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To Investigate the Effect of Light Therapy in Gestation Length and Foal Birth Weight in the Thoroughbred Mare.

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Abstract/Absztrakt

It has already been established that blue light from light emitting diodes directed at a single eye elicits a dose dependent suppression of melatonin in horses (Walsh et al 2012). A suppression in melatonin increases gonadotropin releasing hormone pulse frequency which advances the onset of estrus in mares (Cleaver et al 1991). In this experiment, we carry on from this point, investigating the effects of light therapy from light masks to the mare during her gestation period and the subsequent effect this has on gestation length and birth weight of the foal. We also aim to determine the relationship between administering light therapy via the light mask and rate of coat shedding in mares. Furthermore, we aim to investigate the most recent scientific publications on the topic of light therapy in equines. We trace the evolution of this science and evaluate its application in the equine Industry.

Már megállapították, hogy a fénykibocsátó diódákból származó kék fény egyetlen szemre irányítva a melatonin dózisfüggő elnyomását váltja ki lovakban (Walsh és mtsai. 2012). A melatonin elnyomása növeli a gonadotropin felszabadító hormon impulzusfrekvenciáját, ami előrehozza az ivarzás kezdetét kancáknál (Cleaver et al 1991). Ebben a kísérletben ezen a ponton folytatjuk, és a kancáknak a vemhességi időszak alatt fénymaszkból történő fényterápia hatásait, valamint ennek a vemhesség hosszára és a csikó születési súlyára gyakorolt későbbi hatását vizsgáljuk. Célunk továbbá, hogy meghatározzuk a fényterápia fénymaszk útján történő beadása és a kancák szőrhullásának mértéke közötti összefüggést. Célunk továbbá, hogy megvizsgáljuk a legújabb tudományos publikációkat a lófélék fényterápiája témakörében. Nyomon követjük e tudományág fejlődését és értékeljük alkalmazását a lóiparban.

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Introduction

The mare is a seasonal long day breeder with a natural breeding season in the Northern Hemisphere from late April to October. It has been demonstrated that exposing the mare to extended hours of daylight for 6-8 weeks advances the first ovulation of the breeding season (Palmer and Draincourt, 1985). Due to industry imposed timelines and a demand for foals born soon after January 1st , it is common practice in the Thoroughbred industry to leave mares indoors under barn lights until 11pm nightly during the long winter nights, beginning on December 1st. “Light therapy” or putting mares “under lights” are common terms used to describe the regime of exposing the mare to a lengthened period of daylight. This process mimics the longer days of the horses' natural breeding season but does so by using artificial light. A lengthened period of daylight suppresses the release of melatonin, a hormone secreted by the pineal gland (Fitzgerald and Mc Manus 2000). This suppression releases melatonin's inhibition of gonadotropin releasing hormone (GnRH) and advances the transition into the reproductively active period of the mare. This is done via its regulation of a cascade of reproductive hormones which leads to the onset of estrus (Cleaver et al 1991). Recently, it has been shown that administering low intensity blue light to a single eye causes a dose dependent suppression of melatonin in the horse (Walsh et al 2012), which when incorporated into an automated individual light mask headpiece was shown to be as effective as barn lighting at advancing the breeding season in mares (Murphy et al 2014). Prolactin is a seasonal hormone regulated by increasing day length (Thompson et Hoffman 1997). Worthy et al. 1986, observed that prolactin levels in the blood serum of mares rose significantly during week of gestation, just before parturition and remained high although variable into the early stages of lactation just after parturition. Thompson et Hoffman 1997 observed that there are low levels of prolactin secreted by the mare during the hours of darkness, concluding that light inhibits the secretion. They also concluded that prolactin mediates the onset of vernal hair shedding and that there are low concentrations of prolactin present in the bloodstream of the horse during winter months as well as observing that treatment of mares with prolactin increased the onset of hair shedding within 14 days, with numbers peaking at 28 days. These findings show that the hormone prolactin has a strong circadian and seasonal rhythm. It is the trigger behind horses shedding their winter coats during Spring as the evenings get longer and the photoperiod extends. This extension of bright hours in the Spring is the natural trigger that causes prolactin to be secreted from the anterior pituitary of the animal (Thompson and Hoffman 1997).

Mobile light masks were developed for horses with the aim of advancing the breeding season (www.equilume.com). They release a low intensity blue light into the horses right eye and mimic the appearance of blinkers which mean they can be worn comfortably by the horse. The light mask allows the horse to be maintained outdoors while still receiving the lengthened photoperiod. This means the horse can be in its natural environment, which is advantageous to the horses welfare. As well as advancing the breeding season, the light mask is also thought to advance the shedding of the winter coat potentially via increased circulating prolactin. In this study we will also evaluate the extent of shedding related to mobile light treatment in mares.

In continuation from the Original study in 2014 "*To investigate the effect of light therapy on gestation length and foal birth weight"(Baugh, Equine Science Project)*, we will further discuss the most recent developments in this field. We aim to investigate the effect of light therapy on Mare gestation length, Foal birth weight and Mare and foal Coat quality. In the original study, our main objectives were as follows:

1. To evaluate whether an extended photoperiod during gestation would have any effect on Gestation length.
2. Evaluation of extended photoperiod on foal Birth Weight
3. Evaluation of extended photoperiod on Coat shedding in the mare.

The extended photoperiod was implemented to each individual mare by use of a light mask resembling blinkers worn by the mare, which released a low intensity blue light into the horse's right eye. In the original study, our main results were as follows. With regard to gestation length, application of light Therapy via a blue LED did not show any significant effect on the gestation length of the mare. We concluded that our preliminary study was a good starting point for more detailed research and data collection into the effects on gestation length. It is thought to be of Industry importance in the Thoroughbred breeding industry to be able to safely manage the mare's gestation length with regard to mares that may carry their foal over term or those that may have trouble carrying to term resulting is dystocia and late return to estrus.

With regard to foal birth weight, we concluded that's a lengthened photoperiod during gestation gave rise to on average a heavier foal. Ie." *Light therapy influenced the Foal Birth weight".* This factor is of Industry and welfare importance. It gives us better insight into managing larger foals, dystocia's and uterine health of the mare. With regard to coat shedding, we concluded that a lengthened photoperiod resulted in earlier coat shedding for the mare. On average, 2 weeks earlier shedding for the mare. This was also thought to be an advantageous preliminary finding for further studies. This factor is of valuable industry importance as it is a primary factor in the sales of Thoroughbred horses.

