A SIMPLE AND INEXPENSIVE APPARATUS FOR MEASURING FISH METABOLISM

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A simple, noninvasive, and economical home-made respirometer has been used to determine the standard metabolism of goldfish. The apparatus has been tested on several fishes and has proved its accuracy in determining a mass effect on standard metabolism. The apparatus can be made easily by middle school, high school, and undergraduate students and can be used to introduce them to basic concepts in animal physiology and in biological statistics.

Key words: goldfish; standard metabolism; oxygen consumption; respirometer

In our biology program, we have adapted many physiological and biochemical experiments by giving them an ecophysiological orientation. These experiments are normally linked to wildlife management, marine biology, and aquatic biology. The proposed experiment concerns the last and is carried out in undergraduate animal physiology laboratories.

Many studies in animal physiology concern the impact of physiological, morphological, or environmental factors such as mass, temperature, or chemical pollutants on metabolic rate. Two classical studies are those of Beamish and Mookherjhii (1, 2) on the impact of mass and temperature on the standard metabolic rate of fish. In those experiments, the standard metabolic rate was estimated by oxygen consumption measurements of several individuals at rest from different species of fish. Standard metabolism (or standard metabolic rate) is the metabolic rate of an ectotherm at rest at a given temperature. To determine the standard metabolism, the animal must be calm and inactive and must not be under any form of physical, thermal, or physiological stress (6). Standard metabolism must be distinguished from basal metabolism, which applies only to endotherms, since temperature does not have the same effect on endotherms and ectotherms. Endotherms regulate their temperature with metabolic heat, whereas ectotherms can rely only on external heat sources. Consequently, their corporal temperature follows water temperature, and their metabolic rate increases with increasing temperature (5, 6). Moreover, the mass effect on standard metabolic rate differs in both groups (5).

The metabolic rate in fish is usually estimated by oxygen consumption measurements and requires specialized and onerous equipment. We propose a simple, noninvasive, and economical respirometer to determine standard metabolism in goldfish (Carassius auratus). The construction of the apparatus has been done following the principles of a tunnel respirometer as developed by Brett (3). It can easily be made by middle school, high school, or undergraduate students and used to introduce them to basic concepts in animal physiology and biological statistics. It consists of a dissolved-oxygen meter, usually found in institutions offering biology courses, and a few common items. In addition, the proposed species is inexpensive, easily obtained, demands little maintenance, and is very stress resistant. As a test for our apparatus,
some data have been taken on goldfish of varying body size, and the mass effect obtained (exponent $b$ of an allometric equation) has been compared with that from the classical experiment of Beamish and Mookherjii (1).

**MATERIAL AND METHODS**

Except for the oxygen meter, which should be found in institutions offering biology courses, all the materials (e.g., specimen cups; see Table 1) can easily be found in hardware or scientific/medical supply stores. The couplings are pipe-like pieces that can be screwed through a hole and to which tubing can be linked. The drill pump is sold in hardware stores and can be located locally through a web search. It could be replaced by any low-flow continuous pump.

**Construction of the Respirometer**

Two holes, the size of the couplings, are made in the bottom of the first specimen cup. To ensure adequate water circulation near the electrode, the holes are not allowed to face each other (Fig. 1, no. 1). Two holes (same size as above) are made in the second specimen cup, one in the bottom and the other through the lid (Fig. 1, no. 2). A last hole is made in the lid of the first specimen cup to fit the electrode (Fig. 1, no. 3). The electrode is allowed to reach the center of the cup and then is fastened with hot glue. Couplings are also fastened with hot glue to each hole (Fig. 1, no. 5). As shown in Fig. 1, no. 6, three plastic tubes (7 mm diameter) each 20 cm long are used to link the specimen cups to the pump. This kind of pump is usually activated using an electric drill, but it is not recommended here because the flow it generates would be too high. The pump can be activated by adding a handle or by simply using vise grip pliers. We used a drill pump because it is cheap and easy to find, but any low-flow continuous pump could be used. This pump will allow a gentle circulation of water, ensuring homogeneity of oxygen concentration in the whole set-up without stressing the fish. Water must circulate from cup 2 to cup 1 without passing through the pump.

