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**DESIGN OF A CHAMBER FOR CBW**

**AEROSOL STUDIES WITH RELATIVE HUMIDITY**

**AND PARTICLE CONCENTRATION CONTROL (U)**

by

J. Ho

Project No. 351SQ

February 1989

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RELATIVE HUMIDITY AND PARTICLE CONCENTRATION CONTROL

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DESIGN OF A CHAMBER FOR CBW AEROSOL STUDIES WITH  
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ABSTRACT

→ An aerosol generating system was integrated into a large chamber to regulate three experimental conditions, temperature, relative humidity (RH) and particle concentration during aerosol studies. A commercial heating and cooling unit was used to control temperature. Control of RH was effected by a polymer-based sensing mechanism coupled to a fogging device. Aerosol particle size and numbers were monitored and controlled by a microcomputer-driven detector coupled to an aerosol generator. The system was able to provide precise and reproducible environmental conditions conducive to aerosol studies. Aerosol concentration control was demonstrated over a range of concentrations suitable for different requirements. Under these conditions, a BW simulant aerosol was stable in terms of viability for an extended period of time. As an illustration of the system's potential application, it was used to demonstrate sampling characteristic similarities between a glass fiber filter and a glass impinger. With the features described, the system has significant utility in aerosol studies involving threat detection and protection. This system is easily adapted to drive a dry powder disseminator to generate different CBW simulant aerosols. A patent (Canadian No. 1,222,300; U.S. No. 4,710,887) has been awarded to part of the system.

Chemical Warfare  
Biological Warfare  
Chemical Warfare  
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Simulant

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## INTRODUCTION

Studying aerosols under controlled laboratory conditions has many advantages in that the investigator can reproduce many environmental parameters with ease. This is especially critical if the aerosol of interest is biological in nature, since microorganisms are susceptible to environmental influences. Factors that may require careful monitoring and control include temperature, relative humidity (RH) and light intensity to mention just a few. One of the more difficult parameters to control is particle concentration, as will be explained later. Consequently, most aerosol generation systems used by researchers in biological studies do not have provisions to address this problem.

For example, one of the most commonly used aerosol generating systems is the Henderson apparatus (Anderson, 1966; Druett, 1969). This system may be coupled to a Collison nebulizer (May, 1973) to provide an aerosol source. This source of aerosol is supplied as a continuous stream to the experimental setup with some provisions for controlling the RH by mixing wet and dry air at the output of the generator. In such a system, manual RH control is a difficult task and the concentration of aerosol particles is limited by the air flow capacity of the generator. The output particles flow down a short tube where samples may be taken and the aerosol transit time is the order of seconds. With this system, no provision is made for the aging or environmental equilibration of the aerosol particles. Studies of aerosol interactions with the environment are of critical interest as such interactions are phenomena common to natural aerosols.

For aging studies, where an aerosol must be kept in an environment for extended period of time, the Standard Reference Drum is commonly used (Goldberg *et al.*, 1958). Again, the Collison and the Henderson apparatus may be used to fill a stainless steel drum with a test aerosol and after the source supply is shut off, the drum is rotated to prevent loss of particles due to settling (Wathes *et al.* 1986). Periodically, samples are taken for assessment, without having to stop the drum rotation. Theoretically, rotation of the drum prevents particle loss but in practice, there is always measurable loss so the system does not provide a constant particle concentration capability.

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In biochemical and biological studies, the substrate concentrations before and during an experiment are carefully controlled, as it is important for the experimenter to regulate all factors affecting the outcome of the experiment. For example, in the study of enzyme kinetics, the substrate concentration is kept at saturation levels whenever possible to obtain reproducible test conditions. Similar standards should also be applied to aerosol research involving biological systems. In this case, the substrate concentration is the aerosol concentration. A recent example serves to illustrate this point. As part of some respirator canister quality control tests, failure to implement these procedures was probable cause for disagreement between the results from two test facilities.

Several technically demanding feats must be performed in attempting to regulate the concentration of aerosol particles in a large chamber. First, the sample particle size distribution as well as concentration must be accurately and rapidly measured, for example, within 5 to 10 sec. Then the raw data must be mathematically converted into useful units for comparison to real values (10 to 50 particles/cc) stored electronically, all done within 30 to 40 sec. Feedback control of the aerosol generator can be used to alter the aerosol generating rate to make up for particle losses. The whole cycle is repeated automatically with a cycle time of about 45 sec. A cycling time slower than 45 sec will cause either too much over compensation or too much lag. The perfect system is one which can detect small changes and is able to quickly respond appropriately. Much of these tasks could not have been done economically before the advent of microprocessors and concomitant advances in other areas such as laser light scattering instruments. Due to these advances, instrumentation is commercially available to address each of the problem areas.

### DRES REQUIREMENTS

For DRES applications, a large chamber filled with CBW simulant aerosol is required to study new aerosol detectors, protective devices and aerosol behavior. This chamber must be large enough to accommodate about six people to provide sufficient space for limited physical movement. Other important technical characteristics associated with the chamber are as follows:

- a. Able to maintain a set temperature.
- b. Provide a range of RH conditions.

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- c. Provide a means to select and maintain constant aerosol particle concentration.
- d. Able to support a liquid aerosol generator with potential support for a solid powder system.

This report describes a microcomputer driven aerosol system with generation, measurement and feedback control for maintaining a preset particle concentration threshold. A RH control system is also presented which has critical characteristics relevant to aerosol research. Part of this system has been described in a patent award declaration (Ho, 1987). Operation of this system with a BW spore simulant has been demonstrated.

### MATERIALS AND METHODS

#### AEROSOL CHAMBER

The chamber, also called the "sporelab", measured 28 sq meter of floor space enclosing 90.5 cu meter of air space (Figure 1). For internal biological decontamination, ceiling mounted UV lights could be activated at termination of an aerosol experiment. A high speed venting fan connected to overhead ducting was used for clearing of test aerosols. Banks of UV lights were installed in the ducting to provide a primary biological decontamination. Before the air was exhausted to the outside, a HEPA filter (model 7C23-SLCCD, Flanders Filters Inc. Washington, NC) provided a second decontamination step. Routine biological tests of the exhaust air confirmed filtration effectiveness.