In this paper we also aim to evaluate the most recent research with regard to these main factors. We aim to investigate how the research has developed and what new medical benefits, as well as practical applications this has had for the Thoroughbred industry since this original preliminary paper was written in 2014. We evaluate scientific research on the effect of light therapy on the foal in utero and the potential effects light therapy could have on the foal after birth.

Literature Review

It has been established that light masks have been proven to advance the breeding season in Mares (Walsh et al 2012). This is of great significance to the welfare of the horse as mares can be housed outside in their natural environment. Low intensity blue light to one eye causes a dose dependent suppression of melatonin (Walsh et Al 2012). This suppression of the hormone Melatonin in turn releases melatonin's inhibition on Gonadotrophin releasing hormone (GnRH), thus allowing the development of follicles and the onset of estrus. Thompson and Hoffman stated in 1997 that light inhibits the secretion of prolactin. As discussed before in our introduction, they observed that there are low levels of prolactin secreted by the mare during the hours of darkness. They concluded that prolactin mediated the onset of vernal hair shedding, stating that Prolactin is a seasonal hormone regulated by the environmental factor of increasing daylight (Thompson and Hoffman 1997). In 1986 Worthy et. Al. Stated that Prolactin levels increased in the final week of gestation, just before parturition and remained high, although variable in the early stages of lactation just after parturition. We can conclude from such research that Prolactin is a seasonal hormone, released in a larger extent by an increased photoperiod, with higher concentrations of prolactin in the blood during the extended photoperiod. In a study carried out by Thompson and Hoffman in 1997, a group of mares were treated with the hormone Prolactin. They observed that Prolactin increases the onset of hair shedding within 14 days. In 2017 Murphy et al observed that light treated mares have earlier shedding of their Winter coats. In a study conducted by Lutzer et al in 2022 when a group of mares in late gestation were exposed to blue light therapy to extend the photoperiod it was shown that guard hairs were shorter in foals born to these mares. This difference in guard hairs between treatment and control groups continued until foals were two months old but evened out in treatment and control groups by eight months old. Authors from this study found that foals born earlier in the season were born with longer guard hair compared to those born later in the season.

A previous study carried out by Hodge et al in 1982 aimed to determine the influence of photoperiod on the pregnant and post-partum mare. They studied the effects of an extended photoperiod on reproduction during the periparturient period of 32 mores over

2 years. One of the primary aims of the study was to evaluate the effects of an extended photoperiod regime of 16 hours light on gestation length and foal development. The finding of this study showed that mares exposed to light therapy had a shorter mean gestation length of 10 days compared to the control mares not exposed to light therapy. They found that foals of mares exposed to a long photoperiod were carried for a shorter term, their foal weight tended to be heavier, but not significantly heavier than the foals of the control mares not exposed to an extended photoperiod. The authors also concluded that foal size as determined by body measurements was not affected by an extended photoperiod (Hodge et. Al 1982). From our own study in 2014 on "*The effects of light therapy on gestation length and foal birth weight" (Baugh Equine Science Project),* we found no significant difference between treatment and control groups with regard to gestation length. In 2017, a more advanced study by Murphy et al found a significant difference in gestation length for their group of treatment mares in Ireland, with an on average shorter gestation period in treatment mares being demonstrated. Again in 2022, Aurich et al found that gestation length was shorter in their group of pregnant mares exposed to an extended period blue light treatment. The extended Blue light treatment was administered via a mask that emitted Fifteen hours of continuous light daily, this was the modified version. The original version emitted seven continuous hours daily.

Growth Hormone is a seasonal hormone with a strong circadian rhythm released from the pituitary gland. An experiment to determine the seasonal changes in growth rate, feed intake, growth hormone and thyroid hormone concentrations in young red deer reported that, changes in day length are thought to control somatic growth, antler growth and food intake cycles in red deer (Ryg and Jacobsen 1981). Similarly in our study "*To investigate the effects of light therapy on gestation length and foal birth weight"* we aimed to determine how seasonality, via artificial light therapy administered to the mare, could influence the birth weight of the foal. We found a significant result for foal birth weight, with the treatment group having on average a significantly heavier foal than the control group. In the study conducted by Murphy et al in 2017, where there were two groups of experimental animals to be evaluated regarding foal birth weight. The first group was situated in Ireland, this group showed no significant difference in foal birth weight between treatment and control animals. The second group were situated in Kentucky, USA, this group showed a significantly higher foal birth weight in the group of treatment mares. In a study by Aurich et. Al to determine the effects of blue light on Gestation, parturition and foal maturity when blue light therapy was administered to pregnant mares, no significant results were found with regard to foal birth weight. They found no difference in foal birth weights between the treatment and control groups. Lutzer et al also had the same result in 2022 for their study into the *“Development of foals until one year old when the dam was exposed to blue monochromatic light during late pregnancy”.* An insignificant result with regard to birth weight was also found in this study.

Objectives/Questions

The objective of this Thesis is to evaluate the effect of light therapy on the Horse. In particular, we focus on gestation length in the Thoroughbred mare and foal birth weight and coat quality. We begin by presenting a study carried out in 2014 "*to investigate the effect of light therapy on gestation length and foal birth weight"*. From here we continue by reviewing further scientific publications from 2014 until present. The aim is to assess and understand the developments in light therapy with regard to the horse. We aim to track the progression of scientific research in this field. We trace it from the beginning, where the research investigates the effect of light therapy on the control of reproductive hormones in the mare and the practical application of advancing the breeding season in mares. Later, we see a development in research as we understand the effect that light therapy has on the pregnant mare and the foal in utero. As time progresses, we witness the evolution of light therapy research as it evaluates what effect light therapy can have on the foal in utero and on foal weight and development after birth.

Materials and Methods

1.1 Animals

This study "*To investigate the effect of light therapy on gestation length and foal birth weight" (Baugh, Equine Science Project)* was a preliminary clinical trial carried out on the effects of light therapy on foal birth weight and gestation period of the mare. All experimental procedures were approved by University College Dublin Animal Research Ethics Committee and the University of Kentucky Institutional Animals Health care and Use committee.

