



Research Article

THE EFFECT OF EXOGENOUS MELATONIN ON BLOOD BIOCHEMICAL PARAMETERS IN BLACK BENGAL GOAT

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Abstract: The present study was conducted to see the effect of exogenous melatonin on blood biochemical parameters on Black Bengal goat in the Department of Veterinary Physiology, Ranchi Veterinary college, Birsa Agricultural University, Kanke, Ranchi, Jharkhand. The experimental animals healthy, non-pregnant, non-lactating Black Bengal does (n=21), having average body weight (16.39 kg) and age between (16-24 months), reared under uniform managemental husbandry practices were selected from Instructional Farm of Small Ruminants (I.F.S.R.) for the present experiment during non-breeding season. Selected does were isolated from bucks at least one month prior to start of the experiment. The mean of total serum protein (g/dl) on 0 day was 6.37 ± 0.21 , 6.50 ± 0.16 and 6.56 ± 0.14 (g/dl) in group I to III respectively. The mean of Serum glucose (mg/dl) of group I, II and III on 0 day was 61.2 ± 2.52 , 56.81 ± 1.96 and 54.92 ± 1.44 respectively. Serum glucose was significantly ($p < 0.05$) lower in group II and III in comparison to I from 20 to 40 day. The mean of serum inorganic phosphorus (mg/dl) on 0 day was 4.5 ± 0.25 , 4.77 ± 0.23 and 4.46 ± 0.48 (mg/dl) in group I, II and III respectively. Total serum inorganic phosphorus was significantly ($p < 0.05$) higher in group II and III in comparison to group I from day 20 to 40 in comparison to 0 day. The mean of serum calcium (mg/dl) of group I, II and III on 0 day was 9.54 ± 0.20 , 8.91 ± 0.22 and 8.85 ± 0.12 respectively. On 0 day serum calcium was significantly ($p < 0.05$) higher in group I in comparison to group II and III. The mean of serum magnesium (mEq/l) on 0 day was 0.95 ± 0.03 , 1.02 ± 0.02 and 0.97 ± 0.03 (mEq/l) in group I, II and III respectively. Serum magnesium was significantly ($p < 0.05$) higher in group II and III in comparison to group I at 20 days to the end of experiment. Majority of the biochemical constituent increased in melatonin treated goats except glucose.

Keywords: Black Bengal goat, melatonin, biochemical parameters

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Introduction

Goats (*Capra hircus*) are among the earliest animals domesticated by humans. The Goat population has declined by 3.82% over the previous census and total goat in the country is about (35.17 million numbers) which contributes about 26.40% among livestock (19th livestock census, 2012). Importance of goat in the rural economy is evidenced by its unparalleled economic traits, ability to get acclimatized under diversified agro climatic condition, unsatisfied type in choosing the available forage, high fertility and short generation interval. Black Bengal is a promising dwarf goat for its excellent adaptability, fertility, prolificacy, delicious meat and high quality skin. Animals express both daily rhythms and seasonal cycle in their biochemistry, physiology and behavior. The most obvious biological rhythms are the daily cycle of sheep and wakefulness and seasonal cycle of growth and reproduction. Melatonin (N-acetyl-5-methoxytryptamine) plays role in biologic regulation of cardiac rhythms, sleep, mood and aging. Melatonin has close interaction with hypothalamic pituitary gonadal axis. Its secretion is high during the dark period which indicates that melatonin synthesis and release is regulated mainly by the dark light cycle. Exogenously administered melatonin from continuous slow release expands has been shown to advance the onset of the breeding season in goats by mimicking the stimulatory effect of short days [5]. Goat are practically seasonal breeders, showing peak reproductive activity between June and August and October and January. A study was carried out to investigate the effect of exogenous melatonin on biological parameters in Black Bengal goats.

Materials and methods

The present study was conducted on Black Bengal goat in the Department of Veterinary Physiology, Ranchi Veterinary college, Birsa Agricultural University, Kanke, Ranchi, Jharkhand. The experimental animals healthy, non-pregnant, non-lactating Black Bengal does (n=21), having average body weight (16.39 kg) and age between (16-24 months), reared under uniform managemental husbandry practices were selected from Instructional Farm of Small Ruminants (I.F.S.R.) for the present experiment during non breeding season. Selected does were isolated from bucks at least one month prior to start of the experiment. Bucks were introduced into each group after completion of exogenous administration of melatonin that is one month after the start of experiment. Design of experiment was submitted for approval by the Institutional Animal Ethics Committee vide letter no.-147/528/RVC/IAEC.

