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WATER FLOW DYNAMICS IN THE RESPIRATORY TRACT OF THE CARP (*CYPRINUS CARPIO* L.)

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SUMMARY

Simultaneous measurements of water velocity in the buccal chamber, and buccal and opercular hydrostatic pressure of carp have revealed surprisingly high water velocities. The high flow velocities mean that, at times, the kinetic energy of flow makes a substantial contribution to the total fluid energy. This suggests that there may be unequal distribution of hydrostatic pressures within the buccal chamber. Anatomical examinations showed that fluid channels in the buccal chamber and gill raker sieve are complex and can be expected to vary spatially and temporally throughout the respiratory cycle. It appears that there is a potential for error in many of the previous analyses of 'gill resistance' and energetics of fish breathing based solely on hydrostatic pressure measurements and the simplifying assumption of steady-state conditions.

INTRODUCTION

The breathing apparatus of teleost fishes consists of a buccal force pump separated from paired opercular suction pumps by a continuous gill curtain. The concerted action of these pumps serves to drive a nearly continuous flow of water over the gills as was clearly demonstrated by van Dam (1938) using a glass external shunt connecting the buccal and opercular cavities of a trout. Since flow in the shunt was continuous throughout a breathing cycle he concluded that it must be flowing likewise over the gills. Since van Dam (1938), a large number of studies have been made on the pressure relationships between buccal and opercular pumps during breathing in a variety of fish (see Shelton, 1970, for review), and a consistent finding has been that a fluctuating, but predominantly positive, hydrostatic pressure gradient exists from the buccal to the opercular cavities. However, we know of only one study which has been directed at recording the water velocities which are driven by these pressure changes. Hughes (1975), working on trout, reported: 'we have used a Doppler meter for monitoring the velocity of particles suspended in the water as they pass through the respiratory system and have observed some marked variations in velocity'.

The objectives of the present study were to examine flow velocity at various depths within the buccal cavity of a stationary fish to determine the nature of flow within the buccal activity. These measurements were coupled with anatomical examinations of the buccal chamber which were intended to determine the importance of the geometry

of this chamber on flow characteristics. Pressures within the buccal and opercular chambers were recorded simultaneously with flow velocity to assess the applicability of simple dynamic models to the fish respiratory tract. Finally the effects of hypoxia on pressure-flow relationships were examined.

MATERIALS AND METHODS

Three carp, *Cyprinus carpio* L., weighing 3–3·5 kg, were obtained from a supplier, who netted them in the lower Fraser River drainage system near Vancouver, B.C., Canada. The fish were held in a 200 l container through which was passed a continuous stream of aerated water at a temperature of 19–21 °C. The fish were held for a week without feeding before being used for any experiments. All three fish sustained some superficial abrasions and loss of a few scales as a consequence of capture and handling. These injuries were subsequently attacked by fungus which was eliminated by twice daily administrations of malachite green at concentration of one part by weight in 200 000 parts by weight of water, a level of dosage recommended by van Duijn (1967). The malachite green was washed out of the holding container by running water within 45–60 min of each administration.

Cannulae for hydrostatic pressure measurements consisting of 30 cm lengths of polyethylene tubing (PE 200, 1·4 mm I.D., 1·9 mm O.D., Clay-Adams Inc., New York), heat flared at the implanted end, were placed in the buccal and right opercular chamber of the fish using a method similar to that outlined by Saunders (1961). The openings of the cannulae were at right angles to the axis of water flow. The fish were anaesthetized in a solution of 1:10000 w/vol. MS-222 while the cannulae were implanted. The fish were permitted several days to recover from the implantation procedures.

For experimentation the fish were placed in a container which had provisions for holding them firmly in a head and body clamp, much as in the manner of the apparatus used by Hughes & Roberts (1970). Micromanipulators (W. R. Prior and Co., Hertfordshire, England) were mounted on the apparatus in such a way that instruments could be held and positioned with precision, relative to the head of the fish. The water in the apparatus was continuously aerated and remained at room temperature (19–22 °C). The fish could be rendered hypoxic by bubbling nitrogen gas into the aerating device. Water Po_2 was measured with a polarographic oxygen electrode (Radiometer, Copenhagen) calibrated with humid air and nitrogen.

Pressure fluctuations in the buccal and opercular chambers were measured using two Hewlett–Packard 267b differential pressure transducers. The transducer outputs were amplified and processed using Hewlett–Packard 350-1100 CM carrier preamplifiers, and the results displayed on a Hewlett–Packard 4 channel rectilinear thermal pen recorder. The transducers were calibrated using fluid-filled manometers as pressure references. As the pressures to be measured were very low, it was important that the zero point be accurately located. To avoid errors caused by slight fluctuations in the water level in the apparatus, one active side of each differential pressure transducer was connected to a water filled reference tube, the open end of which was placed below the surface of the water in the apparatus. Thus any fluctuations in water level in the apparatus were automatically compensated for.

