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METABOLIC CONTROL OF LUMINESCENCE IN THE LUMINOUS ORGANS OF THE TELEOST *PORICHTHYS*: EFFECTS OF THE METABOLIC INHIBITORS IODOACETIC ACID AND POTASSIUM CYANIDE

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Summary

1. The treatment of isolated photophores of the epipelagic luminescent fish *Porichthys notatus* Girard with the glycolytic inhibitor iodoacetic acid (IAA) induces light production.

2. This luminescence is not significantly affected by the adrenergic antagonists phentolamine and propranolol.

3. The addition of pyruvate, phosphoenolpyruvate or glyceraldehyde 3-phosphate in saline suppresses the light response to IAA. D-Glucose, D-glyceraldehyde, and 2- and 3-phosphoglycerate have no inhibitory effects.

4. The luminescence of the photophores induced by KCN is inhibited by D-glucose, D-glyceraldehyde 3-phosphate and 3-phosphoglycerate. In contrast, 2-deoxy-D-glucose, pyruvate, phosphoenolpyruvate, D-glyceraldehyde and 2-phosphoglycerate do not affect this light production.

5. These results confirm the view that glycolysis and oxidative phosphorylation are essential metabolic pathways in the photogenic cells. They further indicate that common products of these tightly associated metabolic pathways may regulate the activity of the luminous system in these cells.

Introduction

Isolated photophores of the epipelagic luminescent fish *Porichthys* become spontaneously luminescent a few hours after dissection. This glowing can be prevented by the addition of glucose or pyruvate (Rees & Baguet, 1987*a*, 1988). As treatment of the photophores with potassium cyanide (KCN) elicits bright light production (Baguet, 1975), it has been suggested that photophores are maintained in a nonluminous state by the activity of an energy-consuming inhibitory mechanism; glycolysis could be one of the metabolic pathways supplying energy to this mechanism. Our previous observations suggest that degradation of pyruvate

Key words: bioluminescence, glycolysis, iodoacetic acid, cyanide, fish, photophore.

through the Krebs cycle and oxidative phosphorylation could also be involved in the metabolic control of luminescence in the photogenic cells, the photocytes.

The aim of the present work was to compare the effects of the glycolytic inhibitor iodoacetic acid (IAA) and those of KCN on the luminescent activity of the isolated photophores. We also tested the ability of various glycolytic substrates and intermediates to counteract the action of these metabolic inhibitors.

Our results show that treatment of the photophores with IAA induces a slower light production than that elicited by KCN. The stimulating action of IAA on the photogenic cells has to be ascribed to its blocking action on glycolysis. However, the luminescence evoked by KCN results from the inhibition of oxidative metabolism in photogenic cells and can be suppressed by supplying glycolytic substrates. It is suggested that some common products of glycolytic and oxidative metabolism might control the activation state of the photogenic system in *Porichthys* photophores; adenosine triphosphate appears to be the most likely candidate.

A partial report of these results has already been published (Rees & Baguet, 1987*b*).

Materials and methods

Dissection of the photophores

Five specimens of the midshipman fish *Porichthys notatus*, shipped by Pacific Bio-Marine Laboratories (Venice, California), were kept in aquaria provided with aerated running sea water (18°C).

After anaesthesia with quinaldine (0·037 %) in sea water, strips of six photophores from the mandibular, branchial, gular, ventral and pleural regions (terminology according to Greene, 1899) were excised and immersed in saline.

Recording the light emission

Each of the six photophores was laid in one of six small chambers drilled in a circular plastic plate and filled with 180 μl of saline. The plate rotated around a central axis fixed on the side of the box covering the photomultiplier (RCA 1P21). Thus, each photophore could be successively positioned above the photocathode by rotating the plate.

The light-emitting area of the photophore facing a circular opening was 2 cm from the photocathode. The signal was amplified and displayed on a strip chart recorder. The apparatus was calibrated using a tritium-irradiated phosphor light source (Betalight, Nuclear Enterprises Ltd) emitting on an area of 2 mm^2 and in the same position as the photophore.

