

**Supplementary Table 1: Guideline values for risk assessment of BaP from various subnational, national and international agencies.**

Agency	Category	Value µg/L	Method	Basis for value
WHO (2003)	Health-based guideline	0.7	Oral carcinogenicity in mice using two-stage mutation model. Neal and Rigdon (1967) forestomach tumors	Oral cancer in mice
Cal/EPA (2010)	Public health goal	0.007	Time to tumor model to obtain a lower-bound estimate of dose associated with 10% increased incidence of tumors. Assumed a linear dose-response relationship at low doses. Increased potency factor by 1.7 to correct for early-life exposure. Exposure of 0.052 L/kg bw per day of water (an age-adjusted upper 95th percentile of drinking water consumption for age range for cancer potency correction). q* Cancer potency 1.7 (mg/kg bw per day) <sup>-1</sup> [2.9 (mg/kg bw per day) <sup>-1</sup> when corrected for early-life exposure]	Incidence of oral cavity or forestomach tumors in female mice (Kroese et al., 2001)
Cal/EPA (2010)	Health protective level	4	LOAEL of 5 mg/kg bw per day from subchronic study. Uncertainty factor of 3000 and RSC of 0.1 and drinking water ingestion of 0.044 L/kg bw per day.	Renal toxicity (Knuckles et al., 2001)
U.S. EPA (1994; 2007) Currently under review	Maximum contaminant level	0.2	Used Neal and Rigdon (1967) and Rabstein (1973); both incidence of mouse forestomach tumors from feeding study	Reproductive difficulties; increased risk of cancer (stomach)
NHMRC (2004)	Health-based guideline	0.01	Based on limit of detection, which is slightly less than value derived using a risk assessment from WHO (2003) using Neal and Rigdon (1967).	Mouse tumors
Health Canada (1988, reaffirmed in 2005)	Health-based guideline	0.01	Increased stomach tumors in Neal and Rigdon (1967) feeding study in mice. Used surface area correction and robust linear extrapolation model to estimate lifetime risk associated with 1 µg/L BaP in drinking water of $5 \times 10^{-5}$	Mouse stomach tumors

Supplementary Table 2: All epidemiology studies evaluated.

Study	Case-control/cohort	Exposure route	N	Endpoint	BaP concentration	Outcome	Duration of exposure	Association with BaP
<b>Inhalation</b>								
Arif et al. (2006)	Case-control	Cigarette smoking (comparison of human tissues from cancer patients)	50	Highly lipophilic DNA adducts	Unknown	DNA adducts are present, but are not related to PAHs or BaP	Chronic	Negative
Armstrong et al. (1986)	Case-control	Inhalation among aluminum smelter workers	85 cases and 255 referents	Bladder cancer	1 mg/m <sup>3</sup>	2.3% increase in bladder cancer	Chronic	Positive
Armstrong et al. (1994)	1) Case-cohort; 2) Subcohort	Inhalation in aluminum production plant	1) 338 2) 1138	Lung cancer mortality	1) cumulative < 10, 10-99, 100-199, 200-299, ≥ 300 μg/m <sup>3</sup> -years 2) 342.2 μg/m <sup>3</sup> / 190.1 μg/m <sup>3</sup>	1) Smoking-adjusted rate ratio 1.00, 1.48, 2.23, 2.10, 1.87; significance not reported 2) probability of causation over 50%; 2) probability of causation over 50% first achieved and according to the upper 95% confidence limit	≥ 1 year between 1950 and 1979	Positive
Armstrong and Thériault (1996)	Cohort	From compensation claims from lung cancer patients exposed to coal tar pitch volatiles (occupational exposure)	Not reported	Lung cancer	1) 100 μg/m <sup>3</sup> -years 2) 342.2 μg/m <sup>3</sup> / 190.1 μg/m <sup>3</sup>	1) probability of causation over 50%; 2) probability of causation over 50% first achieved and according to the upper 95% confidence limit	Chronic	Positive
Armstrong and Gibbs (2009)	Cohort	Inhalation from aluminum smelters in Quebec	16 431	Lung cancer	100 μg/m <sup>3</sup> BaP-years of cumulative exposure was	Relative risk = 1.35 (95% CI 1.22-1.51)	at least 1 year of employment	Positive
Bartsch et al. (1998)	Case-control	Lung cancer patients vs. controls	N/A	BPDE-DNA adducts (also PAH DNA adduct)	Unknown	CYP1A1 mutation increases BPDE-DNA adducts	chronic	Positive
Burstyn et al. (2005)	Cohort	Inhalation in asphalt workers across Europe	12 367 males	Ischemic heart disease mortality	Highest exposure group was 273+ ng/m <sup>3</sup> and 2013+ ng/m <sup>3</sup> for cumulative exposure	Significant trend with dose and cumulative exposure; 1.64 relative ratio for average exposure and 1.58 in the cumulative exposure	Minimum of one work season	Positive
Cross et al. (2010)	Cohort	Meat and meat component intake as well as meat cooking by-products	Not reported	Esophageal and gastric cancer	Increased risk estimated for each additional 10 ng/d BaP	No relation for BaP; increased cancer risk was observed for meat consumption	Chronic	Negative
De Flora et al. (1993)	Cohort	Smoking	39 for adducts and 31 for MN	BPDE-DNA adducts and micronuclei (MN not specific to BaP)	Not defined	Not significant for MN; however, BPDE DNA adducts were significant according to number of cigarettes smoked per day	Not defined	Positive
De Stefani et al. (2009)	Case-control	Intake of meat and meat mutagens	846 cases and 846 controls	Lung cancer	1) less than 26.9 ng/g; 2) 27.0-42.1 ng/g; 3) 42.2-51.8 ng/g; 4) 51.9+ ng/g	BaP significantly associated with risk of lung cancer. 1) OD = 1.0; 2) OD = 1.10; 3) OD = 1.49; 4) OD = 2.08	Chronic	Positive
Eingholm et al. (1996)	Cohort	Residents of Copenhagen in comparison with those living in rural areas	927 470 men and 486 130 women	Lung cancer	Up to 10 ng/m <sup>3</sup>	Region had very small effect, and role of BaP is unclear	Chronic	Inconclusive
Friesen et al. (2007)	Cohort	Inhalation at aluminum smelter	6423 males	Bladder and lung cancer incidence, mortality due to myocardial infarction	7.63-122.0 μg/m <sup>3</sup> -year	BaP (and benzene-soluble material) were strongly associated with bladder cancer and lung cancer, but modestly associated with myocardial infarction	More than 3 years of work experience	Positive
Friesen et al. (2009)	Cohort	Inhalation at aluminum smelter	4316 male smelter workers	Mortality and cancer incidence	Unknown	Marginal significance of trend for BaP (also inhalable dust and fluoride exposure)	Chronic	Inconclusive
Gu et al. (2008)	Case-control	Smoking	203 cases and 198 controls	BPDE-induced 9p21 aberrations in cultured peripheral blood lymphocytes (bladder cancer)	Not defined	Increased in individuals with cancer relative to control; BPDE 9p21 aberrations significantly associated with bladder cancer (OR = 5.29, 95% CI = 3.26-8.59)	Not defined	Positive
Gustavsson et al. (1995)	Cohort	Inhalation in Swedish graphite electrode plant	901	Mortality and cancer incidence	In the highest exposure group, cumulative exposure was 33 ug BaP/m <sup>3</sup> -years, mean exposure time was 10.8 years and average exposure level	No excess risk of death from cancer	Average of 10.8 years	Negative
Haugen et al. (1986)	Cohort	Inhalation in coke oven workers	Not reported	BPDE-DNA adducts in lymphocytes	7.3 μg/m <sup>3</sup> on collected particulate matter	One third of the workers had detectable BPDE-DNA adducts	Chronic	Positive
He et al. (1991)	Case-control	Indoor smoky coal exposure	110 cases and 426 controls	Lung cancer mortality	35.60-248.50 μg/100 m <sup>3</sup>	Strong association between BaP exposure and lung cancer mortality	Chronic	Positive
Hemminki et al. (1990)	Cohort	Inhalation in coke workers	91	Aromatic DNA adducts in white blood cells	0.25-90 μg/m <sup>3</sup>	Significant in battery workers in comparison with other jobs; significant in nonbattery workers compared with countryside controls	Current job considered.	Positive
Izotti et al. (1991)	Case-control	Cigarette smoking	39	BPDE-DNA adducts	Unknown	No adducts were detected in samples from nonsmokers or ex-smokers, whereas 84.6% of samples from current smokers exhibited typical fluorescence peaks	Chronic	Positive
Jeng et al. (2011)	Cross-sectional	Inhalation in coke workers	1000	IgA, IgE levels (only endpoint relevant to BaP); other endpoints for all PAHs included malondialdehyde (MDA) and 8-OHdG	Mean concentrations of 1603.06 ng/m <sup>3</sup> in top-oven workers (considered high-exposure group in this study) and 62.47 ng/m <sup>3</sup> in side-oven workers (considered low-exposure group)	IgA and IgE correlated strongly with BaP exposure; MDA was significantly increased, but not 8-OH-dG	At least 1 year of employment	Positive
Junior et al. (1994)	Cohort	Males working in steel foundries	206	BPDE adducts to hemoglobin	Assessed by personal air sampling devices; precise air concentrations not reported	Air concentrations of BaP not associated with levels	Current job considered	Negative
Knox et al. (2005)	Cohort	Home address relative to hotspots for chemical exposure	22 258 cancer-related deaths before age of 16	Cancer mortality	Not defined	Excess risk of cancer-related death within 0.3 km of hotspots for BaP and many other chemicals including PM <sub>10</sub> , benzene and carbon monoxide	Chronic	Positive
Lavoué et al. (2007); Gibbs and Sevigny (2007a); and Gibbs and Sevigny (2007b) (four-part study)	Cohort	Inhalation in workers from aluminum smelters in Quebec	Part 1) 28 910 jobs; Part 2) 5977 workers; Part 3) 9726 workers; Part 4) 5977 workers	All cancer incidence and mortality	0.01-68.08 μg/m <sup>3</sup>	Part 1) looked at exposure; Part 2) causes of death before 1951 significant for bladder cancer, COPD, cancers of stomach, digestive system (unspecified), rectum and rectosigmoid, pancreas and larynx, Alzheimer's disease, cerebrovascular disease; Part 3) cause of death after 1951 significant for COPD and respiratory cancer (also esophagus, rectum and rectosigmoid junction, pancreas, larynx, lung, non-Hodgkin's lymphoma, cerebrovascular disease and asthma; Part 4) overall cancer incidence, significant lung and bladder cancer (BaP specific), laryngeal and buccal cavity cancer also increased with BaP exposure	Chronic	Positive
Mumford et al., 1993	Cohort	Coal or wood smoke during cooking or heating vs. women using natural gas in Beijing	9 Xuan Wei women with chimney; 9 Xuan Wei women without chimney; 9 Beijing controls	DNA adducts in peripheral blood and cord blood white blood cells and placental tissue	19.25 μg/m <sup>3</sup> for smoky coal and 3.24 μg/m <sup>3</sup> for wood	More DNA adducts were reported in exposed women but significance is not reported.	At home throughout lifetime.	Positive
Mori, 2002	Cohort	Inhalation in graphite electrode factory; exposure to coal tar and coal tar pitch volatiles	332 male employees	Mortality, all causes	Unknown	Increased mortality due to lung cancer (SMR=2.62, compared to 2.35 in general population), lymphatic and haematopoietic cancers (SMR=3.46). Relative contribution of BaP unknown.	More than 5 years.	Inconclusive
Nadon et al. (1995)	Case-control	Cancer patients vs. controls in Montreal area	3730 cancer patients and 533 controls	14 types of cancer (esophagus, stomach, colon, rectum, pancreas, lung († oat cell, squamous cell and adenocarcinoma), prostate, bladder, kidney, melanoma of skin and non-Hodgkin's lymphoma)	Unknown (only estimated as low, medium and high according to questionnaire)	Increased cancer cases for stomach, pancreas and prostate; increased lung cancer risk observed in nonsmokers and light smokers only	Chronic	Positive

