Air pollution and thrombosis: an experimental approach

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Key Words

Air pollution – myocardial infarction – thrombosis – ultrafine particles – endothelial injury – diesel exhaust particles – platelet function analyser.

Abstract

Air pollution is associated with cardiovascular mortality. Inhaled ultrafine particles translocate into the blood. Amine-polystyrene ultrafine particles significantly enhance experimental thrombus formation in a damaged hamster vessel and shorten the closure time in the Platelet Function Analyser. Diesel exhaust particles are thrombogenic within one hour of intratracheal instillation and shorten the closure time *ex vivo*. These experimental observations provide a plausible biological explanation for the epidemiologically established link between air pollution and acute myocardial infarction.

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Particulate air pollution is associated with cardiovascular morbidity and mortality [1,2]. Exposure to polluted air for as little as 2 hours increases the risk of myocardial infarction [3]. A plausible biological explanation for these epidemiological observations however is lacking. Most research has centered around the possible consequences of particle-induced pul-

monary inflammation on the heart and other systems. The rationale for these studies is that cytokines and other mediators produced in the lungs are also released in the circulation and exert extrapulmonary effects.

Another line of research, that has not been pursued much so far, consists of studying the possible direct effects of particles that may pass from the lung into the circulation. The rationale for this approach is based on the observation that it is the ultrafine fraction of the particles that probably is most hazardous to health. Ultrafine particles have a diameter below 100 nm, remain airborne for a long time and deposit in greater numbers and deeper into the lungs than larger-sized particles. Their small size makes it conceivable that they translocate from the lung into the blood, and this indeed has recently been demonstrated in the hamster [4] and in man [5], using radioactively labeled ultrafine particles.

We have, therefore, studied the effects of ultrafine particles on a relevant cardiovascular endpoint, namely thrombus formation in a diseased vessel. For this purpose we took advantage of a validated hamster model of endothelial injury, using Rose Bengal and local vessel exposure to green light; thrombus development over the free radical-damaged endothelium is continuously registered over 40 minutes by transillumination [6]. In initial experiments, 60 nm polystyrene particles were administered either intravenously or intratracheally. Although these particles clearly are not pollutant particles, they offer the technical advantage of being well characterized in terms of size and chemical composition, which is not the case for the commonly used complex mixtures of diesel exhaust particles,

concentrated urban air particles or residual oil fly ash particles.

Because most ambient particles are charged, we studied unmodified as well as carboxylate- and amine-polystyrene particles. Only amine-polystyrene particles significantly enhanced thrombus formation in the damaged vessel, whether administered intravenously [7] or intratracheally [8]. This enhancement was already observed within the first hour after administration. Amine-polystyrene particles also shortened in vitro the closure time in the Platelet Function Analyser (PFA-100). Interestingly, while both 60 nm and 400 nm amine-particles caused inflammatory changes in bronchoalveolar lavage fluid following intratracheal instillation and shortened in vitro PFA closure times, only the 60 nm amine-particles enhanced thrombus formation in vivo, indicating that the translocation of

the ultrafine particles into the systemic circulation is a prerequisite for the early enhancement of thrombosis following exposure to microparticles.

Transmission electron microscopy of diesel exhaust particles from the National Institute of Standards and Technology (Gaithersburg, MD, USA) revealed particles 20-50 nm in diameter. In the next study, diesel exhaust particles were therefore instilled into the trachea of hamsters. These experiments confirmed that at relevant concentrations these particles are thrombogenic in vivo already within one hour of administration and that they shorten the PFA-100 closure time ex vivo [9]. These observations provide a first plausible biological explanation for the epidemiologically established link between air pollution and acute myocardial infarction.

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