EFFECTS OF DIFFERENT LIGHT REGIMES ON PROLACTIN CONCENTRATION IN RAT

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ABSTRACT

Prolactin is a peptide hormone which is secreted from hypophysis. Prolactin concentration differs in male and female. Even though prolactin has lactotpope effect, it has disturbances effects on pulsatory secretion of gonadotropins as well as on ovulation. In spite of different plasma level of prolactin in different seasons, the effects of light regime on prolactin secretion still is unclear. Eighty Wistar rats were selected and divided into four groups. Each treatment was exposed in different photoperiod; natural light (control NL), 24 hours light (permanent light LL), 24 hours darkness (permanent darkness DD), and 12 hours light-12 hours darkness (light-dark LD). Blood samples from the heart were taken every 3 month during year. Prolactin concentration was determined using RIA. Data were analyzed using ANOVA and means were compared using Duncan multiple range test. Prolactin concentration in control (NL), dark (DD), light-dark (LD) and permanent light (LL) were 1.10, 1.00, 0.80 and 0.60 ng/ml, respectively. Light regime had a significant effect on prolactin concentration ($p<0.05$). Prolactin was significantly lower in permanent light than other treatments. Means of prolactin in permanent dark and light-dark weren't differ with control ($p>0.05$). Permanent light will reduce the prolactin level. It seems to be one of the causes of short estrous cycles in permanent light is prolactin levels. These finding can be used as a model in human societies.

Keywords: Photoperiods, Prolactin concentration, Lactogen, Mammotropin, Lactotropic hormone, Hypothalamus, Hypophysis, Metabolic performance, Rats

INTRODUCTION

Prolactin is a protein hormone with molecular weight 23000 Dalton. There are three forms of hormone monomer, dimer and multimer. The proportion of each depends on the physiologic, pathologic and hormonal stimulus. Lactogen, mammotropin and lactotropic hormone are the other names of prolactin. Its major role is to stimulate mammary glands. The mammary gland cells proliferated after prolactin injection in mouse (Nadrloo, 2006). Prolactin receptors were identified in liver, kidney, gonads and some other organ. The secretion of prolactin hormone is controlled by two hypothalamic inhibitory and releasing factors. In mammals prolactin secretion is controlled by prolactin inhibitory factor (PIF) and Dopamine.

During pregnancy, the level of prolactin severely increases in women, two to three weeks after pregnancy. Disorder in prolactin secretion is caused by the inhibitory or stimulatory factors of physiologic phases, issues related to hypothalamus, hypophysis, metabolic performance and pharmacological side effect of drugs. It is more related to the problems of hypophysis, microadenoma, macroadenoma and the manner of these diseases development. Amenorrhea (the absence of a menstrual period in a woman of reproductive age), hirsutism (excessive hairiness in women) and infertility are caused due to high levels of prolactin (Yang *et al.,* 1980). Pituitary tumor can increase prolactin concentration, milk let down and menstrual disorder. The hormone concentration in women is usually more than in men. In men, prolactin role is unknown, however high level of prolactin can be increase breast size and cause sexual disability. Suckling stimulates prolactin secretion. It seems that prolactin is involved in the expression of sexual activity. Physical activity can increase prolactin hormone. Prolactin has effect on ovulatory cycle in rats. The cyclic secretion of gonadotropin, estrogen and progesterone is considered as reproductive activities index. The non cyclic secretion of these hormones increased prolactin concentration in uterus secretions (Anisimov, 1971; Irod, 1966; Anisimov *et al.*, 2009). In some species e.g. human, prolactin inhibits LH secretion and ovulation. Hence, prolactin has antigonadotropic effect. In some animal species, milking delays the renewal of sexual cycle. Ovulation and pregnancy was delayed in suckling cows. The amount of prolactin secretion in 24 hours is pulsatory, and its concentration during the day is more than in the night. Photoperiod affects epiphysis and secretions from the suprachyasmatic part of the hypothalamus (Anisimov, 2007; Barbacka-Surowiak *et al.,* 2003; Nadrloo, 2006). Epiphysis can inhibit the prolactin biosynthesis through the production and secretion of melatonin (antagonist gonadotropin) (Chazov *et al.,* 1972). Furthermore, the epiphysis debilitates and inhibits the production and secretion of gonadotropins with melatonin (Lazarev *et al.,* 1976; Khavinson, 2002; Anisimov *et al.,* 2003). Synthetic increase of photoperiod for 2 – 4 hours prolonged the estrus cycle and in some cases may disorder the cycle. In rats, exposure to permanent light causes the rat to enter into obligatory estrus. A similar occurrence happens to women in different climatic zones. Permanent light regime disorders hormonal balance and fastens aging. Its key mechanism is because of change in hypothalamus sensitivity which inhibits estrogen secretion (Yang *et al.,* 1980; Dilman and Young, 1994; Van der Beek, 1996). Exposure to permanent light affected mobility and body temperature of rats (Depres-Brummer *et al.,* 1995) and disordered the estrus cycle (Flecknell, 1996). Light hour's reduction stimulates the production and secretion of melatonin (Baturin *et al.,* 2004), thus light regimes is of significant importance in reproductive physiology due to changes in the season (Cavchenko *et al.,* 1987). There are different photoperiods during the different phases of the year. In the southwest of Iran, light length comes up to 16 hours in summer. Some researches have been conducted on estrus cycle and melatonin concentration in different photoperiod (Van der Beek, 1996; Baturin *et al.,* 2004; Parta Lima *et al.,* 2004; Anisimov, 2007; Anisimov *et al.*, 2009). The effect of different photoperiods on prolactin secretion needs to be re-apprised in the light of recent progress made in science and technology where humans are near permanently exposed to light. This research investigated the effects of exposure to different photoperiod on prolactin concentration in rats.

