J. exp. Biol. (1977), 68, 109–122
With 8 figures
Printed in Great Britain

VARIATIONS IN THE PATTERN OF COUGHING IN RAINBOW TROUT

BY G. M. HUGHES AND R. J. ADENEY*
Research Unit for Comparative Animal Respiration, University of Bristol, Woodland Road, Bristol

(*Received* 8 *December* 1976)

SUMMARY

Recordings of pressure changes within the buccal and opercular cavities of lightly anaesthetized rainbow trout were made simultaneously with movement recordings of the lower jaw and operculum. Coughing movements during the normal rhythm were analysed and in addition coughing cycles evoked by the introduction of dyes into the mouth.

The different patterns recorded could be generally grouped into forward and backward types but a wide range was found. Information regarding the direction of water flow was obtained from analyses of the differential pressure waveforms and this also showed a range in different coughs. It is concluded that many variations of coughing grade into one another and the precise movements producing them and the afferent signals eliciting them remain to be investigated.

INTRODUCTION

The existence of respiratory manoeuvres other than those concerned with ventilation of the gills has been known for many years. One of the earliest detailed investigations was carried out by Kuiper (1907) and he compared his observations on teleosts with the earlier studies of van Rynberk (1905) on the expulsion reflexes of elasmobranchs. He introduced the term 'coughing' and this terminology has been followed in a number of more recent studies, using modern pressure and electromyographic recordings (Hughes & Shelton, 1958; Ballintijn & Hughes, 1965; Ballintijn 1968; Young, 1972, 1974; Hughes, 1975). Kuiper distinguished between periodic coughing movements and those elicited, apparently, by more specific stimuli which he divided into forward and backward coughs. He also distinguished two types of backward cough. In the first type, the jaw and opercular movements are more or less simultaneous. In the second type, he supports the view of François-Franck (1906*a*, *b*) that there is a reversal in the temporal relations of the jaw and opercular movements. It is clear from these studies that coughing can be regarded as a normal part of the respiratory activity of most fishes, but many details regarding its precise nature are yet to be established.

Earlier studies on rainbow trout suggested that coughing was normally absent, but this was probably because of the more excited condition of the animals, for coughing tends to be reduced during hypoxia and other stresses. In more recent studies on the

---

* Present address: Centre for Overseas Pest Research, 56 Grays Inn Road, London WC1X 8LU.

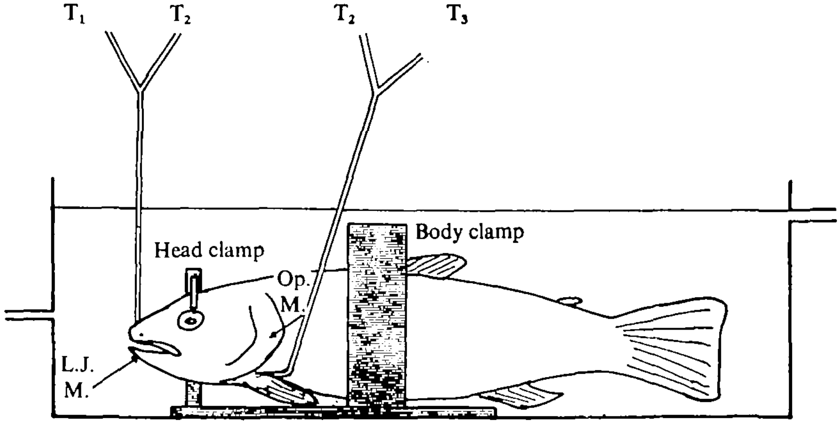

Fig. 1. Diagram to show position of cannulae and placement of movement transducers during recording of ventilatory pressures and movements from a trout restrained and lightly anaesthetized in a through-flow chamber. T_1, T_2, T_3 are connexions to the three manometers shown in Fig. 2. L.J.M., lower jaw movement; Op. M., opercular movement.

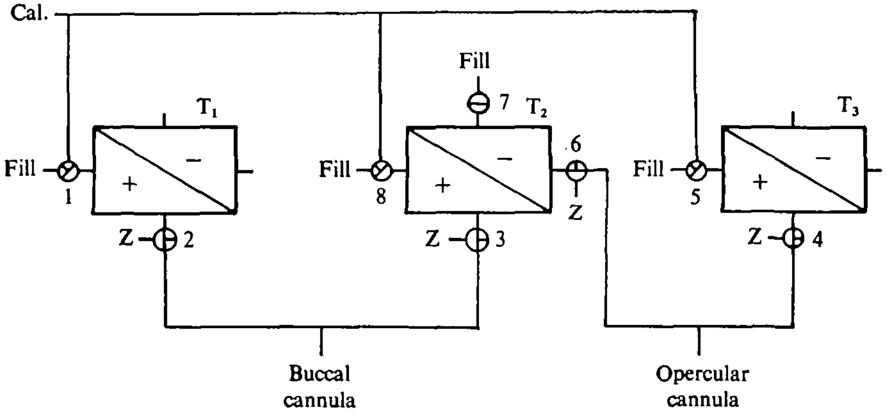

Fig. 2. Diagram of connexions used during simultaneous recording of buccal, opercular, and differential pressures using three Sanborn differential pressure transducers (T_1, T_2, and T_3). Taps 1–7 are 3-way taps with connexions to water in the experimental tank (Z), calibration manometer (Cal), or filling reservoir (Fill) containing boiled distilled water, or to a cannula as indicated in the diagram. The taps are positioned for recording these three pressures.

rainbow trout, emphasis was laid on the types of backward coughing but many differences were apparent (Hughes, 1975). In the present work, these differences have been investigated in more detail by making use of simultaneous recordings of movements and pressure changes. It was necessary, however, to use restrained and lightly anaesthetized fish for such experiments.

