

# Overview of a System for the Computer-Assisted Operation of a Small Animal Inhalation Facility

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Automatic monitoring of the concentration of test gases and other environmental variables in small animal inhalation exposure chambers, coupled with computing capability and feedback control of the concentration of test gas, allows almost fully automatic operation of the chambers with a minimal amount of human intervention. Time-varying exposure profiles may be generated repeatedly with great accuracy, thus allowing a more realistic simulation of real-life exposures than is approached by operating chambers manually at ostensibly constant concentrations of test gases. Carefully conducted, pre-experimental calibration procedures are performed, and daily calibration checks allow statistical control of daily chamber operation and longer term quality control. At the conclusion of each experiment the investigator is supplied with records that document chamber conditions that have been monitored throughout the entire experiment, with estimates of the accuracy that was achieved in creating the specified exposure profile. A purpose of this report is to help to bridge the gap between the practicing inhalation toxicologist and the engineer in order to encourage their cooperation and mutual understanding of the technical problems involved in developing computer-assistance packages for inhalation facilities.

## Introduction

Dynamic flow-through, small animal inhalation exposure chambers (1) are used widely throughout the United States in conducting research in inhalation toxicology. They are most commonly operated by establishing a constant ventilation rate through the chambers and then introducing a test gas or aerosol into the incoming air in order to achieve the desired concentration in the chamber.

Efforts have been made to reduce the degree of human intervention that is required during the daily operation of aerosol exposure chambers by incorporating microprocessor control into the sequencing of the introduction of test substance into the chambers (2).

We have incorporated computing capability into a closed-loop feedback control system that places the generation of any exposure profile entirely under program control. With this advance,

chamber operation is no longer limited to establishing a steady-state concentration of test gas, holding it there, and turning it off after some predetermined interval. Any time-varying exposure profile of one, or a mixture of gases, now may be faithfully reproduced repeatedly with minimal intervention by a technician. Furthermore, continuous sampling of chamber contents with frequent reading of the analytical instruments assures that an accurate record of chamber conditions is obtained that includes relevant variables such as temperature, humidity, airflow, and chamber negative pressure, as well as the concentration of the chemical(s) of interest. Carefully conducted chamber calibration procedures coupled with quality control provide unprecedented accuracy in the monitoring and control of chamber conditions throughout the course of an experiment.

## General Description

A data acquisition system coupled to a system for the feedback control of gas concentration allows exposures in inhalation chambers to be con-

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trolled and documented easily. The technical error rate is likely to be reduced when rote tasks are reassigned from human operators to machines. The capabilities of such a system far exceed those possible with manual operation, both in terms of accuracy of measurements and the complexity of the exposure profiles that may be produced.

Standard automatic data acquisition, whether from the periodic operation of a discrete sampling device such as a gas chromatograph, or from a continuous sampling device such as an infrared gas analyzer, saves time and labor by allowing chambers to be monitored with a minimum of human participation. In addition, such a system records data uniformly, thus eliminating some errors that are likely to occur with manual operation. However, without extensive hand processing, the resulting data represent only a point-by-point index of the gas concentration.

A calibration is performed at several different concentrations in the general range of the desired exposure. From this, an equation is derived that converts an index of gas concentration, generally a voltage or integral of a voltage, to the numerical value of the concentration being measured. This number is stored along with the time of the observation for later evaluation.

The system's computing capability may be used to evaluate the daily calibration by comparing it with previously measured standards and determining how well the calibration data fit the form of the conversion equation. By establishing acceptance criteria, not only are human errors that may be introduced during the calibration procedure detected, but so are many hardware problems. By accumulating daily calibration data over a long period, a very good estimate can be made of the overall accuracy of the measurement system, and equipment errors such as long-term drift may be detected.

The final step in minimizing human participation in chamber operation is to provide a means by which the acquisition system controller can regulate the concentration within the chamber. Thus, once the system has been initialized, the operator needs only to run through a brief calibration procedure and load the chambers, then return at the end of the exposure and unload the chambers. Since the control system constantly checks and adjusts the chamber concentration, the actual exposure will follow the desired profile more consistently than could be expected with human operators. In addition, it now becomes quite easy to change the concentration in virtually any preprogrammed manner. Multiple compound exposures are treated as independent runs

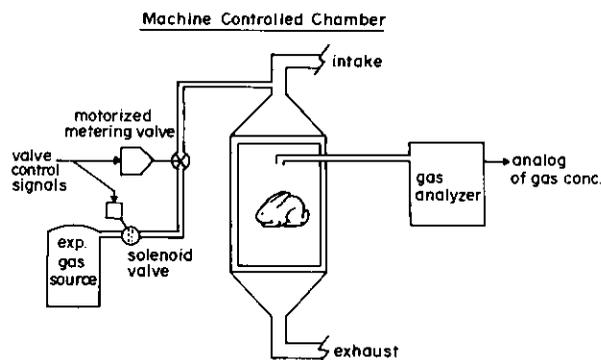


FIGURE 1. A continuous sample of test gas atmosphere is drawn from the geometric center of a dynamic flow-through inhalation chamber. The gas sample goes to an analyzer and the voltage analog of the gas concentration goes to a computer system where it is compared to reference information. Valve control signals are issued by the computer system that may actuate a solenoid-operated on-off valve or turn the stem of a motorized metering valve.

in the same chamber, using the same or different analyzers depending on specific requirements.

The principal elements of the computer-assisted inhalation chamber (Fig. 1) are quite straightforward. A continuous sample is drawn from the geometric center of the chamber and is pumped through the optical cell of an analyzer. The voltage analog of the concentration of the chemical of interest is compared within the computer to the voltage representing the correct concentration as derived from the results of an appropriate calibration procedure. Signals from the computer are translated into revolutions of a stepping motor (MO91-FD-308, Superior Electric Co., Bristol, CT) that is linked to a metering valve. Opening and closing the metering valve regulates the flow of an air-vapor mixture into the chamber inlet.