#### ENVIRONMENTAL CONTROLS

Temperature control was provided by a cooling and heating unit (Lennox model HS18-411-C6P, Lennox Industries Canada, Ltd., Calgary, Alberta). Nominal temperature was set at 22 °C ( $\pm 1.0$  °C) for all seasons and the appropriate heating or cooling demands were regulated by a Honeywell thermostat (Honeywell Controls, Scarborough, Ontario).

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Moisture content (RH) was regulated by a feedback control loop consisting of a Humitemp Controller (model B, Phys-Chemical Research Corp., New York, NY) attached to a PCRC-11 HPB sensor via a 25 ft long extension cable. This controller supplied an on/off signal to another component in this loop, a mechanical aerosol fog generator (model 202, Microsol, Silver Creek Precision Corp., Silver Creek, NY). Ultra pure water (Milli-Q/UF system, Millipore Corp., Bedford, MA, USA) was used to furnish the required moisture to maintain RH above ambient levels. Actual measurement of RH was performed by a chilled mirror type hygrometer (model 1500 Hygrocomputer, General Eastern Instrument Corp., Watertown, Mass.). Very accurate determinations could be obtained with this instrument especially at extreme ends of the RH scale ( $<10$  and  $>80$  %RH). Among all kinds of RH measuring instruments, only this device could measure the low values (5-15%) common to this region, especially in the winter time.

### AEROSOL GENERATION AND CONTROL

An aerosol of *Bacillus subtilis sp globgii* (BG) spores was generated in the chamber by means of a Collision nebulizer (May, 1973) from liquid suspensions (water based) of  $1 \times 10^9$  viable organisms/mL. The experiment setup is shown in Figure 1. Aerosol concentration was monitored by an Aerodynamic Particle Sizer (model APS3300, TSI, St. Paul, MN) running proprietary software on an Apple II+ microcomputer (Apple Computer, Cupertino, CA). The instrument could be set to monitor a narrow size range ( $\pm 0.1 \mu\text{m}$ ) corresponding to the aerodynamic diameter of the particle under study. Measuring time was normally 5 sec, followed by a 40 sec interval for computer processing. This represented a minimum of 45 sec cycling time for the control system. Software control was directed to a solid state switch (model W3PCX-1, Magnecraft Electric Co. Chicago, ILL) capable of supplying a 2 Amp 110 volt power to a solenoid which controlled compressed air (200 kPa) to the Collision. A custom interface card in the computer transferred low voltage control signals to the solid state switch.

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### AEROSOL COLLECTION

The standard All Glass Impinger (AGI) was used to collect aerosol samples at 12.5 LPM (Shipe et al., 1959). The aerosol was also collected by a dry filtration technique. A cost effective borosilicate microfiber (glass fiber) filter was selected for this purpose (Grade GA55, Cat. no GA5537MM, 37 mm diameter, Micro Filtration Systems, Dublin, CA). This filter was chosen for its ease in resuspending collected particles in distilled water as well as its efficiency for collecting small particles (0.3  $\mu\text{m}$ ). For this study, the glass fiber filters with aerosol samples were resuspended in ultra pure water.

### CELL ASSAY

Liquid samples from AGI samplers were plated on nutrient agar after appropriate serial dilutions to enumerate the viable organisms by standard methods. Additional steps were required to resuspend the particles captured on filters. Each filter was homogenized in 20 mL ultra pure water by vigorous shaking. The homogenate was strained through a wire gauze disk to remove fiber strands. The filtrate containing resuspended particles was processed as for the AGI samples. Colonies were counted after overnight incubation at 30 °C.

### DATA ANALYSIS

Analysis of variance of the RH and viable spore replicate data were performed by routines from the RS/1 scientific data analysis package (BBN Software Product Corp. Cambridge, MA).

### RESULTS AND DISCUSSION

#### RELATIVE HUMIDITY STUDIES

During fall and winter, RH measured with the Hygrocomputer in the sporelab usually registered as low as 5 to 15%, a region not easily detected by wet-dry bulb instruments (Ruskin, 1965). Stability of this value was about 0.5% for up to

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several hours. However, the presence of just one person in the chamber could increase this level by about 2% within 5 to 10 min, probably due to moisture from exhaled air. This observation illustrates the sensitivity of the RH detecting device, as confirmed by the experiment shown in Figure 2.

In this case, the ambient RH (16%) was monitored and readings were taken three times within 15 min. Then the Humitemp controller was adjusted to produce and maintain a higher (in steps of about 10%) RH level. After conditions had stabilized, data points were again recorded for a period of 15 min. The cycle was then repeated for the next RH increment until the highest indicated level was obtained. A plot of controller settings versus the measured RH (figure 2) showed a linear relationship as demonstrated by analysis of variance of the replicate readings. The calculated coefficient of regression was greater than 0.99 while standard deviation of the regression slope was a 0.02, suggesting very stable RH conditions at each level. An F value of greater than 1752 at 0.0001 significant level further confirmed good RH stability in the chamber.

As shown in Figure 2, good linearity was observed for up to 65% RH. Beyond that, the curve started to flatten slightly. This curve flattening could reflect the Humitemp sensing element limitations at high RH levels, a common problem with polymer sensors (Ruskin, 1965). However, such a performance shortcoming might not pose operational problems as long as the sensor responded reproducibly, as was demonstrated in this study.

Good temperature stability was also shown in Figure 2. During the time of the study, a constant 22 °C was measure by the precision platinum probe which was part of the Hygrocomputer sensing array.

It was previously shown (Kournikakis *et al* 1986) that trace impurities in water, when aerosolized, could contribute to changes in total particle numbers as well as size distribution. In the RH generating system just described, the addition of moisture to air could alter the naturally occurring (background) particle concentration and/or size distribution. An analysis of selected particle size groups at different RH as shown in Figure 3 revealed that changes did occur to some particles. Shifting the RH from 15 to 25% resulted in a concomitant drop in the 0.55  $\mu\text{m}$  particle num-

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bers. Note that at this RH level, the larger particles ( $0.75\ \mu\text{m}$ ) did not exhibit similar changes. However, shifting the RH from 65 to 75% caused particle numbers to decrease in all three size groups ( $0.55$ ,  $0.65$  and  $0.75\ \mu\text{m}$ ). This observed decrease in fine particle numbers during RH shift could be due to coagulation of smaller size units resulting in larger aggregates (Mercer, 1973).