MATERIALS AND METHODS

Rainbow trout (*Salmo gairdneri*) were obtained from Nailsworth Hatchery, Gloucestershire, and were usually about 30·5 cm in body length. They were anaesthetized in MS-222 before cannulation of the buccal and opercular cavities, as described by Hughes & Saunders (1970). The position of the cannulae and movement

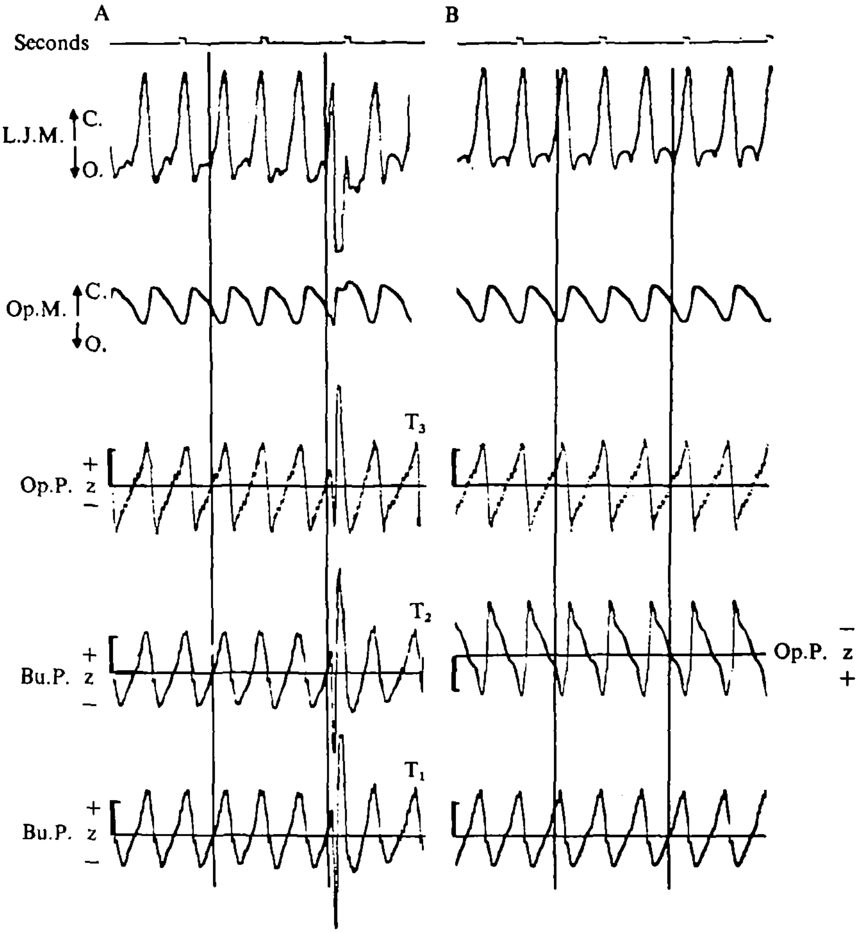

Fig. 3. Rainbow trout. Simultaneous recording of pressure changes in buccal and opercular cavities using three manometers. In A the buccal pressure is recorded on two manometers (T_1 and T_2) and in B the opercular pressure is recorded twice (T_2 and T_3). Notice almost identical waveforms recorded from the separate transducer used for recording the buccal or the opercular pressure and that normally used for recording the differential pressure (T_2). Calibration pressures: +2·0 cm H_2O.

recording levers is indicated in Fig. 1. The fish were held in a clamp similar to that used by Hughes & Shelton (1958) and kept in an experimental tank through which clean, aerated water was re-circulated at a temperature of 17 °C. Movements of the lower jaw and operculum were recorded by means of Grass mechano-electric transducers. Recordings were obtained on a Beckman Type R 6-channel, heated-pen rectilinear recorder. The fish were encouraged to cough by pipetting a small amount of malachite green near or into the mouth or opercular cavity during normal breathing and this also made it possible to observe the direction of the ejected water current.

Pressure recordings were made by means of Sanborn 268 differential pressure transducers; the plumbing arrangement used is shown in Fig. 2. In making such recordings it is important to test that there is no interaction between the transducers, and that they are linear at the sensitivities used. Buccal and opercular cannulae of

PP 120 tubing were cut fairly short (*ca.* 10 cm) and joined to larger bore tubing (PP 270). When the buccal, opercular and differential pressures were recorded simultaneously a 'Y' connector was attached to the end of each cannula. This allowed simultaneous recording of the same pressure on two transducers; care was taken that the two branches of the 'Y' were the same length, thus minimizing any differential modification of the pressure waveforms. Recordings of the same pressure changes on both transducers were found to be almost identical (Fig. 3) and hence the timing of the different pressures could be compared.

