

Cyclic 3',5'-Adenosine Monophosphate Level in the Plasma of the Rainbow Trout (*Salmo gairdnerii* Richardson) Following Adrenaline Administration and Constrained Exercise

The metabolic effects of catecholamines in fish have been the object of numerous investigations and references were already reviewed^{1,2}. However, there have been only a few studies on cyclic 3',5'-adenosine monophosphate (cAMP). NAKANO and TOMLINSON³ showed that muscle cAMP rises in rainbow trout in relation to physical disturbance which increases blood catecholamines. And CUTHBERT and PIC⁴ pointed out an increase of gill cAMP in the mullet treated by adrenaline. In the present work, we have studied the plasma level of cAMP in the rainbow trout after adrenaline injection and after constrained exercise. The hyperglycemic reaction⁵ was studied as a mark of animal response.

Rainbow trout, weighing 160–220 g, were injected i.m. with adrenaline at the rate of 100 µg/100 g body weight. At various time intervals after injection (15, 30, 60 min and 5 h), they were anaesthetized by being placed in a bath containing 0.1 g/l sodium tricaine methane sulfonate (MS 222 Sandoz). Blood was then collected by cardiac puncture with anticoagulant (Anticlot Delagrangé) and immediately centrifugated. For constrained exercise, animals were grasped by the tail with tongs; this causes strong swimming movements with intervals of vigorous struggle agitation. After 5 or 10 min coercion time, animals were anaesthetized for blood sampling. Cyclic AMP was assayed using the competitive protein-binding method of WALTON and GARREN⁶ with a binding protein purified from bovine adrenal glands. Plasma samples were treated with ethanol (8/1) and ethanol extracts dried by a nitrogen flow. Extracts corresponding to 0.05–0.2 ml plasma were assayed in duplicate and cAMP content determined from a calibration curve prepared at the same time with tritiated and non-radioactive cAMP. The protein bound cAMP was isolated by filtration using glass fibre filters (Whatman GF.F). For plasma glucose, a glucose oxidase technique was used (Blutzucker Farbtest Boehringer).

The results are given in the Table. Each experimental series corresponds to 8 animals. Mean values are followed by the standard error. A statistical study was made by analysis of variance according to the F test.

The following points may be emphasized: 1. The normal value for cAMP level in rainbow trout plasma is 25 ± 4 nmoles/l. This value is comparable to the human plasma level⁷.

2. An adrenaline injection causes a strong increase of plasma cAMP. Maximal level is seen at 15 min after

adrenaline injection ($p < 0.01$) and persists for 1 h ($p < 0.01$). After 5 h, cAMP level becomes lower ($p < 0.05$). Owing to animal anaesthesia and blood sampling duration, shorter action times were not used, but the possibility of a more rapid occurrence of cAMP peak must be considered. The time course change in plasma glucose is more gradual and persistent; this evolution suggests that hyperglycemia comes from cAMP increase.

3. After 5 min of constrained exercise, cAMP level is about twice the control value ($p < 0.05$), plasma glucose rises slightly but significantly ($p < 0.01$). A 10 min exercise induces the same change for cAMP ($p < 0.01$), but the hyperglycemic effect is higher ($p < 0.001$). In the same way cAMP increase appears to come before hyperglycemic reaction.

While the effects of adrenaline on tissue cAMP levels have been extensively studied⁸, there are as yet few published data on in vivo effects on blood cAMP. Adrenaline elevates blood cAMP level in man⁹ and in domestic fowl¹⁰. Our study confirms this fact in fish. The increase in plasma cAMP followed by an increase in plasma glucose produced by adrenaline indicates the role of this nucleotide as a second messenger in fish hyperglycemic response. The similar results noted in animals subjected to constrained exercise, also confirms that the general pathway for catecholamine metabolic effects functions in fish tissues.

Résumé. La taux plasmatique de l'AMP cyclique chez la truite arc en ciel (*Salmo gairdnerii* Richardson) d'élevage est pour l'animal normal de 25 ± 4 nmoles/l. L'injection i.m. d'adrénaline provoque une très forte augmentation de ce taux. Au bout de 15 min on observe l'effet maximum qui se maintient au bout d'une h et diminue ensuite au bout de 5 h. L'effet hyperglycémiant plus progressif et prolongé, apparaît comme décalé. Après un exercice forcé de 5 ou 10 min, le taux plasmatique de l'AMP cyclique est doublé tandis que la glycémie s'élève progressivement.

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Cyclic 3',5'-adenosine monophosphate and glucose levels in the plasma of the rainbow trout following an adrenaline injection and a constrained exercise

	cAMP (nmoles/l)	Glucose (g/l)
Control	25 ± 4	0.69 ± 0.04
Adrenaline (15 min)	199 ± 46	1.19 ± 0.08
Adrenaline (30 min)	146 ± 22	1.68 ± 0.17
Adrenaline (1 h)	108 ± 15	1.61 ± 0.08
Adrenaline (5 h)	61 ± 12	1.59 ± 0.11
Exersice (5 min)	42 ± 6	0.86 ± 0.04
Exersice (10 min)	41 ± 2	1.13 ± 0.04

¹ A. DEMAEL-SUARD, Thèse Doc. Sci. Nat. Lyon (1971), No. 32.

² C. PERRIER, H. PERRIER and J. GRAS, *Experientia* 29, 24 (1973).

³ T. NAKANO and N. TOMLINSON, *J. Fish. Res. Bd. Can.* 24, 1701 (1967).

⁴ A. W. CUTHBERT and P. PIC, *Br. J. Pharmac.* 49, 134 (1973).

⁵ H. PERRIER, C. PERRIER and J. GRAS, *C.R. Soc. Biol., Paris* 165, 2141 (1971).

⁶ G. M. WALTON and L. D. GARREN, *Biochemistry* 9, 4223 (1970).

⁷ A. L. LATNER and K. PRUDHOE, *Clin. chim. Acta* 48, 353 (1973).

⁸ E. W. SUTHERLAND and G. A. ROBINSON, *Pharmac. Rev.* 78, 145 (1966).

⁹ J. H. BALL, N. I. KAMINSKY, J. G. HARDMAN, A. E. BROADUS, E. W. SUTHERLAND and G. W. LIDDLE, *J. clin. Invest.* 57, 2124 (1972).

¹⁰ A. A. FRÖHLICH and R. R. MARQUARDT, *Biochim. biophys. Acta* 286, 396 (1972).