Grouping of Animals and Experimental Design

The selected animals were allocated randomly into three groups (viz. group-I to group-III) comprising seven animals in each group. The experiment was conducted as per following schedule:

Group-I (Control): The does in this group were maintained under normal animal husbandry practices and were served as control. **Group-II (Melatonin administration orally, 3 mg/day):** The does in this group were maintained under normal practices and additionally melatonin was administered orally (3mg/day) for one month to each animal.

Oral Administration: 3 mg melatonin was dissolved in 5 ml of water-ethanol (8:2 v/v) and was given orally to each animal. [10]. **Group-III (Melatonin administration subcutaneously, 1mg/day):** The does in this group were maintained under normal practices and 1 mg melatonin was administered subcutaneously daily for one month. Subcutaneous Administration: 1 mg melatonin was dissolved in 2 ml of saline-ethanol (9:1 v/v) and was given subcutaneously to each animal. [14]. Oral / subcutaneous administration of melatonin was done in the morning hours before feeding.

Blood Sampling

Blood sample with and without anticoagulant in vials was collected from jugular vein of each animal day before the start of experiment and thereafter on 20th, 30th, 40th and 50th day. The collected blood samples were taken immediately to the laboratory in ice box. Anticoagulant added blood was used for haematological study. Non anticoagulant added blood samples were allowed to clot at room temperature in centrifuge tubes for 2-3 hours. The clots were eased away from the edge of the vessel and allowed to shrink overnight at 4°C. The serum was pipetted off, centrifuged at 3000 rpm for 10 minutes and stored at -20°C for further analysis of biochemical parameters.

Biochemical parameter

The following biochemical parameters were recorded in all groups before the start of experiment and thereafter on 20th, 30th, 40th and 50th day as per the methods given below -

Total Serum Protein: Total serum protein was estimated by Biuret Method as described by [12]. Tulip diagnostic kit was used for the estimation of Total serum protein. **Serum glucose:** Serum glucose was determined by GOD/POD method as described by [23]. Tulip diagnostic kit was used for the estimation of serum glucose. **Serum Phosphorus:** Serum phosphorus was measured by Molybdate U.V. method as described by [11]. Tulip diagnostic kit was used for the estimation of serum phosphorus. **Serum Calcium:** Serum calcium was estimated by OCPC method as described by [3]. Tulip diagnostic kit was used for the estimation of serum glucose. **Serum Magnesium:** Serum magnesium was estimated by Calmagite method as described by [9]. Tulip diagnostic kit was used for the estimation of serum magnesium.

Statistical analysis

Statistical analysis of the data generated was done by standard statistical procedure as per [21].

Results and discussion

The present experiment was carried out to see the effect of exogenous melatonin administration in different blood biochemical constituents' occurrence of estrus in Black Bengal goats during non breeding season.

The results obtained are here under:

Total Serum protein (g/dl)

The value of total serum protein (g/dl) has been depicted in [Table-1] and [Fig-1]. On 0 day the mean of total protein was 6.37 ± 0.21 , 6.50 ± 0.16 and 6.56 ± 0.14 (g/dl) in group I to III respectively. Among different period serum protein at 50 days was significantly ($p < 0.05$) decreased in group II in comparison to 20 days. A decreasing trend of serum protein was observed in all remaining group from 30 to 40 days but this decrease was statistically non significant. [17] reported that melatonin treatment did not cause a significant change in total protein level in rat, the similar pattern was observed in the research finding also the values obtained in the present experiment are within normal range as reported by [6, 7, 22].

