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INSTRUMENTATION FOR ENVIRONMENTAL MONITORING IN BIOLOGICAL SYSTEMS*

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Man's concern over the quality of his environment is now at an all-time high for he has come to realize that not only is pollution directly detrimental to his well being, but also that it adversely affects the closed system of which he is a part. Such a system must exist in dynamic equilibrium in which an upset by a polluting perturbant will surely have far-reaching effects on all members of the biosphere.

Unfortunately, the number of pollutants is large. Their effects include: toxicity that can be manifested as impairment of health and physical fitness or even mortality, carcinogenicity, mutagenicity, and teratogenicity. Synergism between different pollutants and between pollutants and non-pollutants is likely, but yet to be determined.

Environmental insults can be broadly classified in the following six categories:

1. Trace elements.
2. Pesticides and herbicides.
3. Ionizing Radiation and Radionuclides
4. Asbestos and other microparticulates
5. Gaseous pollutants

It is realized that although the above grouping is somewhat arbitrary, it does provide a suitable working order.

Because of the extensiveness of this classification and the added complication that a given environmental insult might need to be monitored in different components of the biological system (e.g., in urine, blood and/or fat deposits), emphasis is placed upon pollutants which appear to be of immediate threat to man and other living entities.

The term "biomedical instrumentation" implies many categories of measurements, but only those involving environmental insult are included here. Others are generally excluded: for example, diagnostic or therapeutic medical measurements and occupational-safety measurements. An exception is instrumentation developed for other uses which is also applicable as an indicator for problems in the environment. In the following sections, a discussion of each of the selected areas is presented.

Trace Elements

Metals such as chromium, manganese, iron, cobalt, copper, zinc and molybdenum are essential, in small amounts, to the living system. Others seem to be unessential. However, all trace elements are toxic when ingested in high enough concentrations. Of the long list of trace metals, lead, mercury, cadmium, and nickel have been prominently cited in the literature as representing known hazards to human health. Although the amounts of the highly toxic beryllium and of antimony in the environment are small, they

are potentially hazardous and should be closely controlled. Aluminum, barium, strontium, titanium, niobium, zirconium and vanadium appear to have little, if any, harmful effects. The low-level effects of boron, germanium, arsenic, fluorine and selenium on man are yet to be determined. On the other hand, it has been shown that typical concentrations of fluorides found in emissions from such industries as steel, aluminum and ceramics are toxic to plant growth.

A discussion of the significance and of the general principles of detection of the four major metallic pollutants is given below.

Lead

Lead is the second largest (next to iron) single metallic pollutant of the atmosphere. Its effects include damage to the liver, kidney, brain and the central nervous and reproductive systems. Children are particularly susceptible to lead poisoning which is manifested in mental retardation. The organo-metallic alkyl lead is highly toxic and crosses the blood brain barrier. Inorganic forms of lead are less toxic. Lead can enter the human body either orally or by inhalation. Thirty to fifty percent of inhaled lead is absorbed by the lungs, compared to 10% of oral ingestion by the gastro-intestinal tract. Small concentrations of lead have been shown to induce shortening of the life span of mice and rats. Plants incorporate lead directly from leaves.

Several methods are used in the detection of lead in biological systems. These include neutron activation analysis, a combination of chelation and extraction followed by atomic absorption spectrophotometry. Proper experimental techniques are used to minimize interference from other elements which may exist in the biological sample (e.g., iron in blood samples and cadmium in kidney samples). A small number of less sophisticated laboratories still use the dithizone method which involves chelation by dithizone to produce a colored complex soluble in chloroform which has a characteristic absorption spectrum.

Mercury

Mercury is a ubiquitous element in the crust of the earth and in oceans. It is detected in many living systems. When the inorganic form of mercury is either ingested or inhaled, most is rapidly eliminated from the body with only small amounts persisting in the kidney, the liver, the lungs and in the hair. On the other hand, the organic form, methyl mercury, is highly toxic via oral uptake. This form of mercury, which is the most interesting from a toxicological view point, is determined by gas chromatography. As for total mercury, flameless atomic absorption spectrophotometry and neutron activation analyses have been used. There also exists a number of techniques based on wet chemistry and which are slow and less sensitive.

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Recently, a new type of atomic absorption spectroscopy based on hyper-fine structure lines in magnetic fields was developed and used to detect trace amounts of mercury.

Cadmium

Cadmium is released into the air as a result of incineration and disposal of cadmium-containing products such as rubber tires and plastic containers, and as a by-product in the refining of other metals, particularly zinc.

When it enters the body, cadmium is deposited in the kidney, liver, and arteries. It has been implicated in such disorders as osteomalacie and hypertension. Chronic exposure to this metal can induce histological damage to the kidney and the liver. Both atomic absorption and activation analyses techniques are used to quantitatively determine its existance in biological systems.

Nickel

Nickel is detected in almost all living systems. It is found in air and water and constantly occurs in food.

Nickel carbonyl, which is an intermediate in the process of nickel refining, is known to be carcinogenic and a highly toxic agent. Chronic exposure to nickel itself causes nickel dermatitis.