Thirty healthy Thoroughbred mares of various ages and parity were used for this experiment (n=30). The mares were housed in a research farm in Fayette County, Lexington, Kentucky for the duration of the experiment (entire gestation period of the mares). The experiment began on the day the first mare was inseminated (March 23rd) and ended after the last mare had given birth (April 6th). One mare was excluded from the data set as she lost her foal due to a "red bag" foaling indicative of placentitis. This was classified as a reproductive abnormality and was therefore removed from the dataset to avoid interference with normal data. This mare was in the control group and upon her exclusion the new experimental size was 29 mares (n=29). The mares were divided into two groups. Group 1 was the treatment group (n=16) and group 2 was the control group (n=14). All mares in the study had a body condition score of between 4 and 6 using the Henneke body condition scoring system. There was no difference in mean body condition scoring between groups (Group 1 =5.1 +/- 0.8, Group 2 = 5.2+/-0.5, P>0.5). There was no difference in the age ranges of the mares between the two groups (Group 1, 9.4 +/- 4.2 ; Group 2, 10.2 +/- 4.1). Both group 1 and group 2 had ad libitum access to grass as they were kept at pasture for the duration of the study. They also had ab libitum access to hay at night or during the times of sub-freezing temperature, snow or frost. They were given 2.7kg/day mare nuts depending on the individual needs of the mare as assessed by the farm manager and ad libitum access to water for the entire experimental period.

Both groups were maintained at pasture for the entire experimental period. Group 1 received light therapy beginning on December 1st, this corresponded to between days 226 to day 252 of gestation. Light therapy was administered via head worn light masks that provided 50 lux of blue light to the right eye of each mare. The light mask turned on automatically at 16.30h each day and turned off at 23.00h every night during the Winter months. The light masks were designed to activate automatically at dusk. This function saves battery life as the days naturally get longer. Group 2 did not wear light masks and were only exposed to the naturally occurring photoperiod.

Hair loss tests were conducted on the mares using the "stroke technique". This was simply done by stroking the mare from withers to rear with a float, gloved palm and recording the hair loss. Hair loss was recorded by assigning it a number from 1-4. 1 being no hair loss and 4 being severe hair loss. This was done once every week for each mare from January 18th to March 22nd, which coincided with the end of the mares gestation period. The light masks used in this experiment were Equilume TM light masks and were used according to manufacturer's instructions.

1.2 Experimental Protocol

Beginning on 1st December, group 1 (n=16) received light therapy of 50 lux blue light to the right eye from light masks for the remainder of their gestation period. The duration of light emitted by the mask varied accordingly with the changing photoperiods from December 1st onwards. Beginning on the day of Indvidual insemination, group 2 were housed at pasture for the duration of their gestation period and were exposed to the naturally occurring photoperiod only. Beginning on January 18th and ending on March 8th a coat shedding test was carried out for each mare in both the treatment (n=16) and control group (n=13) on a weekly basis, for 8 weeks in total. The mare was stroked in the direction of hair growing along the back, flanks and buttocks using a latex gloved palm. The amount of hair present on the gloved palm was given a score from 1-4. 1, demonstrated no hair loss, 2 demonstrated mild hair loss, 3, moderate hair loss and 4 demonstrated heavy hair loss.

Fig. 1.0: Summary Table - Treatment Group

-	No. animal	Parity	Age	BCS	Breed	Weight M	Weight F	Sex Foal	WF/WM	%Filly	%Colt	Gestation
Treatment	1	3	ukn	6	TB	677	117	FILLY	0.1728	56.56	43.75	340
	2	5	19	6.5	TB	589	100	FILLY	0.1697			344
	3	2	11	5.5	TB	534	102	FILLY	0.1910			349
	4	1	10	4.5	TB	673	107	COLT	0.1589			343
	5	0	8	5.5	TB	632.5	95	COLT	0.1501			342
	6	0	8	6	TB	615	109	COLT	0.1772			361
	7	2	13	4.5	TB	560.5	97	FILLY	0.17306			345
	8	0	12	5	TB	530	94	FILLY	0.177358			355
	9	0	7	5.5	TB	561	103	COLT	0.183601			355
	10	1	6	4.5	TB	442.5	105.2	FILLY	0.23774			339
	11	0	5	3.5	TB	570	111	FILLY	0.194737			346
	12	0	5	4	TB	478.5	95	FILLY	0.198537			353
	13	0	5	4.5	TB	536	108	FILLY	0.201493			330
	14	2	16	6	TB	613	108	COLT	0.176183			347
	15	0	6	4.5	TB	577	105	COLT	0.181976			346
	16	many	ukn	5	TB	558	100	COLT	0.1792			340

Fig 1.1: Summary Table – Control group

	No. animal	Parity	Age	BCS	Breed	Weight M	Weight F	Sex F	WF/WM	%Filly	%Colt	Gestation
Control	1	3	12	5.5	TB	539.5	114	COLT	0.211307	69.23	30.77	358
	2	2	11	5.5	TB	488	104	FILLY	0.213115			337
	3	0	12	5	TB	575	89	COLT	0.154783			362
	4	1	9	4.5	TB	619.5	112	FILLY	0.180791			338
	5	2	8	5.5	TB	559.5	76	COLT	0.135836			329
	6	0	8	6.5	TB	694	94	FILLY	0.135447			358
	7	1	8	5	TB	613.5	110	FILLY	0.179299			347
	8	1	6	5	TB	475	96	FILLY	0.202105			341
	9	0	5	5	TB	608.5	106	COLT	0.174199			358
	10	0	5	4.5	TB	526	77	FILLY	0.146388			340
	11	0	19	5.5	TB	540	82	FILLY	0.151852			360
	12	0	17	5.5	TB	597.5	78	FILLY	0.130544			349
	13	Many	Ukn	5	TB	551.5	99	FILLY*	0.17951			357

Fig 1.0 and Fig 1.1: These tables show information on each animal in this study, including; Ukn (unknown), number of animals, parity, age (years), body condition score, breed (TB=Thoroughbred), weight of mare (weight M) before pregnancy (lbs), weight of

foal(weight F) (lbs), sex of foal, weight of foal expressed as ratio of weight of mare, % fillies born, % colts born, length of gestation for each mare (days) in both the treatment and control groups.

