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An Apparatus for the Continuous Generation of Stock Solutions of Hydrophobic Chemicals*

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A major technical difficulty associated with testing the toxicity to aquatic organisms of chemicals having low water solubility is the production of a stock solution of the chemical without the use of exogenous solubilizers or carriers. For highly toxic hydrophobic chemicals this problem has been approached by passing water through a column of chemical adsorbed to inert supports (CHADWICK & KIIGIMAGI 1968). While this technique is somewhat effective, channelization of water in the column may lead to variation in the concentration of toxicant produced during long term exposure studies. VEITH & COMSTOCK (1975) reduced this variation by continuously recirculating the toxicant solution through the bed of substrate with a submersible pump. For many hydrophobic chemicals of moderate or low toxicity, the difficulties of producing a satisfactory stock solution are compounded by the fact that water concentrations of the chemical that are acutely toxic to fish may be near their limits of solubility. Toxicity testing of these chemicals through continuous flow bioassay methods thus requires large volumes of stock solution of relatively high concentration. In addition, many toxicants are liquids at test temperatures and not amenable to application from a substrate bed.

The apparatus described in this communication is designed to continuously generate a stock solution of hydrophobic chemicals over prolonged periods of time by recirculating water from a 190-L reservoir through a mixing chamber holding the toxicant. It has the advantage of being adaptable to chemicals having a wide array of physical properties and to many types of commonly used diluter systems.

MATERIALS AND METHODS

The stock solution generator consists of a dilution water headbox and the stock toxicant solution reservoir (Fig. 1). The dilution water headbox acts to establish a constant head, and ensures a regulated flow of water both to the main diluter apparatus

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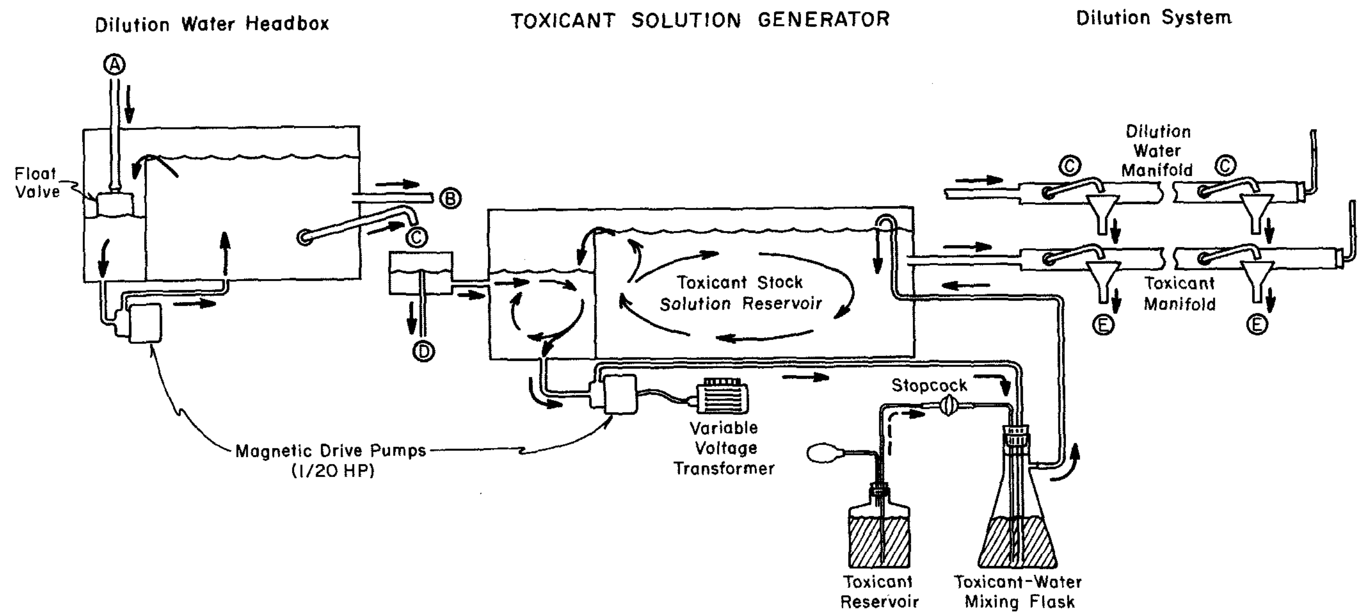