RESULTS

1. *General*

Previous studies have shown that although certain types of cough shown by fish differ, it is not always easy to define them, especially when observed in an aquarium. Sometimes fish show coughing movements which occur at more or less regular intervals and the term periodic cough has been used to describe them. Whether such coughs are low intensity forms of forward or backward coughing remains to be established but in the tench they appear to be a specific type of cough (Young, 1974). In some instances it even seems that the coughs occurring periodically may show larger amplitude buccal pressure changes than those found in coughs initiated by more specific particulate matter in the inspired water.

There are a number of ways of defining backward and forward coughing. That adopted here is as follows. A *backward* cough is when the net expiratory flow of water during these movements is out of the opercular opening. In *forward* coughs the net expiratory water flow is out of the mouth. In both cases not necessarily all the outflow takes place through these respective openings but, in some instances, the water flow is exclusively out of the opercular slits or mouth.

As mentioned above, periodic coughing in the trout may simply be repetitive forward or backward coughing movements which occur at more or less regular intervals in clear water. There is some suggestion that the amplitude of the buccal pressure waveform is greater in the lower frequency coughs of non-polluted water and that their frequency increases in polluted water (Drummond, Spoor & Olson, 1973; Hughes & Adeney, 1977).

2. *Subdivision of the coughing cycle*

The buccal pressure waveform is perhaps the easiest recording to make for the recognition of coughing cycles and consequently a subdivision of the cycle has been based upon it. It is also commonly used in studies of the effects of environmental changes on ventilation patterns. Fig. 4 shows a common type of waveform obtained during a cough and this has been subdivided into four phases A, B, C, and D which are not to be confused with the four phases 1, 2, 3, and 4 into which the normal ventilatory cycle was subdivided on the basis of the differential pressure (Hughes & Shelton, 1958).

Phase A, which may be absent, starts during phase 1 of the normal cycle, usually about half way through phase 1. It begins as the pressure rises above zero and ends when it returns to the zero level.

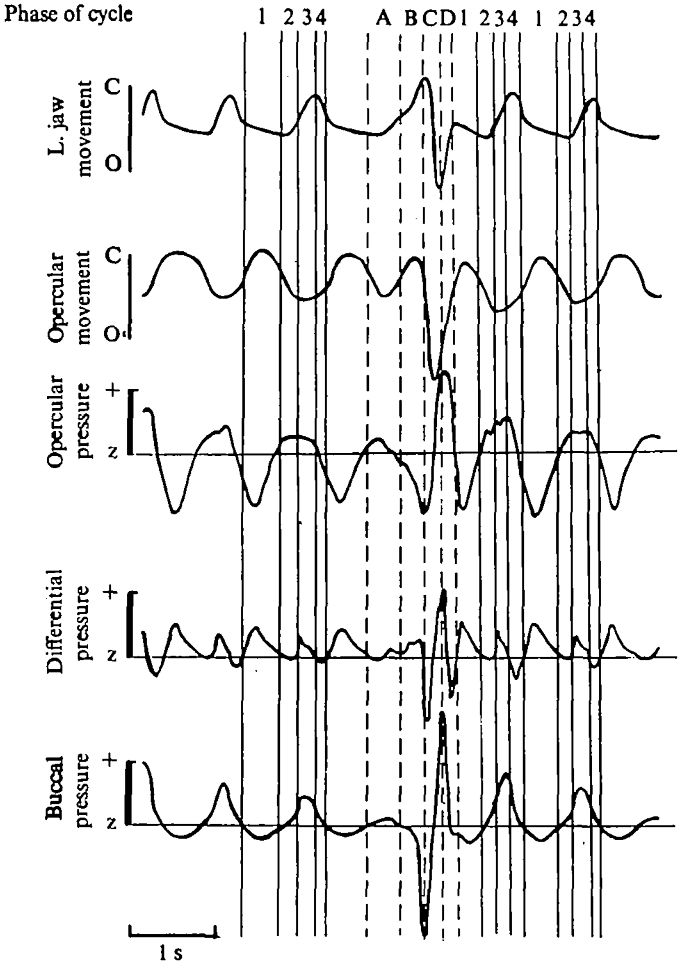

Fig. 4. Typical movement and pressure waveforms during a cough. Subdivided to show the main phases (A, B, C and D). Calibration pressures: +1·0 cm H_2O. For description see text. Phases 1–4 are those defined by Hughes & Shelton (1958).

Phase B begins as the pressure waveform becomes negative and continues until the point of maximum negativity of the cough pressure waveform.

Phase C is the period from this maximum negativity to the point of maximum positivity of the whole waveform. Therefore it is during this phase of the cycle that the maximum overall pressure change occurs within the buccal cavity.

During the final phase (D) the maximum positive pressure declines to below zero and ends at the beginning of phase 1 of the normal cycle.

These phases are not always so readily recognizable as in the typical cycle drawn in Fig. 4, but have been found useful to subdivide different cough waveforms that have been obtained. It has become clear that there are whole families of coughs which grade into one another and in which the particular size of phases A, B, C, and D can vary in a more or less continuous way. Nevertheless the two basic groups of cough resulting in either a net backward or forward flow of the expired water can be recognized

Although a whole range of coughs has been observed among individual fish, nevertheless there are certain features which are commonly found for each of the recordings of pressure or movement. The summary below gives a general indication of the type of recordings obtained for each of the movements and pressures.