Solutions

D-Glucose, potassium cyanide, pyruvate (Merck), (−)phentolamine (Ciba), D-glyceraldehyde, DL-glyceraldehyde 3-phosphate, D(+)2-phosphoglycerate, D(−)3-phosphoglycerate, phosphoenolpyruvate, iodoacetic acid and (±)pr

pranolol (Sigma), and 2-deoxy-D-glucose (Fluka) were dissolved just before use in buffered physiological saline containing (in $mmol\,l^{-1}$): NaCl, 150; KCl, 7·5; $CaCl_2$, 3·5; $MgCl_2$, 2·4; pH 7·3 with Tris–HCl, 20.

Experimental procedure

The photophores were treated with IAA and KCN by adding 20 μl of concentrated stock solutions to the chambers. When the effects of a metabolite were tested on the IAA- or the KCN-evoked luminescence, three of the six photophores received the metabolite at $5{\cdot}5\times10^{-3}\,mol\,l^{-1}$, the concentration of sugars and pyruvate that induced the maximal inhibitory effect on the spontaneous glowing of the photophores (Rees & Baguet, 1988). Metabolites were added 10 min prior to the addition of the metabolic inhibitor. The other three photophores were treated with metabolite-free saline.

Potassium cyanide was applied at a final concentration of $10^{-4}\,mol\,l^{-1}$, a concentration that elicits the maximal light production of the isolated photophores (Christophe, 1987). Since time course and intensities of the luminescence of the isolated photophores vary widely from fish to fish, we attempted to reduce this source of variation by comparing photophores dissected from fish showing comparable light responses. In all our experiments comparing several treatments, each treatment was applied to photophores isolated from the same specimens.

Each mean value is expressed with its standard error (mean ± S.E.M.) and number of experiments, *N*. Significance of differences between means was calculated using Student's *t*-test.

Results

Effects of iodoacetic acid

Treatment of isolated photophores with a final concentration of $5\times10^{-4}\,mol\,l^{-1}$ IAA always elicited light emission (Fig. 1). The mean values of the characteristics of the luminescence evoked by $5\times10^{-4}\,mol\,l^{-1}$ IAA in 22 light organs isolated from five fish are given in Table 1. The phenomenon was very slow: preparations began to light up 12·7 min following application of IAA (latency time, TL), and

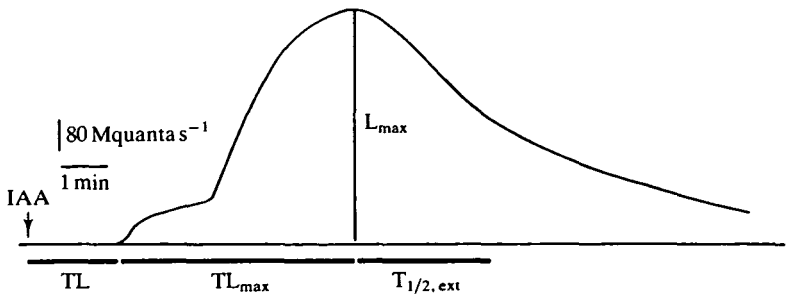

Fig. 1. Typical record of the light emission of a photophore treated with $5\times10^{-4}\,mol\,l^{-1}$ IAA. See text for definitions of abbreviations.

Table 1. *Comparison of the parameters of the luminous responses of isolated photophores treated with IAA or KCN*

	TL (min)	TL_{max} (min)	L_{max} (Mquanta s^{-1})	$T_{1/2,ext}$ (min)
IAA 5×10^{-4} mol l^{-1} ($N=22$)	12·7 ± 2·5	15·7 ± 1·8	543·8 ± 69·1	16·3 ± 1·9
KCN 10^{-4} mol l^{-1} ($N=29$)	0·8 ± 0·12	2·5 ± 0·2	1938·4 ± 184·2	3·3 ± 0·1

Values are means ± S.E.M.
See text for explanations of the abbreviations.
Photophores originated from five specimens. The values of all the parameters are significantly different ($P<0·0005$) between the two treatments.

the maximal intensity (L_{max}) was reached after 15·7 min (TL_{max}). The extinction was much slower than the emission of light: the half-extinction time ($T_{1/2,ext}$), i.e. the time to return to 50 % of the maximal light emission, was 16·3 min. The parameters characterizing the IAA-induced luminescence, although relatively constant among the photophores isolated from different regions of one fish, varied widely from one fish to another.

