

The Origin, Fate, and Health Effects of Combustion By-Products: A Research Framework

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Incomplete combustion processes can emit organic pollutants, metals, and fine particles. Combustion by-products represent global human and environmental health challenges that are relevant not only in heavily industrialized nations, but also in developing nations where up to 90% of rural households rely on unprocessed biomass fuels for cooking, warmth, and light. These issues were addressed at the Seventh International Congress on Combustion By-Products, which convened 4–6 June 2001 in Research Triangle Park, North Carolina. This congress included a diverse group of multidisciplinary researchers and practitioners who discussed recent developments and future goals in the control of combustion by-products and their effects of exposure on human and ecologic health. Participants recommended that interdisciplinary, coordinated research efforts should be focused to capitalize on the important potential synergisms between efforts to reduce the adverse human health effects linked to exposures to combustion by-products and broader efforts to reduce greenhouse gas emissions and save energy through efficiency. In this article we summarize the principal findings and recommendations for research focus and direction. *Key words:* combustion by-products, fine particles, genetic susceptibility, lung pathobiology, metals, polychlorinated dibenzodioxins, polychlorinated dibenzofurans, public health. *Environ Health Perspect* 110:1155–1162 (2002). [Online 26 September 2002]

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Inefficient combustion processes result in incomplete combustion and can emit complex mixtures of organic pollutants, metals, and fine particles due to incomplete combustion. Although this concept generally brings to mind the image of a tall smokestack spewing a dark plume, combustion sources are greatly varied and quite often not related to industrial facilities. Combustion processes include manufacturing processes, residential fuel use for heating and cooking, and open burning, as well as the diverse internal combustion engines found in jets, construction equipment, cars and trucks, lawnmowers, and so on. Not only are combustion processes diverse, but the by-products emitted (defined as products other than the most thermodynamically stable reaction products, i.e., carbon dioxide and water) vary with process efficiency and fuel type.

Indoor and outdoor environments are widely contaminated by complex mixtures of combustion-derived gases and particles (1), and these pollutants create global human and environmental health problems. Because combustion sources are found worldwide, it is not only the residents of industrial cities in developed nations that face the environmental health risks from exposure to combustion by-products. Moreover, by-products of combustion

processes have potential for wide-ranging impacts, not only on human health but on entire ecosystems and the global climate. These issues were addressed at the Seventh International Congress on Combustion By-Products (held 4–6 June 2001 in Research Triangle Park, NC). This congress included a diverse group of multidisciplinary researchers and practitioners who discussed recent developments and future goals in the control of combustion by-products and their effects of exposure on human and ecologic health. In this article we summarize the principal findings and recommendations for research focus and direction. A key need is the development of technologies to provide safe, clean, and efficient energy systems, with the ultimate goal of translating discoveries to public health intervention strategies.

Combustion By-Products: Impacts on a Global Scale

Environmental health policy has primarily focused on issues that are considered global in scale. Current policies emphasize environmental issues that are global in the sense that the environmental impact is the result of worldwide emissions or atmospheric formation of a single pollutant or small identifiable group of pollutants. For example, combustion-generated

hydrocarbons and nitrogen oxides react in the atmosphere in the presence of heat and sunlight to form harmful concentrations of ground-level ozone. But it is not only cities with high numbers of industrial facilities or cars that are subject to high ozone levels, as winds carry nitrogen oxides (NO_x) emissions hundreds of miles away from their original sources (2).

Global climate change. With regard to global warming, the surface temperature of Earth has increased approximately 1.2°C since 1850 (3), with 0.5°C of the change occurring since 1978 (4). Predictions for future changes in our climate vary, but most scenarios predict a steady increase in global temperature throughout the twenty-first century (5). The magnitude of the impacts of global warming on human health, environmental health, and national economies remains unknown. Climate change will likely cause more economic damage in developed countries and more human health damage in less developed countries (6).

There is a growing consensus among many scientists that global warming is, at least in part, the result of increasing levels of anthropogenic greenhouse gases [carbon dioxide (CO₂), nitrous oxide (N₂O), methane (CH₄), chlorofluorocarbon (CFC)] and tropospheric aerosols (7). Industrial, energy-producing, and residential combustion processes contribute significantly to global levels of greenhouse gases (GHG). Burning of fossil

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The steering committee and participants of the Seventh International Congress on Combustion By-Products dedicate the congress and this article to their colleague, Glen R. Cass, who died 30 July 2001. We salute his integrity, his contribution to the field of atmospheric sciences, and his professional achievements.

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fuels is the major source of CO₂ emissions, as well as a major source of aerosols, especially black carbon, organic aerosols, and sulfates (5). Combustion processes are also major emitters of NO_x and sulfur oxides (SO_x) (8).

