

Original Research Article

Influence of light on serum cholesterol levels in complete (by birth) blind people

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ABSTRACT

Background: With blindness normal stimulation of the hypothalamic pituitary axis is reduced. The serum cholesterol is frequently increased the upper limit of normal it is unclear whether the effect is due to complete absence of light, HPA axis function reduction or diurnal variation of melatonin levels.

Methods: A self-administered questionnaire ascertained lifestyle characteristics, including alcohol consumption, cigarette smoking, diabetes mellitus and dietary habits. Inclusion criteria: 50 complete blindness persons (by birth), healthy adults, Age group between 30-50 years. Exclusion area: age <30, Age >50, alcoholic, liver diseases, kidney diseases, diabetes mellitus, thyroid diseases.

Results: A total No of 100 cases were studied by dividing them into two groups controls 50 and cases 50. The results so obtain were compared with 50 healthy controls (excluded consumed alcoholic, liver diseases, kidney diseases, diabetes mellitus, thyroid diseases.). Statistical evaluation was carried out to confirm any deviation from the normal values. The mean serum cholesterol of Cases (298.28 ± 26.82) is having higher level as compared to the mean value of controls (153.38 ± 11.79). This increase is statistically highly significant (<0.0001).

Conclusions: It has been shown from this study with blindness normal stimulation of the HPA is reduced consequently the serum cholesterol is frequently increased the concentration may exceed the upper limit of normal. On this basis we observed increases the upper limit of the cholesterol levels in blind healthy people than subjects with normal healthy light perception.

Keywords: Blindness, Cholesterol, Light, Melatonin

INTRODUCTION

While I was in theoretical study in Tetz text book I came to know the cholesterol levels are high in complete blind people it leads to a curiosity to investigate and take a topic to research that point of view why and how the levels of cholesterol is high in blind people under what circumstances and causes for the same accordingly, I made out through search and study of various books relating to the same topic and it encourage and urged to research the point of cholesterol levels in complete blind

people and its reasons that is the resulted of this research paper.

Cholesterol is found in all cells of the body. The adrenal gland contains 6% cholesterol (by weight). It is a precursor of the adrenal and sex hormones. The brain and spinal cord contain 2% cholesterol in these tissues cholesterol forms part of the lipid "insulation" which separate individual nerve fibers. The precise function of cholesterol in all other cells is unknown, but it is may be

related to the structure and permeability of the cell membrane.

Cholesterol may be synthesized from two-carbon units (acetyl-CoA) in many body tissues, particularly liver, intestine, and skin. Most cholesterol is excreted from the body via the bile, liver cells oxidize the molecule by adding hydroxyl and carboxyl groups (the ring structure remain intact) to form cholic acid. These are excreted in the bile where they are instrumental in the absorption of fats, including cholesterol itself, from the diet. Cholesterol serves as a precursor of other steroids, bile acids, vitamin D3 biosynthesis.

Most aspects of energy metabolism display clear variations during day and night. This daily rhythmicity of metabolic functions, including hormone release, is governed by a circadian system that consists of the master clock in the suprachiasmatic nuclei of the hypothalamus (SCN). The SCN control peripheral timing via the autonomic and neuroendocrine system.

Light is the primary environmental cue that entrains the main circadian clock in the SCN, thus allowing internal rhythms to be adjusted exactly to a 24-hour period on a day-to-day basis. In mammals, retinal ganglion cells perceive the ambient light and transduce this photic signal to the SCN through the retinohypothalamic tract.^{1,2} Through the retinohypothalamic tract the circadian rhythm generated within SCN neurons is entrained to an overt 24-hour rhythm and these coordinated outputs are conveyed to the rest of the body via behavioral, neuroendocrine, and autonomic pathways.^{3,4}

The brain's biological clock, which, in mammals, is located in the suprachiasmatic nucleus (SCN), generates circadian rhythms in behaviour and physiology. These biological rhythms are adjusted daily (entrained) to the environmental light/dark cycle via a monosynaptic retinofugal pathway, the retinohypothalamic tract (RHT).

The effects of environmental light on the hypothalamic-pituitary-gonadal axis is mediated by the pineal gland, through melatonin secretion.^{5,6} Light stimulus from the environment reaches the retina; from here, through a RHT reaches the SCN, then the superior cervical ganglion, and finally the pineal gland, where it exerts an inhibiting effect on the pineal melatonin secretion.

Melatonin is the main mediator that transfer changes in environmental light to human cells. The diurnal variations of melatonin is due to environment light condition. Melatonin production is stimulated by darkness under conditions of darkness melatonin production increases and light reduces the melatonin production.^{7,8}

The darkness activates alpha-1 and alpha2-adrenergic receptors in pineal gland, then it increases cyclic AMP and calcium concentration and activates arylalkylamine

N-acetyltransferase, thus initiating the synthesis and release of melatonin.^{9,10}

Effects of light on melatonin

Light and dark alterations constitute the principal timing signal of melatonin secretion from the pineal gland. Light influences melatonin synthesis in three ways in humans. First, light exposure acutely suppresses elevated melatonin levels. Second, light is able to phase shift the melatonin rhythm. Third, changes in the photoperiod can alter the melatonin secretion.

