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Physical and reproductive features of Mali: An indigenous pig breed of Tripura with special emphasis to impact of weaning on haemato-biochemical parameters

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Abstract
The study was conducted to produce basic information about the physical and reproductive features of Mali breed of pig with special emphasis to changes in hemato-biochemical parameters during weaning of piglets. The haematological parameters were estimated by using an automatic blood analyser. The biochemical parameters were analyzed by using diagnostic kits as per manufacturer's instruction using spectrophotometer. The observations of the study showed mature body weight of female Mali pig is 64.14 ± 2.25 kg with age of puberty and length of estrous cycle are 154.12 ± 1.08 and 20.64 ± 0.13 days respectively. Different physical and haemato-biochemical parameters like body wt. changes (kg), Hb (%), total WBC, lymphocyte count, PCV(%), granulocyte (%), platelet counts, plasma albumin and aspartate transaminase (AST) showed significant (p<0.05) alteration among the pre-weaned and weaned group. The present study has generated the basic information of physical and reproductive features of Mali pig which signifies the importance of conserving this native breed. The current study might also help in designing further elaborate research to understand the variation of different physiological rhythms during weaning stress.

Keywords: Haemato-biochemical, mali, piglets, physical, reproduction, weaning

Introduction
In India, livestock farming has been considered to be the key contributor of agricultural economy. Among livestock species, pig plays an important role mostly in the different communities and tribal masses of North Eastern states and also in other parts of India. Small holder mixed livestock systems contribute significantly toward the increase in livestock production. Tripura, a small state of North-East India is home to a diverse mix of tribal cultures and religious groups with maximum dependency on agriculture and allied activities. Due to its hilly terrain and forest cover, only 27 % of the land is available for cultivation. So, from self sustainability and food safety point of view animal farming is very much essential in Tripura. As per 20th Livestock census [1]. The pig population of Tripura is 2.06 lakh out of which around 50% is of indigenous breeds. Tripura is home to important indigenous breed or desi pig mainly dome and Mali among which Mali has been very recently recognized as breed by National Bureau of Animal Genetic Resources, India. It is featured with good pig genetic resources with distinctly superior production and reproduction characteristics [2]. The Mali breed is mostly reared by tribal population in Tripura and is very much preferred due to certain characteristics such as observed less susceptibility to diseases, easy rearing by traditional rearing system and feeding on local forage and kitchen waste. Mali pigs with positive productive and reproductive performance have a huge potential to improve the livelihood of the poor pig farmers.

In organized piggery, weaning is an important practice for economical and productive management of pig farms. The sow comes to heat immediately after weaning of piglets, starting a new reproductive cycle and also prolonged suckling by piglets reduces body resources of sows, with concomitant time in recovery. However, weaning is also a common stressful event in the life of a piglet, which may lead to low feed intake, weight loss, increased mortality and potentially compromised health [3]. After weaning the young mammals being nutritionally and behaviorally independent from the mother can alter the stress responses in adult progeny [4, 5]. It is reflected in different studies that epigenetic factors occurring in critical

periods of life can have affect on permanent physiological alterations [6, 7, 8] as stresses can alter the circadian rhythm [9]. In swine production, determination of hemato-biochemical parameters is of prime interest in connection with monitoring health and nutritional status [10]. However, the physical, reproductive and haemato-biochemical base line data on Mali pig is very scanty and no report available regarding the physical and haemato-biochemical status of Mali pigs during weaning. Hence, the present study was undertaken to generate a reference data on the physical and reproductive features of Mali and also emphasis was given to represent the alteration of haemato- biochemical parameters during weaning. The presented data can provide the basic information about this pig breed which can be utilized for development of manage mental measures for optimal growth and promotion of animal welfare.

Materials and Methods

Selection of animals

For study of physiological and reproductive features, female Mali Pigs (N=10) were selected from small organized pig farms, Dhalai Tripura, India. The animals were maintained under proper hygienic condition and routine deworming and vaccination were done following the farm protocol. Physical characteristics and performance such as body weight, body measurement, and reproduction performance was studied.

For analysis of effect of weaning on growth, body measurements and hemato-biochemical parameters, two groups namely pre-weaning group and weaned group were formed with 6 (n=6) numbers of Mali piglets in each group irrespective of sex. The piglets were kept with the mother till the weaning of the piglets on 60th day. Thereafter, they were being separated and housed in different sheds.