Human health effects. The effects of combustion by-product pollution on human health are real and need to be addressed by sound scientific research. The practical benefits of efforts to reduce pollutant emissions from combustion processes would be realized immediately (9). Environmental health researchers have estimated that reducing emissions from older coal-fired power plants in the United States could provide substantial benefits to public health, including the avoidance of 18,700 deaths, 3 million lost work days, and 16 million restricted-activity days each year (10). By reducing emissions from nine older coal plants in the Midwest alone, it has been estimated that roughly 300 deaths, 2,000 respiratory and cardiac hospital admissions, 10,000 asthma attacks, and 400,000 person-days of respiratory symptoms could be avoided each year (11). Some scientists have also predicted that reducing GHG emissions in just four major cities (Sao Paulo, Brazil; Mexico City, Mexico; Santiago, Chile; and New York City, USA) could save 64,000 lives over the next 20 years (10–12). It has been suggested that investments in modern technology and air quality control in India and China could lead to reductions in tropospheric ozone and black carbon that would not only improve local health and agricultural productivity but also benefit air quality and the global climate (5).

Numerous studies in urban areas have documented the relationship between urban atmospheric particulate matter pollution emitted by industrial/transportation combustion processes and respiratory and heart diseases (13–16). Less attention has been paid to the significant risk of exposure to harmful combustion by-products resulting from the indoor use of solid fuel for cooking and heating. Simple stoves burning solid fuel (mostly biomass fuel) are used by about half of the world's people (17). Poor ventilation and inefficient combustion result in significant daily exposure of women and young children to fine and ultrafine particulate matter, carbon monoxide, (CO), NO_x, SO_x, a range of volatile organic compounds including formaldehyde, and polycyclic organic matter (18). The World Bank has estimated that indoor air pollution is responsible for almost 50% of the burden of total disease resulting from poor household environments in developing countries (19).

Individual Susceptibility and Predisposition

Environmental health researchers have determined that most human disease results from a combination of environmental exposures and

genetic variation (20,21). However, it is not fully understood why certain people develop disease when challenged with environmental agents and others remain healthy. Thus, a more complete understanding of how genetic characteristics affect the human response to environmental exposures is needed to improve approaches to the prevention and control of environmentally induced diseases. Research addressing individual susceptibility, especially with respect to diseases resulting from exposure to combustion by-products, is progressing on a number of fronts, from molecular biology to epidemiology.

Genetic variations may impact innate immunity—the ability to launch an immune response to toxins, microbes, and microbial products with no previous exposure (22). This may play a role in determining the susceptibility of an individual to an environmental exposure. For example, nearly everyone is exposed to at least low levels of environmental endotoxin. This biologically active lipopolysaccharide (LPS) is a component of the outer membrane of gram negative bacteria and produces airway inflammation when inhaled. LPS has been associated with the development and progression of asthma and other forms of airway disease (23). However, not everyone exposed to LPS will develop airway disease. A broad range of stable, reproducible physiologic responses to inhaled LPS has been documented among healthy, nonasthmatic people (24). Researchers investigating the genetic basis for the physiologic response to LPS have gathered the first direct evidence that mutations in a transmembrane receptor for LPS (*TLR4*) are associated with decreased responsiveness to inhaled LPS (25). These findings suggest that changes in the gene sequence alter the innate ability of the host to respond to environmental stress, as people with the *TLR4* substitutions may be more resistant to localized forms of endotoxin-induced inflammation (e.g., asthma) but more susceptible to a systemic inflammatory response initiated or exacerbated by LPS (e.g., gram negative infections, sepsis, and adverse outcomes).

In home and/or occupational settings, most individuals are exposed to combustion-generated particles, but relatively few people with a compromised respiratory or cardiovascular system respond with increases in morbidity and mortality. As a result, researchers suggest that genetic factors play a major role in determining individual susceptibility to particulate matter (PM) exposure (26). Studies combining genome-wide scans with microarray analysis have provided comprehensive assessments of adverse responses to environmental stimuli. For example, research into individual susceptibility in mice to nickel-induced acute lethal lung injury revealed that response to this metallic constituent of particulate matter and

cigarette smoke is controlled by a region on mouse chromosome 6 with modifier genes suggested on chromosomes 1, 2, and possibly 9 and 16 (27,28). These chromosomal regions are near regions that also have been identified as controlling the response to ozone, suggesting that some of the same genes may be involved in both responses (27). This type of information may help clarify the role polymorphic variation plays in the determination of individual response to environmental exposures. Such knowledge may lead to improvements in preventive medicine, successful treatment interventions, and more effective regulatory policies (29).