## Instruments for Monitoring Inhalation Exposure Chamber Gases

Infrared gas analyzers and some  $\text{NO}_x$  analyzers are continuous sampling devices; gas chromatographs are discrete sampling devices. Each of the instruments may be equipped for automatic sampling and each has its own special merits. We use the infrared gas analyzer for most applications because it is a continuous sampling device and has a wide spectrum of applicability for monitoring gases.

### Infrared Gas Analyzer

The MIRAN (Wilks Scientific, Norwalk, CT) series of infrared gas analyzers are equipped with

optical cells of a volume of 5.6 liters and a variable path length of 0.75–20 m. Most organic chemicals have absorption peaks in the infrared region that are useful for monitoring chamber atmospheres. MIRANs also may be used to monitor ammonia, water, sulfur dioxide and ozone, but usually not oxides of nitrogen. Calibration is performed easily in a closed loop system that includes the optical cell. Since the volume of the calibration loop is known concentration may be computed when aliquots of the chemical of interest are injected into the closed volume. The infrared gas analyzer has a curvilinear concentration-response, but the computations necessary to evaluate such responses are easily done by computer. In our experience, higher polynomial functions have usually provided the best fit to calibration data. We have defined and programmed algorithms that do these computations rapidly and without the need for special knowledge on the part of the system operator (3).

Multicomponent gas mixtures can be monitored by using separate MIRAN IA analyzers for each compound of interest or by using a MIRAN 80 computing, gas analyzer. The MIRAN 80 has a single optical cell and readings are taken sequentially at different, preselected wavelengths on the same sample. The MIRAN 80 is, therefore, a discrete sampling device, whereas the MIRAN IA is a continuous sampling device for purposes of their application to automated systems. The MIRAN 80 offers the advantage of being equipped with a RS-232 interface through which the analyzer may be adjusted (not including path length) under program control.

### Gas Chromatograph

A gas chromatograph (Fig. 2) may be equipped with an air-actuated, automatic sampling valve for use with the computer-assistance system. A sample of the chamber atmosphere is delivered to the inlet line at constant pressure. It is important that the GC inlet pressure selected be held as nearly constant as possible. The detector responds to the total number of molecules impinging on it, a variable that is proportional to pressure. Variations in inlet pressure could result in spuriously high or low detector responses not having any particular relationship to the actual concentration of the compound of interest in the chamber.

The gas chromatograph is a slowly responding, discrete sampling device. It may be suitable for monitoring mixtures of gases that are easily separated on a relatively short column. Because the response time of the gas chromatograph is so long

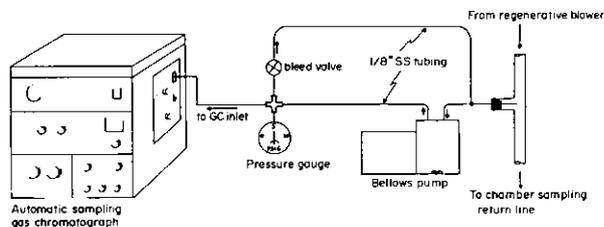


FIGURE 2. A gas chromatograph may be equipped with an air-actuated, automatic sampling valve and used to measure test gas concentration in a computer-assisted inhalation chamber system. A sample of chamber atmosphere is pumped at constant pressure to the GC inlet. A by-pass and bleed valve are incorporated into the gas sample circuit to allow the manual adjustment of inlet pressure.

it is useful only for monitoring nonvarying or slowly varying chamber concentration profiles.

### Oxides of Nitrogen Analyzer

Nitrogen dioxide is monitored most reliably with an oxides of nitrogen ( $\text{NO}/\text{NO}_2/\text{NO}_x$ ) analyzer that operates on the chemiluminescence principle (4). The Monitor Labs (San Diego, CA) nitrogen oxides analyzer, Model 8440E is a continuous sampling device that is well suited to computer-assistance systems for inhalation facilities.

### General Layout of the NIEHS In-House Facility

The general layout of the small animal inhalation facility where the prototype for the computer-assistance package was developed is illustrated in Figure 3. Dynamic, flow-through chambers are continuously sampled from a single point near the geometric center. The validity of measurements taken from a single location is dependent on the validity of the assumption of a practically uniform distribution of gases within the chamber. A general visual assessment can be obtained by smoke testing the chambers under operating conditions. An alternative to smoke testing is multipoint sampling, a procedure that yields more definitive results. Our purpose here is not to explore the technicalities of evaluating chamber dynamics. We assume only that the user has satisfied himself that he is obtaining chamber air samples that represent what the animals are exposed to under experimental conditions.

Materials that form part of the gas path, i.e., chambers, tubing, fittings, valves, etc., are constructed of stainless steel, borosilicate glass, or PTFE (polytetrafluoroethylene). Practically any