Fine particle size dynamics as influenced by RH conditions could be further illustrated by plotting number versus size distribution at different RH levels of 15, 25 and 75% (Figure 4). Shifting from RH 15 to 25% caused a drop in particle numbers in the  $0.5$ - $0.65\ \mu\text{m}$  size range, accompanied by a corresponding increase of  $0.85$ - $1.4\ \mu\text{m}$  particles. In this figure, the error bar or sampling uncertainty was  $\pm 1$  standard deviation from the mean, suggesting that the observed changes were significant. At a humid 75%, the smaller particle size group suffered further decrease along with an overall drop in the larger particles. No appearance of other larger particles were detected. Presumably, at this high RH, much larger aggregates could have been formed but their numbers were below the detection range of the instrument selected for that measurement. For detection of small number populations, greater sampling times would be needed. The data sets in Figure 4 had 20 sec sampling time. No attempts were made at detecting these postulated particles.

The main reason for measuring background particle concentration under different RH conditions was to determine if there were contributing numbers from contaminants in the feed water and spray generator. The above results suggest the observed changes were not related to water impurities but that some ambient particles did increase in number under the influence of RH conditions. However, Figure 5 illustrates that such small increases were insignificant when compared to the magnitude of a typical aerosol spectral plot, in this case, of BG spores. The major peak was due to spore particles while the minor peak was attributed to impurities that originated with the suspension. It was not surprising to find that the ultra pure water contributed no significant particles to the background but the study also revealed that the fog generator itself, did not contribute contaminant particles during operation in the aerosol chamber.

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### AEROSOL CONCENTRATION CONTROL

This illustration of RH induced changes in ambient particle size distribution underscored the importance of performing aerosol experiments in controlled environments. Another critical parameter that must be controlled is particle concentration. In this study, a measuring and generating feedback loop was used to control particle concentrations in the chamber. A particle sizer measured the numbers of a specified size group and instantaneously compared the results with a user selected threshold level, defined as particles/cc aerosol. Depending upon the outcome of this, an electronic signal could be sent to switch the aerosol generator on or off. Figure 6 shows a plot of viable organisms recovered from aerosols generated at different threshold levels. The results indicated increasing viable organisms were recovered corresponding to increasing threshold levels. More data scatter was noted for the higher threshold levels, reflecting less consistency in control for this regions. Fortunately, in order to provide a realistic aerosol, control at level 25 would produced about  $10^5$  viable organisms/L aerosol.

### APPLICATION OF SYSTEM

With the capability to set and maintain aerosol concentrations, it has become possible to use the facility to compare sampling characteristics of two sampling methods, the conventional AGI versus glass fiber filters. The concept of replicate sampling could be applied when a desired aerosol concentration could be maintain for a period of time. The time factor would permit the user to set up and run complex sampling equipment, in this case, with two different sampling techniques. The results of this comparison revealed many useful details, for example, between threshold levels 5 to 30, the two methods gave similar recovery (Figure 6). Beyond level 30, there was considerable data scatter, making direct visual comparison difficult. However, analysis of variance comparison of the two sets of results revealed no difference between their linear regression slopes (table 1). The higher standard deviation value for the filter method would suggest lower overall precision with respect to the AGI method. This observation was consistent with the differences in the two assay procedures. For instance, extra steps were required to extract trapped particles from the filters as described previously. Nevertheless, this study has demonstrated that the filter method could be a feasible alternative to sampling BW

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or other particulate aerosols when liquid based samplers could not be used. Low temperature at sampling sites would be such a situation where filter sampling could overcome freeze up problems associated with liquid based samplers.

The aerosol chamber was designed to provide a way to create stable particle concentrations throughout the duration of an experiment. This capability was dependent upon the coordinated timing function of the whole system to sustain and maintain an aerosol supply. As mentioned earlier, if the system responded too slowly, this would be reflected as fluctuations in measured particle concentrations. From the above results, it was determined that about  $10^5$  viable spores/L aerosol could be generated by the instrumentation without causing unusual sampling errors. Using these criteria, an aerosol was generated to determine concentration stability over time. Figure 7 shows the recovered viable spores in the aerosol chamber over the period on an hour. Sampling of the aerosol commenced after the control electronics had established the desired level (threshold level 25). This condition took between 20 to 30 min from the time the Collision was started and was characterized by a rhythmic solenoid on/off pattern at a rate of about 5 min/cycle. As seen in this figure, the viable numbers were reasonably stable, especially during the latter period of the study.

#### CONCLUSIONS AND SUMMARIES

To meet special requirements in defence CBW aerosol research, a large chamber was successfully designed with the following capabilities:

a. The installation of a heating and cooling unit allowed temperature be set and maintained to facilitate reproducible experimental conditions. This was also a critical feature in preventing heat exhaustion problems during protective clothing studies.

b. A high capacity moisture generating system was designed to provide a wide range of RH to be set and maintained. In particle dynamics studies, this feature would facilitate better understanding of fine particle behavior under various RH conditions, as demonstrated in this report.

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c. Due to commercial availability of laser based particle sizing and measuring instrumentations, it was possible to configure a system capable of providing means to select and maintain a constant aerosol concentration in the chamber. This was achieved by a combination of custom software and hardware interfacing to a standardized laboratory aerosol generator.

d. As an exploitation of the system's ability to set and maintain known aerosol concentrations, the sampling characteristics of a glass fiber filter was compared to that of the AGI. It was found that their sampling performance was comparable and that the filter could be used in place of the AGI under most situations.

e. Although the current system was configured to drive a liquid based aerosol generating system, as demonstrated with BG spores, very little modification would be needed to adapt it for a dry powder system. The same compressed air supply could be used to power such a generator, disseminating solid CBW particles.

Recently, a Canadian and a U.S. patent has been granted to protect the intellectual property describing the feedback control system for controlling aerosol particle concentrations (Ho, 1987).