Epidemiologic studies provide insight to aspects of individual susceptibility not directly related to genetic variability. These studies identify factors such as age, sex, occupation, nutritional status, lifestyle, and coexisting health conditions. These factors must be considered both independently and as potential contributors to complex interactions. For example, boilermakers were studied to evaluate that role of occupational exposure to particulate matter in the development of pulmonary and respiratory diseases. Boilermakers are routinely exposed to boiler fossil fuel ash as well as particles and gases emitted during welding activities. Boiler ash composition varies with the type of fuel (oil, coal, natural gas, or trash) and may include fine particulate matter, polycyclic aromatic hydrocarbons (PAHs), and metals such as vanadium, nickel, iron, zinc, chromium, and arsenic. As dictated by maintenance and construction needs throughout the course of a year, a boilermaker may work at a variety of plants that burn multiple fuel types. The results of a 2-year longitudinal study (30) conducted to establish if exposure of boilermakers to particulate matter is linked to loss of lung function suggest that working at gas, oil, and coal-fired plants is associated with adverse effects on lung function. A short-term prospective study to assess the relationship between continuously measured particles with an aerodynamic diameter (i.e., diameter of a sphere of unit density that has aerodynamic behavior identical to that of the particle of interest) < 2.5 μm and cardiac autonomic function in a group of boilermakers exposed to fuel-oil ash and metal fumes (31) revealed decreased variability in heart rate after occupational exposure, indicating autonomic dysfunction. This finding demonstrates the effects of fine particulate matter on the autonomic nervous system and may provide physiologic evidence for the mechanism linking particulate matter and increased morbidity and mortality.

Combustion By-Products: Concerns and Current Research

The physical state and chemical composition of combustion by-products vary with the type of

combustion source, the efficiency of the combustion process, and the fuels that are burned. Combustion by-products can include CO, NO_x, PAHs, polychlorinated dibenzo-*p*-dioxins (PCDDs) and dibenzofurans (PCDFs), particulate matter, and metals (32). Each of these pollutants has been associated with adverse impacts on human health, the environment, or the global climate (33–35). Researchers and regulators are paying particular attention to several combustion by-products, and a wide range of studies is being conducted to increase our understanding of the contaminant levels, risks of exposure, health impacts, and mechanisms of disease etiology.

Particulate air pollution. Particulate air pollution is characterized by both particle size and chemical composition. Fine particles [those with an aerodynamic diameter < 2.5 μm (PM_{2.5})] are derived chiefly from combustion processes or are formed as secondary combustion particles including sulfates and nitrates (36). Particles in the approximate size range of 0.2–0.5 μm can be transported long distances and readily penetrate indoors, resulting in ubiquitous exposure. Fine particles are small enough to be breathed deeply into the lungs, exposure is widespread, difficult to avoid, and occurs throughout the entire life. The public health significance of fine particulate air pollution is substantial. One estimate of average population decrease in life span due to exposure to fine particulate air pollution for lifelong residents of some of the developed world's more polluted cities is 1–3 years (37).

Because no diagnostic test can prove causality, epidemiologic studies are used to evaluate the relationship between natural variability in exposure and variability in rates of illness (38). Many acute and chronic population-based epidemiologic studies have been conducted, and the overall evidence indicates a probable link between fine particulate air pollution and adverse effects on cardiopulmonary health, suggesting that all individuals who are chronically exposed may ultimately be affected (37). To investigate the physicochemical characteristics of particulate air pollution that effect the cardiorespiratory system and the mechanisms of action, epidemiologists examined the relationship between hospital emergency room data and ambient air monitoring data (39,40). Fine particulate matter was fractionated to study the impacts of sulfates, acidity, water-soluble metals, organic matter (OM), and elemental carbon (EC). The researchers observed a statistically significant, positive association between cardiac dysrhythmia and CO, coarse PM, and EC < 2.5 μm (PM_{2.5} EC), as well as between all cardiovascular diseases and CO, PM_{2.5} EC and OM < 2.5 μm (PM_{2.5} OM). Although covariation of many of the air quality indices may confound such analyses, the study

provides one of the first assessments of PM components in relation to emergency room visits.

Relationships between exposure to fine particles and genetic mutations have just recently been established because epidemiologic studies require much longer monitoring periods and broader monitoring of possible end points (41). It has now been demonstrated that each elevation of exposure to 10 μg/m³ of fine particulate air pollution is associated with an 8% increase in lung cancer mortality and a 6% increase in cardiopulmonary mortality. Thus, the fine particle-associated risk of death due to cancer is apparently greater than the risk due to cardiopulmonary disease. The U.S. Environmental Protection Agency's (EPA) current air standards for fine particle air pollution are based on the older study of cardiopulmonary disease and do not include the greater risk of death due to cancer (8).

Regulatory and industry efforts to control emissions of fine particulate pollutants depend on an accurate assessment of the relative contributions of different types of emission sources to ambient pollutant concentrations. This assessment is based on emission inventories compiled from source data and air quality models. Research has been conducted to improve methodologies for measuring primary emissions from a broad range of stationary and mobile sources and to evaluate post-emission gas to particulate conversions (42–44). Recent modifications of air quality models enable scientists to monitor the fate of particles emitted from different source types and calculate the contributions from each source to ambient air quality for use in the design of emission control programs (45). Advanced development of receptor-source models has complemented these source-receptor methods. Much of the critical work in the advancement of fine particulate matter sampling methods, development of air quality models, and development of defining source apportionment through “fingerprinting” was conducted by the late Glen R. Cass.