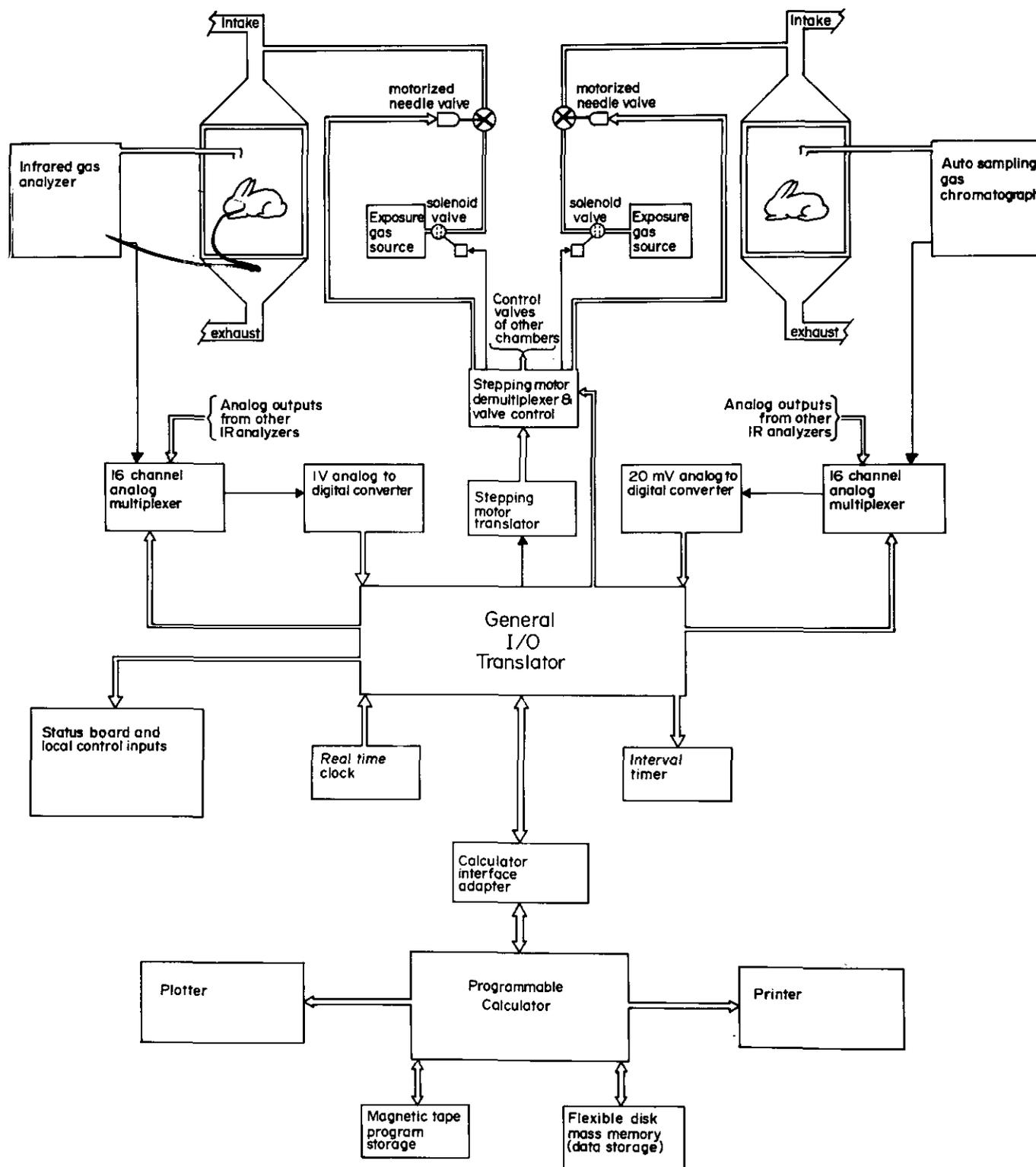


FIGURE 3. The original design of the computer-assistance system for the in-house facility of NIEHS permitted the use of either a GC or infrared gas analyzer as the monitoring instrument. In either case a continuous sample of chamber air is delivered to the analytical instrument. The GC samples the test gas intermittently; the IR gas analyzer samples continuously. The voltage analog of the concentration is compared to calibration data that have been entered previously into the system which then sends signals to a motorized needle valve through which the rate of test gas entering the chamber is regulated. The computer system includes various peripheral devices that are used for purposes that are explained in the text.

other metal or plastic component that would be likely to be used as alternatives eventually will become a source of problems arising from degradation, corrosion or absorption of chemicals.

The sample from the chamber is drawn through  $\frac{1}{2}$ -in. PTFE tubing by means of one or more blowers (SE2 A-14, Rotron, Inc., Woodstock, NY) connected in series. As a safety precaution the blowers have been located between the analytical instrument and the chamber in order to maintain a slightly negative pressure within the analyzer relative to the outside. Blower capacity for any particular assembly should be about 25 L/min. When the chamber air sample is passed through the optical cell of an infrared gas analyzer it remains chemically unchanged and so may be returned directly to the chamber. When a  $\text{NO}_x$  analyzer or gas chromatograph is being used, the analyzer will require only a small fraction of the primary chamber air sample. The unused remainder of the chamber air sample can be returned to the chamber, but the toxic effluents of the analyzers must be disposed of properly elsewhere. In any case, the total volume of the unused chamber air sample may be discarded rather than returned to the chamber, if desired.

The in-house facility at NIEHS is equipped with more or less permanently installed MIRAN IA infrared spectrophotometers. These analyzers must be set manually to the required wavelength, slit width, electrometer settings, and optical cell pathlength. The settings remain constant throughout any given experiment. Running more than one compound in the same chamber requires that additional spectrophotometers be added to the chamber air sampling loop.

The voltage analog of the concentration of the chemical of interest that is detected by the analyzer goes to an analog multiplexer along with similar outputs from other analytical instruments. The analog signals are digitized and sent to the computer by way of a general, digital interface. A similar series of devices converts analog signals from the thermistors, humidity sensors, and differential pressure transducers that are used in monitoring and logging chamber temperature, humidity, airflow, and negative pressure.

An equation that converts the analyzer output voltage to concentration of chemical of interest has been stored in the computer. This equation is used to convert the current analyzer reading into units suitable for comparison with the requirements for exposure profile that also have been stored in the computer. Correction of the chamber concentration is a function of the efferent arm of

the control loop which consists of a motorized metering valve and an appropriate interface to the computer.

The motorized metering valve (Fig. 4) is located between the exposure gas source and the chamber inlet. The computer regulates the chamber concentration by opening or closing the valve as needed. The metering valve should not be used for on-off purposes, however, and so a solenoid-operated valve (Fig. 4, left) is placed between the exposure gas source and the metering valve. The solenoid valve is opened and closed under program control and also closes when electrical power is lost to prevent the continued flow of chemical into the chamber during a power failure.