(1) *Movement of the lower jaw* begins from a fully open position (phase A) and becomes fully closed. During phase B it usually remains in this position but may tend to open slightly. Phase C is variable but if the jaws open it is usually in this phase. During the final phase (D) the jaw returns from its maximum opening or closing to its normal position.

(2) *Movements of the operculum*: during phase A, it is usually abducted but only as far as in the normal cycle. During phase B, however, it opens to its maximum extent and remains maximally expanded during this phase. Phase C is variable. Abduction of the operculum is often rapid in phase D.

(3) The *opercular pressure* changes associated with this movement are generally a slow increase followed by a reduction in pressure during phase A, when it may be either above or below zero pressure. During phase B the negative pressure reaches a maximum and then in phase C it becomes maximally positive. In the final phase (D) there is often a second negative peak which may be as great as the first.

(4) The *buccal pressure* waveform generally begins with a slow rise and fall in pressure about zero in phase A. In some coughs, however, the cough begins with a maximum positive pressure wave. Phase B shows the maximum negative pressure and phase C, by definition of this phase, has maximum positive pressure. During phase D it returns to normal.

(5) The *differential pressure* between the buccal and opercular cavities is very variable, but five distinct peaks can usually be recognised. The first peak is a positive one, i.e. the pressure in the buccal cavity exceeds that in the opercular cavity, and tends to be of low amplitude and long time course. The second peak is very variable, being very pronounced in the forward coughs and less pronounced in backward coughs. This peak is always negative, whereas peak 3 is always positive and may be reduced in height. Peak 4 is negative and usually pronounced in both forward and backward coughs. The fifth peak is similar in form to peak 1 in amplitude, time course and direction.

The relationship between phases A–D and the five peaks of the differential pressure also shows some degree of constancy. During phase A peak 1 has not reached its maximum positivity which occurs during phase B. The differential pressure becomes negative during phase B and reaches its maximum value coincidentally with the maximum negativity of the buccal pressure waveform, at the end of phase B. Thus peak 2 is the result of the difference between the negative pressure in the buccal and opercular cavities. Conversely, peak 3 results from the difference between the positive pressures in these cavities and reaches its maximum at the end of phase C. Peaks 4 and 5 occur during phase D, and the interpretation of these peaks is more complex. Peak 4 is the result of a greater negativity in the opercular cavity than that in the buccal cavity, and peak 5 is the reverse. The coughs shown in Fig. 5 illustrate the variation in size relationships between these peaks of the differential pressure, even during a short burst of coughing and the difficulty in interpreting these pressure waveforms.

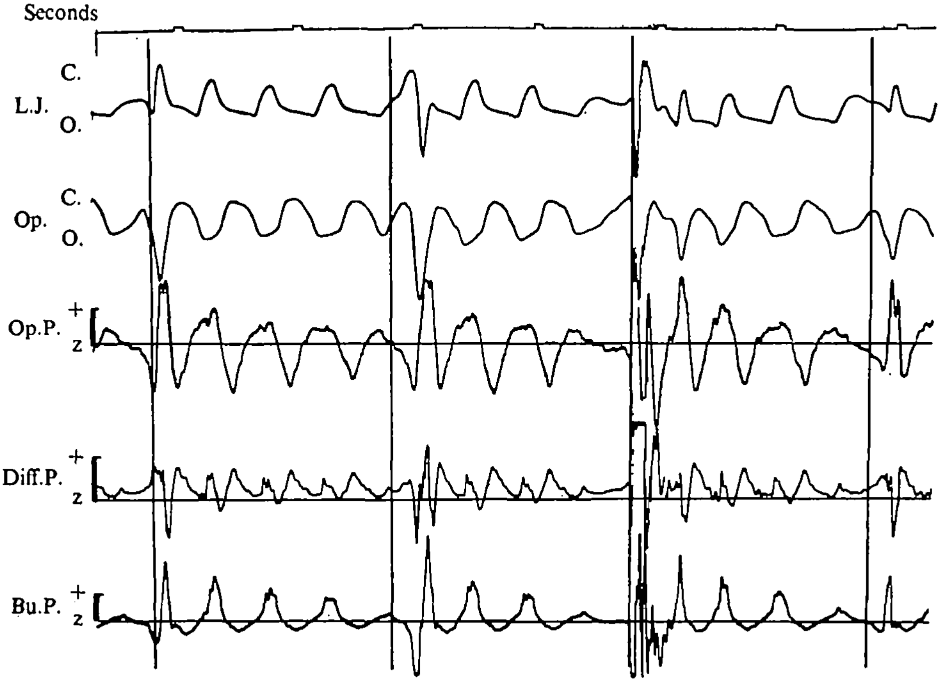

Fig. 5. Extended recording showing lower jaw and opercular movements and the associated buccal, opercular, and differential pressure changes during a sequence of coughs in a rainbow trout. Calibration pressures: +1·0 cm H_2O.

From the pressure waveforms it is possible to make some deductions concerning the likely direction and rates of water flow in and out of the mouth and opercular openings. But, even more than in the normal ventilatory cycles, the interpretation of differential pressure in terms of water flow is very difficult.

