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REVIEW ARTICLE

Melatonin and Environmental Factors in the Reproductive Biology of Amphibians and Reptiles

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Abstract

In wild vertebrates, reproductive cycles are regulated by environmental factors such as photoperiod and temperature, both of which influence the neuroendocrine pathways of their reproductive biology. This article explores how these signals are transduced through the pineal-hypothalamus-pituitary-gonadal axis in amphibians and reptiles, with an emphasis on the role of melatonin in the physiological responses of both males and females. The potentially paradoxical effects of melatonin on the reproductive biology of in amphibians and reptiles, compared to mammals, reveal the complexity of reproductive processes in wild vertebrates. Therefore, there is a pressing need for holistic eco-physiological studies that incorporate biochemical, neuroendocrine, and genomic approaches, among others, to enrich traditional ecological-descriptive research.

Keywords: Melatonin, Pineal, Environment, Amphibians, Reptiles, Biology, Reproduction.

1. Introduction

The ontogeny of vertebrates is intricately influenced by environmental factors such as photoperiod and ambient temperature (Helm et al., 2013). Among the molecules that have evolved to mediate thermoregulation and various physiological processes, melatonin stands out for its multifaceted roles in animal biology. In this context, melatonin is considered a pleiotropic molecule due to its remarkable evolutionary history, which likely includes its involvement in the evolution of both prokaryotic and eukaryotic cells. It is biosynthesized by a wide range of organisms—including bacteria, fungi, plants, and animals—where it functions as a regulator of diverse physiological processes (Zhao et al., 2019; Banerjee et al., 2021; Andronachi et al., 2025).

Melatonin is an indoleamine synthesized from the amino acid tryptophan, which is first converted into serotonin and subsequently into melatonin. In vertebrates, although primarily produced in the pineal gland, melatonin is also synthesized in extrapineal tissues including the retina, skin, gastrointestinal tract, among other tissues and organs (Hardeland 2017).

Efforts to develop compounds that restore skin pigmentation in conditions such as vitiligo led Lerner and colleagues to isolate and chemically synthesize melatonin in 1958 and 1960, confirming its biological effects. However, systematic investigations into

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pineal-derived substances and their influence on pigmentation trace back to MacCord and Allen's pioneering work in 1917. In their seminal paper, they reviewed earlier findings and initiated studies in 1915 on the effects of pineal gland extracts on growth, differentiation, and pigmentation in in three species of anurans (*Rana pipiens*, *R. cantabridgensis* and *Bufo americana*), a fish (*Fundulus* sp.) and a squid (*Loligo* sp.), particularly through their impact on melanophores. Their experiments revealed that varying concentrations of bovine pineal extracts rendered amphibian skin translucent, which later returned to its natural dark coloration.

Over four decades later, Lerner and colleagues successfully isolated the bioactive compound responsible for amphibian skin depigmentation from more than 40,000 bovine pineal glands (approximately 100 kg). They named the indoleamine melatonin—with "mela" referencing its action on melanophores and "tonin" denoting its biochemical origin from serotonin (Reiter et al., 2018).

Beyond its role in pigmentation, melatonin is a potent free radical scavenger and regulator of antioxidant enzymes (Reiter et al., 2016; Banerjee et al., 2021). It also plays a role in reproductive physiology. As an amphipathic molecule, melatonin readily crosses cellular and organelle membranes—including those of mitochondria and the nucleus—allowing it to exert effects independently of receptor mediation. Nonetheless, melatonin also acts through specific receptors (Mel1a, Mel1b y Mel1c for non-mammalian vertebrate; Dubocovich et al., 2010; viviD & Bentley, 2018; Gao et al., 2022), whose spatial and temporal distribution is evolutionarily conserved and linked to the phylogeny of their encoded amino acid sequences, as observed in vertebrate genomes (Li et al., 2013; Denker et al., 2019; Maugars et al., 2020). The presence of melatonin receptors has been demonstrated in both amphibians and reptiles (*amphibians*: Wiechmann et. al., 1986; Wiechmann & Wirsig-Wiechmann, 1993; Tavolaro et al., 1995; Isorna et al., 2004, 2005; *reptiles*: Rivkees et al., 1990; Wiechmann & Wirsig-Wiechmann, 1992, 1994).

2. Melatonin Production and Secretion

In amphibians and reptiles, as in other vertebrates, the light perceived by the eyes—and in some species, by the pineal or parietal organ (Salame-Méndez & Serrano, 2023)—initiates the production and secretion of indoleamines, including melatonin (Besharse & Iuvone, 1983; Skene et al., 1991; d'Istria & Monteleone, 1993; Delgado et al., 1993; Faillace et al., 1995; Falcón et al., 2009). Upon reaching the retina, light triggers electrical impulses that prompt photoreceptors to release neurotransmitters. These signals are transmitted to the suprachiasmatic nucleus of the hypothalamus, the central circadian pacemaker. Neural impulses then travel to the superior cervical ganglion, where neurotransmitter release elicits physiological responses. Postganglionic fibers project to the pineal gland, forming synapses with pinealocytes. At these synapses, dopaminergic and adrenergic neurotransmitters—particularly norepinephrine—modulate melatonin synthesis and secretion.

During the dark phase (scotophase), norepinephrine binds to β-adrenergic receptors on pinealocytes, stimulating the enzymatic conversion of serotonin (5-hydroxytryptamine, 5-HT) into melatonin. This biotransformation involves two key enzymes: N-acetyltransferase (NAT) and hydroxyindole-O-methyltransferase (HIOMT). In contrast, during the light phase (photophase), serotonin biosynthesis is enhanced via tryptophan hydroxylase, converting tryptophan into 5-HT. Notably, HIOMT activity remains relatively stable throughout the photophase, whereas NAT activity exhibits a marked increase during the scotophase. NAT functions as a circadian regulator, effectively serving as a biological clock that governs melatonin synthesis and secretion by the pineal gland (Simonneaux & Ribelayga, 2003).