Serum Glucose (mg/dl)

The value of serum glucose has been depicted in [Table-2] and [Fig-2] Serum glucose (mg/dl) of group I, II and III on 0 day was 61.2 ± 2.52 , 56.81 ± 1.96 and 54.92 ± 1.44 respectively. Serum glucose was significantly ($p < 0.05$) lower in group II and III in comparison to I from 20 to 40 days. Among different periods serum glucose decreased non significantly on day 30 in comparison to the value

on day 20. However, increasing trend of serum glucose was observed in group II and III on 40 to 50 days of experiment. The increase in serum glucose was however non significant. [1] reported blood glucose was significantly lower without change in plasma insulin level when melatonin was administered. The present findings are not in agreement with the finding of [16] and [8] who reported an increase in the level of glucose after the administration of exogenous melatonin

Table-1 Serum protein concentration (gm/dl) of Black Bengal goats in different groups at different periods (Mean $\pm$ S.E)

Period of observations	Group I	Group II	Group III
0 day	6.37 ± 0.21	$6.50^{ab} \pm 0.16$	6.56 ± 0.14
20 days	6.63 ± 0.23	$6.62^a \pm 0.25$	6.60 ± 0.19
30 days	6.34 ± 0.18	$6.36^{ab} \pm 0.14$	6.62 ± 0.24
40 days	6.15 ± 0.18	$6.28^{ab} \pm 0.11$	6.43 ± 0.14
50 days	6.11 ± 0.26	$6.14^b \pm 0.04$	6.46 ± 0.13

Means bearing different superscript vary significantly ($p < 0.05$) within the groups (a, b) and did not vary significantly between the groups

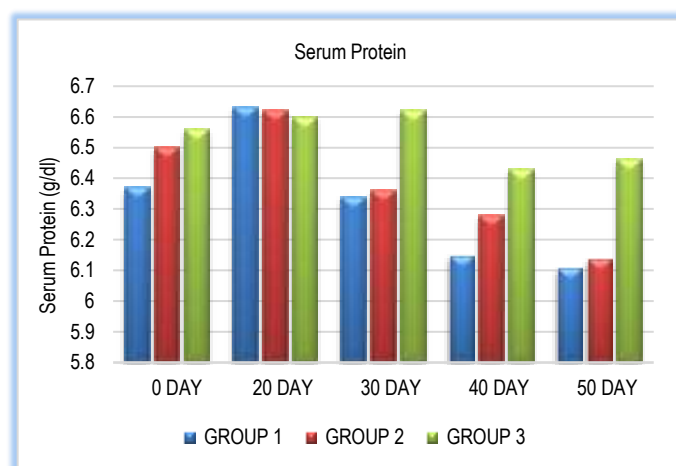


Fig-1 Bar diagram depicting the Mean Serum protein (gm/dl) of Black Bengal goats in different groups at different periods

Table-2 Serum glucose concentration (mg/dl) of Black Bengal goats in different groups at different periods (Mean $\pm$ S.E)

Period of observations	Group I	Group II	Group III
0 day	$61.2^A \pm 2.52$	$56.81^{AB} \pm 1.96$	$54.92^B \pm 1.44$
20 days	$61.05^A \pm 2.37$	$55.11^B \pm 2.08$	$52.9^B \pm 1.33$
30 days	$61.05^A \pm 2.37$	$53.48^B \pm 2.06$	$51.38^B \pm 1.22$
40 days	$61.1^A \pm 2.36$	$54.44^B \pm 2.08$	$52.52^B \pm 1.33$
50 days	$61.1^A \pm 2.31$	$55.71^{AB} \pm 2.04$	$53.5^B \pm 1.28$

Means did not vary significantly within the groups and bearing different superscript vary significantly ($p < 0.05$) between the groups (A, B)

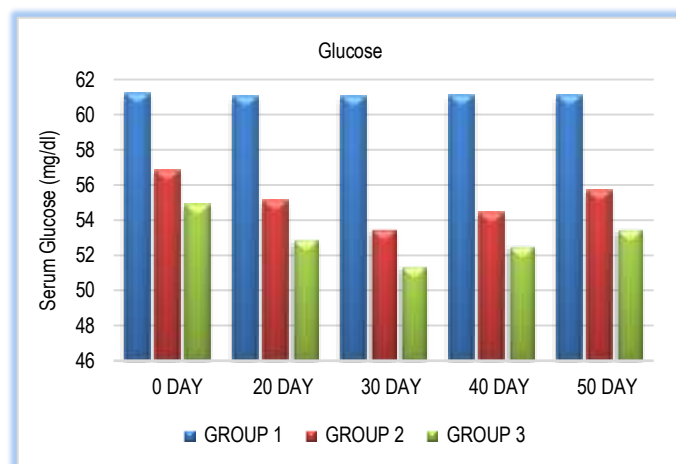


Fig-2 Bar diagram depicting the Mean glucose (mg/dl) of Black Bengal goats in different groups at different periods