Niu et al. (2011)	Cohort	Inhalation in coke oven workers	176 coke oven workers and 48 warehouse controls	Emotional and cognitive function; concentrations of monomine and amino acid neurotransmitters; urinary levels of 1-hydroxypyrene for exposure assessment	Mean concentrations of BaP were 19.5, 185.9 and 1623.5 ng/m <sup>3</sup> at the bottom, side and top of the coke oven, respectively; concentration for controls was 10.2 ng/m <sup>3</sup>	1-OH-Py levels increased; learning and memory decreased; concentrations of norepinephrine decreased; acetylcholinesterase activity decreased; overall, the study shows that exposure to BaP may reduce neurobehavioral function and neurotransmitter levels	Chronic	Positive	
Ovrebo et al. (1995)	Cohort	Inhalation in coke workers	13/- for control, 23/17 for low, 26/18 for medium and 18/15 for high exposure workers (January/June)	Anti-BPDE-DNA adducts and hydroxyethylvaline hemoglobin adducts; urinary 1-hydroxypyrene as a measure of exposure	high=top side workers, medium=side workers, low=maintenance workers	Anti-BPDE-DNA adducts did not correlate urinary marker of exposure or cumulative exposure; nonsignificant increase was observed for hydroxyethylvaline-hemoglobin adducts	Chronic	Negative	
Pan et al. (1998)	Cohort	Inhalation in coke oven workers (top, middle and bottom workers, and controls)	25 males	Urinary 1-hydroxypyrene, leukocyte aromatic DNA adducts, serum p53, glutathione S-transferase M1 (all for PAHs)	Mean BaP level for each worksite of 3.16 µg/m <sup>3</sup> (top), 3.02 µg/m <sup>3</sup> (push side), 0.98 µg/m <sup>3</sup> (coke side), 0.10 µg/m <sup>3</sup> (bottom) and 0.01 µg/m <sup>3</sup> (control); BaP exposure information also available for other PAHs	Serum p53 correlated with cumulative BaP exposure; no correlation was found for DNA adducts and PAH exposure	Chronic	Positive	
Pastorelli et al. (1996)	Cohort	Inhalation of BaP from traffic exhaust	53	BaP diol epoxide adducts with hemoglobin	Estimate of 1-3 ng/m <sup>3</sup> in high-traffic areas	Significant difference observed for high-traffic exposure in nonsmokers	Chronic	Positive	
Perera et al. (1982)	Case-control	Lung cancer patients (normal tissue, tumor tissue and blood samples)	15	DNA adducts	Unknown	Inconclusive due to small sample size	Unknown	Inconclusive	
Perera et al. (1988)	Cohort	Inhalation in Finnish foundry workers	35 + 10 controls	DNA adducts in white blood cells	Low (< 0.05 µg/m <sup>3</sup> ); medium (0.05-0.2 µg/m <sup>3</sup> ); high (> 0.2 µg/m <sup>3</sup> )	Significant for each exposure group	Current job considered	Positive	
Perera et al. (1993)	Cohort	Inhalation in Finnish foundry workers	48	HPRT and GPA mutation; DNA adducts	< 5-60 µg/m <sup>3</sup>	Not significant for any endpoint	Current job considered	Negative	
Phillips et al. (1988)	Cohort	Inhalation in Finnish iron foundry	41	Aromatic DNA adducts in white blood cells	> 0.2 (high), 0.05-0.2 (medium) and < 0.05 (low)	Adducts present in 3/4 (high), 8/10 (medium), 4/18 (low) and 1/9 (controls)	Current job considered	Positive	
Rojas et al. (1995)	Cohort	Inhalation in coke workers (smoking, nonsmoking and controls)	39 exposed, 39 controls	Anti-BPDE-DNA adducts in lymphocytes/monocytes	Ranging from ≤ 0.15 to ≥ 4 µg/m <sup>3</sup> in year of collection	~8 times higher in occupationally exposed individuals	At least 4-6 months prior to blood collection	Positive	
Santella et al. (1993)	Cohort	Inhalation in workers near or in a Finnish iron foundry	48	PAH-DNA adducts in white blood cells	2-60 ng/m <sup>3</sup>	Not significant	Chronic	Negative	
Szczeklik et al. (1994)	Cohort	Inhalation in Polish iron workers	274 (199 coke oven workers and 76 cold-rolling mill workers)	Humoral immunity (IgG, IgA, IgM and IgE concentrations in blood)	0.2-50 µg/m <sup>3</sup> in coke plant workers was 3-5 times magnitude than in cold-rolling mill employees	Decreased IgG and IgA in coke oven workers indicating immunosuppression with BaP exposure	Average of 15 years	Positive	
Tas et al. (1994)	Cohort	Inhalation in two steel foundries and one graphite electrode producing plant	260 (133 controls and 127 exposed)	BPDE adducts on albumin	Not clearly defined	Significantly higher in workers relative to controls; significantly associated with air BaP levels	Current job considered	Positive	
Winker et al. (1996)	Cohort	Inhalation exposure in coke oven workers (one newer/clean facility and another with high levels of PAHs)	24	Immunotoxic effects from blood samples	New facility = 651 ng/m <sup>3</sup> air; old facility = 5396 ng/m <sup>3</sup>	Reduced mitogenic response of T cells to phytohemagglutinin; impairment of B cell activity; reduced oxidative burst in monocytes after stimulation with <i>E. coli</i> ; no effects on lymphocyte subpopulations and immunoglobulin levels in serum	6-30 years	Positive and Negative	
Xu et al. (1996)	Nested case-control	Inhalation in Chinese iron steel complex	610 lung cancer cases, 292 stomach cancer cases and 959 controls	Lung cancer and stomach cancer cases	< 0.84, 0.85-1.96, 1.97-3.2, ≥ 3.2 µg/m <sup>3</sup>	Odds ratio of 0.9, 1.7, 1.3, 1.7 for each dose category, respectively; significant trend	More than 15 years	Positive	
<b>Ingestion</b>									
Anderson et al. (2005)	Case-control	Meat intake and preparation method	193 cases and 674 controls	Pancreatic cancer	Median 0.3-53.7 ng/day	OR = 2.2 (1.2-4.0)	Chronic	Positive	
Butler et al. (2003)	Case-control	Meat intake and preparation method	701 African Americans and 957 Caucasians	Colon cancer	Mean BaP levels of 22.5 and 16.7 ng/day for African American cases and controls; 41.9 and 35.4 ng/day for white cases and controls	Significant association observed in African Americans only	Chronic	Positive	
Cross et al. (2005)	Case-control	Meat intake	29 361	Prostate cancer	≥ 1031.5 ng/day	No association between prostate cancer and BaP intake	Chronic	Negative	
Cross et al. (2006)	Case-control	Meat intake and preparation method	383 controls and 458 cases	Non-Hodgkin's lymphoma	Mean intake of 37.1 ng/day and median intake of 16.8 ng/day	No increased risk	Chronic	Negative	
Ferrucci et al. (2012)	Case-control	Meat consumption	17 072	Distal colon and rectal adenoma	Unknown	Not associated with colon adenoma but significantly associated with rectal adenoma (OR = 1.53)	Chronic	Positive	
Fu et al. (2011)	Case-control	Meat and meat-derived mutagen intake	2386 cases and 1703 controls	Breast cancer	Unknown	Intake of red meat may be associated with breast cancer; however, this correlation does not exist for BaP	Chronic	Negative	
Gunter et al. (2005)	Case-control	Meat intake and preparation method	628 cases and 689 controls	Colorectal adenoma	> 0.29-515.2 ng/day	6% increase of large adenoma per 10 ng/day consumption of BaP	Monitored over the last year	Positive	
Hakami et al. (2008)	Case-control	Intake through diet (staple foods including bread and rice) and water in northern Iran	40 cases, 40 controls from the same area and 40 from a low-risk region	Esophageal cancer	For bread + rice + water (data also presented for each food item separately), total daily intake was 99.0, 91.4 and 70.6 ng/day for cases, controls of the same region and low-risk region, respectively	BaP was significantly higher in cases and controls of the high-risk area, in comparison with the low-risk area; BaP may be associated with esophageal cancer	Chronic	Inconclusive	
Lam et al. (2009)	Case-control	Meat intake	2120 controls and 2101 cases	Lung cancer	Not well defined	Significant association for BaP, heterocyclic amines and red meat consumption	Chronic	Positive	
Li et al. (2007)	Case-control	Meat intake and preparation method	626 cases and 530 controls	Pancreatic cancer	Mean of 69.9 in cases and 61.2 in controls. Median of 42.3 in cases and 37.3 in controls. These differences were not significant.	BaP was a significant predictor of pancreatic cancer	Chronic	Positive	
Sinha et al. (2005a)	Case-control	BaP intake from meat and other foods	146 cases and 228 controls	Colorectal adenoma	Median 5 ng/day from meat and 73 ng/day from food in controls; median 17 ng/day from meat and 76 ng/day from other foods in cases	Trend is significant for both meat and food sources; ORs from meat were 1.19, 1.71, 2.16 and 2.82 and from food were 2.61, 4.21, 2.45 and 5.60 for the second, third, fourth and fifth quintiles, respectively	Chronic	Positive	
Sinha et al. (2005b)	Case-control	BaP intake from meat and processed meats	3696 cases and 34 817 controls	Colorectal adenoma (left-sided descending sigmoid colon and rectum)	0.8-168.1 ng/day	BaP intake associated with marginal elevation of colorectal adenoma risk	Chronic	Positive	
<b>Dermal exposure</b>									
Scheepers et al. (2009)	Cohort	Nurses who apply ointments containing coal tar	35 nurses	Traces of BaP on hands with and without gloves and 1-hydroxypyrene urinary biomarker	33.0 and 16.4 ng/cm <sup>2</sup> on hands	Use of gloves decreased absorption of BaP by 51.5%. Use of suggested protocol reduces absorption of BaP by more than 57%.	Chronic	Positive	
<b>Human cells</b>									
Rojas et al. (2004)		Smoking	22 normal bronchial epithelial cells compared with lung parenchyma in smokers and nonsmokers with lung cancer	BaP BPDE-N2-dG (adducts)	Unknown	Adducts were significantly more elevated in smokers and in bronchial epithelium in comparison with parenchyma		Positive	

