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Gold in the male reproductive tract of rat: A chronobiological study

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Abstract

In a 24 hour study, 10 adult male albino rats (total=60) were sacrificed at every four hour starting from 00:00 hours. Reproductive tissues – testis, epididymis (caput, corpus, cauda), vas deference, seminal vesicle, prostate (ventral, dorso ventral) and coagulating glands were dissected out and the level of gold was measured in each tissue by employing atomic absorption spectrophotometry. Level of gold differed showing peaks and nadirs at different timings, like dorso ventral prostate gland showed the highest amount (3.31µg per gram tissue) at 08:00 hrs and the lowest (0.03 µg per gram) at 00:00 hrs. Fluctuation observed in the level of gold is discussed in terms of chronobiology.

Key words: Chronobiology, Gold, Male reproductive tract, Rat, Time

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Gold was discovered in human semen[1] in 1981 and was shown present in all tissues of human male reproductive organs and accessory glands[2-4]. Level of gold in human semen was reported in some studies[5-7]. Histologically gold has been found in the testis and caput epididymis of rats[8]. In male reproductive system of frog, rat and guinea pig; gold was found by our group (Unpublished data). In female system of the frog gold was seen[9]. In this study an attempt was made to find out the level of gold secreted in reproductive tissues of male rat at different timings of the day, with a purpose to understand if any chronobiological influence is present on its secretion.

Materials and methods

The study was approved by Institutional Animal Ethical Committee of Government Medical College, Surat. During the study period, workers of this project did not wear any gold ornaments to exclude the entry of gold into tissues. Other possible sources of gold like dust, rubber tubing or water stored in plastic carboy were excluded. All glass wares used in the study was cleaned by placing in 6N HNO_3 overnight, rinsing in tap water, freshly prepared glass distilled water and finally double and triple glass distilled water. They, except volumetric items, were dried in hot air oven. Volumetric items were placed on filter paper for drying.

Initially two adult male rats were sacrificed and its tissues - brain, heart, lung, liver, spleen, stomach, skeletal muscle and reproductive tissues were processed for screening procedure to observe if gold was present in it. The instrument employed was Mass Emission Spectroscopy available at Gujarat Forensic Science Laboratory, Ahmedabad.

A total number of 60 male adult rats (breed Charles Foster) weighing 350-450 grams were chosen for this study. The animals were housed at room temperature between 20 °C and 25 °C and were exposed to 12-14 hours of day light. Temperature and light were found to influence the function of the male genital system[10]. The animals were maintained under standard diet and water was given ad libitum as it is essential for fertility[10].

Rats were sacrificed every four hours starting from 00:00hrs (Table1). Animals were anesthetized at fixed timing and tissues from reproductive tract, testis, three parts of epididymis (caput, corpus and cauda), vas deference, seminal vesicle, ventral prostate, dorso-ventral prostate and coagulating glands were separated. Tissue of same nature of animals of one-timing was grouped, blotted with ash less filter paper (Whatman, number 41) and weighed to the nearest milligram. Preprocessing of tissue samples was done by wet oxidation method[11,12]. Estimation of gold was done by employing an atomic absorption spectrometer (Perkin Elmer A 373) available at the Forensic Science Laboratory, Ahmedabad. The wave length of the instrument was maintained at 242.8 nm and slit opening was at 0.7 nm. A gold cathode lamp was in place. Air and acetylene gas were the source of the flame. Each sample was inserted for 10 seconds into instrument. Two consecutive readings were considered for the final value. The amount of metal present in dried material was calculated in terms of total weight of each tissue, with the final reading presented in micro gram per gram tissue. The total study was completed in a week period.

Results

The screening procedure showed gold was not detectable in all tissues except those from reproductive tract.

The results of the total study performed in male reproductive tissues are presented in table 1. Table 2 shows minimum and maximum levels of gold secreted by each tissue during different timings of the day.

Discussion

Studies carried out in human semen of normal and clinical cases suggest that gold may have a role in fertilization[4,6]. Possibly the metal in seminal plasma may be helpful in creating a suitable medium for spermatozoa or which may be entering the cell and participating in the function of motility. Gold was shown inside spermatozoa[3,4]. In one of our experimental study with human semen, it was found that addition of “suvarna bhasma”(gold ash), an indigenous ayurvedic preparation, to it increased sperm motility (Unpublished work).