The computer that is used in the in-house system is a Hewlett-Packard (Palo Alto, CA) model 9825B. One of us (M.P.M.) has written a multi-tasking system for this machine that permits it to service up to nine channels of analyzer data. The analyzers are treated as independent sources of

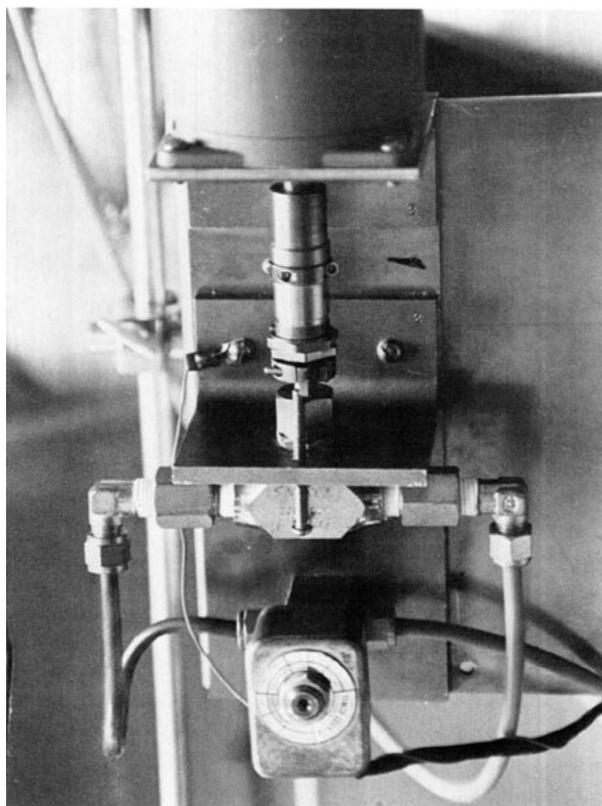


FIGURE 4. The flow of carrier air to a vapor generator, or the flow of test gas from a pressurized cylinder, is regulated with a metering valve (center left) that is coupled to a computer-controlled stepping motor (right) by means of a clutch (center). A solenoid-actuated on-off valve is shown at the far left.

analog data regardless of whether or not two or more are actually linked to the same chamber. The computer cannot handle data from more than one source exactly simultaneously, but it can switch rapidly from one device to another. This switching may be so rapid as to create the impression of practically uninterrupted response to several channels of incoming data. If overall system requirements are anticipated to exceed the ability of a relatively modest computing machine like the H-P 9825B to keep up, increased speed can be built into the system by designing a network consisting of more than one computer. An alternative to this strategy would be to substitute any of a variety of faster, high-powered laboratory computers for the HP 9825B. Of course, networks of the faster machines can be designed as well, as is illustrated in the next section where the on-site contractor-operated inhalation facility at NIEHS is described.

When one, small computer is to be used in the management of as many as nine channels of incoming analyzer data, a primary design consideration revolves around the minimal rates of sampling of the respective channels of data that will be required. In order to make the most economical use of all of the processing power of the computer in handling time-varying signals from the analyzers it was necessary to decide during what phases of the chamber cycle it was necessary to sample the analyzers at higher rates and during what parts of the cycle we could afford to sample at lower rates. We decided that the sampling rate should be higher during periods of changing concentration than during periods of relatively unchanging concentration. Therefore, the computer evaluates the concentration profile requirements for each channel that it is servicing and samples the respective analyzer outputs at a higher rate when the concentration is rising or falling than when it is relatively stable.

The computer system is also equipped with five output devices that may be used in a variety of ways to support operation of the chamber facility. The desk top unit (H-P 9825B) has an integral tape cassette drive and a thermal printer. The principal use of the tape unit is as an input device to provide for automatic restarting in case of power failure. The internal printer can be used to display any data, but since it is only 16 columns wide and thermal printer copy fades with time, it is best suited for the display of on-line diagnostic or monitoring information. Calibration and chamber status data are printed on a character-impact printer (Hewlett-Packard 9871A) that produces permanent hard copy similar to that of a

typewriter. The computer output can also be sent to an X-Y plotter that can be used to construct tables or draw graphs of any kind of raw data or data summaries that are desired. For example, while the chambers are running we display concentration data in a strip-chart mode that simulates a nine-channel strip-chart recorder, thus allowing a visual confirmation of chamber concentration profiles. Likewise, the plotter may be used to display summaries at the end of entire experiments. Mass storage is done on two 8-in. flexible disk drive units (Hewlett-Packard 9885M). These units serve both input and output functions. They are used to hold the programs that comprise the software (instructions to the computer) and also serve as data storage devices.

## General Layout of the NIEHS Contractor-Operated Inhalation Facility

The basic principles that govern the operation of the contractor-operated inhalation facility at NIEHS are the same as those that were described for the in-house facility. The computer-assistance system that has been installed in the contractor-operated facility represents a refinement of the in-house prototype. The system is of a modular design that will permit it to be expanded easily without requiring any basic alterations in design.

The general layout of the contractor-operated facility is illustrated in Figure 5. The system includes three chamber rooms, each of which includes four inhalation chambers. Each room is equipped with a relatively high-speed laboratory computer [PDP 11/34, Digital Equipment Corp. (DEC), Maynard, MA]. A fourth room, the central computer room, is equipped with a fourth PDP 11/34 that is connected to each of the computers in the respective chamber rooms and that serves as a data manager. The central computer room also houses a fifth PDP 11/34 that is available on a standby basis in case of a computer failure anywhere in the system. Each computer in the network uses the DEC RSX-11M operating system upon which the series of tasks that actually monitor and control the facility have been constructed.