Collection of blood sample

Blood samples were aseptically collected from animals in blood/ serum collection vacutainer tubes (4ml) on day 30 from pre-weaned (n= 6) and on day 65 from post-weaned piglets (n= 6). The non-anticoagulated blood samples were kept at room temperature for 1 hour to ensure complete clotting and serum was separated by centrifugation at 1500 rpm for 30 min. The serum samples were stored at –20 °C until further analysis. For study of hematological parameters blood samples were collected in heparinazed vacutainer tubes (2ml).

Haemato-biochemical analysis of samples

Hematological parameters were analyzed by using Auto Haemato-analyzer (Model. BC 2800 vet). All the blood samples collected from pre and post-weaned piglets were subjected for analysis of hematological parameters such as total red blood cell (RBC) count, total leukocyte count (TLC), differential leukocyte count (DLC), hemoglobin % (Hb%), platelet count, packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC).

All the serum samples representing pre and post weaned piglets were used for estimation of important metabolites including total protein, albumin, globulin and enzymes levels like aspartate transaminase (AST) and alanine transaminase (ALT) levels by spectrophotometric (Double Beam Spectrophotometer 2202, Systronics, India) methods using commercially available diagnostic kits (Coral) following standard protocol.

Statistical Analysis

Results were expressed as the means ± SE. A difference with value $p<0.05$ was considered statistically significant. The data were analyzed by Independent students T test to evaluate effect of weaning on hemato-biochemical parameters using Statistical Package for the Social Sciences (SPSS 7.5).

Results and Discussion

The Mali pig in Tripura generally known as desi pig among tribal farmers reared with kitchen wastage and locally available forage. This breed of pig is generally black in color, have short snout which is curved upward and concave. The ear is short and erected and the pot belly is covered with thick hair. The legs are short and hoof is fully covered, sometimes white marking is present just above the hoof Fig.1-4. The bristle is black in color, male is more densely hairy then female and lengths varies from 4-7.7 cm. Shedding of hair is commonly seen in female after 1-2 farrowing. The special reproductive characteristics of Mali pig is given in table 1. They generally show early sexual maturity at around 6 months of age and after furrowing strong maternal instinct with aggressive nature is observed.

Fig 1: Weaned piglets

Fig 2: Natural service in Mali pig

Fig 3: Suckling newborn piglets with sow

Fig 4: Pre-weaned piglets with sow

Table 1: The reproductive parameters of female Mali pig

Sl. No	Parameter	Description (Mean± SE)
1	No. of teat (pairs)	6.07 ± 0.19
2	Mature body wt. (Kg)	64.14 ± 2.25
3	Length of canine teeth (inch)	2.72 ± 0.06
4	Age at puberty (Days)	154.12 ± 1.08
5	Age of Sexual Maturity (days)	189.25 ± 3.37
6	Length of estrous cycle (Days)	20.64 ± 0.13
7	First Farrowing age (Days)	309.5 ± 2.11
8	Litter size in first farrowing (no. of piglets)	5.71 ± 0.24
9	Piglet Mortality (%)	0.42 ± 0.13
10	Oestrous after farrowing (in days)	61.57 ± 2.06
11	Farrowing interval (days)	176.71 ± 2.07

The mean ± SE value of various haematological parameters of both the pre weaning and weaned group of piglets are listed in table 2. The weaned piglets were found to have significantly higher body weight gain, Hb%, TLC, lymphocyte% and PCV. On the other hand, the pre-weaned piglets were found to have higher neutrophil% and platelet count. However, no significant difference was observed between the pre-weaning and weaned groups in values of total RBC count, monocyte%, MCV, MCH and MCHC.