MATERIALS AND METHODS

Permission to conduct this study using rat as an animal model was granted by the ethical and moral committee of the College of Medical Science, University of Lorestan. All ethical principles and considerations involving animal right, welfare, use and disposal of experimental animal were strictly adhered to during the study.

Eighty Wistar female rats obtained from the Animal House of Jondishapour Medical University (JMU) were used. They were kept 5 rats per stainless steel standard cages (replicate) at room temperature of 21 ± 2 °C.

All rats where watered and fed (pellet ration, Shar-e-Kord Animal Feed Industry, Iran) *ad libitum.* The rats were divided into four treatment groups, replicated four times. Rats were placed in different light conditions according to the treatments. The first treatment group rats (control) were exposed to natural light regime (NL), the second treatment group rats were exposed to 12 hours light and 12 hours darkness (LD), the third treatment group rats had 24 hours light (LL), while the fourth treatment group rats were exposed to 24 hours darkness (DD) (Figure 1).

	24 hours	
	12 hours	12 hours
LD	**Light 300 lux**	**Darkness**
LL	**Light 300 lux**	**Light 300 lux**
DD	**Darkness**	**Darkness**
NL	**Day light**	**Night**

Figure 1: Diagrammatic presentation of the illumination patterns for each experimental group

Light was provided by ceiling-mounted daylight fluorescent tubes. Light intensity was measured at the level of the cages with a digital photometer (Gossen Mavolux, Elangen, Germany) and censored using electronic timer for 12 weeks. After 12 weeks of exposure, rats were anesthetized using Ketamin (Flecknell, 1996) and surgically open to exposed the heart. Blood was collected by heart puncture into clean test tube using insulin syringe. Serum samples were separated by centrifugation at 3000 rpm. Serums were kept in -20 °C until hormonally analyzed. Using immune tech kit, the prolactin concentration of samples was determined by radio immunoassay (RIA) (Barkte *et al.,* 1987). Experimental data of were analyzed for their central tendencies using descriptive statistics and for their variations using analysis of variance, and significant means were separated using Duncan's new multiple range test at probability level of P = 0.05. All statistical analyses were done using SAS software.

RESULTS

The average prolactin concentration in each treatment is shown in Figure 2.