Supplementary Table 3: All rodent and *in vitro* studies evaluated

Reference	Health effect category (reproductive toxicity, developmental toxicity, carcinogenicity, mutagenicity, etc.)	Exposure (acute < 1 month, subchronic < 3 months, chronic > 3 months)	Route of exposure (Inhalation, oral, intraperitoneal)	Test material (vehicle, control group, doses, test limit, duration of exposure, dose selection rationale [range-finding study], purity, stability)	Test animals (species, strain, sex, number, age, acclimation, weight, environmental conditions, diet, water)	Clinical observations, necropsy, histopathology, KEY ENDPOINT	Key events (known or speculated)	Findings (MOAs, LD <sub>50</sub> , NOAEL/LOAEL, p-value, relative toxicity compounds)	Quality of study (strengths and weaknesses; use appropriate statistics?)	Notes
Aboutabl et al. (2009)	Cardiovascular toxicity	Acute	i.p. injection	20 mg BaP/kg bw/day for 7 days vs. corn oil control	SD rats n=6 adults	inc. heart rate, bw, cardiac hypertrophic markers, atrial natriuretic peptide, brain neurotrophin. Inc. gene expression of cyp1a1,1b1,2c1,4f4.	MOA/AHR necessity test with inhibitors	only 1 dose tested	weak. Weakness in	single dose MOA
Aboutabl et al. (2011)	Cardiovascular toxicity	Acute	i.p. injection	20 mg BaP/kg bw/day for 7 days vs. corn oil control. With & without soluble hydrolase enzyme inhibitor (14,15-DHET,14,15-EET)	SD rat n=4 adults	inc. heart rate, bw, cardiac hypertrophic markers, atrial natriuretic peptide, brain neurotrophin. Inc. gene expression of cyp1a1,1b1,2c1,4f4.	MOA/AHR necessity test with inhibitors	only 1 dose tested	weak	single dose MOA
Carlson and White (1983)	Cardiovascular toxicity	Acute	i.p. injection	40 mg BaP/kg bw in corn oil for 72 & 48 hours prior to phenobarbital, trichloroethylene or halothane exposure	male new zealand rabbits BaP n=7, control n= 20	BaP weak direct effect to increase number of arrhythmias. BaP somehow facilitates sensitization of myocardium to epinephrine by trichloroethylene to cause arrhythmias	no	no	many weaknesses	N
Genitner and Weber (2011)	Cardiovascular toxicity	Acute	Intranasal	14 days before dosing, surgery to implant BP device and medicated with midazolam, buprenorphine and isoflurane. Anesthetized with isoflurane	male SD rats n=6 group	Thoracic vascular SMCs from congenic AHR <sup>+/+</sup> & AHR <sup>-/-</sup> mice	Identified TGFβ2 & IGF-1 as potential candidates for a AHR alternative pathway for cells to respond to BaP	no	no	poor study, lack details for not a good key study
Kayala et al. (2004)	Cardiovascular toxicity	Acute	<i>In vitro</i>	10 μg BaP to ~125–150 mg/kg bw in intact mouse. Vehicle = DMSO for 24 hours	Human macrophages	Identified TGFβ2 & IGF-1 as potential candidates for a AHR alternative pathway for cells to respond to BaP	postulated	no	no	mechanistic and exploratory
Matsunawa et al. (2009)	Cardiovascular toxicity	Acute	<i>In vitro</i> & intranasal	1 μm BaP for 48 hours	Human macrophages	no stated. More mechanistic study	no	no	no	mechanistic and exploratory
NDiave et al. (2006)	Cardiovascular toxicity	Acute	<i>In vitro</i> & intranasal	<i>In vitro</i> ? Intranasal installation of 500 μg BaP in triacrylin vehicle under anesthesia for 24 hours.	Human primary macrophages. Male adult	CCL1 (chemokine involved in in CV diseases and inflammatory response) altered <i>in vivo</i> lung & <i>in vitro</i> by AHR mechanism. Increased early and transient intracellular calcium <i>in vitro</i>	postulated	no	no	study details not clear
NDiave et al. (2009)	Cardiovascular toxicity	Acute	<i>In vitro</i>	1 μm BaP for 1 hour	Human primary macrophages. Male adult	mechanism of CCL1 induction via AHR vs lipoprotein A	postulated	no	no	mechanistic and exploratory
Owens et al. (2008)	Cardiovascular toxicity	Acute	<i>In vitro</i>	AHR activation with B-NF (1 μm) for 16 hours then BaP for 24 hours	n=3 Primary umbilical cells (HuVEC)	Pro-inflammatory MOA involving ICAM-1, Caveolae leading to increased vascular endothelial adhesiveness which may be a critical step in development of BaP induced atherosclerosis. AHR dependent metabolism of BaP	postulated	MOA but not for key study	mechanistic and exploratory	not useful as mechanistic
Oesterling-Owens et al. (2009)	Cardiovascular toxicity	Acute	<i>In vitro</i>	AHR activation with B-NF (1 μm) for 16 hours then BaP for 24 hours	n=3 Primary umbilical cells (HuVEC)	Flavonoids can protect against BaP induced ICAM-1 mechanism described in Oesterling et al 2008	postulated	MOA but not for key study	mechanistic and exploratory	not useful as mechanistic
Podecharat et al. (2009)	Cardiovascular toxicity	Acute	<i>In vitro</i> & intranasal	<i>In vitro</i> : 1 μm BaP 24 hours. Vehicle ?? <i>In vivo</i> : 500 μg BaP 24 or 72 hours n=37	Primary human macrophages & C57/B6	BaP represses NPC1 expression via AHR in macrophages which may contribute to macrophage lipid accumulation involved in CV disease.	postulated	MOA but not for key study	poor reporting of study details	mechanistic not useful as
Sanyal and Li (2007)	Cardiovascular toxicity	Acute	i.p. injection	200 mg BaP/kg bw or corn oil vehicle PD10, 12, 14 evaluated on PD 20	n=20 dams group 60-day old SD rats	no effect on maternal or fetal survival rates. Decreased maternal BW, placental toxicity, fetal rupture and hemorrhage of blood vessels in skin, cranial and brain tissues.	no	no	no dose response	lowest dose but no stats
Sauzeu et al. (2011)	Cardiovascular toxicity	Acute	i.p. injection	BaP 10 mg/kg bw for 24 hours	4-month old Ahr and Vav3 knock-out mice. N=5-7	Constitutive AHR regulation of Vav3 proto-oncogene to control cardiovascular & respiratory functions does not require AHR activation by BaP	postulated	no	no	response
Kerley-Hamilton et al. (2012)	Cardiovascular toxicity	Chronic	Gavage & in food	Gavage 10 mg/kg bw/day (5 days/week) for 10 weeks n=5. Food: BaP <i>ad libitum</i> in oil-soaked chow (BaP 800 mL/mL corn oil) estimated dose 10 mg/kg bw/week	B6 strain (high affinity AHR) & B6-D2 strain <sup>90%</sup> male white leghorn chickens	B6 strain altered growth rates of body and several organs and induced atherosclerosis and to a greater extent than in B6-D2. BaP had big impact on gene expression of the aorta (immune responses, muscle-specific genes) which support a MOA of inflammation leading to muscle atherosclerosis	postulated	no	no	not sufficient to be key
Pein and Snyder (1988)	Cardiovascular toxicity	Subchronic	i.m. (pectoral injection)	Weekly injection from 4 weeks old to 20 weeks old of 40 mg BaP/kg bw or DMSO, n=6	chickens	Arteriosclerotic plaques in abdominal aorta of all chickens	no	no	no	not sufficient to be key study
Soremen et al. (2003)	Cardiovascular toxicity	Hypothetical MOA linking PAH with biological effects. Help with MOA but not useful as key study for BaP assessment								
Knaapen et al. (2007)	Cardiovascular toxicity	Acute	Gavage	5 mg/kg bw or triacrylin vehicle, 2 treatments 1 week apart	AP0E <sup>-/-</sup> male mice 17 weeks old n=5	increased mild level of atherosclerosis. MOA: vascular pro-inflammatory effects of BaP by AHR-mediated induction of MCP-1 proposed to lead to increased atherogenesis.	postulated	MOA but not for key study	not sufficient to be key study	not sufficient to be key study
Godschalk et al. (2003)	Cardiovascular toxicity	Acute	Gavage	High fat/high cholesterol diet for 25 days. Single oral dose of 5 mg BaP/kg bw or triacrylin vehicle	n=3/reim/diet. Male APOE <sup>-/-</sup> mice	After 4 days: increased BPDE-Dna adducts aorta. Decreased LDL & increased HDL. Direct and lipidperoxidation-induced DNA damage by BaP in aorta.	postulated	no	poor	not sufficient to be key
Pein (1990)	Cardiovascular toxicity	Review paper with MOA, dose range in chicken and mice 0.1–40 mg/kg bw								
Ferguson (2009)	Cardiovascular toxicity	Review paper with proposed MOA: mutation of SMC genes relevant to CVD leads to recruitment of macrophages, buildup of foam cells, fatty deposits in vessel wall and then to atherosclerosis								
Yang et al. (2009)	Cardiovascular toxicity	Chronic	Gavage	Olive oil vehicle, 2.5 mg BaP/kg bw/week for 24 weeks	male C57/B6 mice: APOE <sup>-/-</sup> & APOE <sup>-/-</sup> /overexpress SOD	Increased mean lesion size in aortic tree and root 60% & 40% and increased oxidized lipids. BaP didn't alter lipid peroxidation or atherosclerotic lesions in aorta of any of the APOE <sup>-/-</sup> plus overexpression. Implicate role of ROS in BaP-induced atherosclerosis	no	no	no	mechanistic and exploratory
Curfs et al. (2005)	Cardiovascular toxicity	Chronic	Gavage	5 mg/kg bw or triacrylin control 1 time/week for 24 weeks	Male APOE <sup>-/-</sup> 3 weeks old BaP n=30 control N= 20	increased plaque size but lesion and number unaffected. No oxidative DNA damage or apoptosis in plaques. Plasma lipoprotein unchanged	BaP caused progression of atherosclerosis irrespective	no	no	mechanistic and exploratory
Curfs et al. (2004)	Cardiovascular toxicity	Subchronic	Oral	5 mg/kg bw or triacrylin control 1 time/week for 12 weeks (n=31) or 24 weeks (n=19)	Male APOE <sup>-/-</sup> 5 weeks old	Increased greater than 2-fold T-lymphocytes in plaques and increase TGFβ local response	postulated	no	no	mechanistic and exploratory