Table 2: The average (Mean ± SE) body weight gain and haematological parameters in pre-weaning and weaned Mali piglets**.**

Parameters	(Mean ± SEM) in piglets		Significance (p value)
	Pre-weaning group	Weaned group	
Body wt gain (Kg)	5.857 ± 0.44	9.316 ± 0.67*	*p<0.05*
Total RBC	8.221 ± 0.34	9.335 ± 0.30	*ns*
Hb (gm/dl)	11.816 ± 0.20	14.683 ± 0.22*	*p<0.05*
Total WBC	11.050 ± 0.69	35.316 ± 0.60*	*p<0.05*
Lymphocyte (%)	52.33 ± 1.22	74.00 ± 2.38*	*p*<0.05
Granulocyte (%)	44.16 ± 1.32*	22.16 ± 2.05	*p*<0.05
Monocyte (%)	3.50 ± 0.42	4.33 ± 0.42	*ns*
PCV (%)	37.900 ± 0.67	46.08 ± 0.68*	*p<0.05*
MCV	45.667 ± 1.86	49.566 ± 1.21	*ns*
MCH	14.366 ± 0.66	15.750 ± 0.38	*ns*
MCHC	31.261 ± 0.29	31.850 ± 0.48	*ns*
Platelet count	596.166 ± 45.96*	210.166 ± 7.96	*p<0.05*
* = significant; ns=non-significant			

The serum biochemical parameters are enlisted in table 3 for both the group of piglets. Significantly higher serum albumin and AST level were observed in post-weaned piglets than the pre-weaned stage. However, no significant alteration was found in the level of other studied biochemical parameters between the two groups.

Table 3: The Blood Biochemical parameters in Mali piglets during pre and after weaning period

Parameters	(Mean ± SEM) in piglets		Significance (p value)
	Pre-weaning group	Weaned group	
Plasma Total Protein (gm/dl)	7.14 ± 0.14	6.56 ± 0.145	*ns*
Plasma Albumin (gm/dl)	3.95 ± 0.08	3.25 ± 0.08*	*p*< 0.05
Plasma Globulin (gm/dl)	2.75 ± 0.12	2.65 ± 0.12	ns
A/G ratio	1.45 ± 0.08	1.23 ± 0.06	ns
Plasma AST (IU/L)	21.34 ± 1.81	43.81 ± 1.88*	*p<0.05*
Plasma ALT (IU/L)	31.83 ± 2.11	35.00 ± 2.40	ns
* = significant; ns=non-significant			

Indigenous pig breeds such as Mali, a prominent pig breed of north-eastern states of India, have higher disease resistance as evident by less morbidity, case fatality and mortality observed during outbreaks of infectious diseases and adaptation to the environment due to high genetic diversity as compared to exotic ones although exotic pigs are highly productive. Therefore, it is utmost necessary to conserve this pig breed amid the tendency to cross breed the animals with highly productive exotic breeds. For understanding the potential of the Mali pigs, it is the primary requirement to know the reference values of the physical and reproductive characteristics of the breed. Table.1 represented the physical and reproductive characteristics of Mali breed of pigs. It was found that Mali is a smaller pig breed with physical features of erected short ear, pot belly, concave body back line, full hoof, and short tail with black body coat and sometimes white mark at leg just above hoof.

It was observed in our study that Mali attains sexual maturity at around 189 days. As relevant scientific data on this topic is very scanty, this breed is compared with other local breeds. As per the current finding, the female Mali pigs attain sexual maturity at an early age compared to other indigenous pig breeds of India such as naga local pigs (248.12±34.2 days) [11]. The age at first fertile service was 235±5.21 days and 241.3±2.25 days in ghungroo pig and niang megha pigs, respectively representing higher sexual maturity age

compared to Mali breed pigs [12].

The first furrowing age of Mali pig was found at around 309 days in the current study which is little higher as observed by [2]. However, the first farrowing age of Mali breed is lowest among the other local breeds in India [12, 13, 14, 15, 16, 17].

Weaning of piglets from sow is one of the most stressful events, which affects animal health and growth performance, especially during the first week of post-weaning. This weaning stress reduces the feed intake, lowers growth performance, induces oxidative stress, damages the immune system, and contributes to diarrhea in piglets [18]. Therefore, in the present study, an effort was made to evaluate the effect of weaning on the hemato-biochemical parameters in Mali piglets. Table 2 represented different physiological and hematological parameters of pre-weaning and weaned piglets. There were significant differences found in body weight gain (Kg), Hb%, total RBC, total WBC, lymphocyte (%), neutrophil (%), PCV (%), and platelet count among the groups. Data found in the present study could not be compared with the same breed of animals as no related studies could be traced in the searched literature.