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**Legends**

**Figure 1.** Diagram of aerosol generating and sampling system.

**Figure 2.** Measured relative humidity at various settings.

**Figure 3.** Effect of relative humidity on background particle sizes.

**Figure 4.** Effect of relative humidity on particle size distribution.

**Figure 5.** Comparison of BW aerosol with background particles.

**Figure 6.** BG aerosol concentration at different threshold levels.

**Figure 7.** Stability of aerosol concentration.

**Table 1.** Analysis of variance summary from BG concentration studies.

**Keywords:** CBW aerosol; particle concentration control; microcomputer control; relative humidity control; relative humidity measurement; laser air particle sizer; dry powder aerosol; BG aerosol.

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**TABLE 1**  
**ANALYSIS OF VARIANCE SUMMARY FROM BG CONCENTRATION STUDIES**

|                               | <b>AGI</b>    | <b>FILTER</b> |
|-------------------------------|---------------|---------------|
| <b>REGRESSION COEFFICIENT</b> | <b>0.87</b>   | <b>0.79</b>   |
| <b>SIGNIFICANCE LEVEL</b>     | <b>0.0001</b> | <b>0.0001</b> |
| <b>F VALUE</b>                | <b>146.4</b>  | <b>83.4</b>   |
| <b>DEGREES OF FREEDOM</b>     | <b>22</b>     | <b>22</b>     |
| <b>SLOPE</b>                  | <b>4079</b>   | <b>4622</b>   |
| <b>STANDARD DEVIATION</b>     | <b>337</b>    | <b>504</b>    |

AGI = ALL GLASS IMPINGER SAMPLER  
FILTER = GLASS FIBER FILTER SAMPLER

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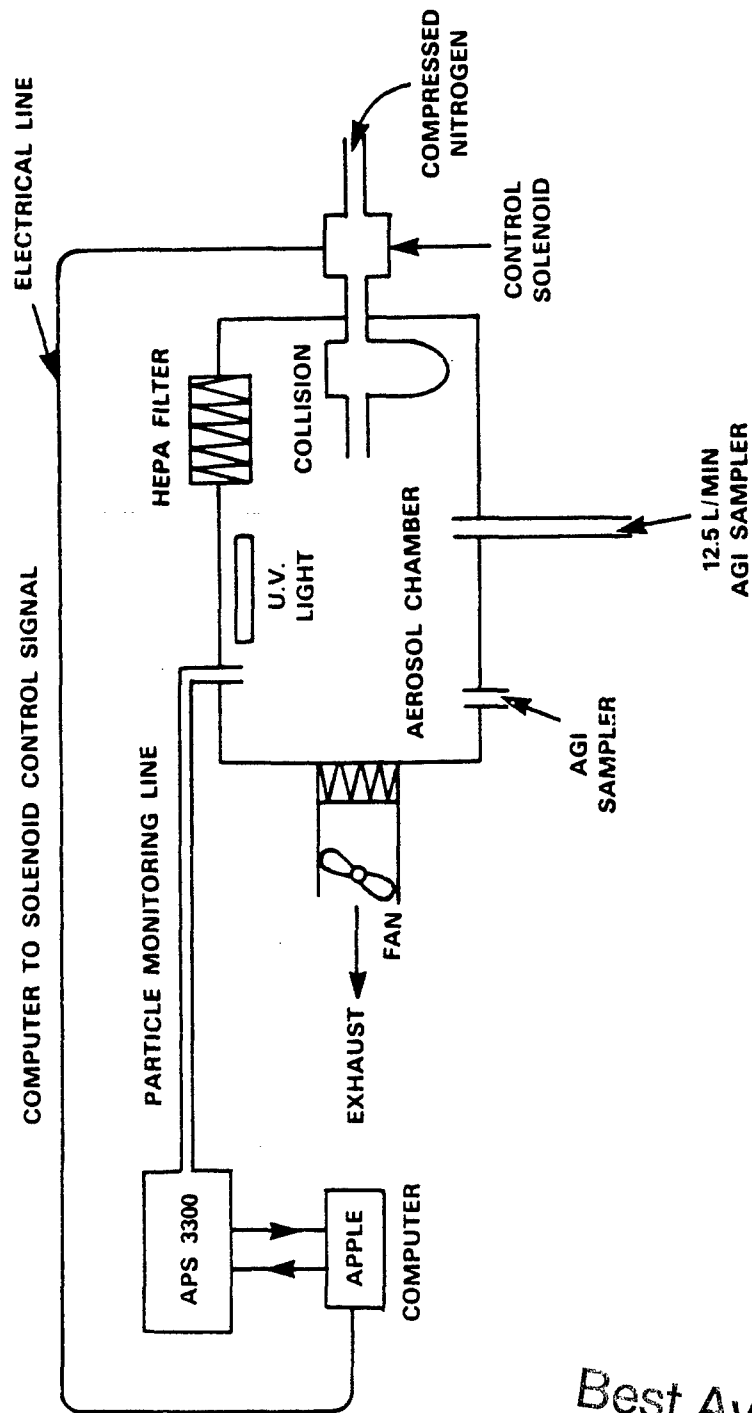