Ramos and Moorby (2005)	Cardiovascular toxicity	Review provides MOA information but no dose-response data				Adducts formed. BaP didn't initiate atherosclerotic plaques but accelerates the progression via a local inflammatory response	postulated	no	no	mechanistic and exploratory
Nakamura et al. (2012)	Developmental/reproductive	Acute	Oral gavage	BaP 0, 2, 10 mg/kg bw (dose selection based on previous study in CD-1 mice)	Male C57/B6 mice	Prenatal exposure to BaP from GD7-16 was dose-dependently associated with sign. 1 testicular and epididymal sperm counts & sperm motility, uterine and placental weights, and litter sizes.	Data demonstrate an important role of AHR in development	Experiment 1	Findings	
Wu et al. (2003)	Development	Acute	Nose-only exposure	BaP (97% pure) purchased from Sigma Chemical Co. (St. Louis, MO). GD 11–21.	Female Ahr <sup>-/-</sup> mice	Increased placental weight and placental length. Birth index (related to # of birth), BaP metabolism (plasma, cortex), relative % of BaP metabolites in plasma and cortex over time, Cyp1a1 and Ahr gene and protein expression was examined in developing pups (post-natal days PND 0-30, sometimes, 60).	Prenatal BaP exposure → Ahr and Cyp1a1 increased	Lower birth index at 75 and 100 μg compared to control	How much	Lengthy explanation
Mickelze and Angevine (1981)	Development	Acute	Oral gavage	BaP from Aldrich; in 0.2 mL corn oil. GD 7–16.	CD-1 female mice; doses 0, 10, 40, 160 mg/kg bw	% pregnant, % viable offspring, mean litter size, reproductive capacity of these animals (treated in utero) later in life.	unknown.	Lower % viable litter at birth; lowered pup weight	TABLE 1 good for BMD	Can't do BMD on the
Shum et al. (1979)	Development	Acute	i.p. injection	BaP from Sigma, dissolved in corn oil. GD 7 or 10.	'Ahr responsive' strain C57BL/6 (responsive to BaP)	Developmental abnormalities were studied (% stillborn, resorptions, malformations, etc.)	Ahr	More abnormalities in C57BL/6 (Ahr-responsive)		
Ball (1970)	Immune	Acute	subcutaneous injection	Acute (one time) subcutaneous injection of BaP (60 and 120 μg) right after birth induced thymic lymphoma formation (56% incidence vs. 1.5% in controls for 120 μg) while no immunosuppression was observed (hemolysis in group of sheep erythrocytes), unlike DMBM (100% thymic lymphoma, about 50% immunosuppression).						
Daynes et al. (1979)	Immune	n.a.	n.a.	This paper looked at cotreatments of BaP and immunosuppressive agents, but not at the immunosuppressive properties of BaP.						
Uroso and Genozian (1980)	Immune	Acute	i.p. injection	i.p. injection of BaP (100 and 150 mg/kg bw) at middle (11–13 days) or late (15–18) gestational days led to marked (about 50%) suppression of the anti-sheep erythrocyte plaque-forming response shortly after birth in the progeny and persisted later into life. Tumor frequency was 76 for mid-gestation and 64% for late gestation compared with						
Uroso and Genozian (1982)	Immune	Acute	i.p. injection	Mice were exposed during gestational development, postnatally or as adults (150 mg BaP/kg bw, once). Animals exposed prenatally were more susceptible to immunosuppression & tumor formation compared with those treated postnatally. Supports the idea that immune deficiency (suppression) influences tumor formation.						
Uroso and Genozian (1984)	Immune	Acute	i.p. injection	Pregnant mice received 150 mg BaP/kg bw during GD 11–17 by i.p. injection. Progeny were assayed for humoral and cell-mediated immune response at various intervals after birth. Immature offspring (1–4 weeks) were severely suppressed in their ability to produce antibody-(plaque-) forming cells (PFC) against sheep red blood cells (SRBC)						
				These data show that <i>in utero</i> exposure to the chemical carcinogen BaP alters development of compounds needed for establishing competent humoral and cell-mediated functions of the immune apparatus and leads to severe and sustained postnatal suppression of the defense mechanism. The immunodeficiency exhibited, particularly in the Tc						
Busbee et al. (1984)	Immune	sheep lymph was isolated and used		A combination of <i>in vitro</i> and <i>in vivo</i> experiments on sheep or sheep lymphocytes. Showed that BaP is taken by lipoproteins. "We propose that lipophilic xenobiotic compounds interact with cells of the immune system via lymphatic lipoprotein transport of potentially mutagenic, carcinogenic, or immunosuppressive agents."						
Dean et al. (1983)	Immune	Acute	subcutaneous injection	Adult B6C3F1 mice were exposed to 10 daily subcutaneous injections of BaP in corn oil for 14 days (oil, 30, 200 and 400 mg/kg bw). Exposures of mice to BaP resulted in a reduced number of IgM and IgG antibody plaque-forming cells						
White et al. (1984)	Immune	Acute	subcutaneous injection	Female B6C3F1 mice, subcutaneous injection of 0, 5, 20 or 40 mg BaP/kg bw in corn oil. On day 11, all mice received sheep red blood cells (SRBC, i.p.). On day 15 animals were sacrificed, spleens dissociated into single-cell suspension and were accessed for the number of anti-SRBC antibody-forming cells producing IgM. Plaques were counted						
Wojdani and Alfred (1984)	Immune	Acute	i.p. injection	C57, C3H and DBA mice, 8 weeks old, were dosed with phytohemagglutinin injection to activate splenic lymphocytes and 24, 96 or 216 hours later were treated with BaP (0, 2.5, 10, 50 mg/kg bw BaP single i.p. injection in oil). Isolated, T-cell-enriched mononuclear cell populations were assayed for Ahr hydroxylase activity, if possible.						
Holladay (1994)	Immune	Acute	gavage	<i>In utero</i> exposure to BaP (0, 50, 100, 150 mg/kg bw/day on GD 13–17 by gavage), and offspring were examined at GD 18. Thymic atrophy, cellular depletion (e.g. CD4+8+ fetal thymocytes). So, BaP → thymic hypoplasia, inhibits thymomatous process, fetal liver hypoplasia, including cells with hematopoietic subpopulations. Da						
Uroso (2008)	Immune	Acute	i.p. injection	Pregnant dams of C3H/Anf mice received 150 μg/kg bw on the 12th day of pregnancy by i.p. injection. On day 18, dams were killed and fetuses were removed. Livers were removed and processed (adherent [macrophages, B cells and others?] and non-adherent cells [T cells] were collected). Then, splenic responder (R) and stimulatory (S) cells were cultured						
Atlan and Sherr (2010)	Immune	<i>in vitro</i> cells		Peripheral blood mononuclear cells (PBMC) were prepared from individual blood donors, depleted of T cells and stained with FITC-labeled CD2-specific antibody and purified (B cells). Plasma cell generation: B cells were plated in irradiated CD40L-transfected L cells. After 4 days of coculture, B cells were harvested and sent to						
Fischer et al. (2011)	Immune	Subchronic	oral subcutaneous injection	Wistar rats, 9–13 weeks old, were fed daily with 150 mg/kg bw for 30 days. Bone marrow and spleen cell numbers were reduced in BaP-treated samples (~70% and 45% reduction, respectively). This is just one of the experiments. Also did some <i>in vitro</i> (cell culture) experiments.						
Smith (2010)	Immune	Subchronic	injection	Compared several models of delayed hypersensitivity using mouse foot pad swelling as endpoint. Female B6C3F1 mice 8–15 weeks of age were challenged with BaP in corn oil sc (subcutaneously) for 14 days at 5, 20 and 40 mg/kg bw. Then, three different antigens were used, and BaP-mediated effect was seen in all three						
Uroso (2008)	Immune	Acute	i.p. injection	In this study, C3H mice were injected once with BaP (150 μg/kg bw) at day 12 of pregnancy and progeny lymphoid tissues were excised during gestation (day 18, GD 18) or at 1 or 6 weeks postpartum. The isolated lymphoid cells were analyzed by flow cytometry/immunofluorescence or assessed for function. In BaP-exposed fetuses, thymic T						
Davila et al. (1996)	Immune	<i>in vitro</i> human cells		Examined the toxic effects of nine different PAHs on human peripheral blood T cell mitogenesis. Found that BaP was highly immunotoxic. Also showed that α-naphthoflavone (ANF), which functions as both an AHR antagonist and an inhibitor of cytochrome P450 activity, was able to block the suppressive effect of BaP. Mitogenesis = induced						
Atlan et al. (2006)	Immune	<i>in vitro</i> human cells		The AHR is a ligand-activated transcription factor that mediates immunosuppression by environmental PAHs. Previous studies demonstrated that activation of mature human B cells upregulates AHR expression, suggesting that human B cells are direct PAH targets. To test this hypothesis and to determine the metabolic requirement						
Galván et al. (2006)	Immune	Acute	i.p. injection	Four-week-old C57BL/6J mice were injected intraperitoneally with 50 mg BaP/kg bw. Mice were sacrificed 12, 24 and 48 hours after intraperitoneal injection with oil vehicle, BaP and bone marrow cells were purified. BaP treatment decreased the pro-pre B-lymphocytes and did not affect the immature B-lymphocytes or mature						
Lee and Uroso (2007)	Immune	<i>in vitro</i> studies with isolated spleen cells		Spleens of C3H/HeJ (source of responder, R cells) and CBY/D2 (source of stimulator cells, S cells) mice, 11–12 weeks of age were removed and made into cellular suspensions. The effect of BaP on the cellular proliferative response in response to allogeneic mixed lymphocyte response (MLR) on concanavalin (only R cells are						