Melatonin production during the scotophase is thus tightly regulated by the daily light–dark cycle through NAT activity. Melatonin secretion may follow at least three distinct temporal patterns: i) a gradual increase during the second half of the scotophase; ii) initiation at the onset of darkness, peaking mid-scotophase, and declining before the photophase begins, iii) a rapid rise immediately after light cessation, with sustained high concentrations throughout the scotophase until shortly before light onset.

3. Effects of Melatonin in Amphibians and Reptiles

Melatonin secretion is influenced by both environmental and body temperature (Reiter 1986; Filadelfi & Castrucci, 1996; Tosini 1997; Lutterschmidt et al., 2003). As a potent antioxidant (Banerjee et al., 2021), melatonin contributes to metabolic regulation under low-temperature conditions, acting as an orexigenic agent (Hattori & Suzuki, 2024). During nighttime or hibernation, it mitigates ischemic physiological states and modulates metabolic capacity (Lutterschmidt et al., 1997; Dun-Xian et al., 2005). Hypothalamic

neuropeptides with orexigenic effects also participate in energy balance and thermoregulation (Chartrel et al., 2003; Navarro et al., 2006; Vigo et al., 2007).

Two key aspects of melatonin function warrant attention. First, melatonin inhibits thyroid activity via thyrotropin-releasing hormone (TRH) (Sarkar et al., 1997) and suppresses adrenocorticotropic hormone (ACTH) by enhancing corticosteroid secretion, which in turn inhibits ACTH through negative feedback. In amphibians, this may influence metamorphosis (Wright 2002; Wright & Bruni, 2004). Second, melatonin negatively impacts reproductive biology (*amphibians*: Kupwade & Saidapur, 1986; Joshi & Udaykumar, 2000; *reptiles*: Haldar-Misra & Thapliyal, 1981; Underwood 1985). Both *in vivo* and *in vitro* studies show that melatonin—alone or in combination with other methoxyindoles and neuropeptides such as arginine vasotocin (AVT)—can inhibit GnRH and gonadotropins secretion, impairing gonadal function in both sexes by reducing gamete production and steroidogenesis, this inhibitory effect is also temperature-dependent (*amphibians:* Delgado et al., 1983, 1992; Kupwade & Saidapur, 1986; Alonso-Bedate et al., 1988, 1990; Paniagua et al., 1990; de Atenor et al., 1994; Pancharatna & Patil, 1997; Udaykumar & Joshi, 1997; d'Istria et al., 2003, 2004; Izzo et al., 2004; Lutterschmidt & Wilczynski, 2012; *reptiles*: Licht 1966; Marion 1970; Haldar-Misra & Thapliyal, 1981; Haldar & Pandey, 1988, 1989; Lutterschmidt et al., 2002).

Melatonin's direct inhibitory effects on the gonads involve alterations in both germinal and interstitial tissues. In interstitial tissue, melatonin suppresses steroidogenesis (Mendieta et al., 1991; d'Istria et al., 2004; Yu et al., 2018; Cipolla-Neto et al., 2022), however, melatonin can also promote the production of androgens (Shankey et al., 2024). In germinal tissue, melatonin binds to estrogen receptors—such as those in spermatocytes and oocytes—blocking estradiol (E_2) from regulating cell maturation (O'Donnell et al., 2001; Carreau et al., 2003; Izzo et al., 2004).

Pineal melatonin secretion may also cease due to desensitization or refractoriness to photoperiod duration. Under short-day conditions, prolonged melatonin production (e.g., temporal pattern type iii) suppresses its own function via: 1) reduced receptor number and/or saturation in neural and gonadal tissues (e.g., hypothalamus), and 2) neurotransmitter and/or receptor turnover (e.g., adrenergic). These phenomena mimic spring–summer conditions with extended daylight ("summer effect"). Conversely, brief melatonin exposure during long days reactivates its function, simulating autumn–winter conditions with shorter daylight ("winter effect") (Dunlap 1993; Reppert & Weaber, 2001). Additionally, melatonin counteracts the gonadal stimulatory effects of broad-spectrum light (e.g., red light), while continuous darkness (e.g., blindness) induces a desensitizing effect on gonads (Joshi & Udaykumar, 2000; Hayasaka et al., 2002; Chiba et al., 2005).

Melatonin production, secretion, and function are regulated by at least two mechanisms: *external:* involving the retina (Cahill & Besharse, 1993; Sakamoto et al., 2000; Mangel 2001; Tosini & Fukuhara, 2002; Ribelayga et al., 2004) and pineal gland (Reiter 1991), and *endogenous*: including: I) NAT activity; II) melatonin feedback (positive or negative) on the pineal gland; III) body temperature; IV) receptor number and/or saturation, and V) neurotransmitter and/or receptor turnover in the pineal gland. Together, these two "clocks" regulate the pineal response to light and temperature, potentially influencing gonadal clock genes and signaling molecules (Della-Ragione et al., 2005).

These mechanisms are essential for understanding the complex reproductive events in amphibians and reptiles—and other vertebrate classes—linked to pineal gland function and melatonin. For example, some urodeles and anurans undergo hibernation, yet their gonadal activity remains functional (e.g., *Rana catesbeiana*), albeit reduced compared to the reproductive season. If melatonin production is more efficient during low-light periods and melatonin negatively affects gonadal function, why do gonads remain active in these species?