Ultrafine particulate air pollution. Scientists have documented health effects of occupational exposure to ultrafine particles (those with an aerodynamic diameter < 0.1 μm) (46), but the impacts of exposure to ultrafine particles in urban air from combustion sources such as automobiles, natural gas combustion, and electric engines remain unknown. Ultrafine particles contribute little to the total particulate mass in urban air but are very high in number, reaching several hundred thousand particles per cubic centimeter. Ultrafine particles 20 nm in diameter have the highest deposition efficiency in the alveolar region of the lungs; ultrafine particles can rapidly penetrate the epithelium so that they can be detected in the interstitium of the

lung shortly after exposure (47). Because ultrafine particles are always present in the urban atmosphere, it is likely that they play a role in causing acute lung injury in sensitive parts of the population.

As a result of their small size, ultrafine particles have a large surface area and such particles may become coated or “enriched” with contaminants in combustion flue gas or the atmosphere including toxic metals (lead, cadmium, arsenic, chromium, and zinc), sulfur, PAHs, partially oxidized hydrocarbons including oxy-PAH (OPAH), and other heteroatom-containing species such as PCDDs and PCDFs (48). Fine particles also stabilize biologically active, organic radicals that induce oxidative stress and DNA damage (49–51). The combination of PM-borne, persistent semiquinone-type radicals or organics and metals can lead to interactions resulting in the generation of reactive oxygen species (ROS) and other secondary, biologically active species. These findings represent a possible mechanism for the genesis of cancer due to exposure to fine particulate air pollution (41).

Research with rodent models (52) indicates that, per given mass, ultrafine particles administered to the lung cause a greater inflammatory response than larger particles and that coagulation of ultrafine particles to larger particles significantly decreases their toxicity. The pulmonary inflammatory potential of ultrafine carbonaceous particles can be significantly enhanced in rodents by gaseous co-pollutants such as ozone. In addition, surface chemistry may play an important role in ultrafine particle toxicity. Susceptibility to the inflammatory effects of ultrafine particles in rats and mice increases significantly with age and/or a compromised/sensitized respiratory tract. It also appears that after deposition in the lung, ultrafine particles are not efficiently phagocytized by alveolar macrophages (47,53). Translocation of ultrafine particles to the liver and of ultrafine dye particles along neurons has been documented, suggesting that organ systems other than the lungs may be impacted by ultrafine particles (54,55).

Elemental speciation of arsenic. Arsenic is emitted into the atmosphere by combustion processes such as coal-fired power generation plants, household use of coal fuel, burning of vegetation, and volcanic activity (56). Arsenic is released into the atmosphere primarily as arsenic oxide (As₂O₃) and exists mainly adsorbed on particulate matter. These particles are dispersed by the wind and are returned to the earth by wet or dry deposition. In well-oxygenated water and sediments, nearly all arsenic is present in the thermodynamically stable pentavalent state (arsenate) (57).

The toxicity of arsenic species varies greatly (58). Inorganic arsenic compounds are highly toxic, with the anionic form, arsine (AsH₃),

being more toxic than cationic arsenite (As^{3+}) and arsenate (As^{5+}). Some organic arsenicals are nontoxic to humans. As a result of this variation, evaluation of total arsenic concentration is inadequate for estimating risk from arsenic present in food and drinking water supplies. Quantitative methods for assessing specific forms of arsenic are necessary for risk assessment studies and relative source contribution estimates. Identification and quantification of the specific forms of arsenic often require the combination of two complementary techniques: efficient separation of the species of interest and detection/quantitation. Systems using liquid chromatography (LC) followed by inductively coupled mass spectrometry (ICP-MS) can be used for elemental speciation at sub-nanogram and sub-picogram levels (59). Evaluation of the LC-ICP-MS system for potential interferences revealed that chloride can contribute minimally to As^{5+} determinations and potential chromatographic interferences from iron could be minimized by the addition of a chelating agent such as EDTA, and this speciation method should satisfy U.S. EPA validation requirements (60).

Metals in particulate matter. $\text{PM}_{2.5}$ particles are generally attributable to emissions derived from combustion sources and are often coated with contaminants from combustion flue gas, including toxic metals (61). Recent studies provide novel insights into the early events in activation of signaling cascades that lead to altered cellular function after exposure to metals present in ambient air PM. Exposure to combustion-derived metal species such as As, V, and Zn is known to dysregulate phosphoprotein metabolism, resulting in the modulation of intracellular signaling cascades and transcription factors. Current research indicates that these PM metals activate epidermal growth factor receptor (EGFR) and increase levels of guanosine triphosphate-bound Ras in human lung cells, and Ras activation is required for Zn-induced signaling (61,62). Although Zn-induced activation of Ras requires EGFR, it can still occur in cells expressing kinase-inactive EGFR or EGFR that is truncated or missing the regulatory autophosphorylation domain. In addition, phosphorylation of EGFR induced by Zn can be inhibited by an Src inhibitor and Src phosphorylation is markedly increased following Zn exposure. These data suggest that EGFR signaling by metal ions does not occur through a direct effect on the receptor, but is due to the activation of Src kinase. These findings show novel mechanisms of action by PM metals and may provide insight relevant to the health effects of PM exposure.