Dent (2007)	Review: Strengths and limitations of using repeat-dose toxicity studies to predict effects on reproductive	Acute	Oral	Four treatment groups (n = 9/group): (1) control, (2) lead (Pb), given 1 g PbCl <sub>2</sub> in drinking water until mating; (3) BaP (10 mg/kg body weight by oral gavage on days 7–16 of pregnancy); (4) combined lead and BaP. Dose selection rationale: lead dose was comparable to blood levels seen in occupational	Male and female F0 Female Bom/NMRI Mice were mated. At 9 weeks of age, F0 females were	Could rodent functional effects be predicted in a sub-chronic study? Yes/No (Reference) If yes, nature of effect: For BaP - Yes (Archibong et al., 2003) Testis: reduced weight; sperm analysis: reduced sperm motility and density (no testicular histology quoted). Also raised LH levels. The chromium compounds, some of the cadmium compounds (cadmium fluoride, cadmium chloride, and cadmium sulphate), hexachlorocyclopentadiene, 1,2-dibromo-3-chloroethane, 2,4-dinitrophenol, and 2,4,6-trinitrophenol, showed a significant indication that the compounds in combination 1 # offspring, # litters, and litter size; results suggest that lead and BaP have synergistic effects on the impairment of fertility	The testicular effects that were predictive of functional effects could be the result of a number of	Intelligent testing strategies therefore need to be developed which can take into account data from a wide variety of	Review focuses on a panel of test chemicals to determine the strengths of their dose selection for lead and BaP marked reduction in the
Kristensen et al. (1995)	Reproductive	Acute	Oral	BaP (0, 10, 40 or 160 mg/kg-day) by gavage on days 7-16 of gestation. Corn oil vehicle, controls given corn oil. Male breeding study: F1 males were placed with 2 virgin untreated females every 5 days for 25 days, at gestation day 14-19, females were sacrificed and # implants, fetuses and resorptions were recorded (F2 young were examined for gross abnormalities). Female breeding study: F1 males	Adult male and female CD-1 Mice; each test group n=30 or n=60 treated pregnant dams. Male breeding study: F1 males	No maternal toxicity or embryolethality at any dose. Decrease pup weight all doses. 160 mg/kg-day of BaP resulted in a reduced percentage of mice that were pregnant, reduced number of viable litters at parturition, and reduced mean pup weight at 20 and 42 days post-parturition. Mean pup weight was also significantly reduced at 20 or 42 days post-parturition in females that received 10 mg/kg-day or greater of BaP. Male offspring of dams treated with 10, 40 or 160 mg/kg-day and then bred with untreated females yielded decreased fertility (10 mg/kg or greater), with almost no pregnant females when the males were exposed to either 40 or 160 mg/kg-day in utero. Similarly, fertility was reduced in female		LOAEL 10 mg/kg/day decreased fertility index in both male and females. Data demonstrate a sensitivity of fetal gonads.	Multi-generational study: exposure of F0 pregnant females for 10.
Mackenzie and Angevine (1981)	Reproductive (bit of developmental)	Acute	Oral gavage	BaP (0, 10, 40 or 160 mg/kg-day) by gavage on days 7-16 of gestation. Corn oil vehicle, controls given corn oil. Male breeding study: F1 males were placed with 2 virgin untreated females every 5 days for 25 days, at gestation day 14-19, females were sacrificed and # implants, fetuses and resorptions were recorded (F2 young were examined for gross abnormalities). Female breeding study: F1 males	Adult male and female CD-1 Mice; each test group n=30 or n=60 treated pregnant dams. Male breeding study: F1 males	A statistically significant ↑ in sperm hyperactivation was observed at concentrations of BaP ≥ 50 µg/mL; Acrosome Halo Test: [BaP] ≥ 25 µg/mL significantly ↓ the percentage of halo formation, indicating an inappropriate (false) acrosome reaction		Conclusion: BaP significantly affected sperm functional- acrosome reaction in vitro, as evidenced by increased	Acute in vitro study, but results seen were statistically
Mukhopadhyay et al. (2010) Fertility and Sterility 94(2): 595-598.	Reproductive	Acute	In vitro (BaP was mixed with media)	Semen collected from 13 fertile, normozoospermic, non-smoking men; spermatozoa were washed and treated with BaP: 12.5, 25, 50, 100 µg/mL. Dose range selection was based on previously described studies and sperm survival studies done in their lab (1 hour exposure)	In vitro study to examine the effects of BaP on sperm hyperactivation and acrosome reaction by computer-assisted semen	Ovaries were analyzed for follicle loss and markers of apoptosis (TUNEL, Caspase 3, Caspase 8, Bax, Bcl-2, Fas and FasL). Cigarette smoke exposure induced a significant ↓ in the number of primordial follicles, but not growing or antral follicles. Mainstream cigarette smoke exposure had no effect on any marker of apoptosis measured. Exposure of ovaries to BaP in vitro resulted in an ↑ in the pro-survival marker Bcl-2, but no change in apoptosis. Significant reductions in the number of follicles in different stages of development in ovaries of mice exposed to cigarette smoke	Data suggest that cigarette smoke-induced follicle loss is not mediated via BaP-induced apoptosis. Findings suggest that a decreased	Good study with in vivo and in vitro components to the research	
Tuttle et al. (2009)	Reproductive	In vivo subchronic (8 weeks) in vitro acute (24 hours)	Inhalation (nose-only)	In vivo: 2 cigarettes/day, exposed 5 days/week for a total of 8 weeks, including the 2-week lead-up period. In vitro BaP: 1-10000 ng/ml (24 h exposure)	Mice and isolated murine ovaries (in vivo in vitro); female C57BL/6J genetic background (B6.129-Gelmt1Tjka, hereafter referred to as Gclm <sup>-/-</sup> ).	We observed no changes in fertility, testicular weights, testicular sperm head counts, or testicular histology and subtle changes in cauda epididymal sperm counts, motility, and morphology in Gclm <sup>-/-</sup> compared to Gclm <sup>+/+</sup> males. Prenatal exposure to BaP from gestational day 7 to 16 was dose-dependently associated with significantly decreased testicular and epididymal weights, testicular and epididymal sperm counts, and with vacuolated seminiferous tubules at 10 weeks of age. Gclm <sup>-/-</sup> males exposed prenatally to BaP had greater increases in testicular weights, testicular sperm head counts, epididymal sperm counts, and epididymal sperm motility than Gclm <sup>+/+</sup> littermates. These results show no effect of BaP significantly inhibited follicle growth & cell proliferation at concentrations of 1.5 ng/ml and higher, an effect attenuated by co-incubation with AHR antagonists. BaP caused a significant ↓ in oestradiol and anti-Müllerian hormone (AMH) output, an effect attenuated by co-treatment with 1 of the 2 AHR antagonists tested.	MOA - AHR agonist; mechanisms of BaP induced follicle toxicity remain unknown. Cross-talk between the AHR and	Results suggest that the adverse effects of BaP on follicle growth, steroidogenesis and AMH output are mediated.	In vitro model best represents in vivo scenario as concentrations
Nakamura et al. (2012)	Reproductive	Acute	Gavage	Dams were treated by oral gavage with 2 or 10 mg/kg BaP (Sigma-Aldrich Supelco, >99.8% purity) in sesame oil daily from GD7 to GD16. Control animals were given the same volume of sesame oil alone	Male Wistar rats (~320 total); age = 6-7 weeks; weight = 130 ± 40g;	Combined effects included: 1 # Spermatozooids, 1 viability, 1 duration of motility, 1 resistance in NaCl and HCl; relative weights of testes, seminal vesicles & epididymis (data ambiguous due to combined dosing of 3 different substances)		Physiologically relevant exposure: cigarette smoke does not increase the rates of apoptosis in the ovaries and by	Good study with in vivo and in vitro components to the research
Neal et al. (2010)	Reproductive	Acute	In vitro (BaP in media)	BaP: 0, 0.5, 1.0, 1.5, 5.0 and 50.0 ng/ml, with or without 5 µmol/l resveratrol (n=6) or 5 µmol/l 3,4-DMF (n=7); dose selection based on physiological concentrations previously measured in human follicular fluid. (5 days)	In vitro study using isolated rat follicles; ovaries (n=6) from 22-26 day old female Wistar rats		MOA - AHR agonist; mechanisms of BaP induced follicle toxicity remain unknown. Cross-talk between the AHR and	Results suggest that the adverse effects of BaP on follicle growth, steroidogenesis and AMH output are mediated.	In vitro model best represents in vivo scenario as concentrations
Pallanaviciene et al. (2006)	Reproductive	Acute	Oral	Cadmium: 0.1, 0.5, 1.92, 4.0 mg/kg; BaP: 0.00015, 0.0015, 33.3 (7 Type in publication?); 10.0 mg/kg; Pyrene: 0.00075, 0.0075, 20.0, 90.0 mg/kg; Exposure duration: 14, 28 & 90 days; Combined exposures performed, oral administration at 1ml/100g BW/day; Vehicle = rape seed oil; 40 groups, 6 control groups (3 water, 3 oil), n=12/10/group	Male Wistar rats (~320 total); age = 6-7 weeks; weight = 130 ± 40g;	Combined effects included: 1 # Spermatozooids, 1 viability, 1 duration of motility, 1 resistance in NaCl and HCl; relative weights of testes, seminal vesicles & epididymis (data ambiguous due to combined dosing of 3 different substances)		Physiologically relevant exposure: cigarette smoke does not increase the rates of apoptosis in the ovaries and by	Good study with in vivo and in vitro components to the research