The computer system is equipped with several peripheral devices that support comparable requirements that have already been described for the in-house facility. Mass storage of programs and data for immediate use is done on hard disk units of 5 megabyte capacity (DEC, RL01). Each computer is equipped with disk drives which al-

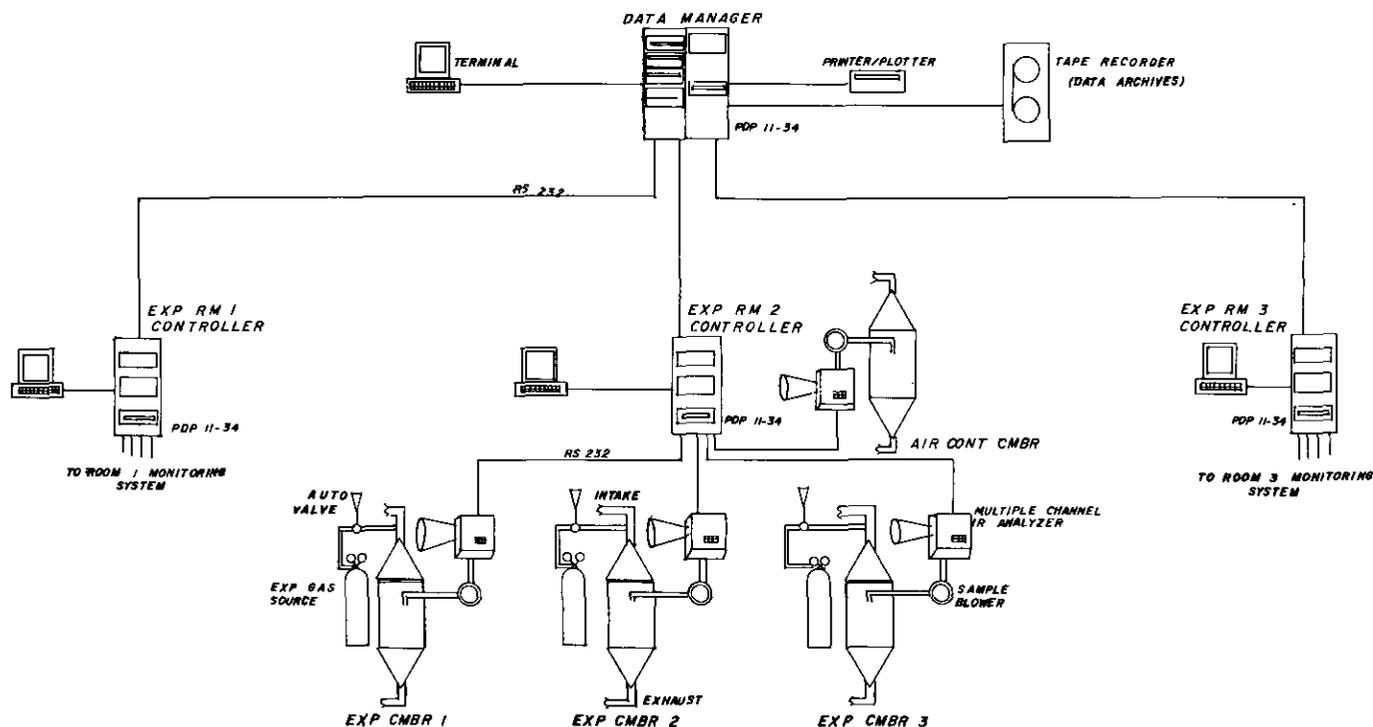


FIGURE 5. The contractor-operated inhalation facility at NIEHS is monitored and controlled by a computer-assistance system that represents an expanded version of the smaller system developed for the in-house facility (Fig. 3). Each of three gas exposure rooms includes four chambers that are serviced by a PDP 11/34 laboratory computer. Each of the three room computer is linked to a fourth PDP 11/34 located in the central computer room. Video terminals are located in each chamber room, the central computer room, and an office. Facility status data are stored on magnetic hard disks and the data archives are stored on magnetic tape. Data are displayed in the form of tables or graphs on a printer or on video display terminals.

low the rapid access of data involved in managing operations for a given room. The large data storage capacity also permits all of the data from an experiment, even one lasting for as long as two years, to be stored for ready access as the experiment proceeds. These data are held on disk for as long as is needed in order to meet all of the requirements of the experiment including review of facility performance and data analysis. When these requirements have been met the data on hard disk are transferred to magnetic tape (DEC, TE 10W) as permanent archives that are held in duplicate in secure areas. These data, once on tape, are not intended for routine retrieval but are available, if they are needed.

The central computer facility is also equipped with a dual flexible disk drive (DEC, RX02) for the input or output of relatively small quantities of data that may need to be transferred to or from other, compatible computer systems. Tables and graphs are made on a DEC LXY11 printer-plotter.

Each exposure room, the central computer room, and an adjoining office are equipped with

DEC VT100 video terminals that allow the technical staff to monitor the data being generated during facility operation.

The exposure chambers are equipped with analyzers appropriate for the chemical being monitored. The infrared spectrophotometers that are used for monitoring organic chemicals are MIRAN 80s. One spectrophotometer is included in the sampling loop of each chamber that is being monitored. For example, when an organic chemical like carbon disulfide is being run in a room, perhaps at zero level (control) and three concentrations, one MIRAN 80 will be used for each of the four chambers in the room. The pathlength has to be set manually on the MIRAN 80, but all of the other settings can be made under program control through an RS-232 interface with which the MIRAN 80 is equipped.

Animal weights are entered into the system automatically. The animals are placed on a balance (PL3000, Mettler Instrument Corp., Hightstown, NJ) and the output is transmitted to the computer system through a BCD interface. Any of a variety of schemes can be devised to call for the

animal weights, e.g., the computer can be programmed to generate a weighing schedule and then prompt the user to place a specific animal on the balance. This kind of approach is efficient to the extent that it eliminates the need for the operator to enter an animal identification corresponding to the weight. Other special data, like food and water consumption, can be entered into the system by a suitable scheme, the simplest of which is the manual entry of numeric data through the keyboard of the terminal. However, the purpose of our documentation is not explore the many facets of the acquisition of data from experimental animals, but rather to describe a monitoring and control system for inhalation chambers. It is important to stress that the data handling capability that is acquired with the installation of a computer-assistance package for the facility can go well beyond the physical operation of the chambers themselves. The uses to which the expanded capacity can be put are limited only by imagination and the availability of the services of talented engineers and programmers.