Gonadal function may be regulated endocrinologically, autocrinally, and/or paracrinally. Opioid-like molecules—such as enkephalins and endorphins—can regulate hypothalamic GnRH (also temperature-dependent; Minucci et al., 1986; Porter & Licht, 1986) and act on interstitial cells with steroidogenic activity (Facchinetti et al., 1993; Hammouche et al., 2009). Thus, steroidogenesis may be stimulated paracrinally via opioids or endocrinologically via gonadotropins induced by opioid-regulated GnRH, resulting in sex steroid synthesis (e.g., E_2). If estradiol displaces melatonin from its receptor, it may restore gametogenesis regulation in both sexes.

This model provides a valuable framework for studying seasonal reproductive neuroendocrinology in poikilotherms. If melatonin suppresses gonadal activity, why are these species reproductively active during hibernation or autumn–winter? Another

intriguing question involves the pineal–thyroid relationship. If melatonin inhibits thyroid function, what is its role in the neuroendocrine regulation of thyroid hormones during metamorphosis?

Environmental factors such as light and temperature clearly influence vertebrate reproductive biology, with direct implications for species survival. While ecological adaptation is evident, the underlying physiological and neuroendocrine mechanisms are more complex. If the pineal gland functions as a neuroendocrine transducer of environmental cues, and melatonin regulates gonadal function, then in some species, the environment–reproduction relationship is well aligned. Species that reproduce in spring–summer exhibit gonadal recrudescence (increased gonadosomatic index), whereas in autumn–winter, gonads regress. Thus, the pineal gland may contribute to population survival by suppressing reproduction during unfavorable seasons.

However, this correlation does not apply universally. Species that reproduce in autumn–winter challenge this model. Therefore, extrapolating reproductive biology findings across species—or even populations—is unreliable. Despite shared processes, physiological and neuroendocrine traits vary by species and sex, and outcomes may be paradoxical or inconsistent.

For example, in adult male water snakes (*Natrix piscator*), pineal activity—assessed via gland weight, histomorphology, and circulating melatonin—peaks during long daylight periods (spring–summer), while gonads remain inactive. Conversely, during short daylight periods (autumn–winter), pineal activity declines, yet gonads are active. This suggests that reproductive activity may be more influenced by temperature than by photoperiod-mediated melatonin, although melatonin does exert a negative effect on testicular function (Haldar & Pandey, 1988, 1989). And in males of the lizard *Calotes versicolor*, after removal of their pineal gland (pinealectomy), they had an early testicular recrudescence, with spermatogenesis maintained for 11 months (Haldar & Thapliyal, 1977). Therefore, these examples, like others (Mayer et al., 1997), would suggest that the pineal-melatonin relationship does not play an important role in reproductive biology in non-mammalian vertebrates.

4. Environmental Signal Transduction Involving the Pineal–Hypothalamic–Pituitary–Gonadal Axis

As previously discussed, reproductive events in vertebrates are modulated by epigenetic factors such as light and temperature (Visser et al., 2010). Seasonal interruption of reproductive function—referred to as dormancy or anestrus in females—is a survival strategy closely linked to photoperiod and nutritional status. The seasonal reproductive cycle, which follows a circannual rhythm with alternating periods of short and long days, parallels the onset of sexual maturity (i.e., puberty in mammals; Nabi et al., 2015). Seasonal reproductive suppression is characterized by hypothalamic dormancy in gonadotropin-releasing hormone (GnRH) secretion, which resumes during a specific window of the reproductive season. Once GnRH reaches a threshold concentration, it stimulates the adenohypophysis to release gonadotropins—follicle-stimulating hormone (FSH) and luteinizing hormone (LH). The sequential action and concentration of these hormones regulate key physiological processes in the ovaries and testes, including gametogenesis and steroidogenesis.

But how is photoperiod transduced into a physiological signal? Light stimulation of the retina initiates photoreception, which is transmitted via neural pathways to the suprachiasmatic nucleus (SCN) of the hypothalamus. From the SCN, neural signals project to the superior cervical ganglion, where presynaptic neurons activate postsynaptic neurons that synapse with inhibitory neurons. These inhibitory neurons contact pinealocytes in the pineal gland, suppressing melatonin secretion. As previously noted, melatonin is synthesized and secreted only during the scotophase of the circadian cycle. Therefore, during the time (hours) of light being recorded via the retina, it activates the excitatory neural pathway at the level of the pineal gland where the inhibitory neurons continue firing, inhibiting the release of melatonin from the pinealocytes. In contrast, during darkness, retinal neurons cease firing, significantly reducing inhibition of pinealocytes and allowing melatonin synthesis and secretion to proceed. Melatonin inhibits GnRH secretion, which prevents the release of gonadotropins and suppresses gonadal function.

Below, and as an example, the pineal-hypothalamus-pituitary-gonad (testicle and ovary) (PHPG) axis is described in an ideal male and female.

In males, GnRH secreted by the hypothalamus binds to receptors on adenohypophyseal gonadotropes, stimulating LH and FSH secretion. LH acts on Leydig cells in the testes, promoting testosterone (T) synthesis via the conversion of cholesterol into pregnenolone. Elevated T levels support intratesticular

concentrations essential for spermatogenesis and enter systemic circulation. Testosterone diffuses into vascular capillaries, binds to androgen-binding protein (ABP), and is transported to Sertoli cells and through testicular vessels into the bloodstream. T then reaches the hypothalamus, exerting negative feedback on GnRH-secreting neurons and desensitizing LH secretion in the adenohypophysis, thereby reducing further T synthesis.

FSH binds to membrane receptors on interstitial cells, promoting their maturation and androgen production. In Sertoli cells, FSH stimulates the synthesis of ABP, inhibin, 5α-dihydrotestosterone (DHT), and estradiol (E_2) via interstitial T. Inhibin circulates systemically to inhibit FSH secretion in the adenohypophysis. DHT and E_2, upon reaching the hypothalamus, suppress GnRH secretion. Thyroid hormones counteract this inhibition by desensitizing the GnRH-releasing mechanism.

