PDFlib PLOP: PDF Linearization, Optimization, Protection

Page inserted by evaluation version www.pdflib.com – sales@pdflib.com

Diurnal Changes in Arginine Vasotocin and Isotocin Plasma Levels in Rainbow Trout Adapted to Freshwater and Brackish Baltic Water

EWA KULCZYKOWSKA

Marine Biology Center, Polish Academy of Sciences, Sw. Wojciecha 5 Str., 81-347 Gdynia, Poland

Arginine vasotocin (AVT) and isotocin (IT) are nonapeptides in teleost fish, produced in separate hypothalamic neurosecretory neurons. The vasotocinergic and isotocinergic axons end in the neurohypophysis, where hormones are stored and released.

AVT is found in all vertebrates, but IT is restricted to teleosts. Although the teleost neurohypophysial hormones were chemically identified in the 1960s, their precise role still remains unclear. AVT seems to participate in osmoregulation, cardiovascular activity, endocrine secretion, reproduction, and probably neurotransmission and neuromodulation processes in the central nervous system in fish. To date, measurement of plasma IT concentrations is limited.

Endocrine and neuroendocrine cells do not release their products continuously, and many display a rhythmicity. For example, the AVT content of the telencephalon and hypothalamus fluctuates within a 24-hour period in goldfish.¹ However, diel changes in plasma concentrations of neurohypophysial hormones have not been investigated. On the other hand, it is well known that the synthesis and secretion of AVT is very sensitive to osmotic stimuli.

The aim of this study was to quantitatively determine plasma AVT and IT levels of rainbow trout adapted to freshwater and brackish Baltic water and to assess possible diurnal variations. The new nonisotopic assay has been applied for hormone measurements.

MATERIALS AND METHODS

Rainbow trout (250–400 g) of both sexes were progeny of single parent spawning. In January and February, animals were kept in tanks at 10–14°C under a natural photoperiod (the dark period occurred between 4 PM and 8 AM). Fish were adapted to freshwater, then to brackish Baltic water (7% salt concentration), and finally to freshwater again. Blood samples for AVT, IT, and osmolality were collected from the dorsal aorta of decapitated, unanesthetized fish. AVT and IT were extracted from plasma by solid-phase extraction. Hormones were determined by gradient high performance liquid chromatography. Quantitative determination of AVT and IT was performed on the basis of a standard curve. The method has been described in detail elsewhere.²



FIGURE 1. (a, b, c) Diurnal variations in plasma arginine vasotocin levels measured in rainbow trout adapted to fresh water, brackish water, and readapted to fresh water, respectively. Dark bar indicates natural dark period during the experiments (January/February). Numbers of animals used are shown in the *bars*. (a) p < 0.05vs 5:00; (**b**) *p* <0.01 vs 5:00.

RESULTS AND DISCUSSION

Unlike AVT, IT levels displayed no diurnal changes. The IT concentrations were significantly lower than those of AVT, except the 5 AM values.

This is the first study presenting the diurnal changes in plasma AVT concentrations in fish (FIG. 1). Fish adapted to each salinity showed a similar rise in plasma AVT at the onset of the dark phase (4 PM) with lowest levels evident at the end of the dark phase (5 AM). The daily patterns of variation in AVT plasma levels in rainbow trout closely matched those found for vasopressin, the mammalian counterpart of vasotocin, and oxytocin in the rat.^{3,4} Moreover, a clear circadian rhythm of vaso-

KULCZYKOWSKA: DIURNAL CHANGES IN AVT AND IT

pressin (AVP) in cerebrospinal fluid, daily changes in AVP neuron activity, and vasopressin mRNA levels in the suprachiasmatic nucleus in mammals have been reported. It is generally presumed that in teleosts the preoptic area is the seat of the "biological clock" homologous with the mammalian suprachiasmatic nucleus. It is notable that the neurons in this area are immunoreactive for AVT. Together these observations suggest that the synthesis and/or release of AVT and IT may be controlled independently and that AVT may participate in the circadian time-keeping system in teleosts.

REFERENCES

- HONTELA, A. & LEDERIS, K. 1985. Diel variations in arginine vasotocin content of goldfish brain and pituitary: Effects of photoperiod and pinalectomy. Gen. Comp. Endocrinol. 57: 397–404.
- KULCZYKOWSKA, E. 1995. Solid-phase extraction of arginine vasotocin and isotocin in fish samples and subsequent gradient reversed-phase HPLC separation. J. Chromatogr. B Biomed. Appl. 673: 289–293.
- 3. GREENLEY G.H. JR., MORRIS M., ELDRIDGE J.O. & KIZER J.C. 1982. A diurnal plasma vasopressin rhythm in rats. Life Sci. **31:** 2843–2846.
- 4. WINDLE, R.J., FORSLING, M.L. & GUZEK, J.W. 1992. Daily rhythms in the hormone content of the neurohypophysial system and release of oxytocin and vasopressin in the male rat: Effect of constant light. J. Endocrinol. **133**: 283–290.