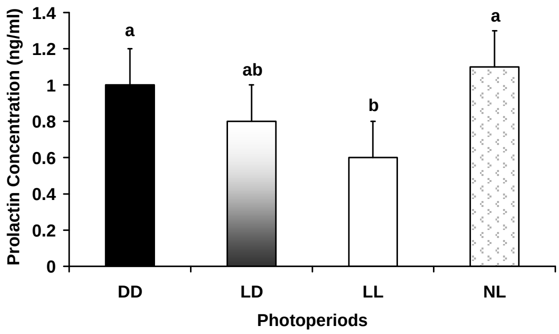

Figure 2: Effect of different photoperiods on prolactin concentration. DD (Dark-Dark), LD (Light-Dark), LL (Light-light) and NL (Natural light)

The highest of prolactin concentration was seen in rats exposed to the natural light regime (NL) (1.1 ng/ml). Rats exposed to 24 hours darkness (DD) had prolactin concentration of 1.0 ng/ml. The least average prolactin concentration of 0.6 ng/ml was recorded in rats exposed to 24 hour of light (LL). The average prolactin concentration in rats exposed to 12 hour light and 12 hour darkness (LD) was 0.8 ng/ml. There were significant differences in prolactin concentration in rats from the different treatment groups when compared to the control. The result of analysis of variance is presented (Table 1).

DISCUSSION

Prolactin hormone is a protein secreted from the pituitary gland. Its receptors have been identified in liver and kidney cells, and in some glands. Prolactin hormone causes the growth and proliferation of the mammary glands. Injection of prolactin has been reported to proliferated the breast cells in rat and stimulate milk production (Mohammadzad *et al.,* 2010). This hormone decreases progesterone at the end of pregnancy and causes parturition to start. Prolactin level in the blood of pregnant female usually increases gradually as pregnancy advances. The prolactin concentration at the end of pregnancy is usually 10 times more than those observed in non pregnant females.

Table 1: Analysis of variance for prolactin concentration in rats exposed to different photoperiodic treatments

Sources of variation	df	SS	MSS	F value	P > F
Photoperiod	3	2/07	0/69	4/22	0/0.01
Error	51	8/29	0/16		
Total	54	10/35			

Key: df: degree of freedom, SS: sum of square, MSS: mean of sum square

Prolactin concentration has been used as sign of amniotic disorder test (ADT) (Taheri Panah *et al.,* 2009). An earlier study indicated that the prolactin level in blood was affected by the duration / length of light and its concentration was high in fall and winter (when lightness period was low) (Anisimov, 2003). Prolactin has a positive effect on maternal instinct. The results of the present experiment indicated that light regime changes the prolactin concentration in rat. The rats exposed to permanent light (24 hour permanent light) (LL) had the lowest level of prolactin. Different results are reported in literatures on the effects of photoperiod on prolactin concentration in animals. Claustrat *et al.* (2008) used urine instead of blood serum to determine the prolactin concentration in rats. They reported that by increasing the light intensity, the concentration of prolactin increased simultaneously after three weeks. For melatonin synthesis, one of the important metabolic components is 6-sulfoxi melatonin. Continuous exposure to light decreased this component (Claustrat *et al.,* 2008). Johnston (2004) reported that prolactin secretion is affected by melatonin, and epiphysis controls its secretions by sending a signal to pituitary gland. In a study conducted by Vinogradova and Cherniva (2006), the concentration of prolactin hormone was high in the blood serum of the rats exposed to the permanent light. In another experiment, male hamsters placed in different light regimes for eleven weeks, had their testis weights significantly decreased in short light period, but no change was observed in long light period (Chandrashekar *et al.,* 1994). Prolactin and FSH injection had no effect on testis weight when rats were placed under different light regimes for two years (Chandrashekar *et al.,* 1994). The length of estrus cycle in lightness-darkness treatment was shorter than the others, but its length in this group increased up to the 14 month. For the rats in permanent light and natural light treatment, short cycle of estrous was observed. The number of estrous cycles and its length in natural light treatment increased in summer and spring (long day seasons). On the contrary, the rat exposed to permanent light showed longer estrous cycle. In the permanent darkness treatment, more short cycles were seen. Finally, it was concluded that absolute (permanent) light disorders the estrous cycle (Vinogradova and Cherniva, 2006). Furthermore, exposing the rats to permanent light advanced puberty (Anisimov, 2003; 2007). The concentration of prolactin in both natural light treatment and permanent darkness (DD) were 1.1 and 1.0 ng/ml respectively, this concentration was decreased to 0.80 ng/ml in the 12 hours light and 12 hours dark treatment, but was not significantly different from the prolactin concentration in rats exposed to permanent darkness. In rabbit, this concentration increased during night hours (Alvarez *et al.,* 2005). In rat it was high in the early hours of the morning (Grattan and Averill, 990). Season is a subject to different photoperiod regimes. The concentrations of prolactin in animals have been studied under the different season. For example, a different frequency of prolactin hormone secretion was registered for horses in different seasons. The concentration of prolactin in ewe was high in short day seasons. In horses in long day seasons, prolactin inhibited LH, but had no significant effect on FSH (Hodson *et al.,* 2010). There are some other factors which also affect prolactin production and secretion. Stimulus generated by paring female and male rats caused the sexual pheromone secretion to increased plasma level of prolactin hormone during pregnancy and milking periods has been reported (Eftekhari *et al.,* 1997).