(*note: *beside the filly foal of mare no.12 in the control group was used in data collection to identify this filly as having a difficult birthing process but has no significance in this experiment*)

Continuing from this point, in 2017 Murphy et al conducted an experiment on pre-partum mares. They administered artificial light via blue LED to extend the photoperiod and observed gestation length, foal birth weight and foal hair coat at birth. In this paper we see the author as expanded the study and we note there are three groups of mares used for experimental analysis.

The First group in situated in Ireland in a commercial breeding farm in Kildare. Here we have Thoroughbred mares that are naturally bred with a selection of Thoroughbred stallions. The mares were kept at pasture during day with ad libitum access to grass. They are brought in each evening and housed in a stable with ad libitum access to hay. The light masks turn on daily at 16.30 hours and turned off at 23.00 hours. The photoperiod mimicked by the masks was gradually extended in motion with the naturally extended photoperiod. It began with 14.5 hours /Day light exposure via a light mask and gradually increased to 16 hours 15minutes light exposure per light mask per day as the natural photoperiod also extended. The control group was comprised of 9 mares. They were kept outside during the day and brought inside in the evening where they were exposed to light therapy using the synthetic lights of the stable. The Second group are situated on a Farm is in Kentucky. Here we see 29 light breed mares inseminated with a mix of semen from two Quarter- horse stallions. The first group on 15 mares were fitted with the light masks as described earlier. The second control group on 14 mares were not fitted with the masks but again were subjected to the natural photoperiod alone. Both groups were kept outside during the experiment with ad libitum access to hay and water. The Third group were also situated on a farm in Co. Kildare, Ireland. The group consisted of Thoroughbred mares. A group of 19 mares are fitted with light masks under the same circumstances as above, beginning with 14.5 hours /Day light exposure via a light mask and gradually increased to 16 hours 15 minutes light exposure per light mask per day as the natural photoperiod also extended. A control group of 27 mares were kept at the same location but were not fitted with light masks. The hairs of both groups were plucked from the mares 48 hours post foaling. The hairs, complete with follicles, are weighed in micrograms and measured. Both experimental and control groups are compared. The overall aims of the study again are similar, aiming to investigate gestation length, foal birth weight and mare hair coat when a light mask is applied to each individual mare emitting 50 lux blue light to the right eye. The masks are fitted to the mares pre-partum.

In 2020 Yoon et al carried out a study on the effect of Equilume light masks on the timing of seasonal ovulation in Thoroughbred mares on South Korea. The study began with the mares being fitted with the masks from a period of November 18th –February 10^{th,} at two farms in Icheon and Sangju, Korea. Mean temperature at the farms over the examination period ranged from -8.8 to -12 degrees Celsius. There were 9 mares in the treatment group and 7 mares in the control group. The Body condition score of each mare was calculated as well as examination of the number and size of uterine follicles on the following dates; January 6th and February 10th. All mares were allowed out to pasture day at night, being occasionally brought into the stable during heavy winds/weather conditions. All mares were allowed to graze ad. Libitum and were fed timothy hay. All mares in the treatment group were fitted with individual masks that automatically turned on each day at 16:30 and off at 23:30. Mares in the control group did not have masks. Body condition score of each mare was assessed on the Henneke scale of 1-9. On Jan 6th and February 10th follicle size and uterine horn score were determined. "*Mares with follicle sizes > 35 mm and uterine horn scores = 3 were classified as having an activate estrous cycle (Hayes et al., 1985)"*

A study by Aurich et al was carried out in 2022 on "The effects of blue monochromatic light directed at one eye of pregnant horse mares on gestation, parturition and foal maturity. 20 warm Blood sport horse mares were used, located on a stud farm In Germany. All mares were in foal with single pregnancies and were available for the study for two consecutive seasons. All mares were bred via artificial insemination that was done within the period of March – June. During the summer months all mares were housed outside. During the winter months all mares were housed in stables with a daily turn out period in a paddock. 14 days before the expected parturition mares were moved to the Stud farms foaling unit. After parturition Foals were closely monitored and a record was kept on each foal's ability and time taken to stand and suckle the mare. It is important to note that in the study both treatment and control groups were exposed to blue light therapy. The control group was exposed to the original blue LED light mask and the treatment group fitted with a modified version of the same mask. Modifications included the extension of the light treatment time emitted daily by the mask. The original mask gave continuous light for seven hours per day. The modified version gave continuous light for Fifteen hours per day. Several parameters were recorded. These include:

1.Foal size and weight and placental surface area: The day after birth several analytical procedures were carried out. The foal was weighed, and its wither height measured. The circumference of the foal's cannon bone was recorded, and the Surface area of the mare's placenta was measured and described.

2.Progestogen analysis: The concentration of progestogen in the mare's plasma on the day after foaling was assessed using ELISA.

3.Hair coat: Mares' hair coat was measured the day after foaling. During December at the beginning of the study an 8 X 6cm rectangular area was shaved on the left side of the mare's chest. Hair grown in this area was subsequently measured the day after foaling. Hair from the foal, including the root was plucked from spina scapula area. Five hairs were collected for each foal and their length and mean length recorded and assessed.

In 2022 Lutzer et al conducted an experiment on the development of foals until 1 year old when the dam was exposed to blue light therapy during late Pregnancy. 40 Warmblood Sport horse foals were used for the purpose of this study, born to 20 mares on two consecutive breeding seasons. The mares were bred by Artificial Insemination to warmblood stallions. During year 1 there were 8 mares in the treatment group, they were exposed to artificial photoperiod via head worn light masks (Equilume). Treatment mares wore the same light masks from 13th December until parturition. The light masks emitted 50 LUX blue LED into the right eye of the mare between the hours of 8.00 - 23.00 daily. During Year 2, 12 mares in the treatment group wore light masks from 16th December and were exposed to the same duration and intensity of light therapy as Year 1 mares. During late pregnancy all mares were housed in the same group. They were housed by night in a stable bedded with straw and had daily turn out to a paddock. Fourteen days before parturition mares were moved to the foaling unit and when the foal was one week old the mare and foal returned back to the group stable. During this time foals were creep fed concentrate. From the month of May onwards the mare and foal were put out to pasture with access to stables if weather conditions turned unfavorable. At six months old the foals were weaned and separated by sex into two groups. During the study the following parameters of the foal were measures and analyzed:

1. Heart Rate, Heart rate viability and hematology

This was determined at 1,6,15,30 days old. Cardiac beat to beat intervals were measured using a portable record system attached to the thorax, this was recorded continuously from 6am –8am. 30 minutes after birth and on days 1,6,15, 30 blood was taken and hematology and Glucose analysis was carried out. Glucose concentration was analyzed using a Dry chemistry analyzer. Body temperature of foals was measured rectally on days 1, 6, 15 and at 1 month old.