In seasonally reproductive males, such as those active during autumn–winter when daylight hours are reduced, pinealocytes synthesize and secrete increased melatonin. This inhibits GnRH secretion, triggering negative regulation of the hypothalamic–pituitary–testicular axis.

In females, during spring–summer, reduced melatonin secretion permits pulsatile or tonic GnRH release from the hypothalamus. Increased GnRH frequency stimulates LH and FSH secretion. Estradiol exerts negative feedback on GnRH and LH release at the hypothalamic and adenohypophyseal levels, respectively, while inhibin suppresses FSH secretion. Following ovulation, residual E_2 and rising progesterone (P_4) levels enhance neurotransmitter-mediated (e.g., norepinephrine) GnRH secretion. E_2 sensitizes gonadotropes to releasing factors that stimulate LH secretion. LH then acts on residual follicular cells forming the corpus luteum, promoting further P_4 production. Together, P_4 and E_2 exert a positive feedback effect on GnRH secretion.

5. Conclusion

The *paradoxical* results of melatonin function in amphibians and reptiles with respect to, for example, that described in mammals (Reiter et al., 2018) are not exclusive to the pineal gland, but are common to several processes involved in the neuroendocrinology of the reproductive biology of wild vertebrates. These findings underscore the persistent challenge of interpreting complex physiological interactions and highlight the need for systematic, multidisciplinary investigations. Integrative approaches—particularly those combining biochemical and genomic methodologies—are essential to deepen our understanding and to complement traditional ecological and descriptive studies. Such strategies will provide a more comprehensive framework for elucidating the complex reproductive mechanisms of the species under research.

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6. References

1. Alonso-Bedate, M., Delgado, M. J. & Carballada, R. (1988). In vivo effect of melatonin and gonadotropin-releasing hormone on testicular function in *Rana temporaria*. *Journal of Pineal Research*, 5(4): 323-3332. doi: 10.1111/j.1600-079x.1988.tb00881.x
2. Alonso-Bedate, M., Carballada, R. & Delgado, M. J. (1990). Effects melatonin on gonadal steroids and glucose plasma levels in frogs (*Rana perezi* and *Rana temporaria*). *Journal of Pineal Research*, 8(1): 79-89. doi: 10.1111/j.1600-079x.1990.tb00809.x
3. Andronachi, V. -C., Simeanu, C., Matei, M., Radu-Rusu, R. -M. & Simeanu, D. (2025). Melatonin: An overview on the synthesis processes and on its multiple bioactive roles played in animals and humans. *Agriculture*, 15: 273. doi: doi.org/10.3390/agricultura15030273
4. Banerjee, A., Chattopadhyay, A. & Bandyopadhyay, D. (2021). Melatonin: an ancient note in a contemporary wrap. *Melatonin Research*, 4(3): 453-478. doi: 10.32794/mr112500105
5. Besharse, J. C. & Iuvone, P. M. (1983). Circadian clock in *Xenopus* eye controlling retinal serotonin *N*-acetyltransferase. *Nature*, 305:133-135. doi: 10.1038/305133a0
6. Cahill, G. M. & Besharse, J. C. (1993). Circadian clock functions localized in *Xenopus* retinal photoreceptor. *Neuron*, 10:573-577. doi: 10.1016/0896-6273(93)90160-s
7. Carreau, S., Lambard, S., Delalande, C., Denis-Galearaud, I., Bilinska, B. & Bourguiva, S. (2003). Aromatase expression and role of estrogens in male gonad: a review. *Reproductive Biology and Endocrinology*, 35:1-6. doi: 10.1186/1477-7827-1-35
8. Chartrel N., Dujardin, C., Anouar, Y., Leprince, J., Decker, A., Clerens, S., Do-Régo, S. J., Vandesande, F., Llorens-Cortes, C., Costentin, J., Beauvillain, J.

C. & Vaudry, H. (2003). Identification of 26RFa, a hypothalamic neuropeptide of the RFamide peptide family with orexigenic activity. *Proceedings of the National Academy of Sciences*, 100:15247-15252. doi: 10.1073/pnas.2434676100

9. Chiba A., Hattori, A. & Iigo, M. (2005). Daily and circadian variations of the pineal and ocular melatonin contents and their contributions to the circulating melatonin in the Japanese newt, *Cynops pyrrhogaster*. *Zoological Science*, 22(1): 65-70. doi: 10.2108/zsj.22.65

10. Cipolla-Neto, J., Amaral, F. G., Soares, J. M. Jr., Gallo, C. C., Furtado, A., Cavaco, J. E., Gonçalves, I., Santos C. R. A. & Quintela, T. (2022). The crosstalk between melatonin and sex steroid hormones. *Neuroendocrinology*, 112(2): 115-129. doi: 10.1159/000516148

11. d´Istra, S. M. & Monteleone, P. (1993). A comparative study of melatonin production in retina, pineal gland and Harderian gland of *Bufo viridi*s and *Rana esculenta*. *Comparative Biochemistry and Physiology Part C*, 106(1): 189-193. doi: 10.1016/0742-8413(93)90271-l

12. d´Istria, M., Palmiero, C., Serino, I., Izzo, G. & Minucci, S. (2003). Inhibition of the basal and oestradiol-stimulated mitotic activity of primary spermatogonia by melatonin in the testis of the frog, *Rana esculenta*, in vivo and in vitro. *Reproduction*, 126(1): 83-90. doi: 10.1530/rep.0.1260083

13. d´Istria, M., Serino, I., Izzo, G., Ferrara, D., De Rienzo, G. & Minucci, S. (2004). Effects of melatonin treatment on Leydig cell activity in the testis of the frog *Rana esculenta*. *Zygote*, 12(4): 293-299. doi: 10.1017/s0967199404002898