Ramesh et al. (2008)	Reproductive	Subchronic	Inhalation (nose-only)	Treatment group = 75 µg BaP/m <sup>3</sup> , 4 h daily for 60 days (1 sperm cycle); Control group = unexposed (UNC). Dose selection rationale: exposure concentration selected as it adversely affected reproductive outcomes in male and female rats in a previous study by these authors and is within a range	Adult Male F-344 rats; age = -12-13 weeks; weight = -340-360g; n=10/group; carbon black used as a carrier for BaP. Like a hybrid (C57BL/6J-CBA/Ca) mice (13 day old mice, n=2)	Blood samples were collected on day 60 (time 0), and subsequently at 24, 48 & 72 hr to assess plasma testosterone (T) and luteinizing hormone (LH). BaP exposure reduced testis weight and caused significant reductions in the components of the steroidogenic and spermatogenic compartments of the testis. Progressive motility and mean density of stored spermatozoa were reduced. Plasma [T] were decreased by two-thirds in BaP exposed rats, concomitant with increased concentrations of LH in BaP-exposed rats. BaP treatment inhibited (p < 0.05) antral follicle development, decreased estradiol output and follicle survival at the 45.0 ng/ml dose. BaP exposure decreased AMH output overall during preantral (p = 0.014) and antral (p = 0.026) follicle development but had no effect on progesterone output or oocyte growth and nuclear maturation in surviving follicles.	MOA: AHR and ARNT discussed	These data suggest that sub-chronic exposure to inhaled BaP contribute to reduced testicular and epididymal	Poor quality of writing was poor (English was not the first language of the authors) Well written paper with many relevant references
Sadeu and Foster (2011) Reproductive Toxicology 31: 402-408.	Reproductive	Acute	In vitro	BaP: DMSO & 0 ng/ml BaP (controls), 1.5, 5, 15 & 45 ng/ml. Dose Selection is representative of follicular fluid concentrations in women exposed to mainstream and/or sidestream cigarette smoke: sub-acute (13 days)	Female C57BL/6N Mice; 6 weeks of age; water and standard mouse chow ad libitum; 1 week acclimatization before	Dose- and time-dependent decrease in # of corpora lutea; 15 weeks of age; water and standard mouse chow ad libitum; 1 week acclimatization before		Data suggest that BaP is an important toxic component of cigarette smoke that adversely affects follicular	The advantage and physiologically relevant aspect of the ions.
Swartz and Mattison (1985)	Reproductive	Acute	Single i.p. injection, studied mice at weekly intervals from 1 to 4 weeks post-injection	BaP: 1, 5, 10, 50, 100 & 500 mg/kg. Vehicle = corn oil (1 acute exposure, studied 1, 2, 3 & 4 weeks post-exposure (designed to study the effect of BaP on ovarian function in intact mice not stimulated with exogenous gonadotropins)	Female Sprague-Dawley rats, n=6 per group; BW = 80-100g; age = 5 weeks; access to purified water and standard chow	Dose- and time-dependent decrease in # of corpora lutea; 15 weeks of age; water and standard mouse chow ad libitum; 1 week acclimatization before		1 week after treatment: threshold of ~1mg/kg, ED50 of 1.6 mg/kg P<0.05	Not corpora lutea present in BP-treated and control mice
Xu et al. (2010)	Reproductive	Subchronic	Oral	BaP: 5 & 10 mg/kg; di-(2-ethylhexyl)phthalate (DEHP): 300 & 600 mg/kg; or combination of BaP & DEHP, control (corn oil). Exposure: alternate days for 60 days; Dose Selection: dietary exposure to BaP and DEHP is likely as both are found in urban and rural water. Doses used are higher than the levels found in the general environment, but may be relevant in areas where the specific BaP in cigarettes: 6-40ng/cigarette; 20 cigarettes in 8 h, could inhale 0.067-0.568 µg of BaP	Female Sprague-Dawley rats, n=6 per group; BW = 80-100g; age = 5 weeks; access to purified water and standard chow	BaP and DEHP exert oestrogenic and suppression of sex hormone (17β-estradiol), secretion and homeostasis, which is associated with prolonged estrous cycles, decreases in ovarian follicle populations and granulosa cell apoptosis involving a PPAR-mediated signaling pathway of action of the two chemicals. [ovarian weight & ovary weight/BW ratios in treatment groups; PPAR-α mRNA & PPAR-γ mRNA & protein expression. With ↑ in the dose of BaP + DEHP, there was a dose-response trend of prolonged duration in the EC and the LH phases of the estrous cycle.	Granulosa cell apoptosis involving a PPAR-mediated signaling pathway of the two chemicals. Some AHR could interfere with observed following	Based on qualitative assessment of the combined toxicity, no interaction effects were observed following	Excellent review with respect to adducts specific to BaP
Zenzes (2000)	Reproductive	review paper	Inhalation (smoking)	BaP in cigarettes: 6-40ng/cigarette; 20 cigarettes in 8 h, could inhale 0.067-0.568 µg of BaP	Human oocytes - A total of 156 (54 smokers & 102 nonsmokers) women undergoing IVF, classified as non-smokers (n = 102).	BaP's reactive metabolic binds covalently to DNA, forming adducts. Smoking-related adducts were detectable in ovarian granulosa-lutein cells, oocytes, spermatozoa & preimplantation embryos. Transmission of altered DNA from smoking by spermatozoa was demonstrated in preimplantation embryos and in association with increased risk of childhood cancer.	Discusses DNA damage with respect to adducts specific to BaP	Excellent review with respect to adducts specific to BaP	Review paper for BaP (focused on tobacco smoke)
Zenzes et al. (1995)	Reproductive	Chronic	Inhalation (smoking)	Women included in study categorized based on smoking frequency. (meiotic maturation of human oocytes) - chronic	Human oocytes - A total of 156 (54 smokers & 102 nonsmokers) women undergoing IVF, classified as non-smokers (n = 102).	Found higher frequencies of diploid oocytes in smokers than in non-smokers, and a very significant dose effect (increased frequency with increased smoking). The observed increased proportion of analysable oocytes in the smoker group suggests an earlier delay in oocyte maturation, compared with non-smokers.		Study shows that external factors, like cigarette smoking, may be hazardous to the viability and function of developing	Good paper with direct relevance to human fertility and BaP alone
Zhao et al. (2011)	Reproductive	Chronic	Oral - Najing city (China) tap water	Control = drinking water free of toxins; treatment = Najing tap water with contaminant levels determined by GC/MS. (90 days)	Male mice, age = 3 weeks old (n=40), weight = 18±1 g	In treated mice, flow cytometry analysis of testicular tissue indicated that the relative percentage of the elongated spermatid (HC) decreased significantly. Also slight increases in the relative percentage of round spermatids (IC) and primary spermatocytes (4C) were noted. The ratios of 4C:2C (diploid germ cells) and 1C:2C increased, and testicular histopathology indicated an expansion of interstitial space and a decreased number and size of Leydig cells in treated mice.		Current study suggests that Nanjing City tap water is toxic to the reproductive system of mice and additional study	Drinking water study, including polycyclic aromatic hydrocarbons
Zheng et al. (2010)	Reproductive	Chronic	Oral gavage	(90 days) Rats were randomly divided into 7 groups; 2 groups received DBP + BaP (DBP+BaP: 50+1 or 250+5 mg/kg/day), 4 groups received DBP or BaP alone (DBP: 50 or 250 mg/kg/day; BaP: 1 or 5 mg/kg/day), and 1 group received vehicle alone (corn oil = control). Dose selection: 1hr.	Adult male Sprague-Dawley rats, age = 5 weeks old; BW = 70-100g; n=16/group	ED2- testicular macrophages (reactive with a differentiation-related antigen present on the resident macrophages) were activated and IL-1β secretion was enhanced. DBP and BaP acted additively, as demonstrated by greater IL-1β secretion relative to each compound alone. These observations suggest that exposure to DBP plus BaP exerted greater suppression on testosterone production compared with each compound alone. DBP and BaP enhances the mRNA and protein expression of IL-1β in testicular macrophages. Exposure to DBP and BaP alters testicular macrophage subset expression and enhances the ability and efficiency of resident macrophages to secrete IL-1β, a significantly suppresses testosterone.	Hypothetical representation of the effects of chronic exposure to DBP and BaP on testicular macrophage function and testosterone production in the testis of rat (figure 6, n = 36). Testis	Findings suggest that co-exposure to DBP and BaP had additive effects on increased mRNA expression of IL-1β, although this did not occur in a dose-dependent manner.	