Figure 1

DIAGRAM OF AEROSOL GENERATING AND SAMPLING SYSTEM

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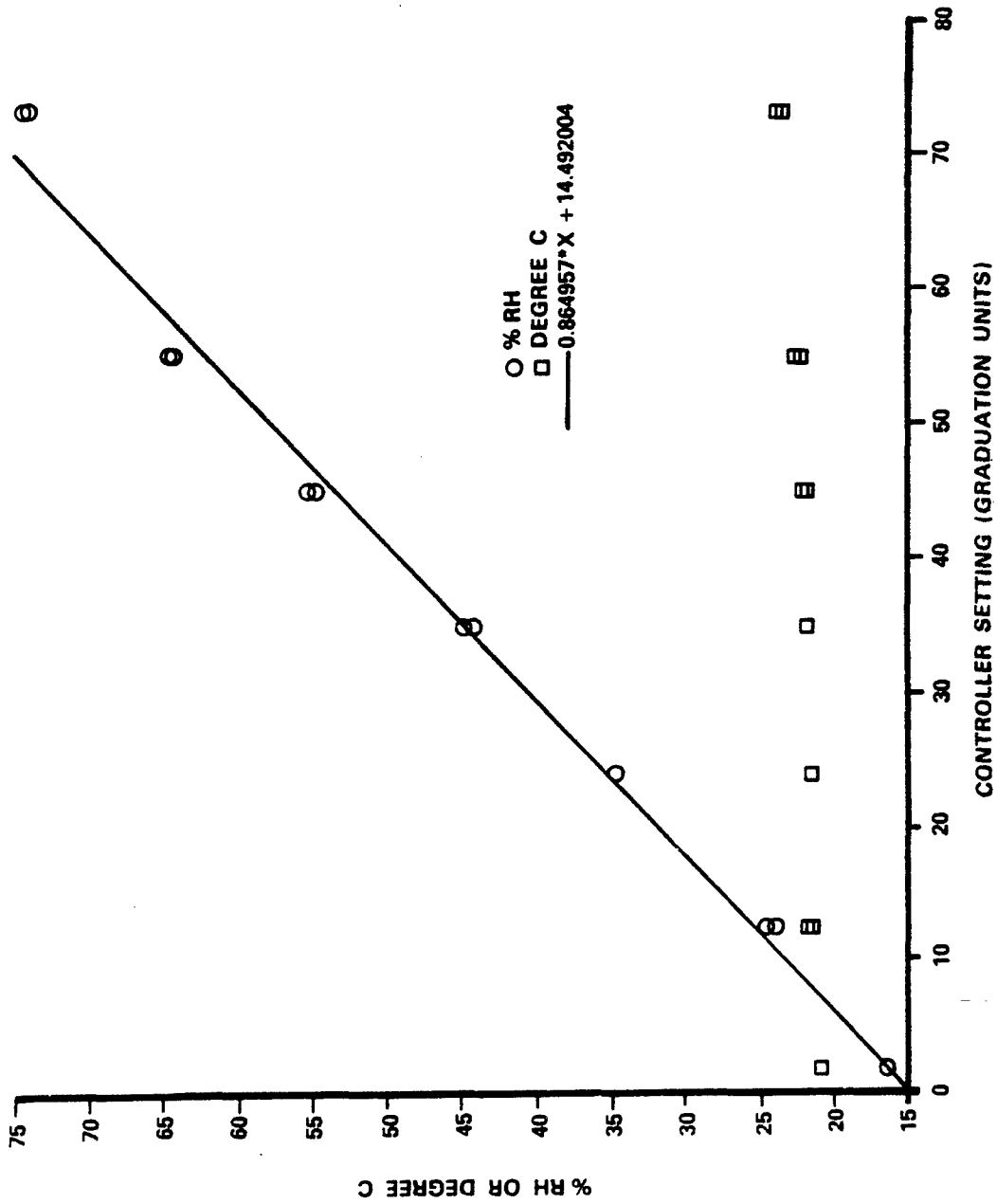


Figure 2

MEASURED RELATIVE HUMIDITY AT VARIOUS SETTINGS

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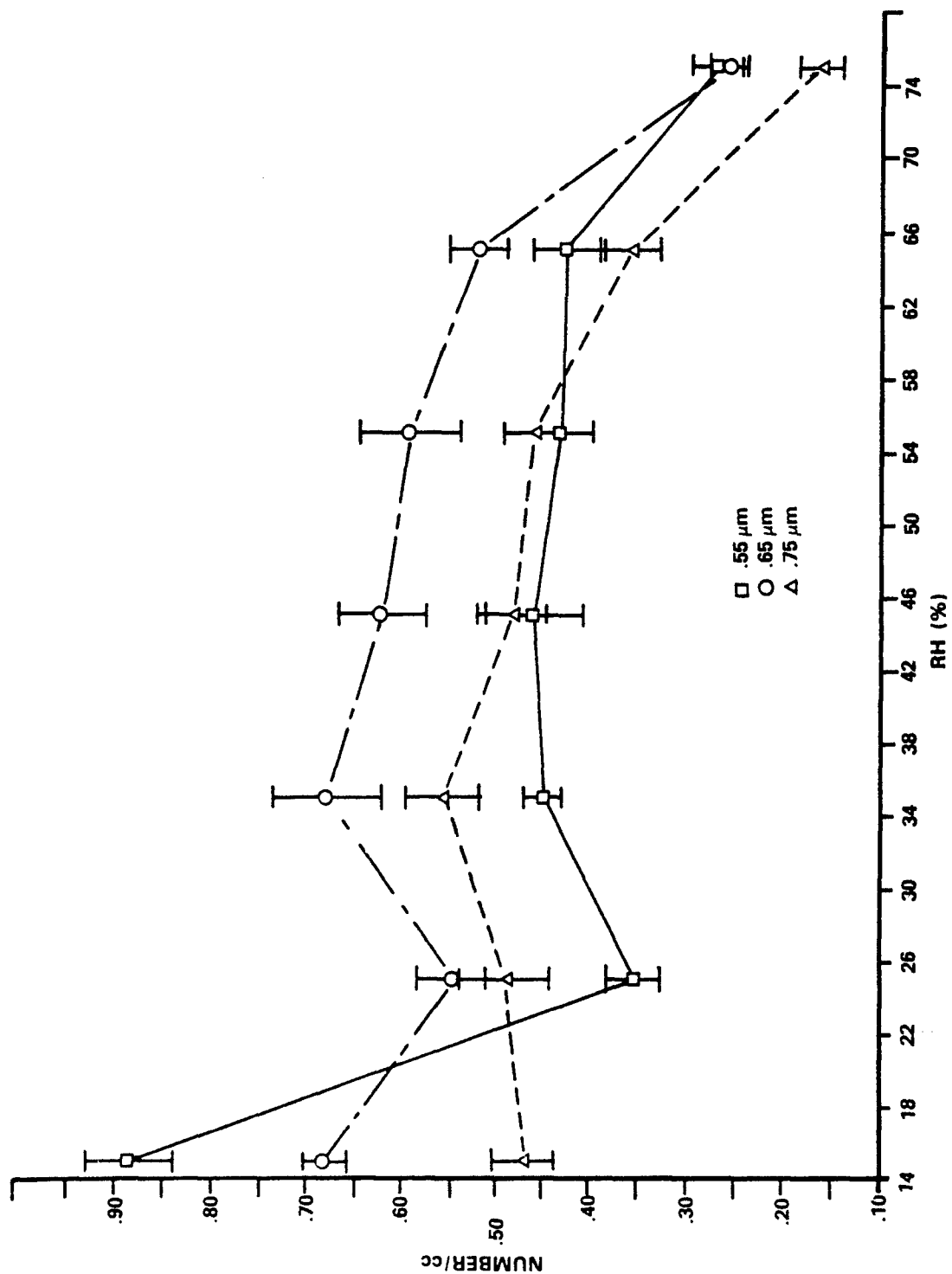