2. Weight, Size, Hair Coat

Weight and size were measured at regular intervals until 12 months old. Weight was measured using an electronic scale and regarding size the following parameters were measured: Height at withers, chest circumference, Distance from fetlock to carpal joint, distance from carpal joint to elbow and cannon bone circumference. Length of Guard hair and hair loss were measured (as described previously) on the same days as weight and size were measured. A minimum of 5 hairs from each animal including their follicles were plucked and their length determined. Hair loss was determined using the "stroking technique" with a latex gloved palm, stroking 3 times and grading on a scale of 1-4.

Results

From our original study in 2014 "To investigate the effects of light therapy on gestation length and foal birth weight" (Baugh, Equine Science Project), In order to construct an ANOVA table, the sample sizes for both the treatment and control groups had to be the same. Group 1 was cut from n=16 to n=12 and group 2 was cut from n=13 to n=12. 12 animals were taken from a random sample of the entire treatment and control populations. Exposure of treatment mares to light therapy during their gestation period significantly ($P<0.05$) increased the birth weight of their foals when a t-test was carried out. Birth weight for group 1 averaged 103.5 lbs, +/- 6.3 and birth weights for group 2, the control group averaged 95.2 lbs +/-13.2. The results of an ANOVA table analysis using age as a blocking factor determined that age of the mare had no significance on birth weight of the foal. It was concluded using a t-test that there was a significant difference between birth weights of the treatment foals and birth weights of the control foals ($P=0.03$). The foals birth weight was also expressed as a percentage of the mares' weight and a t-test was then carried out on this data between the treatment and control groups. The results showed there to be a significant difference ($P=0.03$) with the treatment group again having heavier foals on average.

Fig 1.2 Table of averages

	Weight mean (mare)	D.O.B mean (mare)	Age mean (mare)	Parity mean	Estimated due date mean	Insemination date mean	Date foal mean	Gestation length mean	Foal birth weight mean
Treat	571.688	22 Nov -03	9.357	1.0667	12-March	04-April 2012	15 Mar	345.9375	*103.5125
Ctrl	566.967	07-Mar-03	10,21	0.7857	11-March	03 April 2012	17 Mar	348.7692	95.1538

Fig 1.2: This Table shows the mean values for: Mare weight, mare date of birth, mare age, mare parity, estimated due date, date of insemination, date of foaling, gestation length and foal birth weight for both the treatment and control groups. (Treat = treatment group (group 1), Ctrl = control group (group 2)). * indicates significance.

Fig 1.3: Birth weight averages of treatment and control groups

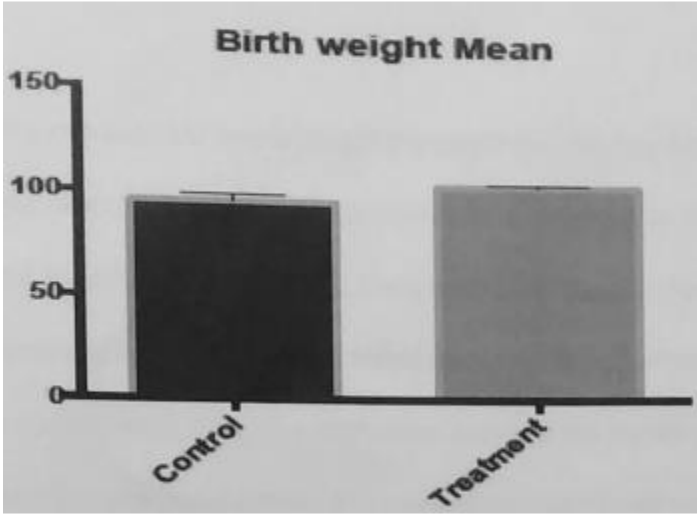

Fig 1.3: Mean Birth Weights of foals from treatment and control groups. Average birth weight treatment group 103.5 lbs +/- 6.3, average birth weight of control group 95.15 lbs +/- 13.2.

*Significant difference (P<0.05) in birth weight. (P=0.03)

Fig 1.4: Birth weight of foal expressed as a percentage of mares weight

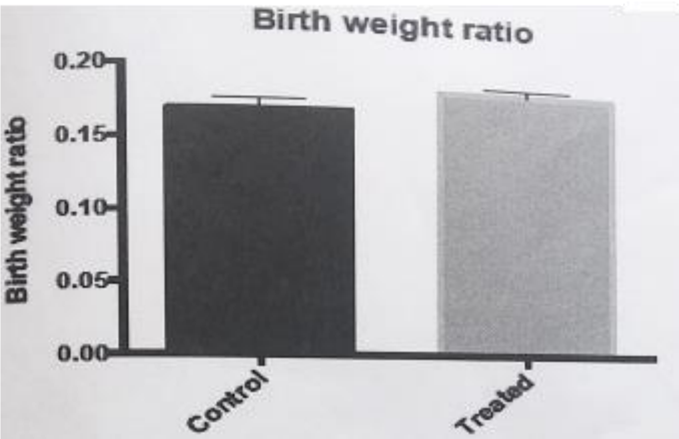

Fig 1.4 * heavier ($p<0.05$) birth weight in treatment group compared to control group. Birth weight means of foals from treatment (n=16) and control (n=13) groups expressed as a percentage of the mares weight. Significant difference ($P=0.03$) was found between the two groups with the treatment group having significantly heavier foals on average ($p<0.05$).

Exposure of treatment mares to light therapy throughout gestation had no significance ($P>0.05$) on gestation length when compared to that of the control group. ANOVA analysis using age as a blocking factor demonstrates that there was no significant difference in gestation length between the treatment and control groups no matter what the age of the mare. P value 0.75 for the blocking factor and P value 0.14 for the interaction between the blocking factor and the treatment. A t-test between treatment and control also showed an insignificant result ($P>0.05$) with a P value of 0.22. Group 1, treatment group had an average gestation length of 346 days +/-7.3 and group 2, control group had an average gestation length of 349days +/- 10.4.