Water velocities were measured using electromagnetic, blood flow transducers (Biotronix Laboratory Inc., Maryland, USA) mounted on slender (1 mm diam.) spring steel rods. The transducers had a maximum diameter of 7 mm. The output from the transducer was passed to a Biotronix BL 610 pulsed logic electromagnetic flowmeter set to a frequency response of 50 Hz. The sensitivity of the electromagnetic flow transducers in Vancouver city water was poor owing to a very low ionic content of the water. This problem was remedied by adding NaCl to the water (3/g l) in the experimental apparatus, which permitted keeping the carp in water which was still hypotonic to their blood.

The velocity probes were calibrated by winching them through a trough of stationary water (taken from the apparatus used for testing the fish) at various velocities in the range encountered within the buccal chambers of the fish. The output versus velocity relationships were linear, and judged sufficiently accurate for the purposes of this study. The probes were calibrated each time the water in the apparatus was changed to avoid any errors due to changes in the ionic strength (conductivity) of the water. The calibration curves always registered a zero response at velocities below 1 cm s^{-1}, and this we believe is a consequence of progressively larger boundary layers of relatively slower moving water which form at lower velocities and which ultimately prevent any significant flow through the minute lumen of the probe. At higher velocities the greater shear forces can be expected greatly to reduce the size and effect of the boundary layers, and thus permit velocity proportional flow through the lumen of the probe.

An experiment was begun by clamping the fish in the holding container and permitting it to recover from the immediate effects of handling over a period of 4–6 h. When the fish had recovered sufficiently that the respiratory pressure fluctuations had remained at a more or less constant level for at least an hour, measurements were taken of the pressure fluctuations in the buccal and opercular chambers. Water velocity measurements were taken at positions immediately outside the mouth, and opercular chambers. Then, at 5 min intervals, the anterior velocity probe was advanced into the fish's mouth in 1 cm increments. It was found that the fish were irritated by the probe at certain depths of insertion. The most serious, in that it caused the fish to struggle, occurred when the probe was placed 1 cm into the buccal chamber, a position which was unique in that it interfered with the closure of the buccal valves. Insertion of the probe 6–8 cm into the mouth also caused some irritation, which was manifested as series of coughing actions for the first few breathing cycles after any movement of the probe. Although this latter irritation effect quickly died away, suggesting a degree of accommodation on the part of the fish, the struggling constituted a serious problem, as it affected the intensity of breathing, generally causing a slight increase in rate, and considerable increase in amplitude of pressures generated. The addition of 1:40000 w/vol. of MS-222 to water prevented the irritation effects without noticeably changing the breathing pressure waveforms. Accordingly, all pressure and velocity measurements were carried out with this analgesic level of MS-222 in the water.

It was initially intended that each pass of the longitudinal flow sensor into the mouth would be followed with another sensor which could detect lateral and vertical currents. We were concerned that, deep in the buccal chamber, water must exit via

the gill slits, and that the major velocity components would be laterally directed. This approach was abandoned after passes into the mouth of the first fish using a lateral flow sensor showed lateral currents to be negligible, except in the deeper regions of the buccal chamber. Even in these deeper regions, from 7 to 9 cm in, lateral velocities were too low to measure accurately with our equipment, generally being less than 1 to 2 cm s^{-1} at all phases of the breathing cycle.

All of the velocities reported in this study refer to velocities along a line parallel to the median longitudinal axis of the buccal chamber. It is quite likely that the velocity profiles will vary in different parts of the buccal chamber away from the mid-line. Consequently great care was taken with the alignment of the velocity probes in order to permit their introduction into the buccal cavity along an axis which would not impinge upon the walls of the cavity, nor interfere with the operation of the buccal valves, save when the probe itself was between the valves. The membranous buccal valves of the carp were sufficiently large as to easily accommodate the spring steel rods and transducer leads and still produce an effective seal. In quiet breathing it was observed that the lips of the carp did not come in contact, the fish relying instead upon the buccal valves to complete the sealing of the buccal chamber during the mouth closure phase of the breathing cycle.

It was not possible to measure velocities within the opercular chambers owing to the bulk of the velocity probes. However, in most experiments, a second velocity probe was placed very near the right opercular opening to permit monitoring of the timing and velocity of water ejected from the opercular chamber.

Once a series of two or more passes into the mouth of the carp with the velocity probe were completed, along with accompanying determinations of pressures and exit velocity, the fish were rendered sufficiently hypoxic that their breathing efforts increased, both in amplitude and frequency. The hypoxic conditions were produced by bubbling nitrogen gas into the water until the oxygen levels were below one third that of air saturation. A pass was then made into the buccal chamber with the velocity probe and recordings of pressures and velocities were made.

After representative recordings had been obtained, the fish were killed and dissected to permit anatomical and dimensional measurements of the respiratory tract. The basic anatomy of the buccal chamber was examined by freezing the freshly killed fish and then sectioning them in a median sagittal plane. The cross-sectional area of the left hand side gill raker sieve of one of the fish was determined by applying and trimming a piece of waterproof paper until a piece was obtained which just covered the sieve. This process was carried out with the sieve in what was judged to be a normally closed and normally open position.