14. de Atenor, M. S. B., De Romero, I. R., Brauckmann, E., Pisanó, A. & Legname, A. H. (1994). Effects of pineal gland and melatonin on the metabolism of oocytes in vitro and ovulation in *Bufo arenarum*. *Journal of Experimental Zoology*, 268(6): 436-441. doi: 10.1002/jez.1402680604

15. Delgado, M. J., Alonso-Gómez, A. L. & Alonso-Bedate, M. (1992). Role of environmental temperature and photoperiod in regulation of seasonal testicular activity in the frog, *Rana perezi*. *Canadian Journal of Physiology and Pharmacology*, 70(10): 1348-1352. doi: 10.1139/y92-189

16. Delgado, M. J., Gutiérrez, P., and Alonso-Bedate., M. (1983). Effects of daily melatonin injections on the photoperiodic gonadal response of the female frog *Rana ridibunda*. *Comparative Biochemistry and Physiology A*, 76(2): 389-392. doi: 10.1016/0300-9629(83)90343-2

17. Delgado, M. J., Alonso-Gomez, A. L., Gancedo, B., de Pedro, N., Valenciano, A. I. & Alonso-Bedate, M. (1993). Serotonin *n*-acetyltransferase (NAT) activity and melatonin levels in the frog retina are not correlated during the seasonal cycle. *General and Comparative Endocrinology*, 92(2): 143-150. doi: 10.1006/gcen.1993.1151

18. Della-Ragione, F., Comitato, R., Angelini, F., D'Esposito, M. & Cardone, A. (2005). Molecular cloning and characterization of the clock gene period 2 in the testis of lizard *Podarcis sicula* and its expression during seasonal reproductive cycle. *Gene*, 363:105-115. doi: 10.1016/j.gene.2005.08.018

19. Denker, E., Ebbesson, L. O. E., Hazlerigg, D. G. & Macqueen, D. J. (2019). Phylogenetic reclassification of vertebrate melatonin receptors to include Mel1d. *G3 (Bethesda)*, 9(10): 3225-3238. doi: 10.1534/g3.119.400170

20. Dubocovich, M. L., Delagrange, P., Krause, D. N., Sugden, D., Cardinali, D. P. & Olcese, J. (2010). International Union of Basic and Clinical Pharmacology. LXXV. Nomenclature, classification, and pharmacology of G protein-coupled melatonin receptors. *Pharmacological Reviews*, 62(3): 343-380. doi:10.1124/pr.110.002832.

21. Dunlap, J. C. (1993). Genetic analysis of circadian clocks. *Annual Review of Physiology*, 55:683 728. doi: 10.1146/annurev.ph.55.030193.003343

22. Dun-Xian T., Manchester, L. C., Sainz, R. M., Mayo, J. C., León, J. & Reiter, R. J. (2005). Physiological ischemia/reperfusion phenomena and their relation to endogenous melatonin production. *Endocrine*, 27(2): 149-157. doi: 10.1385/endo:27:2:149

23. Facchinetti, F., Genazzani, A. R., Vallarino, M., Pestarino, M., Polzonetti-Magni, A., Carnevali, O., Ciarcia, G., Fasano, S., D'Antonio, M. & Pierantoni, R. (1993). Opioids and testicular activity in the frog, *Rana esculenta*. *Journal of Endocrinology*, 37(1):49-57. doi: 10.1677/joe.0.1370049

24. Faillace, M. P., Cutrera, R., Sarmiento, M. I. & Rosenstein, R. E. 1995. Evidence for local synthesis of melatonin in golden hamster retina. *NeuroReport*, 6(15): 2093-2095. doi: 10.1097/00001756-199510010-00033

25. Falcón, J., Besseau, L., Fuentès, M., Sauzet, S., Magnanou, E. & Boeuf, G. (2009). Structural and functional evolution of the pineal melatonin system in vertebrates. *Annals of the New York Academy of Science*, 1163: 101-111. doi: 10.1111/j.1749-6632.2009.04435.x

26. Filadelfi, A. M. & Castrucci, A. M. (1996). Comparative aspects of the pineal/melatonin system

of poikilothermic vertebrates. *Journal of Pineal Research*, 20(4): 175-186. doi: 10.1111/j.1600-079x.1996.tb00256.x

27. Gao, Y., Zhao, S., Zhang, Y. & Zhang, Q. (2022). Melatonin receptors: A key mediator in animal reproduction. *Veterinary Sciences*, 9(7): 309. doi: 10.3390/vetsci9070309

28. Haldar, C. & Thapliyal, J. P. (1977). Effect on pinealectomy on the annual testicular cycle of *Calotes vesrsicolor*. *General and Comparative Endocrinology*,32(4):395-399.doi.org/10.1016/0016-6480(77)90219-2

29. Haldar, C. & Pandey, R. (1988). Effect of melatonin and 5-methoxytryptamine administration on the testis and pineal gland activity of the fresh-water snake *Natrix piscator*. *Archives d´Anatomie, d´Histologie et d´Embryologie Normales et Experimentales*, 71: 85-96. PMID: 3272549

30. Haldar, C. & Pandey, R. (1989). Effect of pinealectomy on annual testicular cycle of Indian chequered water snake, *Natrix piscator*. *General and Comparative Endocrinology*, 76(2): 214-222. doi: 10.1016/0016-6480(89)90152-4

31. Haldar-Misra, C. & Thapliyal, J. P. (1981). Response of reptilian gonad to melatonin. *Neuroendocrinology*, 33(6): 328-332. doi: 10.1159/000123256

32. Hammouche, S., Gernigons, T. & Exbrayat, J. M. (2009). Correlation between ovarian steroidogenesis and β-endorphin in the lizard *Uromastyx acanthinura*: Immunohistochemical approach. *Folia Histochemica et Cytobiologica*, 47(5): S95-S100. doi: 10.2478/v10042-009-0050-y