Mechanisms of chromate mutagenesis. Chromium is a widespread environmental contaminant and a known human carcinogen (63). In the environment, Cr exists primarily

in the trivalent [Cr(III)] and hexavalent [Cr(VI)] forms. Compounds of Cr(VI), but not Cr(III), have been found to be mutagenic and carcinogenic (64). Chromate (CrO_4^{2-}), the major ionic form of Cr(VI), readily penetrates cell membranes (65).

Researchers are using shuttle vector systems in yeast, mammalian cells, and lung tissue of transgenic mice to investigate the mechanisms of chromate mutagenesis (66,67). Chromate is reduced by glutathione, and mutagenesis is influenced by tissue levels of glutathione. Chromate reduction is accompanied by the generation of highly reactive Cr(V), a reactive intermediate, and reactive oxygen intermediates which can cause DNA damage including intra- and interchromosomal deletions and base substitutions. These results support the hypothesis that Cr-induced mutagenesis is due to the generation of reactive oxygen intermediates during the reduction of Cr(VI) by glutathione. In studies conducted with a transgenic mouse system, researchers noted a small but significant increase in mutagenesis in kidney DNA in addition to Cr impacts on lung tissues (68).

Dioxins. PCDDs and PCDFs are ubiquitous environmental contaminants. Dioxins are potent animal toxicants, and the complex mixture of dioxins to which people are exposed is characterized by the U.S. EPA as a "likely human carcinogen" (69). These compounds have never been produced intentionally nor used commercially in the United States, but are by-products of both anthropogenic and natural combustion processes. Historically, municipal and commercial waste incineration, pesticide manufacture and use, and pulp and paper processing have been believed to be the major sources of emissions. Voluntary industry actions, combined with regulatory controls, have resulted in dramatic reductions from these sources in the United States. Currently, uncontrolled burning of residential waste and accidental fires are thought to be among the largest sources of PCDD/PCDF emissions in the United States (70).

PCDD/PCDF formation in combustion processes involves a series of complex mechanisms, and specific aspects of these reactions have been the subjects of controversy. In general, PCDD/PCDF formation is a catalytic reaction of carbon with oxygen and chlorine, strongly dependent on the type and amount of catalyst, temperature, duration of combustion, and the type and amount of carbon (71). On-going research will provide a more complete understanding of dioxin formation to formulate strategies to reduce emissions.

The influence of the polyvinylchloride (PVC) content of the waste stream on the formation of PCDDs/PCDFs during municipal solid waste (MSW) combustion has been debated over the last several years. Special

interest groups (e.g., Greenpeace and Physicians for Social Responsibility) called for de-emphasis on pollution controls and new emphasis on control of the waste stream, replacing PVC with alternative, chlorine-free materials (72,73). A recent study demonstrated that PCDD/PCDF formation is controlled by variation in combustion conditions, not PVC content or the amount of chlorine in the waste (74). Research on methods to reduce PCDD/PCDF emissions from small-scale commercial boilers for energy recovery and waste disposal indicates that if sufficient high-quality fuel is included in the waste stream, dioxin/furan emissions can meet the new European Union directives without the need for cyclone cleaning (75).

The impacts of temperature and residence times on PCDD/PCDF formation have also been examined, and studies conducted in a pilot-scale solid fuel bed reactor revealed different formation pathways for dioxins and dibenzofurans. Chlorination of dibenzofurans occurs after longer dwell times and at lower flue-gas temperatures. Nearly all of the chlorine in flue gas is present as hydrogen chloride, and a catalyst is required for chlorination to occur, suggesting that the activity of the catalyst controls the formation rate of chlorinated furans. Additionally, these studies showed that when bromine is added to the fuel, more brominated than PCDFs are formed (76). A separate study to investigate the impact of increased bromine levels in a waste stream containing flame retardants found the majority of tetra-halogenated dioxins/dibenzofurans formed during combustion were dibromo-dichloro-compounds (77).

Advanced Technologies: Applicability to Research on Combustion By-Products

Questions concerning combustion by-products that remain unanswered include specific issues regarding formation, control, transport, fate, exposure, and human and environmental health impacts. Powerful new analytical methodologies, including microarray technology and nanotechnology, are providing novel approaches to investigate these issues. These same advances are also fueling the growth of new scientific disciplines such as genomics, proteomics, toxicogenomics, and metabonomics. These new paths of investigation have the potential to lead to information, and new research strategies to elucidate toxicologic mechanisms which will provide the answers to some of the questions surrounding the environmental health impacts of combustion by-products.

Nanotechnology, the fabrication of devices with atomic or molecular scale precision (a nanometer is one-billionth of a meter), will allow scientists to build biosensors using semiconductor, organic, and hybrid materials (78).