Although the recording given in Fig. 4 may be described as 'typical', it must not be regarded as the most commonly found type of cough, for, as shown in Figs. 6 and 7, there are many types of waveform. These have been selected to indicate possible relationships between them. It is apparent that any given fish may show quite different types of coughing even when these are elicited in a given sequence of coughing movements. Variations arise because of differences in the relative size and combination of these patterns of buccal and opercular movements and pressure. One of the major variables, however, is the nature of the gill resistance which is well known to undergo active changes during coughing as a result of contraction of filament adductor muscles (Bijtel, 1949; Hughes, 1961; Pasztor & Kleerekoper, 1962; Young, 1972). The precise timing and nature of this activity in each particular type of cough needs to be understood if a complete interpretation is to be given of the water flow in terms of the movement and pressure changes.

3. *Different forms of coughing*

Figs. 6 and 7 show some of the range in pressure and movement recordings obtained during coughing in trout. Fig. 6A and B may be described as purely backward coughs whereas Fig. 7H is regarded as a predominantly forward cough. When inspecting

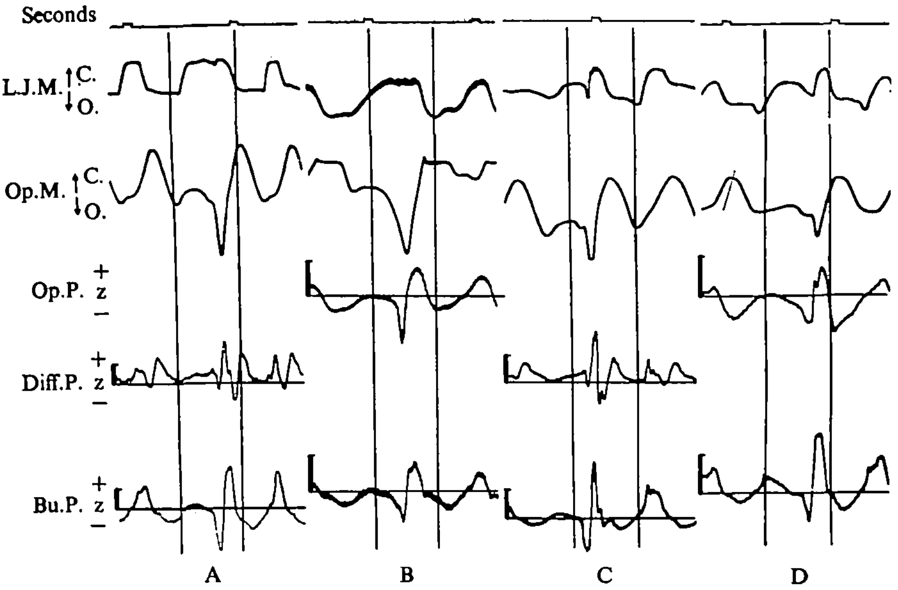

Fig. 6. Rainbow trout. Recording of movement and pressure changes during coughs selected from four different experiments to show backward coughs. The opercular and lower jaw movements and buccal pressure are recorded in all cases, but in A and C the differential pressure is recorded but no opercular pressure. In B and D there is no differential pressure but a recording of opercular pressure is present. For details see text. Calibration pressures: $+1\cdot0$ cm H_2O.

these recordings the general procedure has been to inspect the lower jaw recording movement first and to note whether the mouth is mainly open or closed during the cough. Thus in the first recordings (Fig. 6A, B) the lower jaw is closed throughout almost the whole duration of the cough. In contrast, in that of Fig. 7H the lower jaw remains open for almost the entire cough.

Secondly, the differential pressure is inspected. This may be subdivided, with reference to the peaks 1–5, into five zones. Each zone corresponds to the area under one of the peaks. Where the area of zone 2+4 is greater than 3, a forwardly directed flow of water across the gills is suspected (Fig. 7H) and where the area of zone 3 is greater than 2+4 a backwardly directed flow of water across the gills is suspected (Fig. 6A). It should be noted that in this context it is the flow of water across the gills which is indicated. Its direction may be different from that given in the general definition of the two types of cough (p. 112). Thus water may flow forwards from opercular to the buccal cavity during one phase yet the overall flow is backward.

In the interpretation of these curves it is clear that changes in gill resistance will have a very significant effect. Especially during forward coughing, it is believed that the gill filament adductor muscles are active and hence reduce the resistance to water flow and consequently the differential pressure gradient. Use of the differential curve to indicate qualitatively the direction of flow is probably justified, but a quantitative estimate of flow cannot be made with any certainty, especially during coughs.

Thirdly, the opercular and buccal pressure curves are inspected in detail. As has been indicated, these are not necessarily good guides to the type of cough as, for example, the buccal pressure waveforms shown in Fig. 6A and Fig. 7H are sur-