## Exposure Chambers in Use at NIEHS

Dynamic, flow-through inhalation chambers are available from several manufacturers in the United States (Young and Bertke, Cincinnati, OH, Spengler Bros., Cincinnati, OH, Unifab Corp., Kalamazoo, MI, Wahmann Manufacturing Co., Timonium, MD, Hazleton Systems, Inc., Aberdeen, MD). The chambers that are in use at the National Institute of Environmental Health Sciences are of the design of Hinners et al. (1). The building in which the chambers are located is equipped with an air handling system for the chambers that is separate from the air conditioning system for the building in general. Conditioned air is supplied at a constant temperature and humidity through a series of overhead ducts. The chamber effluent is vented through charcoal scrubbers and into separate exhaust ducts. The scrubber effluent is monitored and the charcoal is replaced as needed. The scrubbers are designed on the "bag-in, bag-out" principle which ensures the safe and efficient handling of contaminated charcoal. The charcoal cartridges can be incinerated. HEPA filters have been installed between the chamber outlets and scrubbers to prevent the discharge of particulates into the scrubbers and the remainder of the exhaust system.

The inhalation chambers in the in-house facility are separated from the instrumentation by a

cement block wall (Fig. 6). The air sample is drawn from and returned to the chambers through PTFE tubing that goes through the wall. This arrangement serves to protect the instruments from contamination incident to animal handling and chamber sanitation. Once the air sample has been delivered to the instrument room it can be made available to whatever analytical instrument is required (not illustrated in Fig. 6).

The ability of the computer system to control the chamber concentration is dependent, in part, on the overall response time of the system. The dynamic response of the total system is a function of the overall time constant (the time required for the concentration to rise from zero to 63% of its programmed limit). The time constant of a chamber is 4 min when it is ventilated at a rate of 25% of its volume per minute. This represents the principal performance limitation of the system. The time constant of the sampling loop, on the other hand, represents the delay between the sampling from the chamber and sensing by the analyzer of the concentration of chemical in the sample. The time constant of the sampling loop is about 20 sec when the loop is ventilated at 25 L/min. The time constant of the sampling loop is so small relative to that of the chamber that it has little effect on overall chamber performance.

The smaller the overall time constant the better will be the control of the chamber. Time constant varies inversely with rate of flow. The rate of flow through the sampling loop can be increased by increasing the diameter of the sampling lines (we use 1/2-in. PTFE tubing; 1/4-in. tubing is too small) and by decreasing the length of the sampling lines. We have placed the chambers and the instruments as close to the wall that separates them as possible in order to minimize the length of the sampling loop. The rate of change of the chemical concentration in the chamber in response to a correction is in inverse proportion to the rate of flow through the chamber, i.e., the higher the flow, the shorter the response time. The cost of decreasing the response time by increasing chamber airflow is realized in a rising rate of consumption of the chemical that is being used, which in turn determines the rate of consumption of charcoal in the effluent scrubber. We have also devised a relatively inexpensive way to improve the response time by circulating the chamber air through a loop that is superimposed on the net flow (Fig. 6). A sample of air is drawn from the bottom of the chamber at a rate approximately equal to that of the total chamber airflow by a blower similar to the ones used to

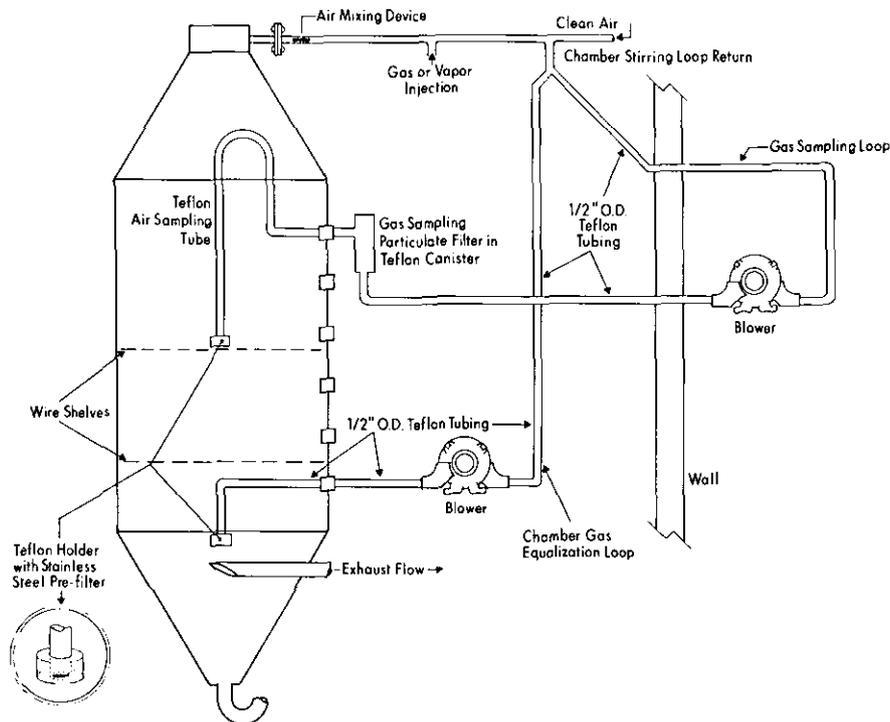


FIGURE 6. The inhalation chambers and the associated analytical instruments in the in-house facility are located in adjacent rooms and on opposite sides of a common wall. They are connected by  $\frac{1}{2}$ -in. PTFE lines. The principal sampling loop is powered by electric blowers that circulate a chamber sample continuously at a rate of 25 lpm. A single sampling port is located at the geometric center of the chamber. The direction of the principal chamber airflow is from top to bottom. The rate of flow is equal to approximately 25% of the chamber volume per minute. A secondary chamber air circulating loop flows at a rate equal to that of the total chamber airflow which increases the rate of equalization of test gas concentration within the chamber.

power the sampling loop and is reintroduced at the top of the chamber along with the return from the sampling loop.

## Chamber Sampling Loop

The chamber sampling loop consists of the volumes enclosed by combinations of the sampling and return lines, the optical cell of a spectrophotometer, the calibration module (Fig. 7) and associated tubing, connectors and valves. The general layout is depicted in Figure 8.