**Supplemental Table 4.** BMD model fit parameters for apical endpoints in Table 3. Benchmark response: BMD<sub>10</sub>/BMDL<sub>10</sub> for quantal data (tumor) and BMD<sub>1SD</sub>/BMDL<sub>1SD</sub> continuous data (Neurotoxicity, developmental, immunotoxicity and mutations).

Reference	Effect	Best Model	AIC	P-value	BMD	BMDL
Chen et al. (2012)	Developmental toxicity	Hill	191.1	1.0	0.09 (0.05†)	0.05 ( 0.02†)
De Jong et al. (1999)	Immunotoxicity	Exponential	333.5	0.6	14.0 (7.6†)	8.9 (4.8†)
Lemieux et al. (2011)	Mutations: forestomach	Linear	148.3	0.6	0.49	0.3
Lemieux et al. (2011)	Mutations: lung	Linear	137.7	0.1	2.2	1.4
Lemieux et al. (2011)	Mutations: liver	Power	121.1	1.0	7.2	4.8
Culp et al. (1998)	Forestomach Tumor	LogLogistic	96.5	0.4	0.83	0.54
Wester et al. (2012)	Forestomach Tumor	Multistage	179.9	1.0	1.54 (0.83†)	0.75(0.41†)
Wester et al. (2012)	Liver Tumor	LogLogistic	127.8	0.5	3.27 (1.77†)	2.36 (1.28†)

† For comparison of rat and mouse BMDLs were scaled from rat to mouse by multiplying rat values by C based on the assumption that the physiological processes scale with body weight to the <sup>3</sup>/<sub>4</sub> power (allometric scaling).