The time course and the intensity of this luminescence were strongly dependent on the concentration of IAA: concentrations below 10^{-4} mol l^{-1} did not generate any light production; concentrations from 10^{-4} to 10^{-3} mol l^{-1} always elicited light production, the shape of which was relatively constant, as shown by the mean values of the parameters (Table 2).

However, with an IAA concentration of 10^{-2} mol l^{-1}, the pattern of the luminescence was affected markedly: the latency of luminescence onset and the

Table 2. *Influence of IAA concentration on the parameters of the luminous response of isolated photophores*

[IAA] (mol l^{-1})	TL (min)	TL_{max} (min)	L_{max} (Mquanta s^{-1})	$T_{1/2,ext}$ (min)
10^{-6}			0	
10^{-5}			0	
10^{-4}	28·9 ± 2·3	24·4 ± 3·4	354·6 ± 96·3	16·9 ± 2·3
5×10^{-4}	22·0 ± 1·3	18·0 ± 3·7	291·3 ± 45·1	26·2 ± 2·6
10^{-3}	22·5 ± 3·7	21·0 ± 3·5	361·8 ± 92·4	15·5 ± 1·1
10^{-2}	5·5 ± 0·3**	6·5 ± 0·4*	972·7 ± 161·2**	7·3 ± 0·8*

Four photophores were tested for each concentration (mean ± S.E.M.); all photophores were dissected from one specimen.
* denotes a value that differs significantly from other values, $P<0·05$; ** $P<0·001$.
See Table 1 for explanation of abbreviations.

time to reach maximal amplitude decreased threefold and the half-extinction time was reduced by half (Table 2). Since all these experiments were carried out in the presence of $5{\cdot}5\times10^{-3}$ mol l^{-1} glucose, these light emissions are distinct from the spontaneous glow that photophores develop in physiological saline (Rees & Baguet, 1988). The time course and intensity of the IAA-evoked luminescence were very different from those of the light responses elicited by treating the photophores with 10^{-4} mol l^{-1} KCN (Table 1): a comparison of the mean values of the parameters shows that the luminescence induced by KCN was much faster and brighter than that induced by IAA.

Effects of adrenergic blocking agents on the IAA-induced luminescence

It was possible that IAA did not act directly on the photogenic cells, but indirectly on some nerve terminals present in the photophores (Nicol, 1957). The neurotransmitter noradrenaline, stored in the nerve varicosities (Anctil *et al.* 1984), could be liberated and could activate adrenergic receptors present on photogenic cell membranes (Christophe & Baguet, 1985); this could lead to the observed light emission.

To test this hypothesis, photophores were incubated for 10 min with 10^{-5} mol l^{-1} phentolamine or propranolol (respectively α- and β-adrenergic antagonists). Phentolamine did not affect the parameters of the light response to IAA (Table 3). Since this α-adrenergic antagonist inhibits the luminescence of the photophores induced by exogenous noradrenaline, adrenaline and phenylephrine (Christophe & Baguet, 1985), it is suggested that α-adrenergic receptors are not involved in the stimulatory action of IAA. However, propranolol reduced by 24 % the intensity of the IAA-induced luminescence, an effect which might be ascribed to nonspecific effects of propranolol.

Table 3. *Effects of the adrenergic antagonists propranolol and phentolamine on the parameters of the luminescence of the isolated photophores induced by* 5×10^{-4} *mol* l^{-1} *IAA*

	TL (%)	TL_{max} (%)	L_{max} (%)	$T_{1/2,ext}$ (%)
Controls	100	100	100	100
Phentolamine (10^{-5} mol l^{-1})	99·59 ± 21·67	106·98 ± 10·93	95·70 ± 8·81	97·40 ± 5·04
Propranolol (10^{-5} mol l^{-1})	104·69 ± 24·15	108·40 ± 12·26	75·30 ± 7·37*	87·35 ± 6·87

Values are means ± S.E.M.