RESULTS

(*a*) *Anatomical examination of the buccal chamber*

The morphology of the buccal chamber is partially summarized in Fig. 1. The drawings are based upon the relaxed configuration assumed by the sectioned fish after they had thawed, and minor differences can be expected in the intact fish. The mouth, when opened, approximates to a flattened oval (section *A–A*, Fig. 1) which can be closed by the lower jaw and lesser movements of the upper lip and

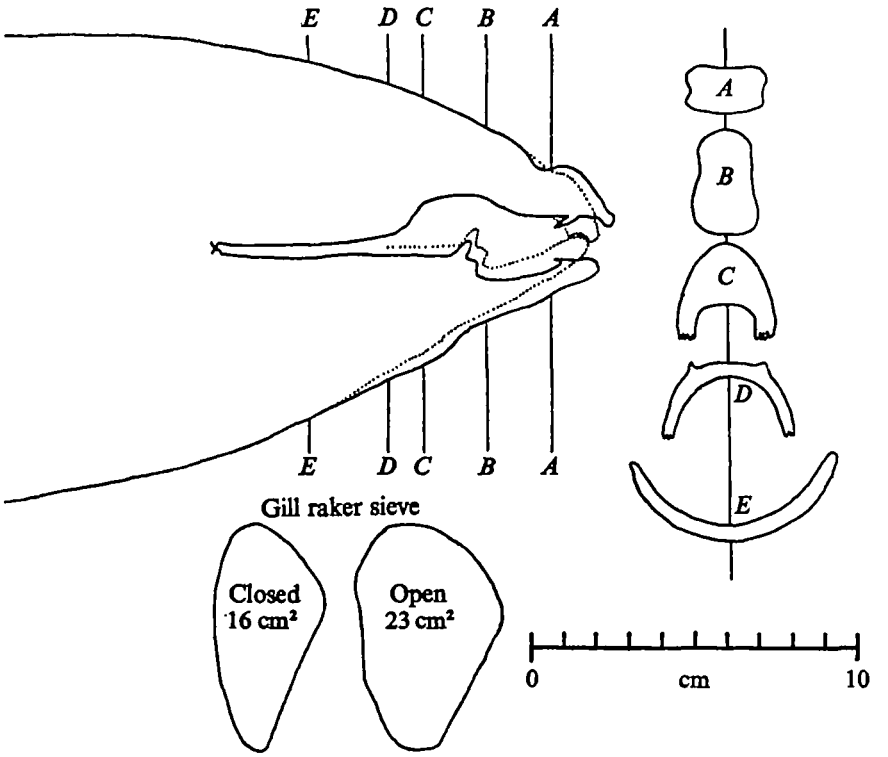

Fig. 1. A median sagittal view of the head and buccal chamber of a 56 cm long, 3·2 kg carp used in this study. Dotted lines indicate the closed configuration of the mouth. *A*, *B*, *C*, *D*, and *E* at right indicate the shape of the relaxed buccal chamber in transverse section at points shown by correspondingly lettered lines on the sagittal view. The extent of the left gill raker sieve in open and closed configuration is shown below.

premaxillary bones. Velar folds of soft tissue in the dorsal and ventral jaws, which are passively activated by water moving out of the mouth, effectively seal the buccal chamber, even when the lips are 2–4 mm apart. There is a large anterior chamber (section *B–B*, Fig. 1), partially filled by the tongue in the posterior regions (section *C–C*, Fig. 1) which does not appear to be capable of reduction to zero volume, in contrast to the more posterior regions of the buccal chamber. The large tongue which bears a firm fleshy papilla on its median antero-dorsal surface, fills the central portion of the chamber converting the passage to an inverted 'U' shape (section *C–C*, Fig. 1). The openings of the gill slits off the ventral regions of the chamber start just after the section *C–C* (Fig. 1) and are present by section *D–D* (Fig. 1). At section *D–D* (Fig. 1) the buccal chamber has become narrow and flattened. The gill arches and their slits sweep outwards and upwards dorsolaterally and are angled posteriorly. The gill rakers guarding the gill slits are well developed and interdigitate to form a sieve which covers most of the venterolateral surface of the buccal chamber by section *E–E* (Fig. 1). The gill raker sieve extends posteriorly to terminate at a point level with the oesophageal entrance. The entire floor, the lateral walls, and the extreme anterior portions of the roof of the buccal chamber are all mobile to varying degrees and can be expected to move during normal breathing.