Potentially, nano-biosensors could provide rapid response time, real-time monitoring, and the capability to analyze biologic phenomena at the cellular level. They may reveal valuable basic information about intracellular responses to the toxicants in combustion by-products and delineate the origins of their adverse health effects. External biosensors may be capable of monitoring emissions of combustion by-products using compact solid state modules.

In addition, nanotechnology is vital to the fabrication of DNA microarrays, which are powerful, high-throughput tools for simultaneously monitoring the expression of thousands of genes or even the entire genome. In a microarray, copies of DNA, each from a separate gene, are printed on “platforms” which can be glass slides, silicon chips, or nylon membranes (79). After exposure to a chemical, gene expression is assessed by measuring the binding of mRNA to the microarray DNA. Generally the mRNA is tagged with fluorescent components, creating patterns of fluorescent dots that reveal which genes are expressed. Microarrays have a sensitivity and dynamic range similar to Northern blot analyses. Microarray slide-to-slide and spot-to-spot variability are less than animal-to-animal variability (80), and assay reproducibility is continually improving.

The vast data derived from studies with microarrays are playing a critical role in the emergence of genomics, toxicogenomics, proteomics, and metabonomics. For example, microarray technologies have provided the tools needed to support the new scientific sub-discipline of toxicogenomics, which combines genomics and bioinformatics to identify and characterize the relationships between chemical exposure and changes in genome-wide gene expression patterns in target cells. Microarrays will likely be used in the immediate future to better address a number of key toxicologic issues, including mode of action, dose-response relationships, chemical interactions, and the use of gene profiling for hazard identification in chemical mixtures and human exposure assessment. Genomics, toxicogenomics, proteomics, and metabonomics have the potential to change the classic risk assessment paradigm. Microarray technologies will allow scientists to move past the current focus on stepwise identification of single biomarkers of exposure to a specific chemical, and characterize gene profiles that identify multiple environmental stressors. This knowledge will allow us to sort out the subtleties of multiple and cumulative exposures as well as investigate the effects of timing, duration, and sequence of exposures (81,82).

Genomics, toxicogenomics, proteomics, and metabonomics will provide significant amounts of scientific data, but this information will be very technical and will seldom provide unambiguous answers to legal and

ethical questions. These data are vulnerable to misinterpretation by scientists, regulators, the media, and the general public. It is critical that scientists and scientific institutions make a commitment to stress effective communication of research findings to policymakers and the general public. It is vital to ensure that information is presented in a fashion that is appropriate for the audience, especially when the information relates to risk. The general public is frequently presented with information that is incomplete and/or misleading. It is critical to solicit and respond to input from stakeholders and community members when designing research programs. This inclusive approach helps build social investment in research results and the public health interventions needed to protect human and environmental health (29,83).

In addition, DNA microarrays will be used to rapidly classify mechanism-based toxicants by characteristic expression profiles and also identify genes that are under similar regulatory control (84). It may be possible to use microarrays to examine the molecular background of differences in individual susceptibility to certain toxic chemical groups (80).

Although an emerging consensus suggests that many of the complex and prevalent diseases that humans develop occur as a result of multiple biologically unique gene-gene and gene-environment interactions, this conceptual framework is limited. In fact, the development of disease in humans, environmental and otherwise, is far more complex. Environmental exposures affect those that are vulnerable temporally (age), spatially (geographically), and by unique circumstance (co-morbid disease, nutritional status, and genetics). Even this paradigm fails to address the complex interaction of endogenous and exogenous risks that ultimately interact to cause disease. Although the recent advances in human and molecular genetics have provided an unparalleled opportunity to understand how genes and genetic changes interact with environmental stimuli to either preserve health or cause disease, without accounting for the temporal, spatial, and other unique components of an individual's microenvironment, our understanding of environmental health will remain incomplete. These concerns are particularly applicable to the health consequences of combustion by-products. Future research efforts need to embrace the complexity involved in etiology and pathogenesis of disease rather simply reduce the model to an understandable, but minimally applicable, gene-exposure-response relationship.

Regulatory and Policy Issues Related to Particulate Matter

Particulate matter is a criteria pollutant, which means that national ambient air quality

standards (NAAQS) have been developed to protect public health, public welfare, and the environment. These standards must be reviewed every 5 years. In 1997 the U.S. EPA published a final rule revising the NAAQS for PM (8). While retaining the PM₁₀ standard levels, the U.S. EPA added new PM_{2.5} standards on the basis of epidemiologic studies that related ambient PM concentrations to various health effects, including premature mortality, exacerbation of asthma, and other respiratory tract diseases, and decreased lung function (85).