Serum inorganic phosphorus (mg/dl)

The value of serum inorganic phosphorus (mg/dl) has been presented in [Table-3] and [Fig-3]. On 0 day the mean of serum inorganic phosphorus was 4.5 ± 0.25 , 4.77 ± 0.23 and 4.46 ± 0.48 (mg/dl) in group I, II and III respectively. Total serum inorganic phosphorus was significantly ($p < 0.05$) higher in group II and III in comparison to group I from day 20 to 40 in comparison to 0 day. Among different periods serum inorganic phosphorus was significantly ($p < 0.05$) higher at 20, 30 and 40 days in group II and III in comparison to 0 day. However, at 50 days serum phosphorus was significantly ($p < 0.05$) lower in group III in comparison to 20, 30 and 40 days. The present finding is in agreement with [15] who reported a significant increase in serum phosphorus levels in ovariectomized rats receiving melatonin as compared to their controls.

Table-3 Serum inorganic phosphorus concentration (mg/dl) of Black Bengal goats in different groups at different periods (Mean $\pm$ S.E)

Period of observations	Group I	Group II	Group III
0 day	4.5 ± 0.25	$4.77^c \pm 0.23$	$4.46^c \pm 0.48$
20 days	$4.36^B \pm 0.24$	$6.27^{Ab} \pm 0.29$	$6.27^{Aab} \pm 0.29$
30 days	$4.04^B \pm 0.21$	$7.42^{Aa} \pm 0.11$	$6.93^{Aa} \pm 0.47$
40 days	$4.30^B \pm 0.27$	$6.49^{Aab} \pm 0.22$	$5.64^{Aab} \pm 0.53$
50 days	$4.41^B \pm 0.21$	$6.21^{Ab} \pm 0.61$	$5.13^{Abc} \pm 0.46$

Means bearing different superscript vary significantly ($p < 0.05$) within the groups (a, b, c) and between the groups (A, B)

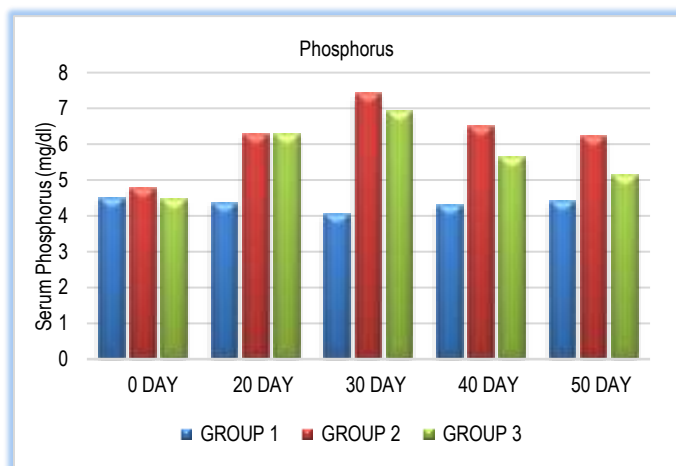


Fig-3 Bar diagram depicting the Mean Serum inorganic phosphorus (mg/dl) of Black Bengal goats in different groups at different periods

Table-4 Serum calcium concentration (mg/dl) of Black Bengal goats in different groups at different periods (Mean $\pm$ S.E)

Period of observations	Group I	Group II	Group III
0 day	$9.54^A \pm 0.20$	$8.91^B \pm 0.22$	$8.85^{Bc} \pm 0.12$
20 days	9.5 ± 0.22	9.22 ± 0.22	$9.12^{bc} \pm 0.15$
30 days	9.58 ± 0.22	9.48 ± 0.22	$9.55^a \pm 0.14$
40 days	9.48 ± 0.22	9.31 ± 0.22	$9.34^{ab} \pm 0.15$
50 days	9.38 ± 0.20	9.01 ± 0.23	$9.0^{bc} \pm 0.15$

Means bearing different superscript vary significantly ($p < 0.05$) within the groups (a, b, c) and between the groups (A, B)

Serum calcium (mg/dl)