All values are expressed as a percentage of those measured for control photophores treated with antagonist-free saline.

Each treatment was applied to six photophores.

* denotes a value that differs significantly from control values ($P<0{\cdot}05$).

See Table 1 for an explanation of the abbreviations.

Effects of glycolytic intermediates on the IAA- and KCN-evoked light responses

The glowing effect induced by IAA suggested that the presence of some glycolytic metabolites could be essential to maintain the photophore in the nonluminescent state. Therefore, we investigated the ability of glucose and some glycolytic intermediates to inhibit the luminescence of IAA-treated photophores. The effects of these compounds were tested in parallel on the KCN-induced luminescence (see Table 4).

Effects on the light response to IAA

Glucose had no effect on the intensity of the response but it affected the latency which increased four- to fivefold (not shown). Pyruvate, which bypasses the assumed glycolytic site of inhibition by IAA, totally suppressed the IAA-induced luminescence. This inhibitory effect of pyruvate could be overcome by KCN: a further addition of KCN to these photophores elicited light production. The inhibitory effect of pyruvate was not observed when we used 10^{-2} mol l^{-1} instead of 5×10^{-4} mol l^{-1} IAA. Phosphoenolpyruvate (PEP), like pyruvate, completely inhibited the light response to 5×10^{-4} mol l^{-1} IAA.

Glyceraldehyde 3-phosphate (G-3-P) totally inhibited the luminescence in response to IAA but D-glyceraldehyde (D-G), and 2- and 3-phosphoglycerate (2-P-G and 3-P-G) did not change the amplitude of the light response.

We also tested whether the IAA-induced luminescence could be reversed by these glycolytic intermediates. In these experiments, the tested compounds were added when the light response had just reached its maximal level. We observed that the compounds which suppressed the IAA-induced luminescence, namely pyruvate, phosphoenolpyruvate and glyceraldehyde 3-phosphate, also induced a rapid and complete extinction (Fig. 2); the three other compounds did not reverse the IAA-evoked luminescence.

Effects on the light response to KCN

The presence of glucose totally inhibited the luminescence induced by KCN; this effect seemed to be mediated by the metabolism of glucose since 2-deoxy-D-glucose, a nonmetabolizable analogue of glucose (Sols & Crane, 1954), had no inhibitory effect on this luminescence; it accelerated the development of the light emission, the time to reach the maximal intensity being reduced by $22{\cdot}5 \pm 6{\cdot}4$ % (not shown). Furthermore, we observed that the inhibition of the KCN-induced luminescence by glucose was reversed by adding IAA to the photophore, suggesting that glucose metabolism through the glycolytic pathway is involved. These observations rule out the possibility that KCN could directly activate the luminescent system, as shown in an isolated system from a luminous worm (Shimomura *et al.* 1964).

As would be expected if KCN inhibited the oxidative metabolism for eliciting the luminescence of the photophores, pyruvate and phosphoenolpyruvate showed no inhibitory action on KCN-induced luminescence. However, it was ver

Table 4. *Effects of some metabolic substrates and intermediates on the maximal intensities of the light responses of photophores treated with IAA and KCN*

	None	GLU	2-D-G	D-GLY	G-3-P	3-P-G	2-P-G	PEP	PYR
IAA (5×10^{-4} mol l^{-1})	100	118·2 ± 22·3	91·4 ± 16·1	98·8 ± 37·7	0*	99·1 ± 12·3	128·5 ± 23·9	0*	0*
KCN (10^{-4} mol l^{-1})	100	0*	124·5 ± 13·4	110·2 ± 6·2	0*	30·8 ± 7·1*	116·7 ± 38·9	97·7 ± 8·8	94·0 ± 13·3

GLU, D-glucose; 2-D-G, 2-deoxy-D-glucose; D-GLY, D-glyceraldehyde; G-3-P, glyceraldehyde-3-phosphate; 2-P-G, 2-phosphoglycerate; 3-P-G, 3-phosphoglycerate; PEP, phosphoenolpyruvate; PYR, pyruvate.

All metabolites were at a concentration of $5{\cdot}5\times10^{-3}$ mol l^{-1} and added 10 min prior to the addition of IAA and KCN.