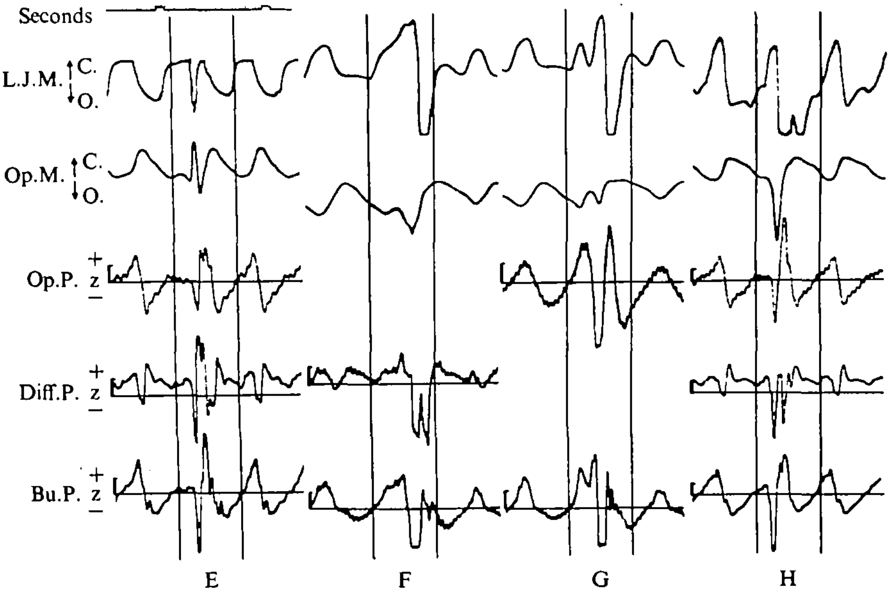

Fig. 7. Rainbow trout. E–H are separate recordings of movements and pressure changes during coughing of different types. This selection shows a range of coughs that are intermediate (E) between forward and backward, to ones that are predominantly forward (H). For details see text. Calibration pressures: +1·0 cm H_2O.

prisingly similar but the coughs are opposite in direction. Thus such analyses emphasize the need to look at all the available waveforms when assessing the likely nature of cough, and these together with any observation of flow of dye in the water give the best overall picture.

Five simultaneous recordings were not always possible and when only four were available two separate coughs have been chosen with similarities in their three common waveforms so that the likely time course of the non-recorded opercular or differential pressure could be estimated. Thus in Fig. 6, A and B are both similar backward coughs, in A the opercular curve is absent whereas in B no differential pressure was recorded but could be derived from the other two. The most characteristic feature of these two coughs is that the mouth is closed continuously during most of the coughing cycle. The operculum is adducted on the other hand for most of this cycle. Coughs of this type are usually low intensity coughs (i.e. there are no violent pressure fluctuations). They may occur very regularly, interrupting the normal ventilatory rhythms. Coughs C and D (Fig. 6) also have a backward flow, the lower jaw in these cases having a somewhat biphasic movement. This biphasic movement is also characteristic of coughs with a large forward flow component.

Fig. 7E is a simultaneous recording of all three pressures. This cough is intermediate between forward and backward forms. The negative and positive components of the differential pressure curve (peaks 2, 3 and 4) are approximately equal. Coughs F and G are a pair of similar coughs and predominantly forwardly directed. The positive phase of the buccal pressure waveform is characteristically about the same size as in normal ventilation, just prior to the commencement of the cough. These

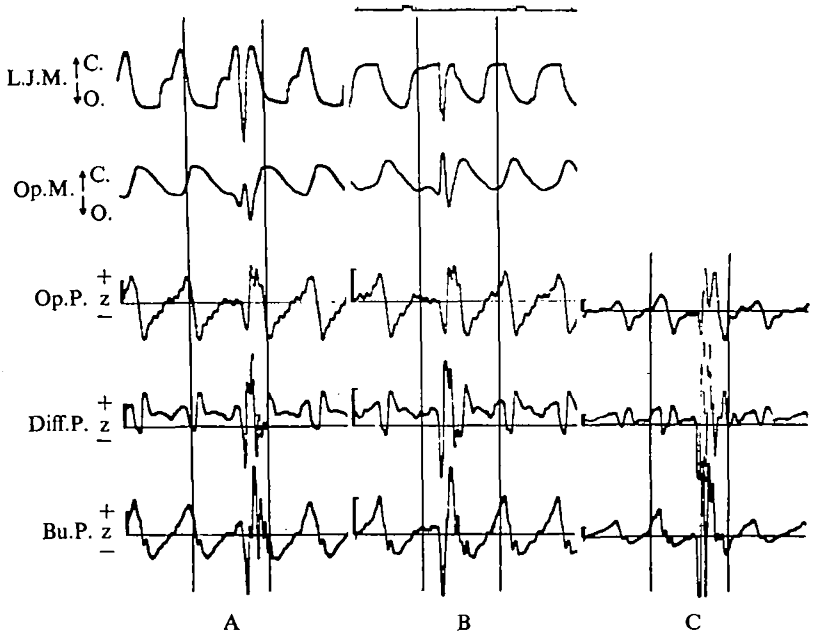

Fig. 8. Recordings of movements and pressures from a rainbow trout that is (A) in the clamp; (B) released from the clamp; and (C) free in the experimental tank. Calibration pressures: +2·0 cm H_2O.

coughs are very often initiated in phase 3 (i.e. at the start of the positive pressure waveform in the buccal cavity). Cough H is predominantly forward, with the lower jaw remaining open for most of the cough. This contrasts with the backward coughs in which the lower jaw remains closed for most of the time. It is also noteworthy that these are high intensity coughs with particularly large pressure fluctuations; the whole of such coughing actions are much more violent.