Figure 1. The stock solution generator supplying dilution water and toxicant solution to one of the several possible diluter types.

and to the reservoir of stock toxicant solution. The stock solution reservoir is constructed of a 190-L glass aquarium that has been separated into an affluent and effluent compartment by a glass partition. Water enters the affluent compartment by gravity flow and is pumped from this compartment by a small magnetic drive pump through a bed of toxicant and into the effluent compartment. The solution is pumped at a rate in excess of that withdrawn from the reservoir and the excess solution flows over the glass partition to be recirculated within the reservoir. The rate of recirculation is adjusted by a variable voltage transformer controlling the recirculation pump (Fig. 1). This pattern of water inflow, recirculation and withdrawal insures that all water entering the apparatus passes through the toxicant at least once before entering the toxicant manifold.

Toxicants may be introduced into the recirculation system in several ways depending on their physical properties. Hydrophobic liquids denser than water are placed in a large (4-L) vacuum flask sealed with a neoprene stopper. The flask is inverted for liquids less dense than water. A piece of glass tubing inserted through the stopper provides an entry port for water which is pumped to the bottom of the vessel and allowed to float up through the chemical. The resulting solution exits from the side arm of the flask and enters the effluent side of the toxicant reservoir. The level of toxicant in the mixing flask is maintained at a uniform volume by connecting a separate toxicant reservoir to the mixing flask by a section of glass tubing fitted with a teflon stopcock. The toxicant reservoir is sealed with a neoprene stopper through which a piece of glass tubing has been inserted. Positive pressure is created in the toxicant reservoir with a gas collecting flask fitted with a one way valve and attached to the glass tubing. When the stopcock is open liquid is forced into the mixing flask, thereby introducing chemical into the mixing vessel without interrupting the operation of the stock solution generator.

RESULTS AND DISCUSSION

The apparatus described herein is particularly well suited for generating stock solutions of hydrophobic chemicals for use in either acute bioassays or for use in prolonged subacute exposure studies. The design of the system is such that it is adaptable to either liquid or crystalline chemicals having a wide array of physical and chemical properties. Furthermore, it is compatible with any type of diluter system that requires a constant supply of stock toxicant of uniform concentration. The constant head reservoir eliminates the need for constant head stock solution bottles commonly employed with many diluter systems.

The concentration of stock solution produced by this apparatus may be increased or decreased by adjusting either the rate of recirculation between the stock solution reservoir and the toxicant reservoir in the rate of withdrawal from the system. The characteristics of performance of the system for a hydrophobic liquid chemical (monochlorobenzene) are illustrated in Fig. 2. For any ratio of recirculation to withdrawal an