The passage of water from the buccal chamber to the gills via the gill slits entails filtering through the elaborate interdigitating gill raker sieve. The gill raker sieve can be expected to change dimensions during a breathing cycle, and the approximate range of dimensional changes which can be expected in the carp examined are shown in Fig. 1. The area of the gill raker sieve when the gill arches were held closed, compared to the area when the gill arches were held apart, much as they would be during deep respiratory movements, varied by *ca.* 40% (Fig. 1). It was possible to extend the area of the gill raker sieve by approximately another 20%, but such an additional extension was not judged to be likely to occur normally. When the sieve was in the closed position the water channels between the gill rakers are very fine and much restricted, and make up only a small proportion of the total sieve area. Thus the 40% increase in total sieve area in the open configuration (Fig. 1) represents a tremendous proportional increase in the total area of water channel. It appears quite possible that the gill raker sieve of this species could present a significant resistance to the flow of water during certain portions of the breathing cycle.

(*b*) *Velocity measurements in the buccal chamber*

The results presented are based upon analysis of measurements taken during 16 experimental passes during normoxia and five experimental passes during hypoxia made upon the three fish. Choice of recordings presented for illustration was based upon clarity of recordings and completeness of the series. The particular breathing cycles shown were chosen on the basis of finding a cycle which started at a similar point on the recording chart.

The shape of the velocity pulse was found to be highly dependent on depth within the buccal chamber (Fig. 2). Just outside the entrance of the mouth the water flow was pulsatile, and water movement occurred for only approximately half of the breathing cycle. At this location there was a small reversal in flow associated with closure of the buccal valves. At a depth of 0·6 cm the probe interfered with the buccal valves and much higher velocities were recorded (Fig. 2). However, when in this position there was a slight change in the pressure profiles recorded from the buccal cavity whereas with the probe in all other positions no detectable change in either buccal or opercular pressure recordings occurred compared with those existing before introduction of the probe. Further into the bucccal chamber, at positions from 2 to 8 cm in, there was a pronounced trend for water velocities to become slower, and more continuous, throughout the respiratory cycle (Fig. 2). Velocity patterns in the anterior regions of the buccal chamber were consistent between experiments and between fish, but there was considerable variation in the velocity patterns in the posterior extremities of the buccal chamber. The variation occurred mainly between individual fish, although it occurred to a lesser extent between experiments on the same fish. The recordings presented in Fig. 2 for normoxic breathing are representative of some of the higher velocities observed, with values about half those also being observed. However, in all cases the patterns were essentially similar, and their analysis leads to the same conclusions as those of the experiment shown in Fig. 2.

Reducing the water Po_2 to 30–45 mmHg was sufficient to stimulate an increase in rate and amplitude of the fish's breathing movements. The effects of hypox