In the present study, we attempted to find out if any change in secretion of gold was present in reproductive tissues of rat at different times of the day. Our results showed a fluctuating level of gold in all tissues studied throughout the day (Table 1). At 00:00 hours, the lowest secretion of gold was seen in five tissues studied - corpus epidydimis, seminal vesicle, ventral prostate, dorso-ventral prostate and coagulating gland. The peak of gold secretion from four of these tissues - testis, ventral prostate, dorso-ventral prostate and cauda epididymis was at 08:00 hours (Table 2).

Table 1: Chronobiological study of gold levels in rat reproductive tissues

	Gold (µg per gram tissue)					
	00:00hrs	**04:00hrs**	**08:00hrs**	**12:00hrs**	**16:00hrs**	**20:00hrs**
Number of animals	6	7	7	9	7	12
Testis	0.51	0.23	0.81	0.71	0.17	0.1
Caput epididymis	1.77	0.83	1.15	1.21	1.4	0.89
Corpus epididymis	0.34	1.04	2.63	2.54	2.7	1.84
Cauda epididymis	0.6	0.31	2.51	1.74	0.57	1.18
Vas deference	1.6	1.68	0.25	1.03	1.16	01.97
Seminal vesicle	0.3	0.41	0.6	0.38	0.84	0.3
Ventral prostate	0.26	0.82	1.71	1.52	0.79	0.53
Dorso-ventral prostate	0.03	1.59	3.31	2.04	0.89	0.7
Coagulating gland	0.07	1.71	1.88	2.01	2.25	0.8

Table 2: showing high and low level of gold present in tissues at two different times

	Gold (ug / gm) at hours	
	Minimum	**Maximum**
Testis	20:00	08:00
Caput	04:00	00:00
Corpus	00:00	16:00
Cauda	04:00	08:00
Vas deferens	08:00	20:00
Seminal vesicle	00:00	16:00
Ventral prostate	00:00	08:00
Dorso-ventral prostate	00:00	08:00
Coagulating gland	00:00	16:00

The changes present in values of gold at different times of the day may be chronobiological in nature. Such changes occur due to light and dark of the day. In man various substances change their concentrations in different tissues according to time of the day[13,14]. In the field of reproduction it was shown that when hamsters were exposed to dark for few weeks, it showed testicular atrophy, cessation of spermatogenesis and involution of male accessory organs and which coincided with low level of prolactin and testosterone in blood. It could be relieved by the administration of prolactin which lead to increase in blood testosterone level and increase in the weight of the testis and accessory organs. The pineal gland as a whole and melatonin in particular are involved with photo periodically controlled cyclical fluctuations in male reproductive function[15,16]. Conditions of light for 12-14 hours per day maintained at temperature between 20 to 25 °C may not be sole reasons for changes in values. Dissections of all animals in this study were done in one week period to exclude chances of any seasonal changes in the level of the element under study[10,17]. It is noticed that five of these tissues studied namely corpus epididymis, seminal vesicle, ventral prostate, dorso-ventral prostate and coagulating gland secreted minimum at 00:00 hrs. Out of which prostate gland (dorsal and ventral) shown maximum level of gold at 08:00 hrs and remaining three tissues secreted maximum at 16:00 hrs (Table 2). More detailed study is essential to explain the disparity seen in secretion of timing of different tissues studied. The factor, time of the day, has influenced the secretion of gold in each tissue studied (Table 1). It is not known if the quality of semen of this species differed according to the time as is known in case of man where a circannual pattern for sperm in semen[18]. Daily fluctuation in semen parameters also exist[19,20]. The superior quality of semen is found at 00:00 hours when volume, total sperm count, total and active motility of spermatozoa are at high[21]. This is likely to be due to the high level of androgen present in peripheral blood around midnight[22,23]. Androgen exists in seminal plasma[10] and testosterone receptors are shown on spermatozoa[24]. Seasonal difference in semen parameters are reported[18,25,26]. Similarly serum LH, FSH, testosterone and inhibin also changes[27]. Reports on seasonal changes in concentration of spermtozoa DNA[28], semen sodium and potassium[17] are available and therefore seasonal changes in secretion of gold is also possible.

Facts given above either from epidemiological or analytical studies revealed a chronobiological influence seen on human semen. Similar changes may also be present in the quality of semen of rat. Among animals chronobiological study of semen are scarce. Gold present in semen at different timings of the day may be an influencing factor for fertility. A conclusion could be reached by conducting more elaborated study.

Conclusion

In conclusion, an influence of time of the day on secretion of gold in male reproductive system of rat is seen in this study.

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Conflict of interest: None

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