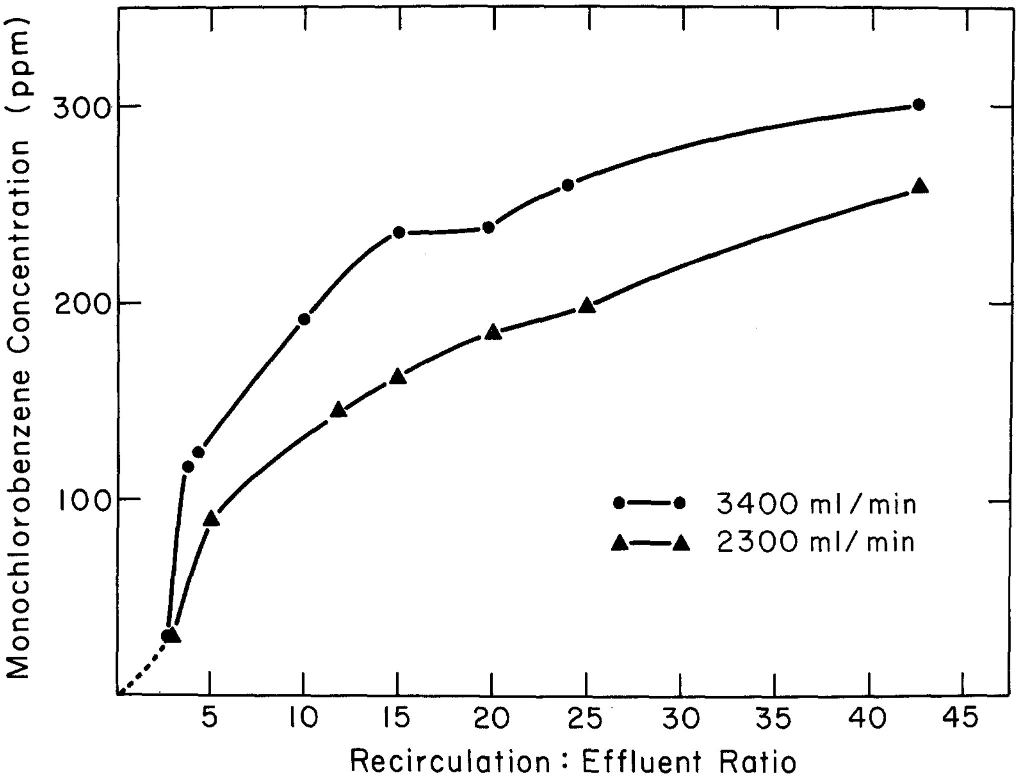

Figure 2. The relationship between toxicant concentration and the ratio of recircultation to withdrawal rate of two recirculation rates, 3400 ml/min and 2300 ml/min.

equilibrium is established between the chemical and the solution of constant and uniform concentration. The equilibirium concentration of monochlorobenzene solution at 15°C generated with a recirculation rate of 3400 mL/min and a withdrawal rate of 78 mL/min (ratio = 43.5) was 293 ± S.E. 10.5 mg/L during a six week period. By changing the flow characteristics of the system a new equilibrium can be established and a stock solution of constant concentration produced. Variation in the concentration of stock solution for any given ratio of recirculation to withdrawal was less than four percent between individual samples after an equilibrium concentration had been established.

The time required for an equilibrium condition to be established in the apparatus varies, depending on the physical properties of the chemical and the rates of recirculation in and withdrawal from the system. For monochlorobenzene stable concentrations were established in the stock solution by 12 h at a recirculation rate of 3400 mL/min and by 18 h at a recirculation rate of 2300 mL/min.

The concentrations of solutions produced in this apparatus were found to vary for a given ratio of recirculation to effluent flow. Generally higher rates of recirculation and effluent flow produced higher concentrations of stock solution than did lower rates of recirculation and withdrawal for the same recirculation effluent ratio (Fig. 2). For example, equilibrium concentrations of monochlorobenzene stock solution consistently were 20-25 percent less for the same ratios of recirculation to withdrawal between 5 and 25 when the rate of recirculation was 2300 than when the recirculation rate was 3400. This difference may not be as apparent for other chemicals and probably is a function of the physical properties of the chemical, particularly vapor pressure and volatility. Our experience with chemicals such as nitrobenzene, toluene, ethylbenzene, monochlorobenzene, o-dichlorobenzene and 1,2,4 trichlorobenzene has been that the optimal ratio of recirculation to withdrawal differs for different chemicals. The production of a stock solution that is less than saturated may be desirable for volatile chemicals such as nitrobenzene since they are less likely to vaporize from a solution that is not saturated. The tendency for chemical loss from the stock solution thereby is reduced and concentrations of chemical reaching the exposure tanks will approach nominal concentrations.

This system is capable of producing stable solutions of single, relatively hydrophobic, organic chemicals over prolonged period of time without the use of carriers or solubilizers. It may not be suited for use with complex mixtures of relatively insoluble chemicals such as crude oil fractions or with technical grade chemical mixtures such as polychlorinated biphenyls because of differences in the water solubility among constituents of the mixture.

REFERENCES

CHADWICK, G., and U. KIIGEMAGI: J. Water Pollution Control Fed. 40, 76 (1968).

VEITH, G., and V. COMSTOCK: J. Fish. Res. Board Canada 32, 1849 (1975).