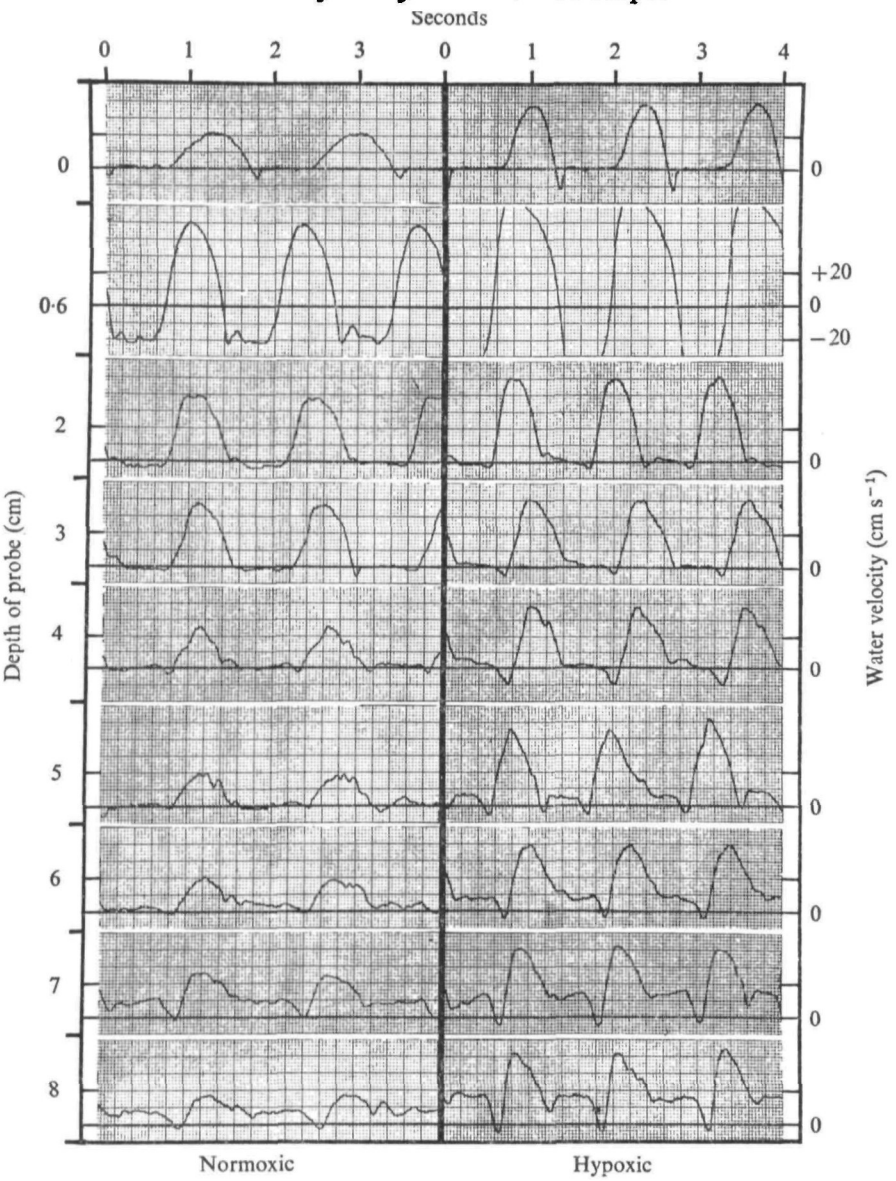

Fig. 2. Water velocity recordings at various depths within the buccal chamber of a carp taken during normoxia and mild hypoxia. The recordings are not simultaneous, but have been selected so that they correspond with each other temporally. Positive deflexions indicate water movement in a posterior direction. Velocity calibration is the same in all cases. The zero depth recordings were taken immediately outside the mouth. Note the conspicuous backflow of water at the 0·6 cm depth which is caused by the velocity probe interfering with closure of the buccal valves.

upon the velocities observed in the buccal chamber are shown on the right side of Fig. 2. Just outside the mouth there was a 90% elevation in the velocity maxima associated with hypoxia. There was a much smaller increase of approximately 30% in the 2–4 cm depth region of the buccal chamber. The effect of hypoxia on flow profiles was much more pronounced in the more posterior regions, where the velocity maxima

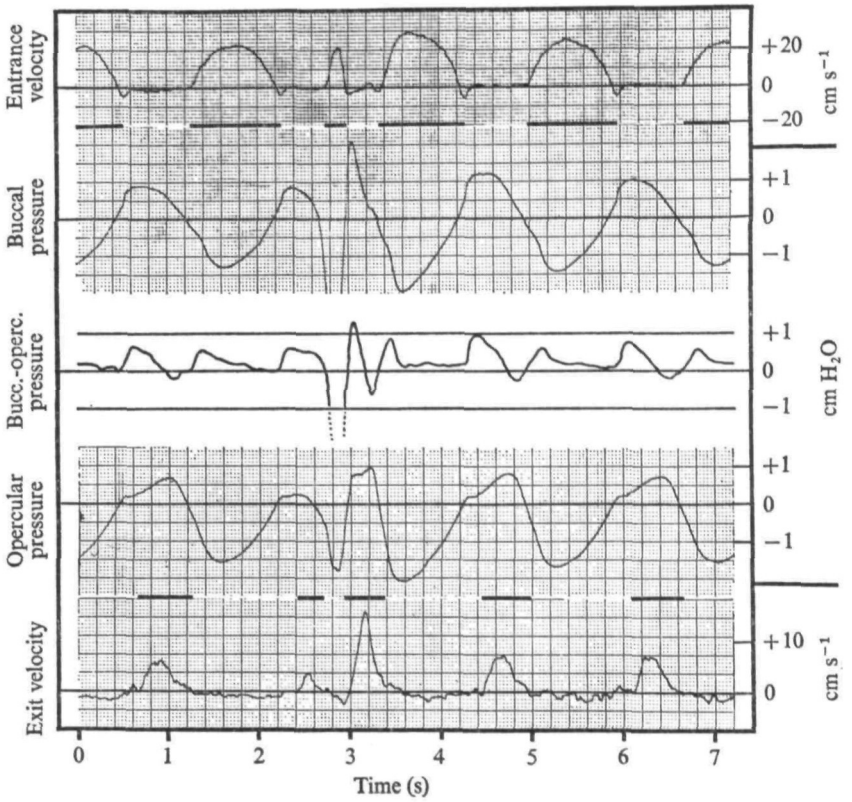

Fig. 3. Simultaneous recording of hydrostatic pressures in the buccal and opercular chambers, and water velocity at the buccal entrance and opercular exit of a carp. The centre trace indicates derived buccal − opercular pressure. A 'cough' occurs between 2·5 and 3·5 s. Calibration of pressures is uniform for the three centre traces, but note differing scales of buccal entrance and opercular exit velocities. Periods when buccal and opercular valves are open is indicated by heavy black bars (accurate to ±0·04 s) near appropriate traces.

increased by as much as 160% by the 8 cm depth (Fig. 2). There was still a brief period of reverse flow in the deep regions during heavy breathing. The variability of the velocity profiles from fish to fish and from experiment to experiment was much less with heavy breathing than was observed with light breathing.