All values are expressed as a percentage of control values measured for photophores treated with the metabolic inhibitor in metabolite-free saline.

* indicates mean values that differ significantly ($P<0{\cdot}001$) from that measured for control photophores (mean ± S.E.M.); $N=6$.

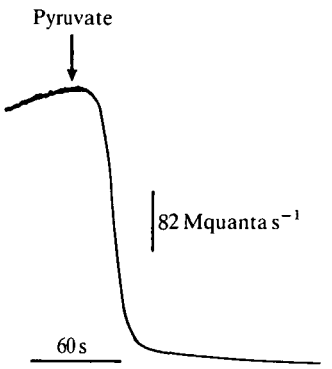

Fig. 2. Record showing the rapid extinction of IAA-induced luminescence following the addition of $5{\cdot}5\times10^{-3}$ mol l^{-1} pyruvate in saline. Pyruvate was added when the luminescence was maximal (arrow).

surprising to observe that G-3-P and 3-P-G could counteract the effect of KCN: the light production was totally suppressed by pretreatment of the photophores with G-3-P and its intensity was reduced by approximately 70% by a 10-min pretreatment with 3-P-G. The inhibition reached $85{\cdot}3 \pm 5{\cdot}5$ % ($N = 6$) when 3-P-G was added 20 min prior to the addition of KCN. It was interesting to observe that the nonphosphorylated form of G-3-P, D-glyceraldehyde, did not affect the KCN-induced light production. Similarly, pretreating the photophores with 2-P-G had no inhibitory effect on the KCN-evoked luminescence.

Discussion

The light-evoking effects of KCN and IAA

Treatment of the isolated photophores of *Porichthys* with IAA concentrations ranging from 10^{-4} to 10^{-2} mol l^{-1} always elicited light emission. Like the spontaneous glowing that results from a metabolic defect (Rees & Baguet, 1987*a*,*b*, 1988), the IAA-evoked luminescence is a slow and long-lasting phenomenon. As adrenergic antagonists have no inhibitory effect on this luminescence, this could be mediated by a direct action of IAA on the photocytes, as has been suggested for the KCN-stimulating effect (Christophe & Baguet, 1982).

Our results strongly suggest that IAA acts on the glycolytic metabolism of the photocytes to induce the luminescence of the photophores. Because pyruvate and phosphoenolpyruvate, the end-products of glycolysis, but not glucose, suppressed and reversed the light response to 5×10^{-4} mol l^{-1} IAA, we can assume that IAA effectively blocks the glycolytic pathway; this inhibition is responsible for inducing the luminescence of the photocytes.

The ability of pyruvate and phosphoenolpyruvate (which is probably itself converted to pyruvate) to inhibit the IAA-induced luminescence might be interpreted as an indirect effect caused by the utilization of pyruvate in the tricarboxylic acid cycle and oxidative phosphorylation. This assumption is sup

ported by the observation that light production following the application of KCN is not inhibited by pyruvate or phosphoenolpyruvate but is inhibited by glucose; this view is confirmed by the observation that KCN reverses the inhibition of the IAA-induced luminescence by pyruvate.

The selectivity of the action of IAA depends upon the concentration used: at concentrations above $1–5\times10^{-3}$ mol l^{-1}, it loses its selectivity (Webb, 1966). Since the luminescence induced by 10^{-2} mol l^{-1} IAA is not inhibited by pyruvate, it probably results from an additional nonspecific effect of IAA. Because this brighter and more rapid luminescence is similar to that observed in KCN-poisoned photophores, it seems possible that a large concentration of IAA should inhibit oxidative phosphorylation, as shown in other organs (see Webb, 1966). As another possibility, IAA, which reacts with thiol groups, could interact with these groups in the luciferin–luciferase system and trigger the luminescence. Such an activation mechanism has been described *in vitro* for the bioluminescent system of the coelenterate *Aequora* (Shimomura *et al.* 1974).