Fine particles behave entirely differently from coarse particles and are much more difficult and expensive to control. Faced with the enormous financial implications of the 1997 NAAQS for PM_{2.5}, the American Trucking Associations, the U.S. Chamber of Commerce, and other state and business groups filed a challenge to the U.S. EPA's action. The U.S. Court of Appeals for the District of Columbia Circuit struck down major elements of the U.S. EPA's two NAAQS standards, but the Supreme Court upheld U.S. EPA's authority under the Clean Air Act to set national air quality standards that protect the American public from harmful effects of air pollution. On 26 March 2002, the District of Columbia Circuit Court rejected the remaining claims that the U.S. EPA's decision was arbitrary and capricious and not supported by the evidence (86). The implementation guideline for the PM_{2.5} standards calls for completion of the scientific review in 2002; designation of nonattainment areas by 2005; submission of state implementation plans by 2008; and state compliance by 2017 (87).

An additional regulatory effort impacting particulate matter emitted as a by-product of combustion processes is the Regional Haze Rules. According to the National Academy of Sciences, the average visual range in most of the western United States, including national parks and wilderness areas, is 100–150 km (about 60–100 miles), or about one-half to two-thirds of the natural visual range that would exist in the absence of air pollution. In most of the East, including parklands, the average visual range is less than 30 km (about 20 miles), or about one-fifth of the natural visual range (88). The primary causes of regional haze are sulfates, organic matter, elemental carbon (soot), nitrates, and soil dust. The Regional Haze Rules are an attempt to tackle the combined visibility effects of numerous pollution sources through a regional, rather than a purely local-source, approach. These rules require all states to make “reasonable progress” in reducing any effect this pollution has on visibility conditions in class I areas and to prevent future impairment of visibility. This regional approach calls for the reduction of non-CO₂ GHGs and is predicted to reduce emissions of key pollutants

including NO_x, SO_x, and CO (87). Such reductions will have a positive impact on our climate and, just as importantly, have an immediate impact on our quality of life (5).

Critical international efforts to minimize or eliminate emissions of combustion by-products are illustrated by the 2001 Stockholm Convention on Persistent Organic Pollutants (POPs). This agreement calls for the elimination of production and use of all intentionally produced POPs and for continuing efforts to minimize emissions of unintentionally produced POPs with the goal of ultimate elimination. The convention requires the establishment and maintenance of source inventories to demonstrate continuing minimization of each country's total emissions. The United Nations Environment Programme is focusing efforts to ensure that all countries compile inventories that are complete and comparable (89). Incineration will undoubtedly be the most widely used method for destruction of POPs.

Ethical Issues Related to Combustion By-Products

As governments attempt to formulate appropriate responses to environmental issues, they must understand and consider a myriad of potential legal, ethical, and social issues. They must acknowledge that air pollution is a global issue and that regional and transoceanic transport of air pollutants impacts neighbors, even in distant regions.

Environmental issues related to combustion by-products must be considered from a global public health point of view. It has been argued that although the common perception is that the biggest environmental problems lie in the industrial world, from the perspective of actual ill health, environmental health impacts are most severe in less-developed nations (90). In addition, the largest burdens of environmentally related diseases (including respiratory infections related to indoor combustion of biomass fuels) in developing nations are borne by children.

Developed nations have a variety of approaches to address these issues in less-developed nations. Many of the environmental health problems in the developing world are tightly interrelated with poverty (17). Developed countries could address these issues by increasing efforts to enhance socioeconomic development, which should result in healthier household environments (90). Additionally, industrialized countries should eliminate and reduce the hazards caused by their own economic and social development and promote new affordable technologies for developing countries. Faced with the reality of finite resources, it is imperative that policymakers focus attention on developing strategies that will yield the greatest impact (6,90).

A series of potential legal, ethical, and social dilemmas evolves from research advances. This concept is illustrated by the possibilities presented by scientific advances in toxicogenomic research. Toxicogenomics will enable scientists to identify genetic variations and their impacts on individual response to environmental exposures (29). Such information could serve as the foundation for the developing strategies to prevent and control environmentally induced diseases. Alternatively, this information contributes to a suite of ethical, legal, and social challenges. Discovery that polymorphism of a specific gene is linked to an environmental disease could lead to discrimination or stigmatization. Health and life insurance companies might discriminate against individuals who carry a gene linked to a disease. Employers might terminate or reassign workers on the basis of such information. Because some polymorphisms are more common in certain populations than in others, it is possible that a particular type of genetic susceptibility might be associated with particular ethnic or social groups. This may threaten the employment and insurance options available to entire groups of people. The identification of a polymorphism that places an individual at a higher risk of developing an environmental disease could result in the individual being stigmatized as carrying a "defective gene" or as already ill, even though an environmental trigger is required for the development of the associated disease and the individual may never develop the disease (29,91,92).

Not only will policymakers have to formulate regulations to prevent discrimination and protect individual privacy, they must confront the challenges of promulgating fair and appropriate regulations based on new information at increased levels of complexity. Regulators will need to determine exposure limits, deciding if limits should be designed to protect everyone at the levels required to protect the most susceptible populations. They will have to address the question of what constitutes an acceptable risk. To address each of these issues appropriately, regulators require in-depth, accurate, and accessible scientific information.