(*c*) *Correlation of entrance and exit velocities with hydrostatic pressures*

A comparison of the velocity of water entering and leaving the respiratory tract, with a buccal pressure, opercular pressure, and buccal−opercular pressure gradient is presented in Fig. 3. This figure also includes a 'cough', a response of the fish to irritation within the pharynx. Coughs were also performed spontaneously on occasion by all the fish tested. The cough shown in Fig. 3 is a relatively mild one compared to many, and was chosen because the pressure traces largely remained on the scale. The frequency of coughing varied widely, from about every tenth breath, to one in several hundred breathing cycles.

During regular breathing cycles the movement of water into the mouth occurred in close association with the negative phase of the buccal and opercular pressure cycles

and diminished when the buccal pressure began to rise. During backflow there was a small jump in the buccal pressure trace associated with the closure of the buccal valves. Virtually all of the positive phase of the buccal pressure cycle was associated with the zero velocity phase of the inspiratory water velocity trace (Fig. 3).

The expulsion of water from the opercular chamber was nearly coincident with the positive pressure phase of both the opercular and buccal chambers. The negative phase of the opercular pressure cycle was associated with a very low anterior directed velocities (Fig. 3) which coincided with a slight anteriorly directed movement of the opercular valve, a velar flap of tissue on the posterior margin of the operculum, which occurred during expansion of the opercular chambers. These anteriorly directed water currents were probably caused by the forward motion of the opercular valve which maintains an effective seal on the opercular chamber during this phase of the breathing cycle.

The intake velocity of water entering the mouth showed a good correlation with the magnitude of negative pressure in both the buccal and opercular chambers (Fig. 3). The expulsion of water from the opercular slits occurred at much lower velocities than those associated with inspiration which is indicative of the larger cross-section of the opercular slits. The opercular expulsion velocity tended to lag behind the rise of pressure in the buccal and opercular chambers, but otherwise the pressure–velocity relationship was quite close during the expulsion phase of the cycle (Fig. 3). The buccal – opercular pressure was highly variable but predominantly positive with a short period of negative pressure associated with the last stages of opercular expulsion. This period was completed before movement of water into the mouth commenced. The magnitude of the buccal – opercular pressure, with the exception of the cough, was never greater than + 1 cm of H_2O over the breathing cycle (Fig. 3).

The 'cough' in Fig. 3 shows it to consist of an accelerated breathing cycle with an emphasized expulsion of water from the opercular slits indicated by the high peak of the exit velocity trace. The subsequent reduction in the contained volume of the respiratory tract is illustrated be the pressure traces for buccal and opercular chambers, which show a compensatory increase in the negative pressure phase immediately following the cough (Fig. 3). With the cough there was a considerable alteration in the relationship between pressures and velocities observed. In the cough shown in Fig. 3, changes in pressure in the buccal chamber were much greater than those associated with normal breathing, and yet the velocity of water entering the mouth had a lower maximum than that encountered normally. The sudden and large drop in pressure in the buccal chamber coincided with a faster acceleration of water entering the mouth than that associated with a normal breathing cycle. Undoubtedly inertial effects could limit maximum entrance velocity during the brief period of negative buccal pressure but it is likely that an increase in inflow resistance is also being observed (i.e. the mouth is only opening slightly). On the other hand expulsion from the opercular cavity during the cough was characterized by short duration and nearly normal ejection pressure yet greatly increased outflow velocities, clearly opercular outflow resistance fell markedly.