Calibration requires that a closed loop of known volume be assembled, into which known quantities of the chemical of interest are injected at known pressure. The calibration loop and chamber sampling loop both must include the analyzer. The use of a six-port valve with  $\frac{1}{2}$ -in. bore that was fabricated entirely from PTFE (Nacom Industries, Garden Grove, CA) (Fig. 9) provided an efficient means for switching the analyzer between the calibration loop and the

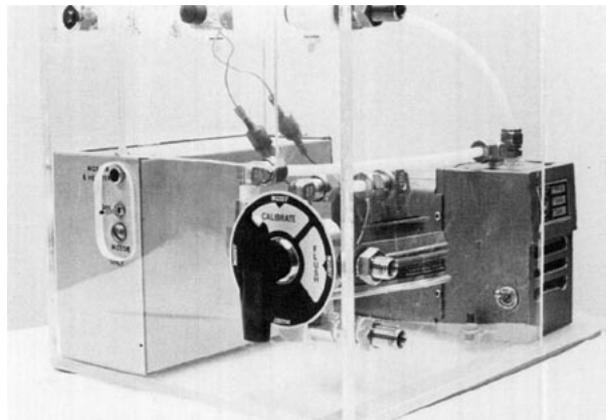


FIGURE 7. We designed a calibration module to be used in the closed-loop calibration of the infrared gas analyzers used in the gas inhalation facilities at NIEHS. A four-port valve is illustrated in the foreground. A stainless steel bellows pump is shown toward the rear and the zero-gass filter is in between. The injection port is shown in the upper left center. The wires lead from a power supply to heating coils that are wrapped around the injection port.

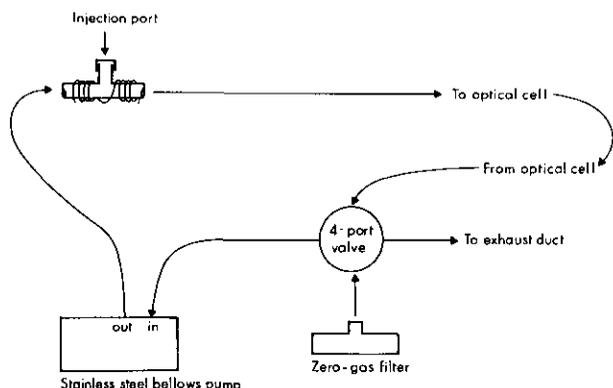


FIGURE 8. This is a schematic diagram of the calibration module that is illustrated in Fig. 7. Gas is circulated through the calibration loop by a stainless steel bellows pump. Gas or vaporizable liquid standards are injected into the heated injection port and circulated through the optical cell of the analyzer. Setting the four-port valve in one position puts the module in the closed-loop calibration mode. Setting it the other position purges the system with zero-gas.

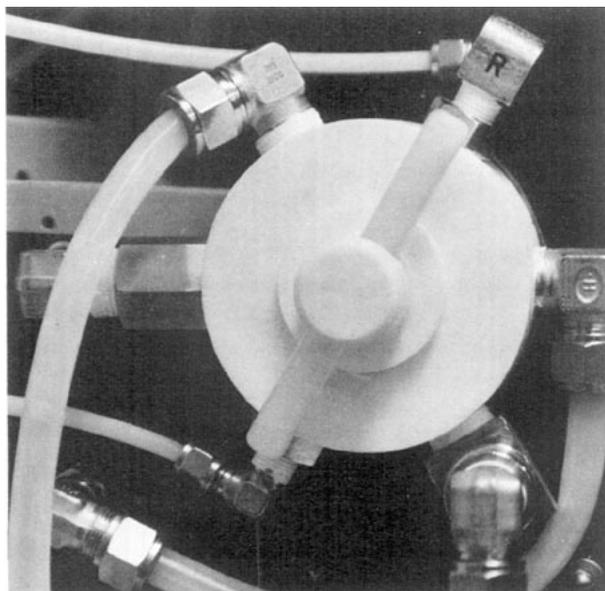


FIGURE 9. A six-port valve with  $\frac{1}{2}$ -in. orifices was fabricated entirely from PTFE. This valve permits the rapid switching of the gas analyzer from the calibration configuration to the chamber sampling configuration without requiring the disconnecting and reconnecting of tubing fittings.

sampling loop. The use of the valve proved to be superior to other means of interchanging the lines, including that of quick-disconnect fittings. When calibration is required the 6-port valve is set in one position. When chamber sampling is required it is set in the other position.

The calibration module (Fig. 7) was designed to

facilitate the process of closed-loop calibration. Figure 8 is a schematic diagram of the calibration module. The module includes a stainless steel four-port valve (SS-43YF2, Whitney Co., Cleveland, OH) that is set in one position to flush the optical cell of the spectrophotometer with room air that has been drawn through a "zero-gas" filter (R51, American Optical Corp., Southbridge, MA). Set in the other position the valve connects the injection loop (T-tube with rubber septum) to the optical cell. A stainless steel bellows pump (MB-41, Metal Bellows Co., Sharon, MA) that is mounted inside the calibration module is included in the airflow circuits in both valve positions for the purpose of rapidly circulating zero-air or the calibration mixture through the system. The injection port is heated when necessary by nichrome coils that are wrapped around the glass "T" on either side of the side-arm that holds the septum.

The volume of the various components of the sampling loop other than the optical cell of the infrared gas analyzer (5.6 L) and the bellows pump can be estimated by filling the components with water from a calibrated reservoir.