Fig 1.5: Gestation length mean between treatment and control

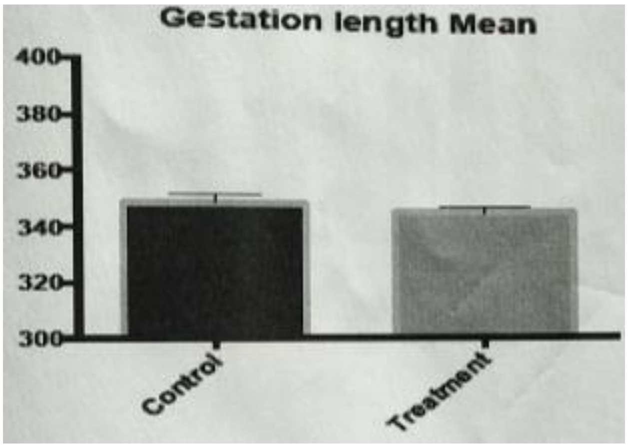

*Fig 1.5 s*hows the difference in gestation length between treatment (n=16) and control (n=13) groups. The x axis represents the treatment and control groups, the Y axis represents the days of gestation. There was no significant difference found between the gestation length of the treatment and control groups. Average gestation length for treatment group = 346 days +/- 7.3. Average gestation length for control group = 349 days +/- 10.4.

Hair loss data from the mares for a period of ~ 3 months (18th Jan – 8th March) was taken for both groups, treatment (n=16) and control (n=13) and the results were plotted using a graph. Hair loss was rated on a scale of 1-4, where 1 represents no hair loss and 4 represents heavy hair loss. The following are the results obtained from the hair loss data.

Fig 1.6: Hair loss data

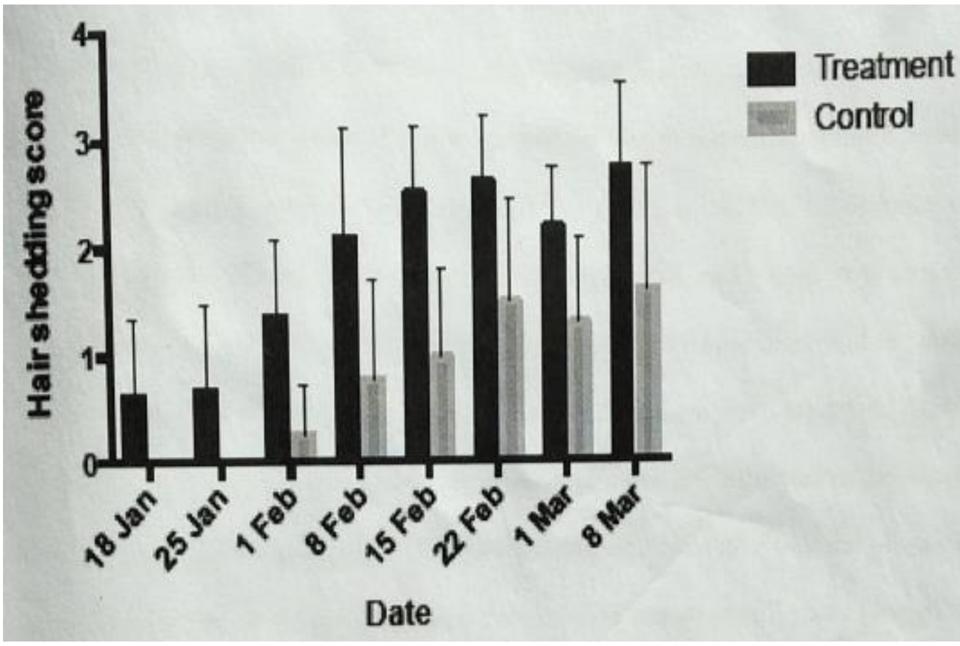

Fig 1.6: The treatment group (n=16) began to shed their hair ~ 2 weeks earlier than the control group (n=13) and thereafter, shed their hair at an increased rate compared to the control group. Hair loss was ranked on a scale from 1-4. 1 being no hair loss, 4 being heavy hair loss.

Murphy et al obtained the following results from their study in 2017 "Artificially extended photoperiod administered to pre-partum mares via blue light to a single eye: Observations on gestation length, foal birth weight and foal hair coat at birth". With regard to the first group (situated in Ireland), they found that, Gestation length on average was shorter in the group of mares fitted with light masks to those that were not. There was no difference in the mean Foal Birth weight between the two groups. Results from the Second Group (Kentucky) were as follows; No difference in number of foals sired by sire X in comparison to Sire Y. Birth weight was higher in group 1(those fitted with light masks) when compared with control group 2. However, within a group there was no difference in Birth weight between foals (fillies or colts). Gestation length: No difference in gestation length between group 1 (light masks) and control group two or between male and female foals. Regarding the Third Group (Ireland- coat quality) "*A correlation matrix was compiled to assess the associations between coat growth dynamics and all other variables"(Murphy et al 2017). In* 2020 Yoon et al obtained results that were negative for both Body condition score and uterine follicles. Neither group of mares demonstrated follicles greater than 35mm on either 6th January or 10th February. The study considered several factors that scientifically may have led to such a result. Taking into account Body condition score, Age of mare, and environmental conditions. Age and Body condition score were even among groups but unmistakably there were some environmental factors that were noted. These included sub-zero temperatures in February with a minimum average temperature of -7.2 to -12.8 degrees Celsius. The study concluded that although light masks have been proven to advance the breeding season of mares (Murphy et al 2014) their effects in different environmental conditions have not been explored. As the authors of this study stated, *"Temperatures differed from Walshe's study".*

Aurich et al obtained the following results from their study on the effect of blue light therapy in pregnant mares on parturition and foal maturity. All mares and foals were healthy throughout the experimentation period and after parturition. There were no complications during pregnancy or parturition in those mares exposed to blue light therapy. Gestation length was shorter in mares exposed to blue light therapy versus the control group. The placental surface of the treatment and control groups showed no significant difference. The hair length was significantly lower in newborn foals of those mares exposed to Blue light therapy versus the control group of mares. The foals of those mares exposed to light therapy stood faster than the control group foals. However, there was no significant difference between time of birth to first suck from mare's udder between foals of treatment and control groups. There was no significant difference between cannon bone circumference and Body weight between foals from treatment and control groups. *"Foals born to mares exposed to blue LED light were smaller at birth and had a markedly shorter hair coat but did not differ in weight from foals born from control pregnancies." (Aurich et al).*