Figure 3

EFFECT OF RH ON BACKGROUND PARTICLE SIZES

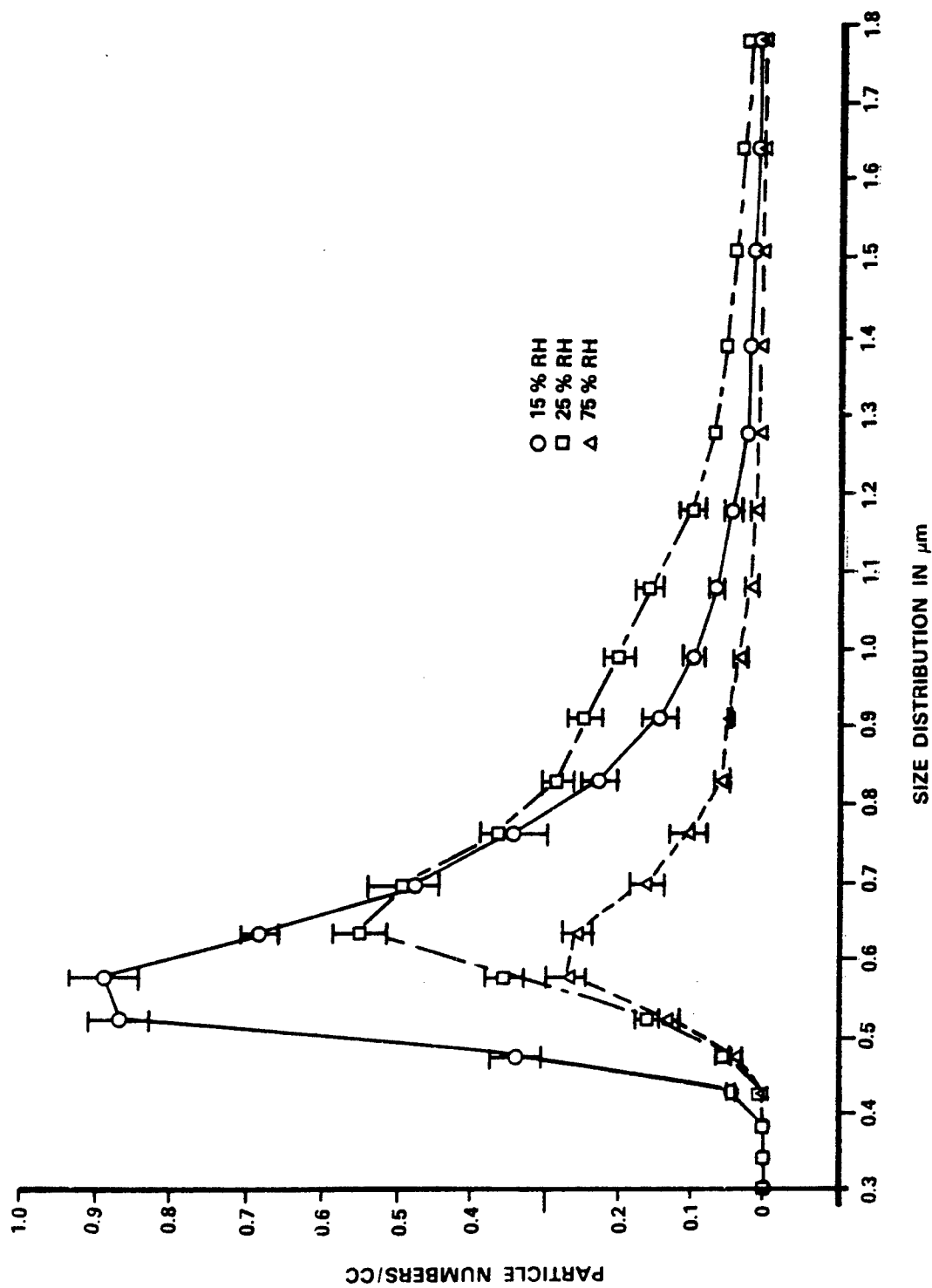


Figure 4  
EFFECT OF RH ON PARTICLE SIZE DISTRIBUTION

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