A peristaltic dosing apparatus for the chronic exposure of aquatic organisms to pollutants

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Abstract

A dosing apparatus which is suitable for the chronic exposure of aquatic organisms to pollutants is described. Water flows are adjusted by altering the head height and delivery tube dimensions. A peristaltic pump is used to meter toxin input. Failsafe controls ensure that undilited toxin does not reach the test organisms in the event of a water supply interruption and that excessive toxin dilution does not occur in the event of an electrical power failure. The apparatus was used to provide constant seawater/toxin flows for periods of up to four weeks and was found to be reliable and easily maintained.

Introduction

The traditional method of assessing toxicity by means of static, acute LC 50 determinations has many limitations (Brown, 1976). In recognition of this, aquatic toxicologists now tend to use chronic sublethal effects on the more sensitive stages of the life cycle in order to formulate criteria for toxic effects and, where practicable, complete life cycle multigeneration tests are being favoured.

To perform a chronic toxicity test it is essential to have a reliable means of dosing the toxin to experimental aquaria over an extended period. Numerous devices have been designed to achieve this (e.g. Abram, 1960; Alabaster and Abram, 1965; Stark, 1967; Mount and Brungs, 1967; Burke and Ferguson, 1968; Connor and Wilson, 1972). Although electrical pumps, especially peristaltic pumps would seem suitable for incorporation into dosing systems, most investigators appear to have concluded that they are "too undependable" (Sprague, 1970). Connor and Wilson (1972), however, decided that this had led to unnecessary complications and they designed a system incorporating two electrical pumps with failsafe controls. Although their apparatus was greatly simplified in comparison with earlier designs it was evident that further simplification and refinement was possible. This led to the design and construction of a new dosing apparatus.

Design

The apparatus (Figure 1) was designed to supply a constant flow of seawater containing fixed quantities of toxin to twelve test aquaria, each of 10 ℓ capacity.

Seawater is introduced from an overhead reservoir into a valve chamber (A). A filter (B) containing synthetic filter wool (Filtron) removes any large solids which may be present. The valve chamber is connected by means of a flexible PVC hose (C) (12 mm internal diameter) to a constant head chamber (D). Contained within the valve chamber is a float valve (E) with a positive buoyancy of approximately 1,5 kg. Construction details of the valve chamber, filter, constant head chamber and float valve are given in Table 1. As seawater enters the valve chamber the float rises and, on reaching the tapered inlet, tends to restrict further seawater inflow. Twelve outlets constructed of perspex piping (4 mm external diameter, 1,5 mm internal diameter and 10 mm long) load off from the constant head chamber to provide seawater flows to the test chambers (F) (10 ℓ glass aquaria with 4 mm internal diameter overflow pipes). Both the constant head and valve chambers are mounted on retort stands and their heights are therefore adjustable. By adjusting the relative heights of the constant head and valve chambers it is possible to reach a situation where a constant head of seawater will be maintained. This will occur when the upward pressure of the float valve regulates seawater inflow such that it exactly balances the outflow from the constant head chamber. An overflow pipe leads from the constant head chamber to waste. This is primarily a safety measure but serves also to provide an upper limit to the water head height. In practice the water level was maintained some distance below the overflow pipe to avoid seawater wastage.

The components which have been described so far serve only to provide a constant head of filtered seawater. Alternative methods are obviously available, such as the overflowing reservoir described by Connor and Wilson (1972) or a simple ballcock of the type used in toilet cisterns. The former has the disadvantage of requiring an electrical pump to recycle water that has overflowed whilst the latter is somewhat bulky and would require a considerably larger vessel in which to operate. The system which has been described was found to be compact and reliable.

Having established a constant head it is relatively simple to provide a constant flow of seawater, provided that some sort of regulator is present. Connor and Wilson (1972) devised a system of flow adjustors for regulating their flow rates. The regulator used in this instance was simply a length of narrow bore flexible PVC tubing. Despite the complexities associated with describing liquid flow (Starling, 1961), it may be assumed that the flow rate of a liquid through a tube is essentially a function of the density of the liquid, the diameter and length of the tube and the head height. It follows that, with a liquid of reasonably constant density, such as seawater, the flow rate can be altered merely by altering the head height or by substituting tubes of different internal diameters or lengths. This principle has been exploited in the design of the peristaltic doser to provide constant flows of seawater at predetermined rates. A wide range of accurate bore narrow gauge PVC tubes are available from suppliers of autoanalytical equipment. In practice it was found that a standard tubing of 1,3 mm internal diameter, a head height of approximately 80 cm and a tube length of about 100 cm provided flow rates of the required order (ca 1 500 mℓ h^{-1}). This tubing is designated as G in Figure 1.