In a study carried out in 2022 to determine the effect of blue light therapy on foal development until 1 year old when the mare was subjected to blue light therapy the results were as follows*:* There was no difference in foal body temperature at any time between treatment and control groups. There was no difference in foal blood glucose parameters at any time between treatment and control groups. There was no difference in HRV (heart rate viability) and RMSSD (Root mean square of successive differences) between treatment and control groups. As expected, height and size of foal in both treatment and control groups increased during the first 12 months of life but there was no significant difference demonstrated between the two groups. No effect on weight, height, carpus to fetlock length, chest circumference, cannon length was demonstrated. Guard hairs were shorter in foals born to blue LED treated mares. This difference persisted until 2 months old but after this time guard hair length decreased in control foals and on average evened out between both groups by 8 months old. Authors found that foals born earlier in the season were born with longer guard hair compared to those born later in the season. Hair loss changed overtime, hair coat in individuals 4 months and older was not affected by blue light.

Discussion/ Conclusion

One of the factors known to influence gestation length is parity, or number of previous foals the mare has had. Parity influences uterine size with multiple parities resulting in expansion of the uterus. Smaller foals in a larger uterus will have a longer gestation period than a larger foal would. This is since parturition is initiated when the fetal hypothalamic-pituitary-adrenal axis is stimulated due to the stress of confinement, nutritional insufficiency and/or hypoxia (Pashan and Allen 1979). It has already been demonstrated that light therapy can advance the onset of estrus in mares, and so, we now aim to determine whether light therapy affects the gestation length of the mare and subsequent birth weight of the foal when the mare is exposed to a lengthened photoperiod. We recall the study by Hodge et al in 1982 into the influence of photoperiod on pregnant and post-partum mare. They found that Foals of the treatment group had a shorter gestation period and on average, a heavier birth weight although it was not significantly heavier. Photoperiod is an environmental variable that we expose the mare to. In 1996, Rossdale and Short stated that "the length of pregnancy in the mare is known to be influenced both by the genotype of the fetus and the environment of the mother". The environmental factor in this case is that we are exposing the mare to an extended photoperiod. An extended photoperiod can increase the levels of sexual hormones in the body causing ovulation, development of the corpus luteum (CL) and the subsequent release of progesterone from the CL which plays a major role of the variation in gestation length and appears to be independent of nutritional factors (Howell and Rollins 1951). Upon the suppression of melatonin by light, the hypothalamus secretes Gonadotropin releasing hormone (GnRH) which in turn causes the pituitary gland to secrete Follicle stimulating hormone (FSH) and luteinizing hormone (LH). FSH secreted by the anterior pituitary stimulates the maturation of ovarian follicles. LH secreted by the anterior pituitary gland stimulates ovulation and development of the corpora lute once ovulation has been achieved, simultaneously other seasonal hormones are released such as prolactin and growth hormone. Prolactin concentrations increase in spring as the natural photoperiod extends. Prolactin is the primary hormone involved in vernal hair shedding (Thompson and Hoffman 1997). We recall the study by Ryg and Jacobsen in 1981 to investigate the seasonal changes in growth rate, feed intake, growth hormone and thyroid in young red deer. They reported that the hormonal changes in young red deer are thought to control somatic growth, antler, growth and food intake. This is due to a strong circadian rhythm and seasonality of growth hormone. In this experiment "*To investigate the effects of light therapy on gestation length and foal birth weight" (Baugh, Equine Science Project 2014)* we used low wavelength blue light from LED's in a light mask as our source of light therapy. The results indicate that foal birth weights in the mares exposed to light therapy from 1st December of their gestation were significantly higher than the foals from the control mares not exposed to light therapy. Blue light from light emitting diodes causes a dose dependent suppression of melatonin in the horse (Walsh et al 2012). This suppression of melatonin causes a reduction in the inhibition of GnRH pulse frequency from the hypothalamus (Cleaver et al 1991). This means the secretions of gonadotropins from the pituitary axis are increased. It is probable that light therapy increases levels of maternal hormones which are signaling messages to the foal in utero. It is interesting to note that 60-80% of fetal development occurs in the last trimester of pregnancy, this would suggest that light therapy administered to the pregnant mare at this stage of her gestation would be of most benefit to the developing fetus. Lutzer et al carried on from this point in 2022 when they administered light therapy to mares in late pregnancy and studied the effects of this on the development of the foal until one year old. The authors found that foals born to light treated mares were born with shorter guard hairs and that any differences in size and weight of foals between treatment and control groups had evened out by the time the animals were eight months old. We also determined from this experiment in 2014 "*To investigate the effect of light therapy on gestation length and foal birth weight"* that light therapy had no effect on the gestation length of the treatment mare when compared to that of the controls. Age can have a significant effect on the gestation length of the mare as well as the sex of the foal. Colt pregnancies have been shown to be significantly longer than fillies (Davies et al 2002). However, when we took age into account and blocked for age using an ANOVA analysis, we still found no significance (Baugh 2014, Equine Science Project). This could be surprising as it had already been determined that the size of the foal has a significant effect on the gestation length of the mare and previous studies have determined that mares exposed to a lengthened photoperiod had on average a 10-day shorter gestation than control mares of the same study (Hodge at al 1982). However, our insignificant findings could be attributed to a reduced sample size to accommodate ANOVA analysis. For future experimentation, a larger sample size and a more uniform mare age may determine a more significant result for gestation period. We see that Murphy et al did such in their study in 2017. They conducted their study with a larger sample size of mares, having 3 different study groups in total and a significant result in terms of gestation length was obtained. The group of mares located in Ireland showed a significant result for gestation length with the mares fitted with light masks having an on average, shorter gestation length.

The results of our hair loss data demonstrated that light therapy advanced the onset of hair shedding in our treatment group by ~ 2 weeks compared to the control group. Prolactin is the hormone that mediates the onset of hair shedding. It is inhibited by darkness and thus there are low levels of circulating prolactin during the winter months (DL Thompson et al 1997). The longer days of spring brings with it a lengthened photoperiod which increases the secretion of prolactin in the horse, mediating the onset of vernal hair shedding. In this experiment, when the control group caught up with the treatment group and began to shed their hair, the treatment group were shedding their hair coat at an increased rate when compared to the control group. This means that when treated with light therapy, mares will lose their coats sooner and faster than if they were not exposed to light therapy. This advanced and rapid hair loss results in a better-quality coat as the coats are shed out over a shorter period of time.

