

# Comparative Evaluation of Hormonal Status Between bTB Affected and Non-affected Dairy Cattle in Chattogram, Bangladesh

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**Abstract— Background:** A cross-sectional study was done on dairy cattle in Chattogram, Bangladesh to compare the hormonal status between bTB (Bovine Tuberculosis) affected and apparently healthy cattle. **Methods:** The bTB was diagnosed by Caudal Fold Tuberculin Test (CFTT) followed by Comparative Intradermal Tuberculin Test (CITT). **Results:** By OIE recommended ( $\geq 4$  mm) cut off value, 49 cattle were found positive from 846 cattle and 44 good quality serum samples were used for hormonal analysis. Same numbers of serum samples from apparently healthy cattle were used to compare the values. The study revealed that, thyroxine (T4), triiodothyronine (T3) and free triiodothyronine (FT3) level was found moderately higher (T4,  $p=0.165$ ; T3,  $p=0.082$  and FT3,  $p=0.139$ ) but insignificant in bTB positive cattle. Serum cortisol levels were slightly higher ( $p=0.349$ ) in tuberculin positive cattle than the tuberculin negative cattle. **Conclusion:** All of these are preliminary findings and need further analyses to correlate with novel biochemical features and associated risk factors.

**Keywords—** Bovine Tuberculosis, Hormone, Caudal Fold Tuberculin Test, Dairy Cattle.

## I. INTRODUCTION

In developing countries, Tuberculosis (TB) is considered one of the common illnesses, which affect all classes of society, and zoonotic TB (animal origin human TB) is an important public health concern [1, 2]. Pasteurization of milk is not well practiced in most developing countries and 10-15% human TB is animal origin (*Mycobacterium bovis*) [3]. In Bangladesh TB remains a major public health problem and ranks fourth in the world for both prevalence and mortalities. In 2014, due to TB about 81,000 Bangladeshi died and 1, 96,797 were new cases though case notification rate is very low (53%) (USAID TB Care II Project, Bangladesh). Due to population density the demand of milk is more in Bangladesh and to fulfill these excessive demand intensive dairy farms with high yielding cross breeds and their crosses are increasing especially in urban and peri-urban areas which are more susceptible to bTB [4, 5, 6]. One billion people of the world are infected with tubercle bacillus and decreased immunity increases the development of active TB. Malnutrition, diabetes, prolonged use of corticosteroid drugs, gastrectomy, and immunosuppressive drugs are the probable risk factors for developing active TB [7, 8].

Biochemical analyses of blood serum are very useful to get insight in the metabolic and health status of animals. During diagnostic procedure, it is very useful to compare the values obtained from ill animals with normal values in healthy animal [9]. In TB infection, the activated immune cells release cytokines. The released cytokines subsequently activate the hypothalamic-pituitary-adrenal axis leading to the production of glucocorticoids and dehydroepiandrosterone (DHEA) [10].

Generally cortisol, estradiol, prolactin, thyroid hormone in plasma will be altered in tuberculous patients [8]. In human beings, a lot of information are available about the endocrine responses of TB infected patients, especially thyroid gland, adrenal glands, gonads, pituitary gland etc. [1]. In dairy cattle, definitive information is not available about comparing hormonal profiles between normal and bTB infected cattle [11].

On the basis of above discussion, the study was conducted with a reasonable sample size to give an idea about the hormonal parameters between tuberculin reactor and non-reactor dairy cattle, which might be the enhancing diagnostic accuracy.

## II. MATERIALS AND METHODS

### Study Area and Study Design

Three important dairy cattle areas namely- Double Mooring (urban), Shikolbaha (peri-urban) and Raozan (rural) of Chattogram district, Bangladesh were selected for this study. Double Mooring is an important part of Chattogram metropolitan areas and it has the highest number of intensive dairy farms with high yielding cross breeds (N=415) in compare to other metropolitan areas and the major source of milk for the city (DLO, DLS of Chattogram, Personal Communication, 2018). Among all peri-urban areas of Chattogram metropolitan area, Shikalbaha has the highest number of dairy cattle farms (N=400). Raozan is 32 km far from Chattogram metropolitan area. It has (N=150) numbers of intensive dairy farms with high yielding exotic breeds (DLO, DLS of Chattogram, Personal Communication, 2018). A farm consisting of at least 15 exotic cattle breed was defined as the smallest sampling unit. Accordingly, a total of 96 farms were enlisted among which 65,

21 and 10 farms were belonged to Double Mooring, Shikolbaha and Raozan, respectively. Individual animal of 6 month or more was considered as eligible for investigating bTB in this study. Therefore, all animals met the selection criterion were brought under the study and the distribution of animals by study site was 538 in Double Mooring, 200 in Shikolbaha and 108 in Raozan. From the total of 846 tested cattle, 49 cattle were found bTB positive by OIE recommended ( $\geq 4$  mm) cut off value. From 49 cattle, 44 blood/serum samples were used for hormonal analysis. 05 samples were missed due to the farmers non-cooperation and some others technical problems. Contrarily, 44 blood/ serum samples of CITT negative cattle ( $\geq 4$  mm) were used to compare the hematological, biochemical and hormonal parameters between tuberculin test positive reactors and non-reactors.