33. Hardeland, R. (2017). Melatonin-more than just a pineal hormone. *Biomedical Journal of Scientific and Technical Research*, 1(4): 994-997. DOI: 10.26717/BJSTR.2017.01.000351

34. Hattori, A. & Suzuki, N. (2024). Receptor-mediated and receptor-independent actions of melatonin in vertebrates. *Zoological Science*, 41:105–116. doi.org/10.2108/zs230057

35. Hayasaka, N., LaRue, S. I. & Green, C. B. (2002). In vivo disruption of *Xenopus* clock in the retinal photoreceptor cells abolishes circadian melatonin rhythmicity without affecting its production levels. *Journal of Neuroscience*, 22(5):1600-1607. doi: 10.1523/JNEUROSCI.22-05-01600.2002

36. Helm, B., Ben-Shlomo, R., Sheriff, M. J., Hut, R. A., Foster, R., Barnes, B. M. & Dominoni, D. (2013). Annual rhythms that underlie phenology: biological time-keeping meets environmental change. *Proceedings of Royal Society B*, 280: 20130016. http://dx.doi.org/10.1098/rspb.2013.0016

37. Isorna, E., Delgado, M. J., Guijarro, A. I., López-Patiño, M. A., Alonso-Bedate, M. & Alonso-Gómez, A. L. (2004). 2-[^{125}I]-Melatonin binding sites in the central nervous system and neural retina of frog *Rana perezi* regulation by light and temperature. *General and Comparative Endocrinology*, 139(2): 95-102. doi: 10.1016/j.ygcen.2004.07.015

38. Isorna E., Guijarro, A. I., Delgado, M. J., López-Patiño, M. A., Pedro, Nd. & Alonso-Gómez, A. L. (2005). Ontogeny of central melatonin receptors in tadpoles of the anuran *Rana perezi*: modulation of dopamine release. *Journal of Comparative Physiology A*, 191: 1099-1105. doi 10.1007/s00359-005-0032-2

39. Izzo, G., d´Istria, M., Serino, I. & Minucci, S. (2004). Inhibition of the increased 17beta-estradiol-induced mast cell number by melatonin in the testis of the frog *Rana esculenta*, in vivo and in vitro. *Journal of Experimental Zoology*, 207(Pt3):437-441. doi: 10.1242/jeb.00786

40. Joshi, B. N. & Udaykumar, K. (2000). Melatonin counteracts the stimulatory effects of blinding or exposure to red light on reproduction in the skipper frog *Rana cyanophlyctis*. *General and Comparative Endocrinology*, 118(1): 90-95. doi: 10.1006/gcen.1999.7443

41. Klein, D. C., Auerbach, D. A. & Weller, J. L. (1981). Seesaw signal processing in pineal cells: homologous sensitization of adrenergic stimulation of cyclic GMP accompanies homologous desensitization of beta-adrenergic stimulation of cyclic AMP. *Proceedings of the National Academy of Sciences*, 78(7): 4625-4629. doi: 10.1073/pnas.78.7.4625

42. Kupwade, V. A. & Saidapur, S. K. (1986). Effect of melatonin on oocyte growth and recruitment, hypophyseal gonadotrophs, and oviduct of the frog *Rana cyanophlyctis* maintained under natural photoperiod during the prebreeding phase. *General and Comparative Endocrinology*, 64(2): 284-292. doi: 10.1016/0016-6480(86)90015-8

43. Lerner, A. B., Case, J. D. & Takahashi, Y. (1958). Isolation of melatonin, the melatonin gland factor that lightens melanocytes. *Journal of the American Chemical Society*, 80: 2587. doi: 10.1021/ja01543a060

44. Lerner, A. B., Case, J. D. & Takahashi, Y. (1960). Isolation of Melatonin and 5-Methoxyindole-3-acetic acid from bovine pineal glands. *Journal of Biological Chemistry*, 235(7): 1992-1997. PMID: 14415935

45. Li, D. Y., Smith, D. G., Hardeland, R., Yang, M. Y., Xu, H. L., Zhang, L., Yin, H. D., Zhu, Q. (2013). Melatonin receptor genes in vertebrates. *International Journal of Molecular Sciences*, 14(6): 11208-11223. doi: 10.3390/ijms140611208.

46. Licht, P. (1966). Reproduction in Lizards: Influence of temperature on photoperiodism testicular recrudescence. *Science*, 154: 1668-1670. doi: 11.1126/science.154.3757.1668

47. Lutterschmidt, D. I., Lutterschmidt, W. I. & Hutchinson, V. H. (1997). Melatonin and chlorpromazine: Melatonin and chlorpromazine: Therma selection and metabolic rate in the bullsnake, *Pituophis melanoleucus*. *Comparative Biochemistry and Physiology C*., 118(3): 271-277. doi: 10.1016/S0742-8413(97)00106-0

48. Lutterschmidt, D. I., Lutterschmidt, W. I., Ford, N. B. & Hutchison, V. H. (2002). Behavioral thermoregulation and the role of melatonin in a nocturnal snake. *Hormones and Behavior*, 41(1): 41-50. doi: 10.1006/hbeh.2001.1721

49. Lutterschmidt, D. I., Lutterschmidt, W. I. & Hutchison, V. H. (2003). Melatonin and thermoregulation in ectothermic vertebrates: a review. *Canadian Journal of Zoology*, 81(1): 1-13. doi: 10.1139/Z02-189

50. Lutterschmidt, D. I. & Wilczynski, W. (2012). Sexual dimorphic effect of melatonin on brain arginine vasotocin immunoreactivity in green treefrog (*Hyla cinerea*). *Brain Behavior and Evolution*, 80(3): 222-232. doi: 10.1159/000341238,

51. Mangel, S. C. (2001). Circadian clock regulation of neural light responses in the vertebrate retina. *Progress in Brain Research*, 131: 505-518. doi: 10.1016/s0079-6123(01)31040-3

52. Marion, K R. (1970). Temperature as the reproductive cue for the female fence lizard *Sceloporus undulatus*. *Copeia*, 1970(3): 562-564. https://doi.org/10.2307/1442286.