It may be concluded that the coughing patterns of trout are very variable but that they follow a general trend from backward to forward coughs (i.e. from that shown in Fig. 6A to that of Fig. 7H). It is particularly evident in the movement of the lower jaw; the opening phase (C) becoming larger and progessively earlier, Fig. 7E being an intermediate pattern between the two extremes. The characteristic feature of trout cough patterns, then, is their variability which derives from a whole spectrum of movement patterns and their associated pressure changes and their interrelationships. The range of coughing patterns may reflect differences in the type of stimuli responsible for their occurrence. Variations are more striking than is the existence of precisely defined types and possibly reflect differences in the peripheral input and the part of the respiratory cycle during which the cough begins. However, there are certain common features (e.g. all coughs are interruptions of the normal ventilatory rhythm involving oscillatory pressure changes of various kinds).

DISCUSSION

Although during many of the experiments discussed in this paper coughs were elicited by administration of foreign dyes such as malachite green (a dye sometimes used in the treatment of fish infections), coughing is certainly a naturally occurring phenomenon, especially in many cyprinoid fish which show regular coughing movements. Though it is not so common in fish living in clearer and more rapidly flowing waters interruptions of the ventilatory rhythm can occur, as for example in trout.

The frequency of coughing has been found to increase when the environment is in a more polluted condition and this has been used to monitor levels of pollutants (Drummond *et al.* 1973; Hughes, 1976).

Different species show different forms of coughing pattern although there are certain common features and earlier workers have distinguished several distinct types of coughing. Kuiper (1907) described three types of cough, one forward and two backward; Ballintijn (1969) only distinguished one type of backward cough and Young (1974) described one forward and one backward type, and a periodic cough. Young believes that the Ballintijn type of cough, and Kuiper's first type, are similar to her periodic cough. The cough shown in Fig. 6A bears some resemblance to these coughs, but the timing of the lower jaw and opercular movements are different.

The relationship between the opercular movements and buccal pressure also differs from these coughs. The main difference, however, is that the cough shown in Fig. 6A does not occur periodically but only in response to a specific stimulus. The forward coughs described by Young (1974) differ from those found in the trout in the following ways:

(*a*) the mouth always starts by closing and remains shut during the first part of the cough cycle;

(*b*) the negative peak of the buccal pressure waveform is always greater than the negative peak of the opercular pressure.

The negative pressure is always much larger than the negative peak in the normal buccal pressure. From Young's records it would appear not to be greatly different. There are similarities, however, particularly in the opercular movements and the later movements of the lower jaw. During backward coughing in the trout the lower jaw remains closed for most part and contrasts to the situation in the tench, where the lower jaw opens and then closes.

It is evident that differences in coughing patterns occur between the trout and the tench. This is not a surprising conclusion when it is remembered that the ventilatory pressure and movement waveforms and their timing are quite different in these two fish (Ballintijn, 1969). The anatomy of the buccal and opercular cavities is also very different.

'*Periodic cough*'

Young (1971, 1972, 1974) in her studies on tench, described coughs which occurred at regular intervals which she termed a 'periodic cough'. Kuiper (1907), Hughes & Shelton (1958) and Ballintijn (1969) also described regular coughs in cyprinoids. These coughs occurred spontaneously without any apparent stimulation. In the trout a similar coughing behaviour occurs. The periodic coughs described for the tench are constant in type and of low intensity. In trout their frequency is much lower, usually less than one cough per minute and the buccal pressure amplitude is significantly greater than that of the ventilatory pressure amplitude. This suggests that these coughs are of a high intensity. The regular coughs of trout, although they may be termed 'periodic', are not strictly homologous with the periodic coughs described by Young (1974) for the tench.

It is evident that coughing can be classified in a number of different ways. In two of them the main criteria are the frequency with which they occur and the direction of the water flow through the respiratory system. The former is a fairly clear difference, although for regular coughs of fairly low frequency it is difficult to determine whether or not each individual cough is evoked by particulate material in the environment. Coughs initiated as a result of a specific external stimulus are clearly of this type. The direction of water flow in to the mouth and out of the opercular cavities is also not such a clearly defined criterion, for in many coughs there is some efflux of water through the other opening. Thus if coughing is stimulated by pipetting of dye near to the mouth it usually evokes an immediate forward ejection of water followed later by backward ejection of dye from the opercular openings. When dye is injected into the opercular cavity it often gives a backward ejection current.

Just as in normal ventilation it is possible to find different patterns of muscular activity among different individuals, and also in the relative dominance of the buccal and opercular pumps, similar variations in coughing are frequently found. In neither case is a single fixed pattern of neuronal firing responsible for the pattern of overt movements. Gradation of the responses may be due to local sensory feedbacks concerned with regulating the mechanical relationships and/or the functional relationship between the water and blood flow involved in gas exchange. Interference with these normally regulated patterns might result from 'foreign inputs' and their effects will doubtless depend on the spatial and temporal nature of the input, as well as its nature. All these considerations suggest a very plastic system from which a whole range of output patterns can be expected, each of which is presumably related to the specific input pattern and the central background against which it is expressed. Evidently, coughs are not rigidly fixed motor patterns and can vary according to the peripheral input. Nevertheless, certain regions of the hind brain appear to be specifically concerned with coughing (Shelton, 1959). An individual fish may show a predominance of either forward or backward coughs over a certain period, but at other times they may be mixed. The possibility must be borne in mind, however, that future research may show how some more specific outputs can be initiated regularly by more controlled forms of afferent stimulation. Analyses of this kind have been possible in relation to the mechanical stimuli involved in the co-ordination patterns of locomotory systems. Thus while 'running' and 'walking' may be distinguished by certain specific features – these may be somewhat artificial and normally the locomotory patterns grade one into another, and in certain cases a whole spectrum of gaits may be distinguishable.