**Oxygen Consumption Measurements**

**Preparation.** Seventeen goldfish ranging from 1.73 to 12.80 g were kept at room temperature and fed daily. To avoid any artifact owing to digestive metabolism, fish were starved for 24 h before physiological measurements. To facilitate filling with water, the system was immersed with all lids open in a basin of well oxygenated water at room temperature (20°C). Air was then evacuated by activating

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**Table 1**

List of materials and construction cost ($Can)

<table>
<thead>
<tr>
<th>Item</th>
<th>Number</th>
<th>Cost ($Can)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oxygen meter</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Transparent plastic specimen cups, 100 ml</td>
<td>2</td>
<td>20.00/100</td>
</tr>
<tr>
<td>Brass (or plastic) couplings</td>
<td>4</td>
<td>10.00</td>
</tr>
<tr>
<td>Flexible “Nalgen” plastic tubing, 20 cm × 7 mm</td>
<td>3</td>
<td>2.00</td>
</tr>
<tr>
<td>Drill pump (or any low-flow continuous pump)</td>
<td>1</td>
<td>15.00</td>
</tr>
<tr>
<td>Hot glue dispenser</td>
<td>1</td>
<td>0.05</td>
</tr>
<tr>
<td>Hot glue stick</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Basic tools</td>
<td></td>
<td>Total (approx.) 28.00</td>
</tr>
</tbody>
</table>
the pump. The lid holding the electrode was then carefully put in place without trapping air, and the oxygen meter was calibrated to 100% O₂ saturation. This condition was obtained by bubbling with air into the water.

**Measurements.** Before measurements, fish were kept in pet shop plastic bags. To avoid stress, which increases metabolism, each fish was transferred quickly and gently by pouring the plastic bag into the aforementioned basin. The fish was then caught with a landing net and released in the respiration chamber (cup 2) without taking it out of the water. The lid was quickly closed and the apparatus kept immersed for a few minutes for the fish to acclimate to the respiration chamber. The apparatus was then pulled out of the water and dried with paper towels to detect leaks before oxygen consumption measurements. Measurements were taken at 2-min intervals for a total duration of 10 min. Before the recording of each datum, the pump was activated by continuous motion for 1 min (a rate of one-half turn of the handle per second). This allowed the standardization of the oxygen concentration throughout the respirometer without stressing the fish. The pump could be activated continuously, but it is not necessary, because a complete turn-over of the water is made by 1 min of pumping. Some stress may come from the transfer of the fish to the respirometer and can be reduced by using an automatic pump and leaving the fish overnight for a period of rest at low-current speed. But because this kind of pump is not always available and pumping by hand overnight is not possible in the usual duration of a physiology course, the experimenter must keep in mind the possibility of an artifact caused by stress. For this reason, the mass of the fish was measured after the respiration session.

**RESULTS AND DISCUSSION**

Mass-specific metabolism usually decreases with an augmentation of mass. Consequently, metabolism (not mass-specific metabolism) follows an allometric equation of the type \( Y = a \cdot M^b \) (Fig. 2): where \( Y \) is the metabolic rate, \( M \) is the mass, \( a \) is a constant, and \( b \) is always <1 (5, 6). A logarithmic transformation of this relation gives a linear model with a slope of \( b \), as shown by our data (Fig. 2). The regression line obtained in our experiment is significant (\( P < 0.000; r^2 = 0.7867 \)) and has a slope of 0.910 mg O₂ h⁻¹ g⁻¹. A \( t \) test for independent samples \( [t_{0.05;28}] \) has been used to compare our slope with the 0.913 mg O₂ h⁻¹ g⁻¹ obtained by Beamish and Mookherjii (1) at a similar temperature (~20°C). No significant difference has been found between the two methods, meaning that our apparatus can be used to detect a mass effect with a precision comparable with that of more sophisticated equipment.

In biology teaching, this set-up can be used for a great variety of applications. We used fish of different sizes to show a potential mass effect. This demonstration can thus be used to introduce students to the concept of allometry as well as to basic statistics in biology. For statistical convenience, we suggest pooling the results of many students in a course. Although mass is a factor that is easy to study, the principle of pooling results can be used to show the effect of many other factors. For example, measurements can be made with varying temperature or with some pollutant concentration. Measurements can also be made before and after the fish are fed, allowing the teacher to demonstrate the so-called specific dynamic action, which is the metabolic rate augmentation owing to digestive activity (4). Evidently, the experiment can be made with different fish species as well as different taxonomic groups (e.g., crustaceans, mollusks).
The respirometer we propose herein has shown an efficiency for the determination of oxygen consumption that is quite comparable with that of more sophisticated equipment. It has the advantage of being inexpensive, easy to make, and user friendly.

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