Conclusion: According to the finding of the present study and other researches, light – light caused the serum prolactin level to decrease in rat. This can affect some phenomena such as reproduction disorder, like early puberty, estrous cycle disorder, decreasing of growth and number of alveolar mammary glands. This result can be considered as a model for further research in human communities which receive significant amount of light during 24 hours.

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REFERENCES

ALVAREZ, P., CARDINALI, D., CANO, P., REBOLLAR P. and ESQUIFINO, A. (2005). Prolactin daily rhythm in suckling male rabbits. *Journal of Circadian Rhythms,* 3(1): doi: 10.1186 /1740-3391-3-1.

ANISIMOV, V. N. (2007). Biology of aging and cancer. *Cancer Control,* 14: 23 – 31.

ANISIMOV, V. N., AILAMAZYAN, E. K., BATURIN, D. A., ZABEZHINSKI, M. A., ALIMOVA, I. N., POPOVICH, I. G., BENIASHVILI, D. S., MANTON, K. G., PROVICIALI, M. and FRANCESCHI, C. (2003). Light regimen, anovulation, and risk of malignant tumors of the female reproductive system: the mechanisms of link and prevention. *Journal of Obstetric and Woman Diseases,* 52: 47 – 58.

ANISIMOV, V. N. (1971). Blastomogenesis in rats with persistent estrus. *Voprosy Onkologii,* 17(8): 67 – 75.

ANISIMOV, V. N., SIKORA, E. and PAWELEC, G. (2009). Carcinogenesis and aging. *Open Longevity Science,* 3: 1 – 10.

ANISIMOV, V. N. (2003). *Molecular and Physiology Mechanism of Aging*. Nauka, St. Petersburg, Russia.

BARBACKA-SUROWIAK, G., SUROWIAK, J. and STOKŁOSOWA, S. (2003). The involvement of the suprachiasmatic nuclei in the regulation of estrous cycle in rodents. *Reproductive Biology,* 3: 99 – 129.

BARKTE, A., MATT, K. S., STEGER, R. W., CLAYTON, R. N., CHANDRASHEKAR, V. and SMITH, M. S. (1987). Role of prolactin in the regulation of sensitivity of the hypothalamic-pituitary system to steroid feedback. *Advances in Experimental Medicine* and *Biology,* 219: 153 – 175.

BATURIN, D. A., ANISIMOV, V. N. and PAPOVICH, I. G. (2004). Effect of depression photoperiod on homeostasis, life span and growth in transgenic mouse. *Oncology*, 50: 332 – 338.

CLAUSTRAT, B., VALATX, J. L., HARTHÉ, C. and BRUN, J. (2008). Effect of constant light on prolactin and corticosterone rhythms evaluated using a noninvasive urine sampling protocol in the rat. *Hormone and Metabolic Research*, 40(6): 398 – 403.

CAVCHENKO, O. H., CTRELTSOVA, N. A. and DANILIVA O. A. (1987). Effect of 2-3-oxtriptamin and some model of photoperiod on hypophysis male rat. *Physiology Journal,* 37: 480 – 482.

CHANDRASHEKAR, V., MAJUMADAR, S. S. and BARTKE, A. (1994). Assessment of the role of follicle-stimulating hormone and prolactin in the control of testicular endocrine function in adult djugarian hamster (*Phodopus sungorus*) exposed to either short or long photoperiod. *Biology of Reproduction,* 50(1): 82 – 87.