The value of serum calcium has been depicted in [Table-4] and [Fig-4]. Serum calcium (mg/dl) of group I, II and III on 0 day was 9.54 ± 0.20 , 8.91 ± 0.22 and 8.85 ± 0.12 respectively. On 0 day serum calcium was significantly ($p < 0.05$) higher in group I in comparison to group II and III. Among different periods serum calcium was significantly ($p < 0.05$) higher in group III on 30 days in comparison to 0 and 20 days. In other group serum calcium increased non significantly on 30 days in comparison to 0 and 20 days whereas serum calcium was significantly ($p < 0.05$) decreased in group III at 50 days in comparison to 30 days. In all other group serum calcium decreased non significantly on 40 days in comparison to 30 days and this trend was continued up to end of experiment. The increase in serum calcium following melatonin treatment is similar to the finding of [18] and [1] who

reported an up regulation of melatonin to blood calcium level [18]. Probably based on the present of existed evidence for the pineal control of the secretion of parathyroid hormone and calcitonin, confirmed by the ultra-structural and functional changes observed in parathyroid gland after pinealectomy [13].

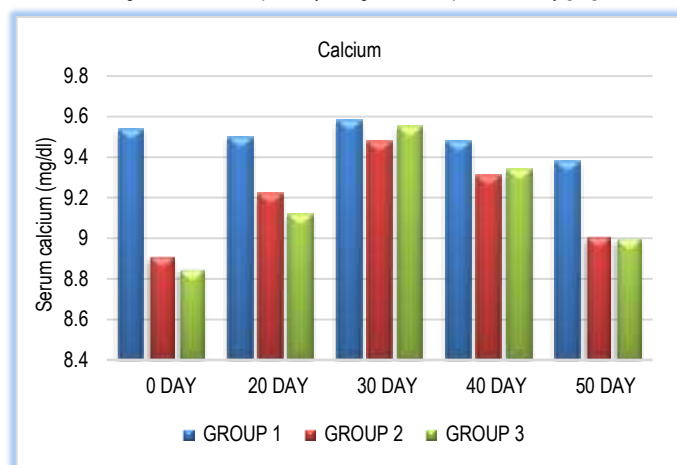


Fig-4 Bar diagram depicting the Mean Serum calcium (mg/dl) of Black Bengal goats in different groups at different periods

Table-5 Serum magnesium concentration (mEq/liter) of Black Bengal goats in different groups at different periods (Mean $\pm$ S.E)

Period of observations	Group I	Group II	Group III
0 day	0.95 ± 0.03	$1.02^{bc} \pm 0.026$	$0.97^c \pm 0.03$
20 days	$0.93^B \pm 0.02$	$1.07^{Ab} \pm 0.01$	$1.04^{Ab} \pm 0.02$
30 days	$0.94^B \pm 0.02$	$1.13^{Aa} \pm 0.01$	$1.13^{Aa} \pm 0.01$
40 days	$0.94^B \pm 0.02$	$1.07^{Ab} \pm 0.01$	$1.04^{Ab} \pm 0.01$
50 days	$0.92^B \pm 0.02$	$1.00^{Ac} \pm 0.01$	$1.00^{Abc} \pm 0.01$

Means bearing different superscript vary significantly ($p < 0.05$) within the groups (a, b, c) and between the groups (A, B)

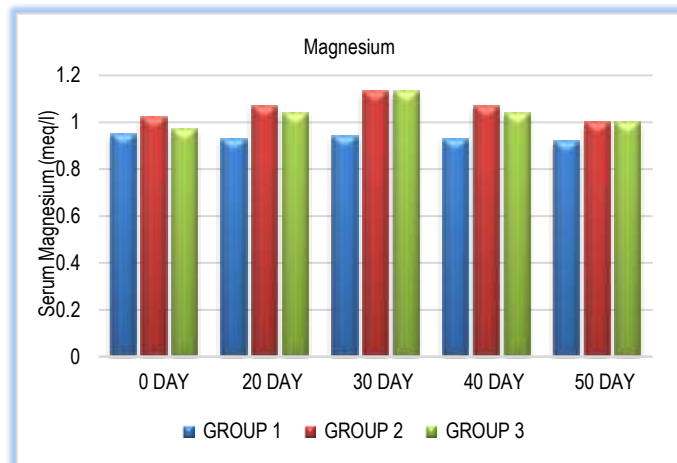


Fig-5 Bar diagram depicting the Mean Serum magnesium (mEq/liter) of Black Bengal goats in different groups at different periods