The significant results of this study(Baugh, Equine Science Project, 2014) showed that exposing the mare to light therapy during her gestation significantly increased the birth weight of the foal. We can exclude nutrition as a contributing factor as both the treatment and control groups were fed the same diets. The only varying environmental factor between the two groups was the exposure of the treatment group to light therapy from December 1st onwards. This corresponded to between days 226 and 252 of gestation.

The primary aim of this study was to determine if light therapy had a significant effect on the gestation length of the mare and birth weight of the foal. A secondary aim was to determine the difference in coat shedding in those exposed to light therapy and those unexposed. This study demonstrates that mares exposed to a lengthened photoperiod during their gestation period give birth to heavier foals. These results could provide breeders with valuable information on how to control foal birth weight, excluding genetics and nutrition as contributing factors. However, it would prove valid to backup this preliminary study with further research into the effect of light therapy on gestation length by altering the experimental design to have more uniformity in traits relating to the experimental units such as age, parity and an increased population size. Reassessing experimental design according to these factors could give rise to more significant results in relation to gestation length. This study also proved successful in determining the rate of coat shedding when mares are exposed to light therapy from light masks. Our findings demonstrate that mares fitted with light masks began to shed their coats about two weeks before those with no light masks and continued to lose their coats at a faster rate thereafter. (Baugh, Equine Science Project 2014)

Murphy et al advanced the study from this point in 2017. Having a larger sample size of mares and a larger overall study they obtained a significant result for gestation length, with treatment mares having an overall shorter gestation length. In summary, this study describes the effects of exposing Thoroughbred mares to low intensity blue light during their gestation period. The study demonstrates a decrease in gestation length in groups of mares exposed to such light. These such mares have previously had prolonged gestation periods. This can be implemented as both beneficial to welfare and Industry. This study also demonstrated a significant effect on birth weight and demonstrated a finer, lighter hair coat of foals born to mares exposed to light therapy during the gestation period. The author says the preliminary results are promising but more work is needed.

"An important area for further study is to determine what other aspects of foetal developmental physiology are influenced by the photoperiodic stimulus perceived by the mare." (Murphy et al 2017).

As the research continues from this point, we begin to look at the effect of light therapy on parturition and foetal development. Aurich et al looked at blue light therapy administered to the pregnant mare and how this could affect mare gestation, parturition and foal maturity. The authors of this study concluded that the onset of foaling can be advanced by the use of Blue LED light aimed at one eye and such advancement has no negative consequence on the foal. They concluded at blue light therapy in the treatment group resulted in the birth of slightly smaller foals with a shorter hair coat than that of the control group. The authors stated that artificial light treatment can reach the fetus and speed up its development. *"demonstrating that artificial light directed at the mare does reach its fetus and accelerates fetal maturation*.

In 2022 Lutzer et al further explored the effect of blue light therapy on foals that were born to mares exposed to blue light therapy. The authors of this study evaluated the development of these foals until they turned one year old. The authors concluded that foal size between control and treatment groups will equalize within the first 12 months of life and that the initial difference in guard hair between control and treatment groups will not persist beyond 12 months old. The authors continued by stating that from this evidence it would highly suggest that Blue LED light administered to the mare to advance parturition or to decrease the interval from parturition to first ovulation will have no negative effects on the foal and growing weanling and yearling.

It is important to note that while Light therapy can successfully manipulate the mare's natural circadian rhythm to a more advantageous timeline for breeders and industry professionals to bring about early onset to estrus, the environment in which the mare is housed must also be advantageous to her internal cycle. In a study carried out by Yoon et al in 2020 we see a negative result for early onset to estrus when blue light therapy was introduced to a group of mares. The authors concluded that the harsh , cold winds in South Korea on that particular year during the experimentation period of November to February may have had a negative effect on the reproductive hormonal function of the mares. It is the authors opinion that while the masks have been proved to bring about the early onset of the breeding season, they have not been tested in such environmental conditions as those previously described. For future studies the authors of this paper wish to conduct a similar experiment, application of light masks with simultaneous hormonal assays to detect hormonal changes and analyze their mechanisms. With such further studies the authors may find a way to make the masks function to their full capability in a new and more challenging environment. This finding demonstrates to us how the external environment the mare is housed in is an important contributing factor to optimizing the effects of light therapy on the Internal hormonal environment of the mare.

Summary

In this paper we discuss the development of light therapy technology in mares. It had previously been determined that exposing the mare to an extended photoperiod of 6-8 weeks could advance the breeding season by causing ovulation at a faster rate (Palmer and Draincourt 1985). In 2014 Murphy et al concluded that automated individual light masks were shown to be as effective as traditional barn lighting in advancing the breeding season in mares. We concluded from our study in 2014 *"To investigate the effect of light therapy on gestation length and foal birth weight" (Baugh, Equine Science Project)* that light therapy administered via individual blue LED light masks increased foal birth weight, increased the rate of coat shedding in the mare but had no significant effect on gestation length. However, a similar but more extensive study carried out by Murphy et al in 2017 found a that in a group of mares housed in Ireland, gestation length was on average, shorter in those mares fitted with light masks compared to those that were not. In this paper we aim to examine how this research has evolved. We see more extensive and specialized studies being carried out. Aurich et al (2022) look at the effect of blue light in pregnant horse mares on gestation, parturition and foal maturity. They find that gestation length was shorter in mares exposed to light therapy and that although foals born to light treated mares were smaller, they did not differ in bodyweight from those foals born to mares not subjected to light treatment. They also found that foals born to light treatment mares stood faster and had a shorter hair coat when born to those foals on non-light treated mares. In 2022 Lutzer et al track the development of foals born to blue light treated mares until one year of age. They conclude that there was no difference in normal newborn foal parameters between treatment and control groups and that the initial shorter hair coat and size differences had equalized between treatment and control groups by the time foals reached 8 months old. We also note how external environment can contribute to the internal hormonal environment of the mare (Yoon et al 2020).

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