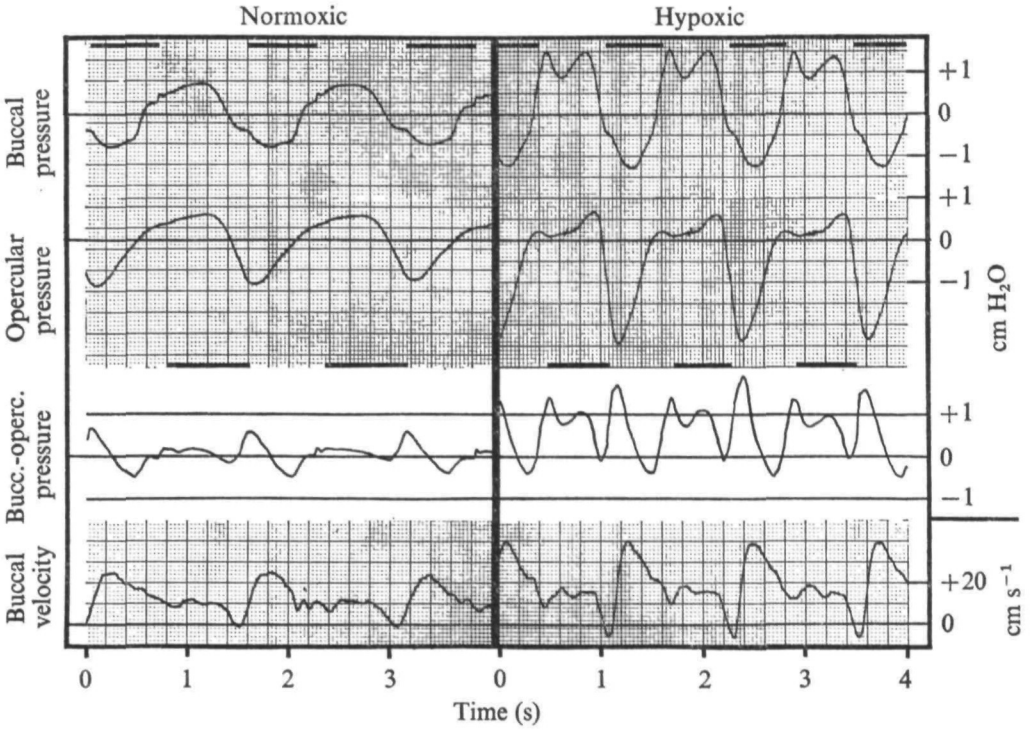

Fig. 4. Simultaneous recording of hydrostatic pressure in the anterior buccal and opercular chambers, and water velocity at a depth of 8 cm into the buccal chamber of a carp, during normoxia and mild hypoxia. Derived buccal − opercular hydrostatic pressure differential is also included. The position of the velocity sensor is approximately mid-way between the buccal and mid-opercular openings. Period when buccal and opercular valves are open is indicated by black bars near appropriate recordings. (Timing accuracy: ±0·04 s.)

(*d*) *Correlation of deep buccal velocity with hydrostatic pressures*

A comparison of the cyclic velocity profiles (obtained at a depth of 8 cm into the buccal chamber) with hydrostatic pressure fluctuations within the buccal and opercular chambers, as well as the pressure differential between these two sites, is shown in Fig. 4. A depth of 8 cm was chosen as a basis of comparison as it was very nearly mid-way between the pressure measuring sites. It was also about the closest practicable approach to the gills that could be made with the techniques used. A similar comparison for more forceful breathing occurring during hypoxia is also shown in Fig. 4.

It can be seen that the buccal − opercular pressure record shown in Fig. 4 resembles the same type of record shown in Fig. 3. During normoxia a velocity peak in deep buccal water flow was associated with minimum buccal pressure, but lagged considerably behind the buccal − opercular pressure peak (Fig. 4). Other corresponding features of the buccal − opercular pressure and deep buccal flow show a similar lag and consequently if deep buccal flow is a good estimate of gill flow, then inertial effects must be of considerable importance in determining dynamics of flow through the gills. Inertial effects may be, in part, responsible for other alterations in dynamics of flow across the gills. The more pronounced differential pressure reversal, which occurred just after peak flow, generated velocities of approximately one half of

maximum whereas the lesser reversal at the end of the respiratory cycle halts or generates small but distinct back flow (Fig. 4). Apparently the earlier reversed pressure gradient is incapable of totally decelerating the rapidly moving water during this phase of respiration.

The response to hypoxia was largely a magnification of all recorded profiles as the amount of water breathed increased, although the positive pressure phase in both chambers was characterized by a double peak. The buccal – opercular pressure was greatly increased at most phases of the breathing cycle. There was a marked decline in positive pressures in the opercular cavity. Since the flow of respiratory water is much greater during hypoxia, the reduction in opercular pressures suggest a drop in opercular outflow resistance as was observed during the cough reflex. Such a drop in opercular outflow resistance would be expected since it was confirmed by visual observation that the gape of the opercular slits was much greater during the expulsion phase of hypoxic breathing.

DISCUSSION

The analysis of the breathing mechanics of fish is difficult because it involves the measurement of the unsteady flow of a dense fluid through a non-uniform system which is ill defined. The compliance of the respiratory tract is variable, both spatially and temporally, and certain resistive elements (such as the gill filaments) are mobile, both passively and actively, throughout a breathing cycle (Pasztor & Kleerekoper, 1962; Saunders, 1961). In view of this, an analysis of the hydrodynamics of breathing in fishes must involve not only pressure recordings but also measurements of water flow profiles and the associated dimensional changes of the respiratory tract. Unfortunately, the present study provides little information on these dimensional changes except that they are potentially important.

Although the intake of water is in phase with the negative phases of both buccal and opercular pressures and expulsion with positive phases of these pressures there is no doubt that deep buccal flow velocities do not correlate well with the 'differential' pressure across the gills. The considerable lag between differential pressure and peak flow velocity implies that inertial effects cannot be neglected as they have been in determinations of 'gill resistance' from measurement of area mean differential pressure and volume flow rate (Alexander, 1967; Davis & Randall, 1973; Hughes, 1966; Hughes & Saunders, 1970; Muir & Buckley, 1967). Furthermore, in view of the mobility of virtually all of the structures in the ventral and lateral regions of the respiratory tract, the assumption of constant fluid resistance, required for simple averaging of the differential pressure waveforms, is not soundly based.