Effects of glycolytic metabolites

All the enzymes of glycolysis have been detected in fish tissues (Walton & Cowey, 1982). IAA has been shown to inhibit this pathway preferentially at the level of glyceraldehyde 3-phosphate dehydrogenase (G-3-PDH), the enzyme that catalyses the transformation of glyceraldehyde 3-phosphate (G-3-P) into 1,3-diphosphoglycerate (Webb, 1966).

To evaluate the degree of selectivity of IAA in the photocytes, we calculated the amount of inhibition produced by some glycolytic substrates or intermediates on the IAA-evoked light emission. As these compounds were added exogenously, it could be argued that their effects were mediated extracellularly. Several arguments support the view that these compounds enter the photogenic cells.

As discussed above, the induction of luminescence by IAA and KCN results directly from an intracellular effect of both metabolic inhibitors. Indeed, since extracellular metabolism of the exogenously applied metabolites appears unlikely, we can assume that they enter the photocytes to exert their effects on the IAA- and KCN-induced luminescences. The phosphorylated compounds show selective effects on the IAA- and the KCN-induced luminescences. 2-P-G and 3-P-G have no effect on the IAA-induced luminescence, but 3-P-G specifically inhibits the KCN-induced light emission and, although PEP has no effect on the KCN-induced luminescence and, although PEP has no effect on the KCN-induced light emission, it totally suppresses the IAA-evoked luminescence. Such selective effects of these metabolites could probably not be ascribed to nonspecific action at the extracellular side of the plasma membrane, and probably reflect their respective abilities to be metabolized in such poisoned cells.

Surprisingly, G-3-P totally suppresses and reverses the effects of IAA. As IAA normally blocks its conversion to pyruvate, G-3-P should not affect the luminescence. It is possible that G-3-P could protect G-3-PDH against inactivation by IAA. Such a substrate protection mechanism of this enzyme has been recorded *in*

vitro in several tissues (see Webb, 1966). However, as G-3-P may also be able to reverse the luminescence induced by IAA, this hypothesis may not be helpful.

As a second possibility, G-3-P could exert its inhibitory effect *via* some metabolic pathway other than glycolysis, e.g. the pentose phosphate shunt, and restore normal metabolic activity of the cells. This hypothesis appears very doubtful since glucose, which should still be metabolized, would be expected to act like G-3-P and suppress the IAA-evoked luminescence; this was not observed.

A third possibility, then, is that G-3-P could act directly on the luminous system and inhibit its activation. For example, G-3-P could be essential in the control of the functional state of luciferin or luciferase. This kind of interaction has been demonstrated in the *Renilla reniformis* luminescent system: 3',5'-diphosphoadenosine (PAP) controls the level of luciferin available for the luminous reaction (Cormier *et al.* 1974). Further experiments are necessary to clarify this situation.

We have shown that 2-P-G and 3-P-G have no inhibitory effect on the IAA-induced luminescence. Since they enter the glycolytic pathway after G-3-PDH, we expected that both compounds would impair IAA action, as do phosphoenolpyruvate and pyruvate.

Because spontaneous glowing, which results from a lack of glycolytic substrates, is inhibited by 3-P-G but not by 2-P-G (J. F. Rees & F. Baguet, unpublished results), we suggest that 2-P-G is not metabolized by the photocytes. Its apparent inability to be metabolized is not yet understood and probably does not result from a lower membrane permeability for 2-P-G than for 3-P-G, which are very similar molecules.

The lack of inhibitory effect of 3-P-G cannot be explained by the inability of this triose to enter the cellular metabolism. Since the light response evoked by KCN treatment is inhibited by 3-P-G but not by pyruvate, it seems possible that 3-P-G could affect light emission without being metabolized into pyruvate by glycolysis. This result suggests that 3-P-G could be metabolized by the neoglucogenetic pathway, which runs the opposite way from glycolysis, and converted to G-3-P.

G-3-P, which inhibits the luminescence in response to IAA, could be the messenger mediating the 3-P-G effect. As a consequence, since the enzyme G-3-PDH, which is involved in both glycolytic and neoglucogenetic pathways, is inhibited by IAA but not by KCN, this could perhaps explain why the inhibitory effect of 3-P-G did not appear after poisoning the photophore with IAA.

How does metabolism control the luminescence in photocytes?