Atomic absorption spectroscopy and activation analyses techniques are suitable methods for the detection of this element.

X-ray fluorescence spectroscopy is used for the detection of trace amounts of heavy metals, however this technique lacks the needed sensitivity for biomedical samples.

Pesticides and Herbicides

In man's effort to maximize the agricultural output of his land and to control disease vectors, he resorted to the use of synthetic compounds such as chlorinated hydrocarbons (e.g., DDT, Aldrin and Diel-drin), organophosphates (e.g., parathion and malathion), acylanilides, phenylcarbamates and phenylureas. The assumption has been that the potential hazard of such compounds is limited to accidental poisoning. However, the accumulating experimental evidence indicates otherwise.

Organic compounds constitute the bulk of chemical pesticides in current use. Distinctions may be drawn between natural and synthetic organics; however, commercial synthesis of natural organic pesticides (botanicals) is by no means precluded. The chemical origin of organic pesticides thus has little bearing on their effect on the environment *except* in one important sense: synthetic pesticides *may* be produced which are much more resistant to environmental degradative processes than natural organic compounds. *In fact, nondegradability is the most important single aspect of environmental contamination by pesticides.* For this reason primary emphasis is given to those synthetic pesticides which persist in the environment.

In the detection of chlorinated hydrocarbons and other pesticides a combination of gas-liquid chromatography and gas chromatography-mass spectrometry, as well as thin-layer chromatography are used. Wet

chemical techniques have been used. These same techniques can also be used for measuring pesticides which are proposed to substitute for DDT (e.g., carbaryl [1-naphthyl-N-methylcarbanate] known as Sevin).

Ionizing Radiation and Radionuclides

Until the beginning of this century, man's only exposure to ionizing radiation was derived from naturally occurring radioactive sources. With the discovery of x-rays, radium, and with the eventual discovery of the processes of fission and fusion, sources of ionizing radiation became potential polluters.

Studies have shown that the major man-made source of ionizing radiation to the total population in the U.S. is from diagnostic and therapeutic x-rays. The estimated doses received from such sources almost equals the dose received from natural sources. Other sources of man-made radioactive pollutants include effluents from nuclear power plants, from fuel reprocessing plants, and from medical and industrial uses of radioisotopes.

Generally, the detection techniques for radionuclides and ionizing radiation are quite well in-hand. Capabilities exist for *in-vivo* total-body monitoring as well as for particular samples such as blood, urine, and bone marrow.

Asbestos and Other Microparticulates

Asbestos is known to lead to harmful structural changes in lung tissue. It is released to the air during the construction of buildings and has been found in drinking water.

"Asbestos" is a generic term for a number of hydrated silicates that, when crushed or processed, separate into flexible fibers made up of fibrils. Although there are many asbestos minerals, only six are of commercial importance: chrysotile, a tubular serpentine mineral, accounts for 95% of the world's production; the others, all amphiboles, are amosite, crocidolite, anthophyllite, tremolite, and actinolite. The asbestos minerals differ in their metallic elemental content, range of fiber diameters, flexibility or harshness, tensile strength, surface properties, and other attributes that determine their industrial uses and also affect their respirability, deposition, retention, translocation, and biologic reactivity.

The most meaningful data concerning the nature and identification of microparticulates may be obtained from analysis of single individual particles. Combinations of electron diffraction, transmission microscopy, and microprobe chemical analysis yields information concerning the morphology, structure, and chemistry of the particulate examined. A single electron microscope grid may be used for diffraction and microscopy. This grid may in turn be mounted on a plastic substrate and chemical analysis performed with a micro-analyzer. In this way the physical and chemical nature of the particulate may be determined and identified.

Gaseous Pollutants

The techniques for measuring carbon monoxide are based on the fact that oxyhemoglobin (O_2Hb) in oxalated blood is completely reduced in the presence of small amounts of sodium hydrosulfite, whereas carboxyhemoglobin ($COHb$) is not. The spectral absorbance curves

of O₂Hb and COHb are different. Thus the ratio of the absorbance at 555 nm and 480 nm is directly proportional to the percentage of COHb in blood.

In the case of sulfur dioxide (SO₂), no laboratory technique exists for determining the extent of exposure to this noxious gas. However, it has been found that the concentration of S-sulfonate in blood plasma increases as a function of exposure. Consequently, biological monitoring of plasma S-sulfonate may provide a useful measurement technique to verify and quantify SO₂ exposure.

To sum up, the status of instrumentation for monitoring the effects of pollutants in biological systems varies from one pollutant to the next. Techniques are well in hand for detecting trace metals, pesticides

and herbicides, and ionizing radiation. This is not the case for gaseous and liquid pollutants. Asbestos and other microparticulates present a unique problem where lung tissue samples have to be analyzed to ascertain the extent of exposure. However, once a sample is obtained, rather sophisticated techniques such as electron microscopy and electron microprobe analysis are used to identify the degree of exposure to this potentially serious pollutant which seems to be more prevalent in U.S. waterways than previously suspected.

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