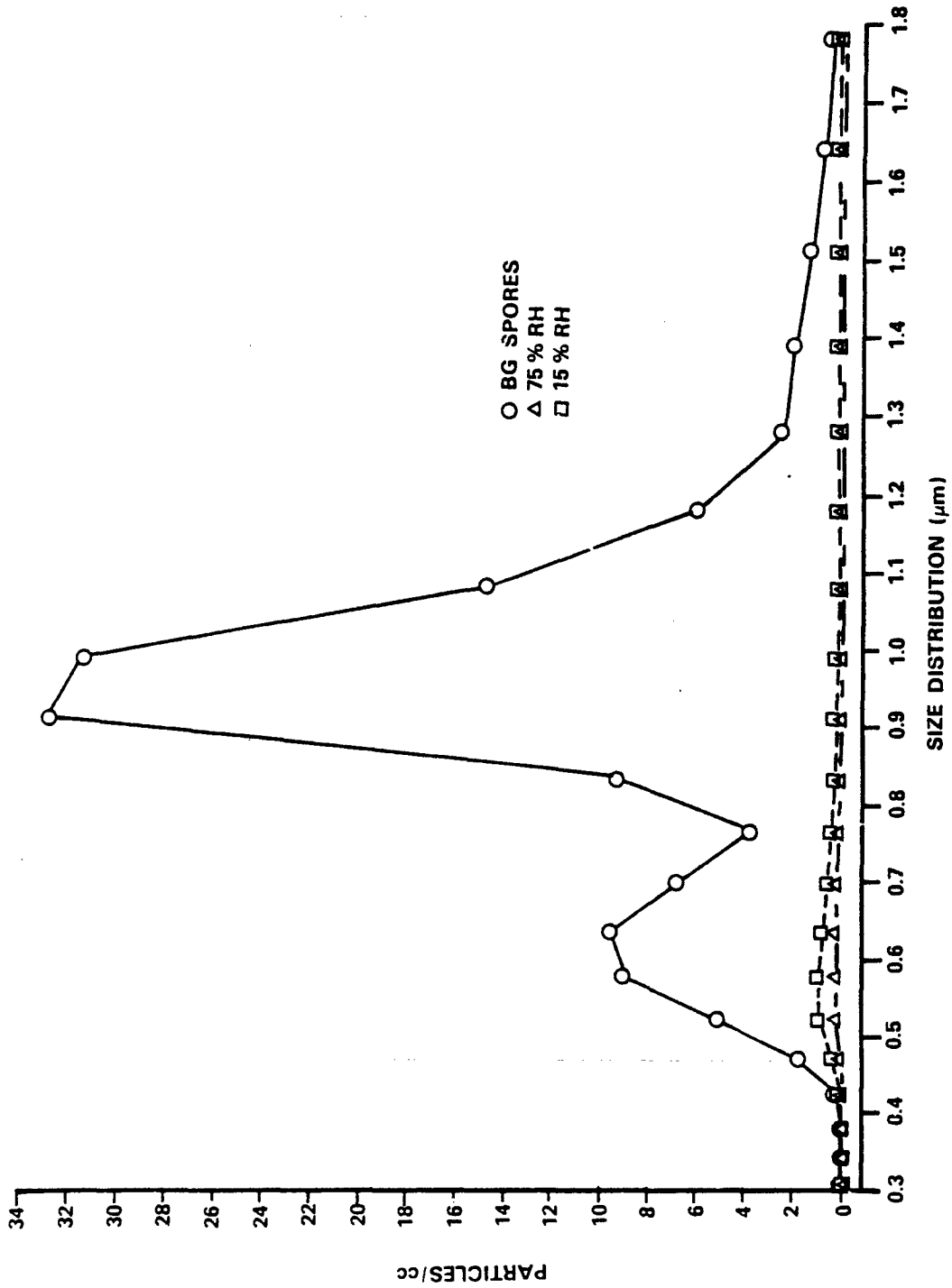


Figure 5  
COMPARISON OF BW AEROSOL WITH BACKGROUND PARTICLES

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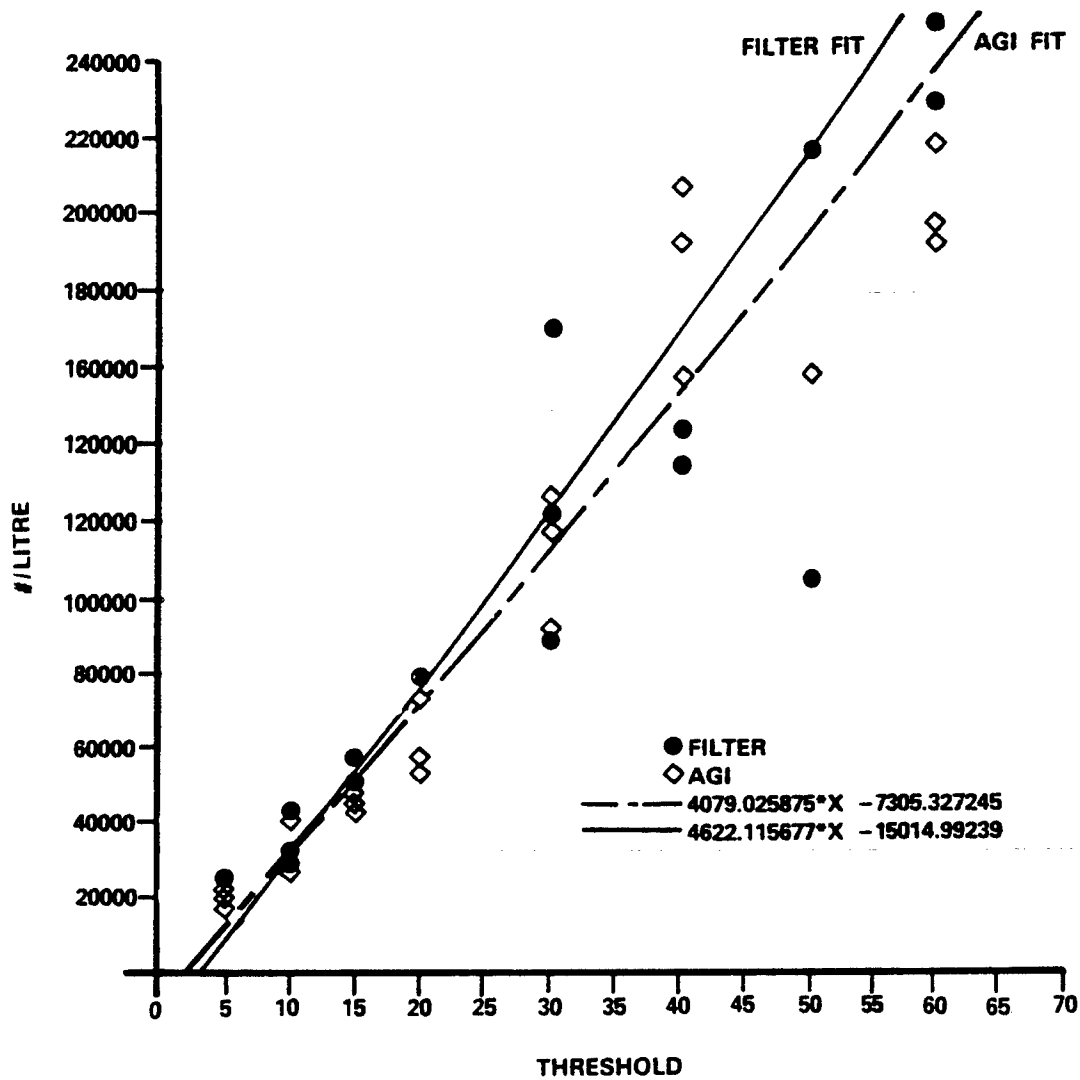