Figure 10 is a schematic diagram of the gas generation and delivery system. The use of a motorized metering valve to control of the rate of introduction into the chamber of the chemical of interest allows the user to switch between liquids and cylinder gases with a minimum of effort. When cylinder gases are used they may be introduced directly from the outlet of a low pressure regulator through  $\frac{1}{4}$ -in. tubing into the inlet side of the metering system. The gas is delivered through  $\frac{1}{4}$ -in. tubing from the outlet of the metering valve directly to the chamber inlet. The user will have to do some preliminary computations in connection with flow rates and desired concentrations in the chamber in order to decide what concentration of chemical of interest is to be used from cylinders under pressure. The motorized metering valve will be found to have an optimal working range and therefore a cylinder of 100% chemical of interest may not always allow optimal regulation at the concentrations that are programmed for the chamber, in which situation the chemical should be purchased as a mixture with a suitable diluent. Another way to help to improve the match between the required rate of delivery of the gas or gas mixture and the position of the motorized metering valve is to use metering valves with internal orifices of different diameters.

When liquids are to be vaporized prior to introduction into the chamber inflow, a carrier gas is

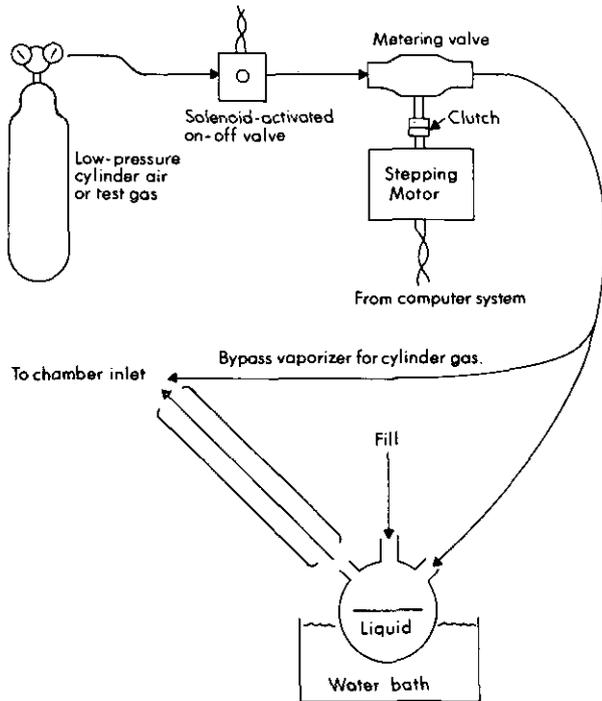


FIGURE 10. This is a schematic diagram of the test gas delivery system. Carrier air or test gas from pressurized cylinders is delivered at low pressure to the solenoid-actuated on-off valve and then to a motorized metering valve. When the test gas is delivered directly from a cylinder the gas is injected into the chamber inlet by bypassing the vapor generator.

used, usually compressed air of breathing quality. A bank of three cylinders is connected to an ordinary two-stage regulator through a tubing manifold. The air is delivered to low pressure regulators attached to each chamber. From the low pressure regulator (< 5 psi) the air goes through the metering system and then through a vapor generator prior to introduction into the chamber inflow. The vapor generator is constructed of ordinary laboratory glassware with ground glass fittings lined with PTFE sleeves and sealed with silicone stopcock grease. We immerse the round-bottomed reservoir flask in a water bath that may or may not be heated. Either way, it confers on the system a certain amount of thermal stability at whatever vapor pressure that we select for the liquid in the vaporizer.

The initialization of the system includes, among other things, selecting optimal conditions for gas delivery that may be summarized as follows: pressure of carrier air (liquids) or cylinder gas; temperature of water bath (liquids); diameter of metering valve orifice; concentration of chemical of interest in cylinder under pressure.

Mass flow controllers are superior in concept to motorized metering valves. They are expensive, however, and at the time of this writing manufacturers had backlogs of orders that would be expected to result in long delays of shipment.

## Monitoring Other Properties of the Internal Environment of Chambers

Temperature, humidity, airflow, transmural pressure (chamber negative pressure, and barometric pressure are monitored and stored along with the concentration of the chemical(s) of interest. Temperature is measured with thermistors, humidity with a digital humidity analyzer (Model 911, EG&G, Waltham, MA), transmural pressure and airflow (pressure drop across an orifice plate) are monitored with differential pressure transducers (Style IOR transmitter, Model 1ORB32A2, Flo Tek, Inc., Newburgh, NY), and barometric pressure with a digital barometer (Model DB99, Validyne, Northridge, CA).

The transducers are calibrated and the data are stored for retrieval later by means of tasks that have been written specifically for the purpose. Measurements are taken at intervals that are defined by the requirements of the investigator. Temperature, humidity, pressure, airflow and barometric pressure are ordinarily quite stable and their time constants are relatively long. Therefore, their values are recorded only 12 times per hour.

## Alarm Conditions

Computer-assisted facilities usually operate with no human intervention during routine chamber runs. Sometimes, however, situations arise that are beyond the control of the computer at which time alarms are triggered to alert operators to the need to give attention to the system. Our systems operate only during normal working hours, and so alarm conditions are signalled with lights and audio alarms. Designers of similar systems can equip them to trigger any kind of alerting device that suits the needs of the institution including telephone dialers, lights and audio alarms.

We have established acceptable operating ranges for all of the measurements. Temperature and humidity out of range, and losses of chamber negative pressure, airflow, or carrier gas pressure (pressure transducer, Style 10R, Flo-Tek, Inc, Newburgh, NY). All trigger alarms that summon

operators to correct the problem. Since the operators are relieved of the necessity of checking meters and gages frequently, many man-hours are saved. Also, a permanent record of all of the measurements is kept for the purpose of documenting the conditions of exposure.

## Epilogue

A purpose of our description of a computer-assistance package for a small animal inhalation facility is to bridge the communications gap between inhalation toxicologists and the engineers with whom they must collaborate in order to be successful in implementing modern technology in support of the needs of inhalation toxicology. Each new generation of a computer-assisted inhalation toxicology facility will be expected to be unique in its own right. We would not encourage exact duplication of our system but, rather, would hope that the general pattern would be used to facilitate steady progress in the advancement of inhalation technology. We hope that we have presented a description to toxicologists of what they

can expect from computer technology, and a description to engineers of some of the technical requirements of inhalation toxicology.

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