] 2000; China	43 Pesticide factory workers; 20 high exposed and 23 with no or very low exposure	Occupational exposure to ethyl parathion and methamidophos	Exposure associated with reduction in sperm concentration and motility, but not sperm morphology. Adjusted means for exposed and non-exposed workers were 28.5 and 49.4 million sperm/mL ($p = 0.01$), respectively, for sperm concentration; and 64% and 74% ($p = 0.03$), respectively, for percentage of motile sperm	Confounders considered: age, abstinence period, current smoking status
Swan et al.,[41] 2003; United States	86 male partners from couples attending prenatal clinic	Urinary levels of pesticides or metabolites (IMPY, 2,4-D, 1N, TCPY, others)	Elevated odds ratios (95% CI) for below reference semen parameters associated with high- exposure groups for alachlor mercapturate 30.0 (4.3–210); IMPY 16.7 (2.8–98); atrazine mercapturate 11.3 (1.3–99); 1-naphthol 2.7 (0.2–34); and TCPY 6.4 (0.5–86)	Small study size contributed to wide confidence intervals; odds ratios were unadjusted for potential confounders
Meeker et al.,[42] 2004; United States	272 males from couples attending infertility clinic	Urinary levels of insecticide metabolites (1N, TCPY)	Inverse association between urinary carbaryl metabolite (1-naphthol) and sperm concentration, motility. IQR increase in 1N associated with 16% decline in sperm concentration and 3.8% decline in motile sperm. Suggestive inverse association between chlorpyrifos metabolite (TCPY) and sperm motility	Confounders considered: age, BMI, abstinence time, smoking status, race, season

IMPY, 2-isopropoxy-4-methyl-pyrimidinol; 1N, 1-naphthol; TCPY, 3,5,6-trichloro-2-pyridinol; CI, confidence interval; BMI, body mass index.

OR = 2.7; 95% CI to 0.2, 34), and 3,5,6-trichloro-2-pyridinol (TCPY; chlorpyrifos metabolite; OR = 6.4; 95% CI to 0.5, 86).

Using urinary biomarkers for exposure to low environmental levels of pesticides commonly encountered among the general population, Meeker et al[42] studied 272 men who were partners of an infertile couple. They found inverse associations between urinary levels of 1-naphthol, a metabolite of both carbaryl and naphthalene, with sperm concentration and motility. They also found a suggestive inverse relationship between the urinary metabolite of chlorpyrifos and sperm motility. Compared with the lowest tertile, the ORs for medium and high tertiles of 1-naphthol were OR = 4.2 (95% CI, 1.4 to 13.0) and OR = 4.2 (95% CI, 1.4 to 12.6) for below reference sperm concentration (< 20 million/mL), and OR = 2.5 (95% CI, 1.3 to 4.7) and OR = 2.4 (95% CI, 1.2 to 4.5) for below reference sperm concentration (< 50% motile sperm). In multiple linear regression analyses, an interquartile range increase in 1-naphthol (1.8 to 5.0 μg/L) was associated with a 3.9% (95% CI; −7.3 to −0.5%) decline in motile sperm and a 16% (95% CI; −29 to +1.0%) decline in sperm concentration. An interquartile range increase in chlorpyrifos metabolite (TCPY; also 1.8 to 5.0 μg/L) was associated with a 2.2% (95% CI; −5.1 to +0.7%) decline in motile sperm.

In summary, there are human data supporting an association between contemporary-use pesticide exposure and altered semen quality, though it is somewhat limited. Although the human data are suggestive, they are mostly derived from occupational studies involving simultaneous exposure to several pesticides. The relationship between reproductive health and exposure to specific contemporary-use pesticides, and/or low-level mixed exposures through diet and residential use among the general male population, are not well understood.

CONCLUSIONS

The limited human data on these emerging environmental contaminants, and in certain instances inconsistent data across studies, highlight the need for further epidemiological research on these contemporary-use emerging chemicals. In addition to the chemicals discussed in this article, there are additional classes of chemicals that require further study regarding their relation with human semen quality. These chemicals include alkylphenols such as 4-nonylphenol, bisphenol A (BPA), and fluorinated organic compounds such as perfluorooctane octanoate and perfluorooctane sulfonate. Alkylphenols are used as surface-active agents in cleaning/washing agents, paints, and cosmetics, whereas BPA is used in the manufacture of polycarbonate plastics and epoxy resins. The perfluorinated compounds are used to make fabrics stain resistant/water repellant and in coatings on cookware and other products. Although human exposure to these chemicals has been demonstrated, the data on health effects in humans remain very limited.

In framing the future research agenda on the relationship between environmental chemicals and male fertility, the following should be considered. Research should be directed toward (1) the study of chemicals that have the opportunity for widespread human exposure; (2) the study of subpopulations with high exposure; (3) the identification of vulnerable subpopulations, which is critical to our understanding of risk and the protection of public health; (4) the study of how exposure-response differs across life stages (e.g., in utero versus peripubertal versus adult exposure); and (5) research on the underlying mechanisms by which chemicals may alter human fertility.

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