53. Maugars, G, Nourizadeh-Lillabardi, R. & Fin-Arne, W. (2020). New insights into the evolutionary history of melatonin receptors in vertebrates, with particular focus on teleosts. *Frontiers in Endocrinology*, 11: 538196. doi: 10.3389/fendo.2020.610274

54. Mayer, I., Bornestaf, C. & Borg, B. (1997). Melatonin in non-mammalian vertebrate: Physiological role in reproduction? *Comparative Biochemistry and Physiology Part A*, 118(3): 515-531. 10.1016/s0300-9629(96)00468-9

55. McCord, C. P. & Allen, F. P. (1917). Evidence associating pineal gland function with alterations in pigmentation. *Journal of Experimental Zoology*, 23(1): 207-224. doi: 10.1002/jez.1400230108

56. Mendieta, E., Salame, A, Herrera, J. & Antón-Tay, F. (1991). Melatonin inhibition of androgen biosynthetic pathway in Leydig cell (LC)-enriched cell fractions from normal adult rats. *Molecular Andrology*, 3: 319-329. https://www.researchgate.net/profile/Arturo-Mendez-4/publication/295126798

57. Minucci, S., Di Matteo, L., Pierantoni, R., Variale, B, Rastogi, R. K., Chieffi, G. (1986). In vivo and in vitro stimulatory effect of a gonadotropin-relasing hormone analog (HOE 766) on spermatogonial multiplication in the frog, *Rana sculenta*. *Endocrinology*, 119(2): 731-736. doi: 10.1210/endo-119-2-731

58. Nabi, G., Amin, M., Khan, A. & Kami, M. (2015). Endogenous signals and mammalian puberty onset: A review. *Journal of Biology and Life Science*, 6(1): 1-14. doi:10.5296/jbls.v6i1.6032

59. Navarro, V. M., Fernandez-Fernandez, R., Nogueiras, R., Vigo, E., Tovar, S., Chartrel, N., Le Marec, O., Leprice, J., Aguilar, E., Pinilla, L., Dieguez, C., Vaudry, H. & Tena-Sempere, M. (2006). Novel role of 26RFa, a hypothalamic RFamide orexigenic peptide, as putative regulator of the gonadotropic axis. *Journal of Physiology*, 573(Pt1): 237-49. doi: 10.1113/jphysiol.2006.106856

60. Pancharatna, K. & Patil, M. M. (1997). Role of temperature and photoperiod in the onset of sexual maturity in female frogs, *Rana cyanophlyctis*. *Journal of Herpetology*, 31(1): 111-114. doi: 10.2307/1565338

61. Paniagua, R., Fraile, B. & Sáez, F. J. (1990). Effects of photoperiod and temperature on testicular function in amphibians. *Histology and Histopathology*, 5(3): 365-378. PMID: 2134391

62. Porter, D. A. & Licht, P. (1986). Effects of temperature and mode of delivery on responses to gonadotropin-releasing hormone by superfused frog pituitaries. *General and Comparative Endocrinology*, 63(2): 236-44. doi: 10.1016/0016-6480(86)90161-9

63. Reiter, R. J. (1986). The pineal gland: An important link to the environment. *Physiology*, 1(6): 202-205. doi: 10.1152/physiologyonline.1986.1.6.202

64. Reiter, R. J. (1991). Pineal melatonin: cell biology of its synthesis and of its physiological interactions. *Endocrine Reviews*, 12(2): 151-80. doi: 10.1210/edrv-12-2-151

65. Reiter, R. J., Mayo, J. C., Tan, D. X., Sainz, R. M., Alatorre-Jimenez, M. & Qin, L. (2016). Melatonin as an antioxidant: under promises but over delivers. *Journal of Pineal Research*, 61(3): 253-278. doi: 10.1111/jpi.12360

66. Reiter, R. J., Dun-Xian, T & Ramaswamy, S. (2018). Historical perspective and evaluation of mechanisms by which melatonin mediates seasonal reproduction in mammals. *Melatonin Research*, 1(1): 59-77. doi: 10.32794/mr11250004

67. Reppert, S. M. & Weaver, D. R. (2001). Molecular analysis of mammalian circadian rhythms. *Annual Review of Physiology*, 63: 647-76. doi: 10.1146/annurev.physiol.63.1.647

68. Ribelayga, C., Wang, Y. & Mangel, S. C. (2004). A circadian clock in the fish retina regulates dopamine release via activation of melatonin receptors. *Journal of Physiology*, 554(Pt 2): 467-482. doi: 10.1113/jphysiol.2003.053710

69. Rivkees, S. A., Conron, R. W. Jr. & Reppert, S. M. (1990). Solubilization and purification of melatonin receptors from lizard brain. *Endocrinology*, 127(3): 1206-1214. doi: 10.1210/endo-127-3-1206.

70. Sakamoto, K., Oishi, K., Shiraishi, M., Hamano, S., Otsuka, H., Miyake, Y. & Ishida, N. (2000). Two circadian the mammalian retina. *Neuroreport*, 11(18): 3995-3997. doi: 10.1097/00001756-200012180-00018

71. Salame-Méndez, A. & Serrano, H. (2023). Neuro-endocrinology of amphibians and reptiles: An overview. *International Journal of Zoological Investigations*, 9(2): 1-32. doi: 10.33745/ijzi.2023.v09i02.001

72. Sarkar, S. N., Sarkar, K., Bhattacharyya, S. & Das, P. (1997). Melatonin action on thyroid activity in the soft-shelled turtle, *Lissemys punctata punctata*. *Folia Biologica*, 45(3-4): 109-12. PMID: 9643166.