Figure 6

BG AEROSOL CONCENTRATIONS AT DIFFERENT THRESHOLD LEVELS

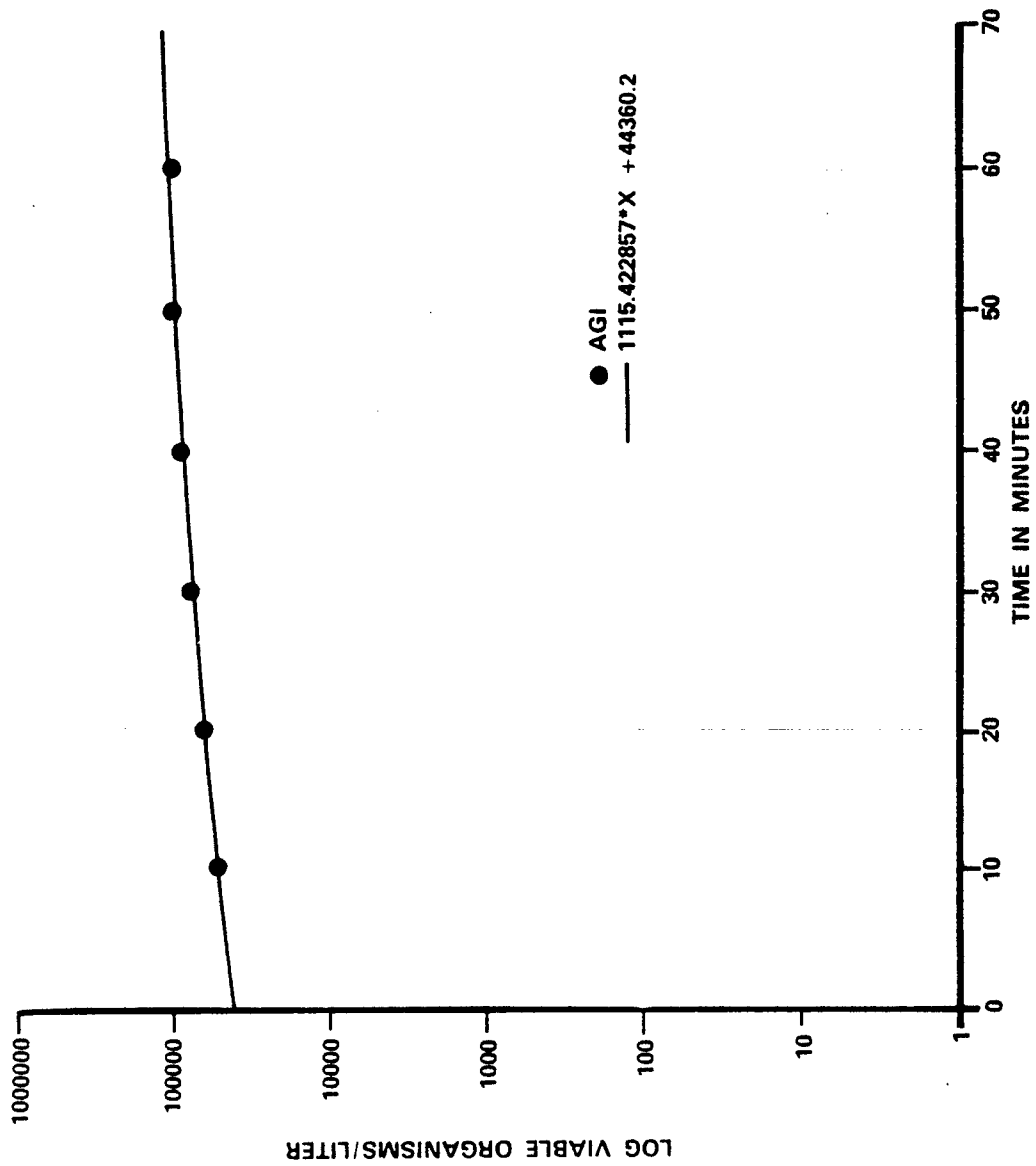


Figure 7  
STABILITY OF AEROSOL CONCENTRATION

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An aerosol generating system was integrated into a large chamber to regulate three experimental conditions, temperature, relative humidity (RH) and particle concentration during aerosol studies. A commercial heating and cooling unit was used to control temperature. Control of RH was effected by a polymer-based sensing mechanism coupled to a fogging device. Aerosol particle size and numbers were monitored and controlled by a microcomputer-driven detector coupled to an aerosol generator. The system was able to provide precise and reproducible environmental conditions conducive to aerosol studies. Aerosol concentration control was demonstrated over a range of concentrations suitable for different requirements. Under these conditions, a BW simulant aerosol was stable in terms of viability for an extended period of time. As an illustration of the system's potential application, it was used to demonstrate sampling characteristic similarities between a glass fiber filter and a glass impinger. With the features described, the system has significant utility in aerosol studies involving threat detection and protection. This system is easily adapted to drive a dry powder disseminator to generate different CBW simulant aerosols. A Canadian patent (No. 1,222,200) has been awarded to part of the system.

14. KEYWORDS, DESCRIPTORS or IDENTIFIERS (technically meaningful terms or short phrases that characterize a document and could be helpful in cataloguing the document. They should be selected so that no security classification is required. Identifiers, such as equipment model designation, trade name, military project code name, geographic location may also be included, if possible keywords should be selected from a published thesaurus, e.g. Thesaurus of Engineering and Scientific Terms (TFST) and that thesaurus-identified. If it is not possible to select indexing terms which are Unclassified, the classification of each should be indicated as with the title.)

Keywords: CBW aerosol; particle concentration control; microcomputer control; relative humidity control; relative humidity measurement; laser air particle sizer; dry powder aerosol; BG aerosol.

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