Our results clearly show that the light reaction is under metabolic control in photogenic cells. They further indicate that some metabolite, directly or indirectly, inhibits the luminous system. This metabolite appears to be produced in both glycolysis and oxidative phosphorylation. Since adenosine triphosphate is the major product of these metabolic pathways, we suggest that it could be implicated in the regulation of the light-emitting reaction. It could act either as an energy source to an inhibitory mechanism of the luminous system or as a reagent in th

luciferin–luciferase system. No-one has yet studied the possible role of ATP in preventing the light reaction *in vitro*.

As discussed above, the inhibitory effect of G-3-P probably does not result from an indirect action of ATP, but G-3-P could possibly interact with the components of the luminous system. *In vitro* experiments studying the interactions of G-3-P with the isolated luminous system are necessary.

It is tempting to conclude temporarily that metabolism might provide some compounds necessary to maintain the photophore in a nonluminous state. It is likely that the physiological processes triggering the luminescence of the photophore could somehow prevent the activity of these metabolic products. Experiments are in progress to specify the nature of the metabolic control of luminescence in *Porichthys* photophores.

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References

ANCTIL, M., DESCARRIES, L. & WATKINS, K. C. (1984). Distribution of ^{3}H-noradrenalin and ^{3}H-serotonin in photophores of *Porichthys notatus*. *Cell Tiss. Res.* **235**, 129–136.

BAGUET, F. (1975). Excitation and control of isolated photophores of luminous fishes. *Prog. Neurobiol.* **5**, 97–125.

CHRISTOPHE, B. (1987). Contribution à l'étude du contrôle de la photogénèse des photophores du poisson téléostéen *Porichthys notatus*. Thesis, Université Catholique de Louvain.

CHRISTOPHE, B. & BAGUET, F. (1982). Potassium cyanide stimulation of isolated photocytes from a luminescent fish. *Archs int. Physiol. Biochim.* **91**, P88.

CHRISTOPHE, B. & BAGUET, F. (1985). The adrenergic control of the photocyte luminescence of the *Porichthys* photophore. *Comp. Biochem. Physiol.* **81**C, 359–365.

CORMIER, M. J., HORI, K. & ANDERSON, J. M. (1974). Bioluminescence in coelenterates. *Biochim. biophys. Acta* **346**, 137–164.

GREENE, C. W. (1899). The phosphorescent organs in the toadfish *Porichthys notatus* Girard. *J. Morph.* **15**, 667–696.

NICOL, J. A. C. (1957). Observations on photophores and luminescence in the teleost *Porichthys*. *J. microsc. Sci.* **98**, 179–188.

REES, J. F. & BAGUET, F. (1987*a*). Glucose metabolism and luminescence control of *Porichthys* photophores. *Archs int. Physiol. Biochem.* **95**, P6.

REES, J. F. & BAGUET, F. (1987*b*). Iodoacetic acid-induced luminescence in *Porichthys* isolated luminous organs. *Archs int. Physiol. Biochem.* **95**, P76.

REES, J. F. & BAGUET, F. (1988). Metabolic control of spontaneous glowing in isolated *Porichthys* photophores. *J. exp. Biol.* **135**, 289–299.

SHIMOMURA, O., BEERS, J. R. & JOHNSON, F. H. (1964). The cyanide activation of *Odontosyllis* luminescence. *J. cell. comp. Physiol.* **64**, 15–22.

SHIMOMURA, O., JOHNSON, F. H. & MORISE, H. (1974). Mechanism of the luminescent intramolecular reaction of aequorin. *Biochemistry, N.Y.* **13**, 3278–3286.

SOLS, A. & CRANE, R. K. (1954). Substrate specificity of brain hexokinase. *J. biol. Chem.* **210**, 581–595.

WALTON, M. J. & COWEY, C. B. (1982). Aspects of intermediary metabolism in salmonid fish. *Comp. Biochem. Physiol.* **73**B, 59–79.

WEBB, J. L. (1966). Iodoacetate and iodoacetamide. In *Enzyme and Metabolic Inhibitors*, vol. 3 (ed. J. L. Webb), pp. 1–283. New York: Academic Press.