#### Laboratory Test

Different parameters of hormone (TSH, T<sub>3</sub>, T<sub>4</sub>, FT<sub>3</sub>, DHEA and Cortisol) was tested by using hormonal kit (Human Gesellschaft für Biochemica und DiagnosticambH, Max-Planck-Ring 21. 65205 Wiesbaden, Germany). For hormonal parameters ELISA Reader (Model: 2100 – C, Spain), Microplate washer (Model: 2600 – C, Spain), Autoclave (Model: SX – 500, Japan) were used.

#### Statistical Analysis

Laboratory data obtained were entered into Microsoft Office Excel-2007 and transferred to the software STATA/IC-13 for analysis. Student t-test was applied to compare the results of the hormonal parameters between comparative groups. The results were presented as mean, standard error, 95% confidence interval and p-value. The p value of 0.05 or less was set as a cut off.

### III. RESULTS

Of the total 846 cattle, 49 cattle was positive for bTB and the remaining are negative in this study. Table I, shows that the comparison of different hormonal parameters between bTB positive and bTB negative group of cattle. None of hormonal parameters was differed significantly ( $p \leq 0.05$ ) between comparative groups in the present study.

### IV. DISCUSSION

In this study, the thyroid hormones, viz TSH, T<sub>4</sub>, T<sub>3</sub> and FT<sub>3</sub> were evaluated between tuberculin reactor and non-reactor cattle. In all cases, the estimated values were statistically insignificant. A prospective study of 114 patients with equal number of age and sex matched controls with same dietary habits revealed that there was no significant difference with the baseline thyroid profile of the general population [12] but a modest increases of thyroid hormones (T<sub>3</sub> and T<sub>4</sub>) were reported in other study [8]. Modest increased T<sub>4</sub> ( $p=0.165$ ), T<sub>3</sub> ( $p=0.082$ ) and FT<sub>3</sub> ( $p=0.139$ ) levels were found in this study. Very recently, the other researchers compared cortisol, triiodothyronine (T<sub>3</sub>) and thyroxin (T<sub>4</sub>) values in TB suspected and healthy elephants and found no apparent differences [13]. Decreased T<sub>4</sub> and T<sub>3</sub> values and unaffected TSH values were reported from 266 confirmed pulmonary tuberculosis (PTB)

cases [14]. The adrenocortical functions were studied in tuberculin positive dairy cattle of intensive dairy farm and the findings compared with those in tuberculin negative cattle. In this study, serum cortisol levels were slightly higher ( $p=0.349$ ) in tuberculin positive cattle than the tuberculin negative cattle. Higher serum cortisol values but different levels (both significant and insignificant) were also reported from some studies done on active tuberculous human patients [10, 15, 16, 17]. There was considerable variation in serum cortisol concentrations between TB suspect and healthy elephants, but overall means were not significantly ( $p > 0.05$ ) different [13].

TABLE I: Comparison of serum hormonal parameters between tuberculin reactor and non-reactor cattle with *Mycobacterium bovis* infection (performed by CFT test followed by CITT) (N=44 positive and 44 bTB negative animals).

Variable	Category	Mean $\pm$ SEM	95% CI	p value
TSH (mIU/L)	Positive	1.4 $\pm$ 0.01	1.3-1.4	0.686
	Negative	1.4 $\pm$ 0.01	1.3-1.4	
T <sub>4</sub> ( $\mu$ g/dl)	Positive	7.5 $\pm$ 0.4	6.7-8.3	0.165
	Negative	6.6 $\pm$ 0.5	5.7-7.6	
FT <sub>3</sub> (pg/ml)	Positive	2.03 $\pm$ 0.1	1.8-2.3	0.139
	Negative	1.8 $\pm$ 0.1	1.6-2.0	
T <sub>3</sub> ( $\mu$ g/dl)	Positive	1.9 $\pm$ 0.1	1.7-2.2	0.082
	Negative	1.6 $\pm$ 0.1	1.4-1.9	
DHEA ( $\mu$ g/ml)	Positive	1.6 $\pm$ 0.04	1.6-1.7	0.485
	Negative	1.6 $\pm$ 0.02	1.6-1.6	
Cortisol (ng/ml)	Positive	7.2 $\pm$ 1.3	4.7-9.8	0.349
	Negative	5.8 $\pm$ 0.8	4.2-7.4	

### V. LIMITATIONS

During the collection of blood, some physiological conditions of the cattle such as before meal or after meal, physical exercise, stress, temperature etc. were not considered. Incentives to farmers were not provided in this project. For this, all farms owners and attendant were not fully motivated and cooperated. For this, some CITT positive cattle were missed from collection of blood sample. Some manufacturer's instructions were violated. The power of this study was low, because only 44 blood samples were collected from CITT (OIE recommended cut-off value based) positive cattle.

### VI. CONCLUSION

From the best of our knowledge, this is the first study in Bangladesh where the hormonal parameter were compared between bTB affected and non-affected groups of dairy cattle. The results of the study declared that there is no significant difference among the hormonal parameters between this groups. Further studies should be needed to correlate with novel biochemical parameters and associated risk factors of bTB affected dairy cattle.

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