Synthesis of melatonin

Melatonin is a small (molecular weight 232.3) indoleamine secreted rhythmically with increased synthesis during the dark period of day. Alterations in environmental lighting are signalled as multisynaptic neural inputs by the central nervous system via peripheral nerves into a hormonal output of the pineal gland. In mammals, circadian photoreceptors of the retina convert light and darkness into signals that are sent directly to the SCN through the main pathway, the RHT.^{11,2} From the SCN, neuronal projections make synaptic connections in the PVN of the hypothalamus descending onward through the medial forebrain bundle to the intermedialateral cell column of the spinal cord from where pre ganglionic fibers reach the superior cervical ganglia; sympathetic postganglionic noradrenergic fibers from the superior cervical ganglia innervate the pineal gland through the *nervi conarii*.^{12,13} The production of pineal melatonin occurs in response to noradrenergic stimulation which produces a cascade of biochemical events within the pinealocytes. The *N*-acetyl transferase (NAT) activity represents a key regulatory step in melatonin synthesis.

Relation between melatonin and cholesterol production

Melatonin a pineal and gut secretory product, due to its antioxidant activity along with its effect on the aging gall bladder myocytes, inhibits gallstone formation. Melatonin reduces the biliary levels of cholesterol by inhibiting cholesterol production across the intestinal epithelium and by increasing the conversion of cholesterol to bile acids.¹⁴

Melatonin exerted a beneficial effect by increasing the HDL/total LDL cholesterol ratio. These findings suggest that the hypocholesterolemic effect of melatonin may work through the augmentation of endogenous cholesterol clearance mechanisms. This is accompanied by the lowering of the cholesterol fraction associated with low density lipoproteins.¹⁵

Melatonin itself has been shown to inhibit LDL receptor activity and cholesterol synthesis in human mononuclear leucocytes. Melatonin also influences lipoprotein lipase (LPL) activity, a key regulatory enzyme in circulating TAG clearance, in adipose tissue.¹⁶ LPL is activated by

insulin; a lower nocturnal insulin sensitivity could, therefore, be associated with lower LPL activity and relatively impaired plasma TAG clearance. Insulin also suppresses very low-density lipoprotein (VLDL) secretion by the liver; nocturnal insulin resistance could, therefore, also be associated with higher circulating TAG levels of hepatic origin.¹⁷

The circadian nature of cholesterol synthesis has been well established in animals. In nocturnal species such as the rat, hepatic 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, the rate limiting enzyme in the cholesterol synthesis pathway, peaks in activity at midnight and reaches a nadir at noon, miittinen observed in healthy humans diurnal periodicity in plasma levels of cholesterol precursors squalene and lanosterol, with peak levels occurring at night while plasma LDL and HDL cholesterol values remained relatively constant over 24h.^{18,19} Fluctuations in other pathways of cholesterol metabolism further suggest that cholesterol production may not be constant through the day. Diurnal variation in bile acid production has been observed in humans, with peak activity occurring in the early morning.²⁰

METHODS

The study was last from 18 July 2015 to 31st March 2017 in complete blind (by birth) Male subjects with age between 30-50, at Gayatri Vidya Parishad Institute of Health Care and Medical Technology (GVPIHC and MT), Marikavalasa, Visakhapatnam. The Written consent of participants was taken prior to study.

A self-administered questionnaire ascertained lifestyle characteristics, including alcohol consumption, cigarette smoking, diabetes mellitus and dietary habits.

Inclusion criteria

It included 50 complete blindness persons (by birth), Healthy adults, Age group between 30-50 years.

Exclusion criteria

It excluded age <30, age >50, alcoholic, liver diseases, kidney diseases, diabetes mellitus, thyroid diseases.

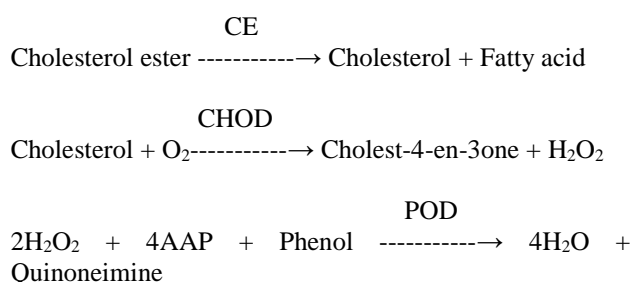
Sample collection

Fasting blood samples (from 18th July 2015 to 31st March 2017) were collected from complete blind (by birth) 50 healthy male subjects. (not to consumed alcohol, smoking) and 50 healthy controls (not to consumed alcohol, smoking). 5ml of venous blood was collected from each subject in fasting conditions and dispensed into lithium heparin bottles. Plasma was obtained by centrifugation for 5 min at 3,000 rpm and separated into plain bottles for analysis concentrations were assayed at the institution, based on the IFCC (International Federation of Clinical Chemistry) methods with a Semi

Auto analyser (Lab life CHEM MASTER, RFCL) using commercial reagents (ERBA for cholesterol); the normal ranges are Cholesterol: 140-250mg/dl).