Having allowed for constant seawater flows, the next step is to provide for the precise metering of toxins into the flowing

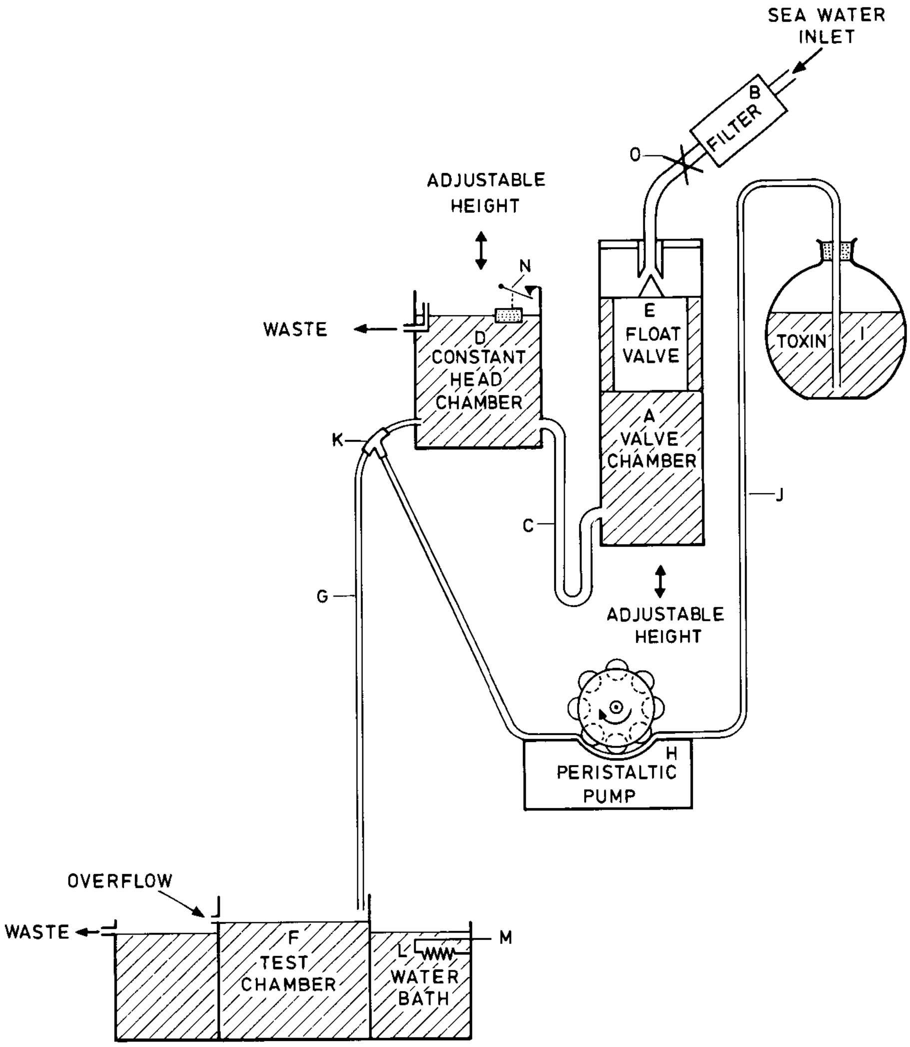

Figure 1
Diagrammatic representation of peristaltic doser.