Conclusion

Research on toxic combustion by-products may be characterized by its complexity: the number of agents, their interactions, their sources and origins, and their environmental fates. Powerful advances in technology and models have been achieved that now allow the study of this difficult problem, a problem that has been previously considered too complex and onerous to be addressed in regulations for all but a few sources and a few pollutants. Recommendations for research focus and direction identified during the Seventh

International Congress on Combustion By-Products are presented below.

Recommendations for research policy priorities are

- Interdisciplinary, coordinated activities should be developed among the governmental agencies responsible for public health, worker health, environmental and industrial engineering, environmental monitoring, biomedical research, health policy, and respiratory medicine. This is needed to more effectively target research that will reduce risks to both human health and the environment.
- Dialog should be promoted among policy makers, researchers, nongovernmental agencies, and research sponsors to address regional and local exposure issues. Coordinated, multisite, multipollutant research efforts are needed to specify and quantify the health effects of exposures to combustion by-products, as they are not adequately represented by exposure to criteria pollutants such as ozone.
- New combustion research programs should be designed to capitalize on the important potential synergisms between on-going international programs including efforts to reduce greenhouse gas emissions and those to reduce health-damaging emissions from solid fuel stoves. Such programs need to integrate the efforts of policy makers and economists as well as engineers, scientists, and health researchers.
- Additional attention needs to be paid to improve development and implementation of measurement and monitoring techniques. Improved diagnostics could lead to smart systems, better understanding, and efficient control/prevention strategies for better use in public health policy decision-making.
- Policy makers should focus their attention on the development of strategies to address the complex issues surrounding environmental health, specifically from the standpoint of its ethical, legal, and social implications.

Recommendations for a research framework are

- An initiative on the origin and mechanism of genotoxicity of fine particles should be undertaken in light of the recent epidemiologic evidence for the correlation of exposure to airborne fine particulate matter and increased risk of lung cancer, as well as the paucity of published research on the source of this correlation.
- Advanced methods are needed for characterization of the bioavailability of particle-associated pollutants. Fine particles can act as effective delivery systems for toxic chemicals, but the bioavailability of the toxins depends on the nature of their interaction with the particles. Advanced methods for characterizing the nature of association and binding of stable molecular as well as radical species with various types of particles are essential.

- Research is needed to increase our understanding of mechanisms of pollutant formation and stabilization at the gas-particle interface. Research on the rate of formation of gaseous pollutants and precursors as well as development of mechanisms of gas-particle interactions to form pollutants are strongly indicated. This will require development of more detailed mechanistic models and methods for identifying gas-phase and surface intermediates as well as products.
- Improved computational methods for modeling gas and particle properties are needed. Detailed reaction kinetic mechanisms of pollutant formation and fate require accurate thermochemical and reaction rate parameters. Development of reliable *ab initio* methods of calculation of these properties are necessary as semi-empirical methods appear unreliable.
- Rapid microarray techniques should be applied for assessing potential health impacts of individual pollutants and mixtures. The varied and complex products formed in combustion systems can potentially induce a variety of health impacts alone or in combination in a manner that has previously defied characterization. Application of the rapidly advancing field of microarray technology to screening for various biologic end points of individual pollutants and mixtures should be used to address this poorly understood but critical component of environmental health.
- Additional research should be conducted to increase understanding of the composition, sources, and health impacts of fine and ultrafine particles. Atmospheric studies should determine the composition of fine and ultrafine particles, including nano-particles; determine the ratios of elemental carbon to organic carbon in airborne particulate matter, identify the main sources for fine, ultrafine and nano-particles, and identify alterations of fine, ultrafine and nano-particles by time and by location.
- Research in toxicology should be advanced to determine the fate of fine and ultrafine particles during inhalation and after deposition (dosimetry); determine translocation rates to other extrapulmonary tissues and influence of particle chemistry on such translocation; evaluate specific cellular and molecular mechanisms via both animal and *in vitro* studies; develop and use compromised animal models; and to apply toxicogenomics and proteomics in mechanistic studies.
- Epidemiology studies should include prospective cohort studies of specific compromised people (e.g., cardiovascular diseases, respiratory including chronic obstructive pulmonary disease (COPD), asthma). Clinical studies with surrogate or concentrated ambient particles should include the elderly

and subjects with cardiovascular disease, asthma, and COPD, as well as include measures for endothelial dysfunction and other vascular parameters in addition to cardiac and respiratory end points.

- Advanced techniques are needed for speciation of toxic metals. The oxidation state as well as chemical form of metals affects their toxicity, increasing the importance of improved methods for speciation of metals, especially those associated with particles and surfaces.
- Concerted and focused efforts should be made to fulfill the potential of biomarkers to significantly reduce the burden of exposure and disease and to protect individuals from the uncertainty of risk, specifically in the area of exposure and effects of combustion by-products.
- Efforts should be focused to develop reliable and sufficiently widespread environmental and health monitoring systems. This is required both to effectively target actions to reduce outdoor pollution and to evaluate their effectiveness.

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