NOTE

New Ampoule Type Water Sampler for Microbiological Study in Aquatic Environments

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For more effective microbiological investigation of aquatic environments, the authors devised an aseptic water sampler using glass ampoules for sampling vessels. This sampler was named Ampouler TS-3.

1. Preparation of Sampling Vessels

Each glass ampoule has a capacity of 14 ml (about 12 ml is the most suitable volume for sampling). The ampoules were washed in boiling water containing detergent or immersed overnight in 1 N HCl, rinsed by tap water and distilled water, then evacuated by a handy pump (Yamato, Mini-vac) at appropriate pressures, which differed with sampling depth*.

Thus, to obtain about 12 ml sample water in each ampoule, 110 mmHg, 210 mmHg and 310-410 mmHg were usually used for

sampling at 0-5 m, 10-20 m and 20-28 m depth in Lake Kizaki, respectively. Then the ampoules were hermetically sealed after the stem of the ampoule was heated and bent rectangularly and elongated to form a capillary tube (Fig. 1). The ampoules were autoclaved before use. They could withstand water pressures of up to $12 \text{ kg} \cdot \text{cm}^{-2}$ (i. e., about 110 m depth in freshwater). Careful preparation of the glass capillary tube allowed use even at over $20 \text{ kg} \cdot \text{cm}^{-2}$

2. Construction of Sampler

Figure 2 shows a schematic representation of the Ampouler TS-3, which consists of a cylinder to hold ampoules (Part III) that is fused with a capillary cracker (Part IV), and a head cap (Part I). Each of the parts is made of stainless-steel.

The ampoule container (Part III) holds three ampoules. To fix these ampoules in the container, an adapter (also shown at the right in Fig. 2) is inserted between them in the middle after they are set inside. A head cap (Part I) is tapered in order to cushion the cracking impact by the messenger. A cylindrical stainless-steel messenger with a ringed cutter (also shown at the upper part in Fig. 2) is used to snap off the capillary tube. A bottom (Part IV) shown in Fig. 2, is used for the sampler only when sampling the lowest site in a serial sampling system; a flange (or wider plate) at the bottom of the sampler stops the messenger. Such a flange is removed from the bottom of a sampler used for upper sites in a serial sampling system.

3. Sampling Procedure

Autoclaved glass ampoules, prepared as described before, are inserted in the sam-

Fig. 1. Ampoules prepared for sampling vessels.

* Remaining pressure in the ampoule (P_0) was calculated by the following equation:

$$P_0 = (V'/V_0) (P' + 73.6 X)$$

where V' : remaining space in ampoule after aspiration of water sample (suitable volume, 2 ml)

V_0 : full ampoule volume (14 ml)

P' : atmospheric pressure in mmHg at sampling site

and X : water depth in m

Fig. 2. A schematic representation (side view) of the Ampoule TS-3 with those of an adapter and a messenger.

plers. As shown in Fig. 3, the capillaries of the three ampoules are set at an angle of 120° (Fig. 3a), and an adapter is inserted between them (Fig. 3b). After the container is capped (Fig. 3b), samplers are held at the desired water depth (cf. Fig. 3c). A cylindrical messenger (Fig. 3b) is sent to snap off the capillaries of the ampoules in the first sampler. After snapping them off, the messenger descends to the second sampler, and similarly snaps off its capillaries. The messenger then stops at the lowest sampling site. When a messenger snaps off the capillaries, evacuated glass ampoules aspirate the surrounding water in three directions in a few seconds.

Appropriate evacuation corresponding to a sampling depth (see footnote in P. 341), leaves a certain air space in the ampoules after water sampling. This air space prevents ingress of any contaminants, since when the sampler is drawn up the water pressure decreases. After retrieval, the capillaries of ampoules are resealed by heating with a handy burner.

4. Comments

Several types of water sampler have been used for microbiological study in aquatic environments (ZOBELL, 1946; RODINA, 1965; SOROKIN and KADOTA, 1972). The most simple apparatus is a glass bottle with a

Fig. 3. Handling of the Ampouler TS-3.

(a) A sampler with inserted ampoules (left) before insertion of an adapter (center) and capping of a head (right).

(b) A sampler with inserted ampoules and an adapter before being capped with a head along with the lowest capped sampler (left) and a messenger (right).

(c) Vertical holding of the Ampouler TS-3. In (b) and (c), the lowest sampler has a rubber tube ring on the bottom flange for a shock absorbance of the messenger.

removable stopper, but it usually requires two cables for suspending the apparatus and for removing the stopper. It is only useful for shallow water because the hydrostatic pressure makes it impossible to remove the stopper in deeper water.

The most commonly used type of microbiological sampling apparatus has a slightly evacuated glass bottle or a deflated rubber bulb to which a capillary tube inlet is jointed (ZOBELL, 1941). Recently, two modifications of this type of sampler were devised by YANAGITA (1978) for surface and deep water collection.

For large volume (3-5 l) sampling, NISKIN

(1962) devised a bellows-type sampler using a sterile plastic bag, which was initially deflated between two large metal hinges and inflated; it took in sample water by a messenger-triggering mechanism.

For comparatively small volume sampling, there are two types of sampler, one with syringes the other with glass ampoules for sampling vessels. JANNASCH and MADDUX (1967) developed the former type of sampler and constructed a special sampling device to prevent contamination from the sampling gear or the hydro-wire. The ampoule-type sampler devised by ISSACHENKO was cited by RODINA (1965). SOROKIN (1964) devised

the same type of sampler for serial sampling by sequential messenger releasing mechanism.

The newly devised sampler, Ampouler TS-3, rather resembles SOROKIN's sampler (the same use of evacuated glass ampoules for sampling vessels and the device of serial sampling), but has a stainless-steel cylinder in which three ampoules are inserted and only one cylindrical messenger to snap off the glass capillary.

The small volume of sampling water is compensated by three-way sampling system. Contamination from the hydro-wire is minimized by the cylindrical messenger.

The "Ampouler TS-3" has been employed in our laboratory for extensive microbiological studies on Lake Kizaki since 1976. The results of these studies were reported by TERAJ (1979), KATO and SAKAMOTO (1979, 1981), and TERAJ and YOSHIOKA (1983).

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摘 要

陸水域における従属栄養細菌群の計数、種構成の把握などの微生物学的研究のために、無菌試料採取容器として減圧封入ガラスアンフルを用いた小型採水器を考案した。

この採水器 (Ampouler TS-3) は、従来の微生物用採水器と比較して、次の3点を新しい特徴とする。

- 1) 3方向から同時に採水することにより、少量 ($3 \times 10 \sim 12$ ml) の採水で試水を代表させる。
- 2) 深度に応じたアンフルの減圧を行ない採水後アンフル内に空間を残し、試水回収の際の汚染を防ぐ。
- 3) 採水器の外側を落下するメッセンジャーがアンフルの毛管を割る単純な方法で、異なる深度の採水を連続して行なえる。

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