73. Shankey, N. T., Igo, B. L., Grossen, T. L. & Cohen, R. E. (2024). Melatonin treatment during the breeding season increases testosterone in male green anole lizards (*Anolis carolinensis*). *Hormones and Behavior*, 166: 105655. doi: 10.1016/j.yhbeh.2024.105655

74. Simonneaux, V. & Ribelayga, C. (2003). Generation of the melatonin endocrine message in mammals: A review of the complex regulation of melatonin synthesis by norepinephrine, peptides, and other pineal transmitters. *Pharmacological Reviews*, 55(2): 325-395. doi: 10.1124/pr.55.2.2

75. Skene, D. J., Vivien-Roels, B. & Pevet, P. (1991). Day and nighttime concentrations of 5-methoxytryptophol and melatonin in the retina and pineal gland from different classes of vertebrates. *General and Comparative Endocrinology*, 84(3): 405-411. doi: 10.1016/0016-6480(91)90088-n

76. Tavolaro, R., Canonaco, M. & Franzoni, M. F. (1995). Comparison of melatonin-binding sites in the brain of two amphibians: an autoradiographic study. *Cell and Tissue Research*, 279: 613–617. doi: 10.1007/BF00318173

77. Tosini, G. (1997). The pineal complex of reptiles: physiological and behavioral roles. *Ethology Ecology and Evolution*, 9: 313-333. doi: 10.1080/08927014.1997.9522875

78. Tosini, G. & Fukuhara, C. (2002). The mammalian retina as a clock. *Cell and Tissue Research*, 309: 119-126. doi: 10.1007/s00441-002-0578-z

79. Udaykumar, K. & Joshi, B. N. (1997). Effect of exposure to continuous light and melatonin on ovarian follicular kinetics in the skipper frog, *Rana cyanophlyctis*. *Biological Signals*, 6(2): 62-66. doi: 10.1159/000109110

80. Underwood, H. (1985). Annual testicular cycle of the lizard *Anolis carolinensis*: effects of pinealectomy and melatonin. *Journal of Experimental Zoology*, 233(2): 235-242. doi: 10.1002/jez.1402330210

81. Vigo, E., Roa, J., Pineda, R., Castellano, J. M., Navarro, V. M., Aguilar, E., Pinilla, L. & Tena-Sempere, M. (2007). Novel role of the anorexigenic peptide neuromedin U in the control of LH secretion and its regulation by gonadal hormones and photoperiod. *American Journal of Physiology. Endocrinology and Metabolism*, 293(5): E1265-1273. doi: 10.1152/ajpendo.00425.2007

82. Visser, M. E., Caro, S. P., van Oers, K., Schaper, S. V. & Helm, B. (2010). Phenology, seasonal timing and circannual rhythms: towards a unified framework. *Philosophical Transactions of the Royal Society B*, 365: 3113–3127. doi:10.1098/rstb2010.0111

83. viviD, D. & Bentley, G. E. (2018). Seasonal reproduction in vertebrates: Melatonin synthesis, binding, and functionality using Tinbergen's four questions. *Molecules*, 23(3): 652. doi: 10.3390/molecules23030652

84. Wiechmann, A. F., Bok, D. & Horwitz, J. (1986). Melatonin-binding in the frog retina: autoradiographic and biochemical analysis. *Investigative Ophthalmology and Visual Science*, 27: 153-163. PMID: 3484734

85. Wiechmann, A. F. & Wirsig-Wiechmann, C. R. (1992). Asymmetric distribution of melatonin receptors in the brain of the lizard *Anolis carolinensis*. *Brain Research*, 593(2): 281-286. doi: 10.1016/0006-8993(92)91319-A

86. Wiechmann, A. F. & Wirsig-Wiechmann, C. R. (1993). Distribution of melatonin receptors in the brain of the frog *Rana pipiens* as revealed by *in vitro* autoradiography. *Neuroscience*, 52(2): 469-480. doi: 10.1016/0306-4522(93)90173-d

87. Wiechmann, A. F. & Wirsig-Wiechmann, C. R. (1994). Melatonin receptor distribution in the brain and retina of a lizard, *Anolis carolinensis*. *Brain, Behavior and Evolution*, 43(1): 26-33. doi: 10.1159/000113622

88. Wright, M. L. (2002). Melatonin, diel rhythms, and metamorphosis in anuran amphibians. *General and Comparative Endocrinology*, 126(3): 251-254. doi: 10.1016/s0016-6480(02)00012-6

89. Wright, M. L. & Bruni, N. K. (2004). Influence of the photocycle and thermocycle on rhythms of plasma thyroxine and plasma and ocular melatonin in late metamorphic stages of the bullfrog tadpole, *Rana catesbeiana*. *Comparative Biochemistry and Physiology*, 139(1): 33-40. doi: 10.1016/j.cbpb.2004.06.012

90. Yu, K., Shou-Long, D., Tie-Cheng, S., Yuan-Yuan, L. & Yi-Xun, L. (2018). Melatonin regulates the synthesis of steroids hormones on male reproduction: a review. *Molecules*, 23(2): 447. doi:10.3390/molecules23020447

91. Zhao, D., Yu, Y., Shen, Y., Liu, Q., Zhao, Z., Sharma, R. & Reiter, R. J. 2019. Melatonin synthesis and function: Evolutionary history in animals and plants. *Frontiers in Endocrinology*, 10: 249. doi: 10.3389/fendo.2019.00249