Serum magnesium (mEq/litre)

The value of serum magnesium (mEq/l) has been presented in [Table-5] and [Fig-5]. On 0 day the mean of serum magnesium was 0.95 ± 0.03 , 1.02 ± 0.02 and 0.97 ± 0.03 (mEq/l) in group I, II and III respectively. Serum magnesium was significantly ($p < 0.05$) higher in group II and III in comparison to group I at 20 days to the end of experiment. Among different periods serum magnesium at 20 days was significantly ($p < 0.05$) higher in group II and III in comparison to 0 day. At 30 days of experiment serum magnesium was significantly ($p < 0.05$) higher in group II and III in comparison to 0 and 20 days of experiment, whereas at 40 days serum magnesium was significantly ($p < 0.05$) lower in group II and III in comparison to 30 days and this trend was continued up to 50 days in group II. Our finding is in agreement with the finding of [4] who reported that magnesium concentration was significantly higher in the group of rats receiving melatonin compared with control.

In many respects' magnesium metabolism is specific in relation to other micro elements. The regulatory mechanisms of magnesium flow have not been discovered up to now. Inside the cell magnesium act as a catalyst to all reactions that use ATP. In this manner magnesium affects all endergonic and exergonic processes in the organism [2]. The present research finding is not in agreement with finding of [20] who reported plasma magnesium did not show significant changes to the treatment of melatonin.

Summary

Serum glucose was significantly ($p < 0.05$) lower in group II and III in comparison to I from 20 to 40 days. Among different periods serum glucose decreased non significantly on days 30 in comparison to the value on day 20. Total serum inorganic phosphorus was significantly ($p < 0.05$) higher in group II and III in comparison to group I from day 20 to 40 in comparison to 0 day. Among different periods serum inorganic phosphorus was significantly ($p < 0.05$) higher at 20, 30 and 40 days in group II and III in comparison to 0 day. Among different periods serum calcium was significantly ($p < 0.05$) higher in group III on 30 days in comparison to 0 and 20 days. In other group serum calcium increased non significantly on 30 day in comparison to 0 and 20 days where as serum calcium decreased significantly ($p < 0.05$) in group III at 50 days in comparison to 30 days. Serum magnesium was significantly ($p < 0.05$) higher in group II and III in comparison to group I at 20 days up to the end of experiment. Among different periods serum magnesium at 20 day was significantly ($p < 0.05$) higher in group II and III in comparison to 0 day. At 30 days of experiment serum magnesium was significantly ($p < 0.05$) higher in group II and III in comparison to 0 and 20 days of experiment, whereas at 40 days serum magnesium was significantly ($p < 0.05$) lower in group II and III in comparison to 30 days and this continued up to 50 days in group II.

Conclusion

Majority of the biochemical constituent increased in melatonin treated goats except glucose.

Application of research: Study of Blood Biochemical Parameters in Black Bengal Goat

Research Category: Veterinary Physiology

Abbreviations:

RVC- Ranchi Veterinary College, DLC- Differential Leucocytic Count, EDTA-Ethylene Diamine Tetra Acetic Acid, BAU- Birsa Agricultural University

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***Research Guide or Chairperson of research:** Dr R.K. Verma

University: Birsa Agricultural University, Kanke, Ranchi, 834006, Jharkhand

Research project name or number: MVSc Thesis

Author Contributions: All authors equally contributed

Author statement: All authors read, reviewed, agreed and approved the final manuscript. Note-All authors agreed that- Written informed consent was obtained from all participants prior to publish / enrolment

Study area / Sample Collection: Department of Veterinary Physiology, Ranchi Veterinary College, Birsa Agricultural University, Kanke, Ranchi, 834006, Jharkhand

Experimental animal: Black Bengal goat

Conflict of Interest: None declared

Ethical approval: Ethical approval taken from Department of Veterinary Physiology, Ranchi Veterinary College, Birsa Agricultural University, Kanke, Ranchi, 834006, Jharkhand.

Ethical Committee Approval Number: 147/528/RVC/IAEC

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