**Supplementary Table 5: Custom gene list for PCR arrays, including housekeeping genes (denoted by one \*) and controls (denoted by two \*\*).**

<b>Gene</b>	<b>Syml Refseq #</b>	<b>Official Full Name</b>
Bax	NM_007527	Bcl2-associated X protein
Bcl2	NM_009741	B-cell leukemia/lymphoma 2
Blk	NM_007549	B lymphoid kinase
Blnk	NM_008528	B-cell linker
Btk	NM_013482	Bruton agammaglobulinemia tyrosine kinase
Cd19	NM_009844	CD19 antigen
Ccnb2	NM_007630	Cyclin B2
Ccnd1	NM_007631	Cyclin D1
Ccng1	NM_009831	Cyclin G1
Cd19	NM_009844	CD19 antigen
Cd3g	NM_009850	CD3 antigen, gamma polypeptide
Cd40lg	NM_011616	CD40 ligand
Cd8a	NM_001081110	CD8 antigen, alpha chain
Cd8b1	NM_009858	CD8 antigen, beta chain 1
Cdkn1a	NM_007669	Cyclin-dependent kinase inhibitor 1A (P21)
Cxcr5	NM_007551	Chemokine (C-X-C motif) receptor 5
Cyp1a1	NM_009992	Cytochrome P450, family 1, subfamily a, polypeptide 1
Cyp1b1	NM_009994	Cytochrome P450, family 1, subfamily b, polypeptide 1
Cyr61	NM_010516	Cysteine rich protein 61
Dock2	NM_033374	Dedicator of cyto-kinesis 2
Gadd45a	NM_007836	Growth arrest and DNA-damage-inducible 45 alpha
Gadd45g	NM_011817	Growth arrest and DNA-damage-inducible 45 gamma
Gsta1	NM_008181	Glutathione S-transferase, alpha 1 (Ya)
Gsta2	NM_008182	Glutathione S-transferase, alpha 2 (Yc2)
Mdm2	NM_010786	Transformed mouse 3T3 cell double minute 2
Mgmt	NM_008598	O-6-methylguanine-DNA methyltransferase
Nqo1	NM_008706	NAD(P)H dehydrogenase, quinone 1
Pdgfa	NM_008808	Platelet derived growth factor, alpha
Pmaip1	NM_021451	Phorbol-12-myristate-13-acetate-induced protein 1
Polk	NM_012048	Polymerase (DNA directed), kappa
Sesn2	NM_144907	Sestrin 2
Srxn1	NM_029688	Sulfiredoxin 1 homolog (S. cerevisiae)
Tnfrsf10b	NM_020275	Tumor necrosis factor receptor superfamily, member 10b
Trp53inp1	NM_021897	Transformation related protein 53 inducible nuclear protein 1
Ugt1a9	NM_201644	UDP glucuronosyltransferase 1 family, polypeptide A9
Vegfa	NM_009505	Vascular endothelial growth factor A
Vegfc	NM_009506	Vascular endothelial growth factor C
Zmat3	NM_009517	Zinc finger matrin type 3
Gusb*	NM_010368	Glucuronidase, beta
Hprt*	NM_013556	Hypoxanthine guanine phosphoribosyl transferase
Gapdh*	NM_008084	Glyceraldehyde-3-phosphate dehydrogenase
MGDC**	SA_00106	Mouse Genomic DNA Contamination
RTC**	SA_00104	Reverse Transcription Control
PPC**	SA_00103	Positive PCR Control





















**Supplementary Table 8.** BMD modeling parameters for the lowest BMD<sub>low</sub> values at the 10th percentile of all genes in a pathway for different approaches for POD derivation.

Issue	Approach	Gene count	IPA Pathway	# probes with B	BMD <sup>a</sup>	Gene <sup>a</sup>	BMD List	BMD List	# probes with B	Log <sub>10</sub> (BMD) List	Log <sub>10</sub> (BMD) List
Liver	Metagenomics key event preceding the committed step	34(24)	Cell Cycle: G1/S Checkpoint Regulation	18	8.1	1.0	11, 2245452; 6, 1417514.2; 4, 3388; 38, 4101; 39, 3633	22770, 2773, 0, 51	145702, 72885, 44, 91553, 27, 74456, 33, 744893, 71, 107176, 47, 408112, 42, 468374, 32, 625013, 74, 622873, 61, 21817, 87, 738441, 44, 622087, 40, 612486, 67, 720287, 40, 91771, 61, 104479, 38, 629198, 61, 682766		
	Metagenomics lowest MCM-associated pathway	34(24)	Cell Cycle: G1/S Checkpoint Regulation	18	8.1	1.0	4, 32357; 6, 45232; 10, 11, 1854548; 5, 1417514.2; 4, 3388; 38, 4101; 39, 3633	42279, 2773, 0, 51	149702, 72885, 44, 91553, 27, 74456, 33, 744893, 71, 107176, 47, 409112, 42, 468373, 32, 625013, 74, 622873, 61, 21817, 87, 738441, 44, 622087, 40, 612486, 67, 720287, 40, 91771, 61, 104479, 38, 629198, 61, 682766		
	Metagenomics lowest pathway	34(24)	Neuron Signaling	7	0.3	0.2	12, 73002; 9, 27238; 1, 18725; 0, 157628; 22, 6304; 35, 4455; 31, 109148; 47, 9182	32963, 4692, 0, 51	147844, 47, 296784, 26, 38, 370287, 46, 691987, 44, 68602, 17, 629028, 73, 652294, 48, 820274, 43, 644694		
Lung	Metagenomics key event preceding the committed step	28(24)	Cell Cycle: DNA/DNA Damage Checkpoint Regulation	5	14.8	1.7	11, 8376; 18, 1444; 1, 8376; 48, 17802; 10, 7244; 37, 1056; 37, 7734	10760, 2414, 0, 18	332483, 28, 888742, 22, 33, 849187, 25, 773489, 37, 448109, 61, 63866, 11, 67119		
	Metagenomics lowest MCM-associated pathway	28(24)	Cell Cycle: DNA/DNA Damage Checkpoint Regulation	5	14.8	1.7	11, 8376; 18, 1444; 1, 8376; 48, 17802; 10, 7244; 37, 1056; 37, 7734	10760, 2414, 0, 18	332483, 28, 888742, 22, 33, 849187, 25, 773489, 37, 448109, 61, 63866, 11, 67119		
	Metagenomics lowest pathway	28(24)	Cellular Effects of Silencing (Via)gna	9	15.7	2.1	12, 9776; 18, 4502; 1, 8376; 48, 17802; 10, 7244; 37, 1056; 37, 7734	10560, 5163, 0, 18	4, 2358; 119, 882873, 22, 40, 47072, 31, 365746; 63, 647724; 33, 405614; 26, 88438; 34, 878652; 36, 046556; 50, 193724; 66, 227593		
Fore stomach	Metagenomics key event preceding the committed step	28(24)	IGF Signaling	40	11.4	7.4	7, 51405; 10, 9897; 2, 26767; 20, 670; 23, 6247; 32, 6252; 7, 4956; 67, 47387; 7, 308	1424, 3481, 0, 16	2278717, 17, 382953, 23, 38, 65343, 26, 764106; 38, 793848; 57, 877971; 44, 68418; 40, 918496; 34, 685842; 42, 87828; 22, 686971; 37, 835487; 41, 853786; 31, 598078; 45, 89308; 67, 833543; 40, 434833; 26, 69346; 56, 688843; 25, 699202; 38, 388448; 71, 818848; 39, 638767; 19, 253196; 38, 614993; 48		
	Metagenomics lowest MCM-associated pathway	28(24)	IGF Signaling	40	11.4	7.4	7, 51405; 10, 9897; 2, 26767; 20, 670; 23, 6247; 32, 6252; 7, 4956; 67, 47387; 7, 308	1424, 3481, 0, 16	2278717, 17, 382953, 23, 38, 65343, 26, 764106; 38, 793848; 57, 877971; 44, 68418; 40, 918496; 34, 685842; 42, 87828; 22, 686971; 37, 835487; 41, 853786; 31, 598078; 45, 89308; 67, 833543; 40, 434833; 26, 69346; 56, 688843; 25, 699202; 38, 388448; 71, 818848; 39, 638767; 19, 253196; 38, 614993; 48		
	Metagenomics lowest pathway	28(24)	Protein/Lipid Degradation (V) (Mammalian, via Side Chain)	6	16.1	4.5	16, 7734; 16, 4843; 6, 394146; 7, 8; 9, 85744; 8, 65012; 37, 873; 38, 641654; 8954	10560, 0, 7442, 0, 28	11, 1028454; 420712; 3, 21, 383281; 0, 841424; 12, 56976; 26, 368895; 24, 038787; 38, 64989		

<sup>a</sup> In mg BAP/kg bw per day  
<sup>b</sup> Similar to Thomas et al., (2011) but corrected BMD-BMD<sub>low</sub> at the 10th percentile rather than median.