CHAZOV, E. I., ISACHENKOV, V. A., KRIVOSHEEV, O. G., TUZHILIN, V. D. and DEGTIAR, V. G. (1972). Biosynthesis of pituitary hormones. 3. Effect of epiphyseal extracts and melatonin on biosynthesis of the growth hormone and prolactin in rat pituitary. *Voprosy Mediùsinskoĭ Khimii,* 18(1): 3 – 7.

DEPRÉS-BRUMMER, P., LÉVI, F., METZGER, G. and TOUITOU, Y. (1995). Light-induced suppression of the rat circadian system.

American Journal of Physiology, 268(5 Pt 2): R1111 – R1116.

DILMAN, V. M. and Young, J. K. (1994). *Development, Aging and Disease. A New Rationale for an Intervention Strategy.* Harwood Academic Publishers, Chur, Switzerland.

EFTEKHARI, H., ROWHANI, A. H., BABAPOUR, V., PARIVAR1, K. and OMRANI, G. R. (2007). Effect of pheromones on the plasma level of prolactin during pregnancy and lactating periods in female rat. *Physiology and Pharmacology,* 11(2): 107 – 114.

FLECKNELL, P. (1996). *Laboratory Animal Anesthesia.* Academic Press, New York, USA.

GRATTAN, D. R. and AVERILL, R. L. (1990). Effect of ovarian steroids on a nocturnal surge of prolactin secretion that precedes parturition in the rat. *Endocrinology,* 126(2): 1199 – 1205.

HODSON, D. J., TOWNSEND, J., GREGORY, S. J., WALTERS, C. and TORTONESE, D. J. (2010). Role of prolactin in the gonadotroph responsiveness to gonadotrophin-releasing hormone during the equine annual reproductive cycle. *Journal of Neuroendocrinology,* 22(6): 509 – 517.

IROD, Y. A. (1966). Follicular cycle of ovary and dishormonal in cancer. *Medicine,* 136.

YANG, J., RICHARDS, J., GUZMAN, R., IMAGAWA, W. and NANDI, S. (1980). Sustained growth in primary culture of normal mammary epithelial cells embedded in collagen gels. *Proceedings of the National Academy of Science, USA,* 77(4): 2088 – 2092.

JOHNSTON, J. D. (2004). Photoperiod regulation of prolactin secretion: Change in intra pituitary signaling and lactotroph heterogeneity. *Journal of Endocrinology,* 180(3): 351 – 356.

KHAVINSON, V. KH. (2002). Peptides and ageing. *Neuroendocrinology Letters,* 23(Suppl. 3): 144.

LAZAREV, N. I., IRD, E. A. and SMIRNOVA, I. O. (1976). *Experimental Models of Endocrine Gynecological Diseases*. Meditsina, Moscow, Russia.

MOHAMMADZAD G. F., FAGHANI, M. and KHAJE JAHROMI, S. (2010). Protection effect of prolactin in histological variation in testied rat by busoulfan treatment. *Fertility and Infertility,* 11(2): 79.

NADRLOO, Z. (2006). *Prolactin.* First Conference of Medical Scientist, Tehran, Iran.

PARTA LIMA, M. F., BARACAT, E. C. and SIMONES, M. J. (2004). Effect of melatonin on the ovarian response to pinealectomy or continuous light in female rats. *Brazilian Journal of Medical and Biological Research*, 37(7): 987 – 995.

TAHERI PANAH, R., DAVUDI, Z., ENTEZARI, A., HOSSEINI, M. S. and KHODAKARAMI, N. (2009). Detection prolactin and B-hCG in vaginal liquid in determination of parturition starting (rupture of ammonic sac). *Medical Journal of Shahid Sadughi Yazd University,* 11(2):

VAN DER BEEK, E. M. (1996). Circadian control of reproduction in the female rat. *Progress in Brain Research,* 111: 295 – 320.

VINOGRADOVA, I. A. and CHERNIVA, I. V. (2006). Effect of photoperiod on estrous cycle dynamic and prolactin in rat. *Gerontology,* 19: 60 – 65.