The measurements of water velocity within the buccal cavity suggest that respiratory water flow becomes progressively less pulsatile at deeper sites within the cavity during quiet breathing. Undoubtedly a portion of buccal inspiratory flow will be stored in the chamber itself which can lead to a reduction in flow velocity with depth (assuming uniform cross-sectional area of the buccal chamber from anterior to posterior), while between inspirations there will be an increase in flow velocity with depth as a larger portion of buccal compression progressively contributes to flow. Even so, the effectiveness of depulsation is probably less than recordings suggest, for water velocity and not volume flow rate is recorded and no accounting is taken of the simultaneous

changes in the dimensions of the system. For example, deep in the buccal chamber where the depulsation appears greatest (Fig. 2), noticeable peaks in the velocity pattern occur which coincide with a phase of the breathing cycle when the buccal chamber may be nearly maximally expanded. In this case the period of maximum velocities and maximum cross-sectional area for flow coincide, which means that a proportionally greater peak volume flow must be occurring during inspiration than the velocity recordings alone would suggest.

The present data also reveal that the contribution made by kinetic energy to the total fluid energy in large fish can be significant. In this study normoxic water velocity maxima reached approximately 38 cm s^{-1} at a depth of 2 cm into the buccal chamber as compared with maximum velocities of 16 cm s^{-1} at an equivalent phase of the breathing cycle at a depth of 8 cm. Since total fluid energy consists of the sum of both pressure and kinetic energies, then at 38 cm s^{-1} the kinetic energy contribution to the total fluid energy is 0·74 cm H_2O. At 16 cm s^{-1} the kinetic energy contribution is equivalent to a manometric pressure of 0·13 cm H_2O. A regional velocity head difference of the magnitude of the example just outlined suggests very strongly that hydrostatic pressures are not uniform throughout the buccal chamber and further that kinetic energy effects can be quite marked, equalling in some cases the pressure energy. This emphasizes the need to investigate whether hydrostatic pressures are virtually uniform throughout the buccal and opercular chambers, since an assumption of spatial pressure uniformity and negligible flow resistance within the buccal and opercular chambers is inherent in virtually all previous studies which attempt to examine 'gill resistance' on the basis of fluctuating hydrostatic pressure measurements in the buccal and opercular chambers of stationary fish (Ballintijin, 1972; Davis & Randall, 1973; Hughes & Saunders, 1970; Muir & Buckley, 1967; Shelton, 1970).

To reduce the complexity of fluctuating pressure versus time waveforms some workers have used steady-state approximations (Alexander, 1967; Davis & Randall, 1973; Hughes, 1966; Hughes & Saunders, 1970; Jones & Schwarzfeld, 1973; Muir & Buckley, 1967). These approximations can lead to a number of errors for, as pointed out above, kinetic energy effects alone can be quite significant. However, most of the data which has been utilized in steady-state approximations has been obtained from fish which are about one tenth of the weight of the carp used in this study. It may be that the maximum flow velocities encountered would be less than those attained by the large carp, in which case the kinetic energy effects would be less significant. Other errors in this approach arise from the fact that the energy costs of generating pulsatile flow will undoubtedly exceed those of pumping a given volume of water at a constant rate (Ballintijn, 1972) and from disregarding dimensional changes. The pressure–flow relationship of a viscous fluid through a vessel is extremely sensitive to the cross-sectional area of the vessel. In the case of fish both the pressures generated and the dimensions of the respiratory tract are continuously fluctuating Therefore, in terms of potential for water movement, there is a vast difference in a situation where the pressure maxima coincide with dimensional or cross-sectional area maxima, as compared with the case where pressure maxima coincide with dimensional minima. Obviously, any adequate analysis of the hydrodynamics of fish ventilation can only be made when the data on water velocities and cross-sectional areas are obtained in addition to hydrostatic pressures.

It is of interest that the nature of the spontaneous coughs of carp in the present study were reversed relative to the coughs described by Hughes & Morgan (1973). Basically, the coughs of the carp in the present study (Fig. 3) consisted of an accelerated, and forceful, breathing cycle with pressure waveforms which appear similar to those described by Ballintijn (1969) for the same species, and which occur if opercular abduction is slight. Examination of the water velocity records during a cough (Fig. 3) reveal a smaller than usual in take of water into the mouth, with no back flow, followed by a much faster exit of water from the opercular slits. These records suggest that the motions performed by the fish would serve to send a forceful pulse of water through the respiratory tract in a normal direction, and that this pulse may be preceded by a short, but abrupt, reversal of flow over the gills. Such an action could be effective in clearing the gills of any accumulated detritus or mucus. Furthermore, if the gill filament abductor muscles were to contract in the early phase of the cough cycle, then the gill cleaning function would be greatly enhanced. This is because such an action would emphasize the functional effect of any momentary backflow of water over the gills carrying free any materials which had accumulated on the upstream surface of the gills. The completion of abduction of the gill filaments would then swing them clear from the subsequent posteriorly directed water currents, thus permitting free passage of any particulate matter past the gills and out through the opercular openings.

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