Such views on the co-ordination of the ventilatory and locomotory movements are further indicated when the range of variations between individuals is considered. In the case of trout, ventilatory movements vary both in their overall pumping effects and in the detailed nature of the active musculature and their timing (Ballintijn & Hughes, 1965). Such flexibility is perhaps consistent with what might be expected on general evolutionary grounds, as the salmonids are usually thought of as more primitive or generalized fish. Comparable flexibility will not necessarily be expected in some of the more specialized mechanisms of more advanced teleosts where the ventilatory mechanism and consequent co-ordination patterns are designed for more specific actions.

The situation with respect to trout ventilatory patterns is perhaps more complex, for these fish appeared to have more consistent pressure and movement waveforms when they were first analysed (Hughes & Shelton, 1958) and for that reason detailed analysis of this fish was preferred to those on tench which was undertaken first of all. Analysis from electromyography indicated, however, that this consistency was rather more superficial as the same movements and ventilatory pressures could be recorded when different patterns of muscle activity were involved (Ballintijn & Hughes, 1965). The situation in other fishes can clearly be quite different and there are suggestions that in the tench there are greater variations in ventilatory pressures but that the muscle patterns are more constant. During coughing this can give rise to the appearance of a more specific pattern of activity which can therefore be classified more readily into several distinct types (Young, 1974). In the carp, however, the situation is comparable with the trout. The number of muscles taking part in the movements is very variable and depends on the depth of ventilation. The shape of the movement and pressure recordings, however, is largely the same irrespective of the number of active muscles (Ballintijn, 1969).

This work was supported by a grant from the Natural Environment Research Council. We wish to thank Dr Sheila Young for her helpful discussions.

REFERENCES

BALLINTIJN, C. M. (1969). Muscle co-ordination of the respiratory pump of the carp (*Cyprinus carpio* L.). *J. exp. Biol.* 50, 569–91.

BALLINTIJN, C. M. & HUGHES, G. M. (1965). The muscular basis of the respiratory pumps in the trout. *J. exp. Biol.* 43, 349–62.

BIJTEL, J. H. (1949). The structure and the mechanism of movement of the gill filaments in Teleostei. *Arch. néerl. Zool.* 8, 267–88.

DRUMMOND, R. A., SPOOR, W. A. & OLSON, G. F. (1973). Some short-term indicators of sublethal effects of copper on brook trout, *Salvelinus fontinalis*. *J. Fish. Res. Bd Can.* 30, 698–701.

FRANÇOIS-FRANCK, CH. A. (1960*a*). Méchanique respiratoire des poissons téléostéens. I. Technique des explorations graphiques. *C. r. Séanc. Soc. Biol. Paris* 60, 962–4.

FRANÇOIS-FRANCK, CH. A. (1960*b*). Technique des prises de vues photo et chrono-photographique dans l'étude de la mécanique respiratoire des poissons téléostéens. *C. r. Séanc. Soc. Biol. Paris* 60, 965–7.

HUGHES, G. M. (1961). How fish extracts oxygen from water. *New Scientist* 11, 346–8.

HUGHES, G. M. (1975). Coughing in the rainbow trout (*Salmo gairdneri*) and the influence of pollutants. *Revue Suisse de Zoologie* 82, 47–64.

HUGHES, G. M. (1976). Polluted fish respiratory physiology. In *Effects of Pollutants on Aquatic Organisms* (ed. A. P. M. Lockwood). Cambridge University Press.

HUGHES, G. M. & ADENEY, R. J. (1977). Effects of zinc pollution on the cardiac and ventilatory rhythms of rainbow trout and their response to environmental hypoxia. *Water Research* (in Press).

HUGHES, G. M. & SAUNDERS, R. L. (1970). Response of the respiratory pumps to hypoxia in rainbow trout (*Salmo gairdneri*). *J. exp. Biol.* 53, 529–45.

HUGHES, G. M. & SHELTON ,G. (1958). The mechanism of gill ventilation in three freshwater teleostean fishes. *J. exp. Biol.* 35, 807–23.

KUIPER, T. (1970). Untersuchungen über die Atmung der Teleostier. *Arch. ges. Physiol.* 177, 1–107.

PASZTOR, V. M. & KLEERKOPER, H. (1962). The role of the gill filament musculature in teleosts. *Can. J. Zool.* 40, 785–802.

RYNBERK, VAN G. (1905). Recherches sur la respiration des poissons. *Arch. Ital. Biol.* 46, 183.

SHELTON, G. (1959). The respiratory centre in the tench (*Tinca tinca* L.). I. The effects of brain transection on respiration. *J. exp. Biol.* **36**, 191–202.

YOUNG, S. (1968). The activity of respiratory neurones in fish observed with chronically implanted electrodes. *J. Physiol., Lond.* **200**, 85–6*P*.

YOUNG, S. (1971). EMG activity in tench (*Tinca tinca* L.) gill lamellae and its association with coughing. *J. Physiol., Lond.* **215**, 37–8*P*.

YOUNG, S. (1972). Electromyographic activity during respiration and coughing in the tench (*Tinca tinca* L.). *J. Physiol., Lond.* **227**, 18–19*P*.

YOUNG, S. (1974). Coughing in fish, a study of the expulsion reflexes. PhD. thesis, University of London.