Estimation of serum cholesterol (CHOD-PAP (with LCF) method)

Cholesterol esterase hydrolyses esterified cholesterol to free cholesterol. The free cholesterol oxidized to form hydrogen peroxide which further reacts with phenol and 4-aminoantipyrine by the catalytic action of peroxidase to form a red colour quinoneimine dye complex. Intensity of the colour formed is directly proportional to the amount of cholesterol present in the sample.²¹ The estimation of cholesterol involves the following enzyme catalyzed reaction:



Allow the Table 1 containing reagent bottles to attain room temperature add the 20ml of aqua-4 of each vial. Swirl to dissolve. Do not shake vigorously.

Table 1: Reagent composition (single step reagent).

Reagent
Cholesterol esterase
Cholesterol Oxidase
Peroxidise
Sodium phenolate
4-Aminoantipyrine
Phosphate buffer
Lipid clearing Agent

Table 2: Assay procedure.

Pipette into tubes marked	Blank	Standard	Test
Working reagent	1000µl	1000µl	1000µl
Distilled water	20µl	-	-
Standard	-	20µl	-
Test	-	-	20µl

Mix well Table 2 containing blank, standard, test, reagent samples tubes, incubate 37°C for 10minutes. Aspirate blank followed by standard and test and note the values.

RESULTS

During the period from 18th July 2015 to 31st March 2017 in the Department of Medicine, Gayatri Vidya Parishad Health Care and Medical College, Marikavalasa,

Visakhapatnam. A total No of 100 cases (men) were studied by dividing them into two groups controls 50 (men's) and cases 50 (men) and observation made were tabulated.

Table 3: Cholesterol (mg/dl) levels in men in both cases and controls.

Groups	Cholesterol Mean \pm SD	Z - value	P- value
Cases (n=50)	298.28 \pm 26.82	34.968	<0.0001
Controls (n=50)	153.38 \pm 11.79		

The above Table 3 shows that mean serum CHOLESTEROL of Cases (298.28 \pm 26.82) is having higher level as compared to the mean value of controls (153.38 \pm 11.79). This increase is statistically highly significant (<0.0001).

DISCUSSION

Melatonin production in blind

The retinal projection innervating the circadian pacemaker originates from a subset of retinal ganglion cells and is known as the retinohypothalamic tract.^{2,22} Lesion of the optic nerves results in free running of the circadian rhythm and blindness.²³

Blind individuals may have abnormal retinal processing and/or a defective RHT and, therefore, may not be capable of photic entrainment.²⁴

A few studies have demonstrated that the retinal circadian clock regulates many retinal functions such as retinal gene transcription, visual processing and photoreceptor viability, because retina is the only source of photic input to the SCN and peripheral tissues of the body, it is suggested that the interaction between retinal clock and SCN clock plays a key role in the circadian organization of entire organism.^{25,26}

Melatonin rhythm normally peaks at night both in animals and in humans²⁷ Blindness affects melatonin secretion significantly. Blind patients show increased day-time melatonin levels or more complex changes in circadian rhythmicity.²⁸⁻³⁰

Circadian control of lipid biosynthesis

Circadian genes do not only control lipid absorption but are also involved in the regulation of lipid biosynthesis. It was long ago observed in rodents that cholesterol synthesis showed a circadian pattern in the liver and in the intestine, being higher during the night and lower during the day.³¹⁻³⁸

This regulation was mainly achieved through the circadian expression of α -hydroxy-3-methylglutaryl-CoA reductase (HMG-CoA reductase).^{31-35,39}

Together with light entrainment cholesterol biosynthesis and HMG-CoA reductase expression appeared to be strongly entrained also by food ingestion, as shown in several rodent models in both the intestine and the liver and also in a human study.^{32-34,40}

CONCLUSION

With blindness normal stimulation of the HPA is reduced consequently the serum cholesterol is frequently increased the concentration may exceed the upper limit of normal. On this basis we observed increases the upper limit of the cholesterol levels in blind healthy people than subjects with normal healthy light perception. The above said few studies have the evidence to the elevated levels of cholesterol due to the lack of environmental light and abnormal rhythm disorders of melatonin production and HMG Co-A reductase activity but further studies are required. Several authors have recently supported a pivotal role of Melatonin in the regulation of both cholesterol and triglyceride metabolism in presence of environmental light.

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Ethical approval: The study was approved by the Institutional Ethics Committee

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