TABLE 1
CONSTRUCTION DETAILS OF THE VALVE CHAMBER, FILTER, CONSTANT HEAD CHAMBER AND FLOAT VALVE

Reference	Component	Item	Materials and dimensions
A	Valve chamber	(i) Main body	Perspex piping (133,6 mm external diameter, 123,6 mm internal diameter) 500 mm long and capped with perspex end plates.
		(ii) Outlet pipe	Perspex piping (13 mm external diameter)
		(iii) Valve seat	Rigid PVC piping (17 mm external diameter, 9,5 mm internal diameter) machined to accept tapered valve.
B	Filter	(i) Main body	Perspex piping (70 mm external diameter, 60 mm internal diameter) 140 mm long, blanked off with a perspex cap at one end and threaded at the other end to accept a removable perspex lid with "O" ring seal.
		(ii) Inlet/outlet pipes	Perspex piping (13 mm external diameter).
D	Constant head chamber	(i) Main body	Perspex (6,4 mm) 320 x 170 x 210 mm deep.
		(ii) Inlet/overflow pipes	Perspex piping (13 mm external diameter).
E	Float valve	(i) Float	Perspex piping (100 mm external diameter) 210 mm long, blanked off with perspex sheet. 4 perspex ribs, 10 mm wide, attached vertically to the float at 90° intervals enable a close fit in the chamber while allowing the passage of water.
		(ii) Valve	Nylon cone machined to accept an "O" ring seal for mating with the valve seat.

seawater. This is achieved by means of a twelve channel peristaltic pump (H). The toxin to be tested is stored in a 6 ℓ flat bottomed flask (I). It is withdrawn from the flask though a 0,5 mm internal diameter PVC tube (J) by the pump and passed via a connector (K) (A10 platinum connector manufactured by Elkay Products Inc., Worcester, Massachusetts, USA) to mix with the seawater flowing through tube G. Stainless steel syringe needles are used to connect the pump tubes to the transmission tubing. A mixing chamber, as incorporated in many established designs, was found to be unnecessary as adequate mixing occurred during the passage of the seawater and toxin from the connector to the aquarium.

The toxin flow rate can be adjusted by altering the rotation speed of the peristaltic pump or by substituting pump tubes of different diameters. The desired concentration of toxin reaching the test chamber (F) is obtained by the interplay of three variables, namely the concentration of toxin in the toxin reservoir (I), the seawater flow rate and the toxin flow rate. Considerable flexibility is thus available for the adjustment of toxin concentrations.

The organisms to be tested are held in the test chambers (F) which, in turn, are contained in a water bath (L) constructed of epoxy coated asbestos and measuring 270 x 70 x 24 cm deep. The water bath can be maintained at a constant temperature by means of a 1 kW thermostatically controlled immersion heater (M) which is activated by a contact thermometer. Water circulation in the constant temperature bath is achieved by means of two inclined airlifts (80 cm PVC pipes of 6 cm diameter containing airstones and inclined at an angle of about 10° up the water column).

Two failsafe devices are incorporated into the system. A float switch (N) (Type 10M, manufactured by Girdlestone, Suffolk, England) cuts off the electrical supply to the peristaltic pump when the water level in the constant head chamber falls, and so prevents concentrated toxin entering the test aquaria if the seawater supply is interrupted. A solenoid valve (O) (catalogue no. 8222 B94 SA manufactured by Ascoreg Automation, Johannesburg, South Africa) which remains open when energised, cuts seawater supply in the event of an electrical power failure thus ensuring that excessive dilution of toxin in the test chambers does not take place. Both failsafe devices are self correcting in that they are automatically restored to their original status when the power or water supply is resumed.

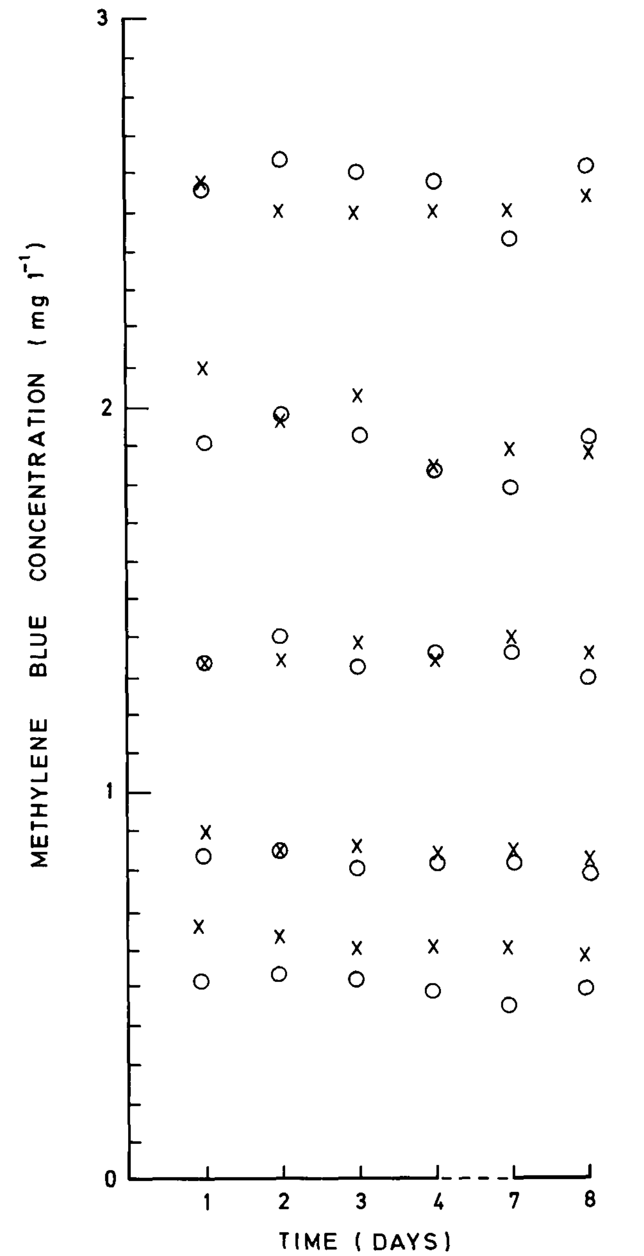

Figure 2
Variations in expected and measured concentrations of methylene blue in five aquaria using the peristaltic doser. O denotes expected result, X denotes measured result.

Operation

The peristaltic doser was used in assessing some chronic effects of cadmium and mercury on the prawn, *Penaeus indicus* (McClurg, 1984). It was found to be reliable and easily maintained.

Routine maintenance was limited to ensuring that the desired flow rates of seawater and toxin were being maintained. The seawater flow rates were checked by diverting the flows to beakers for a set period (usually 15 minutes) and measuring the volumes of seawater obtained. A statistical analysis of flow rates measured twice daily for 6 tubes over 4 days yielded coefficients of variation of 2,39; 2,26; 2,38; 1,82; 1,58 and 2,41 per cent. It was found that after about 4 days the flow rates tended to decrease due to partial blockages of the tubes with cyanobacterial slime. The slime formed on the inner walls of the tubes and, on flaking off, tended to clog the tubes and cause erratic flow decreases. Although slime formation could be permanently prevented by measures such as ultraviolet irradiation, it was found to be more practical to remove the slime by blowing compressed air through the tubes periodically or by replacing the tubes at least every four days.

The toxin flow rates were determined by diverting the pump flows to pre-weighed specimen tubes for one hour and determining the masses of the accumulated toxin. The masses were converted to volume by applying the appropriate specific gravity. Toxin flow rates were found also to be consistent. A statistical analysis of flow rates, taken twice daily over six consecutive days from tubes with nominal pumping rates of 1,8, 6,0, 9,6 and 13,8 $m\ell\,h^{-1}$, yielded coefficients of variation of 4,91, 3,62, 3,21 and 2,09 per cent for the four tubes respectively. The pump tubes were replaced every seven days, regardless of condition, to minimize the risk of reduced toxin flows arising from their physical deterioration.

Having established that reasonably consistent flows of toxin and seawater could be expected from the tube outlets, tests were carried out using the water soluble organic dye, methylene blue, to confirm that this would result in consistent toxin concentrations in the test aquaria. A concentrated solution of the dye was placed in the toxin reservoir and delivered via the peristaltic pump to mix with five identical seawater flows entering five separate aquaria. The dye flow rate to each aquarium differed through the use of pump tubes of different diameters. By taking the flow rates of seawater and dye into account it was possible to predict the dye concentrations in the aquaria. These theoretical concentrations were compared (Figure 2) with the concentrations that were measured using a Pye Unicam SP 1800 spectrophotometer at a wavelength of 663 nm. There is good agreement between the two sets of values indicating accurate and consistent dosing.

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