Although the current study has shown a non-significant increase in RBC number in the weaned group but an significant increase in RBC number at the end of post weaning period of 60 days was reported and justified with the fact that early postnatal ontogeny and the extremely rapid organ development was responsible for the formation and maturation of RBC [19]. In the same study it was also reported that the life span of 120 days contributed the increase number of RBC in weaned group.

It was reported that, piglets were born with less efficient [20] and incompletely developed immune system at birth [21]. Although better nutrition, animal handling and breeding technology had reduced weaning stress but its adverse effects could not be completely eliminated [22]. Weaning associated deleterious effect on intestinal barrier function [23, 24, 25] allows increased epithelial permeability of toxins, bacteria, or feed-associated antigens results in inflammation [18]. In the current study it was found significant alteration in total WBC number, lymphocyte and granulocyte percentage among the pre-weaned and weaned groups. It was reported that WBC and lymphocytes count increased on days 2, 10 and 27 post-weaning [26] whereas in another research it was found that the white blood cell count of piglets increased on day 0 to days 4 and 11 post-weaning [27]. Further it was reported that the number white blood cells (WBC) increased significantly in two months aged piglets compared to one-month-old piglets [19]. As the concentration of WBC reflects the porcine health status [22] the increase in the WBC count may be due to occurrence of inflammation during weaning [18]. Therefore, both the increase of RBC and total WBC number might have contributed to the increase in PCV (%) of weaned grouped animals observed in present study.

In the present study, it was observed that the granulocyte % was reduced significantly with subsequent rise in lymphocyte count in the weaned group of piglets compared to pre-weaning stage. It was reported that the trend of neutrophil and lymphocyte percentage alters with the age of piglets and as the piglets gets older from birth the neutrophil percentage declines with subsequent rise in lymphocyte count from 6 weeks and above [28]. In another study, it was mentioned that lymphocyte counts were very significantly higher in between 9 and 15 weeks aged piglets than in preceding and subsequent periods [29]. Therefore, the higher lymphocyte count with low neutrophil in Mali pigs after weaning might be due to its generalized resistance against disease conditions beside the persistence of weaning stress.

Higher platelet count was observed in present study in the pre-weaned piglets than the weaned group, which might be due to the autophagy ability of the platelets [30]. Reduced iron store in the liver of new born piglets and low iron content in the milk of sow [31] may lead to less hemoglobin concentration in the circulation as observed in the current study, further this iron deficiency may lead to normal or higher platelet count as found in human children [32].

A significant decrease in the serum albumin level and significant increase in serum AST level was observed in weaned group compared to pre-weaning stage. No significant differences were observed in serum total protein (gm/dl), serum globulin (gm/dl), A/G ratio, plasma ALT (IU/L).

Weaning may result in poor feed intake, particularly during the initial first week [18]. Albumin being a negative acute phase protein, its lower level may be the result of inflammation due to weaning. Further, chronic stress may lead the release of glucocorticoids [33, 34] resulting into a negative nitrogen balance and increased nitrogen in the urine [35] which might have also contribute to the reduced level of albumin in the weaned group.

AST being an intracellular enzyme [36], damage to the body tissue can lead to increased serum level [37]. AST is present in all tissues except bone and this enzyme is found in exudates, and transudates in proportion to the cellular damage [38]. Recent studies had shown that weaning causing the oxidative stress may lead to enterocyte apoptosis and cell cycle arrest in small intestine in post weaning piglets [39, 40]. This kind of disrupted intestinal function and increased permeability could lead to impairment of liver function [41, 42]. The increased level of AST and ALT, which are specific indicator of hepatic damage may be due to leakage of the enzymes in the circulation [43]. Although ALT showed a non-significant increase in the post weaned group but the significant rise of AST is observed which justifies the previous studies.

Conclusion

Mali, an indigenous pig breed of Tripura with its unique physical and reproductive feature can serve as a genetic pool for livestock breeders. The present study has generated the baseline data of physical and reproductive features of Mali pig which has been very recently recognized as pig breed of India. This breed is traditionally maintained by the tribal people due to its less disease prone and better feed utilization ability with strong maternal behaviour. The impact of haemato-biochemical study on weaning will provide an important clue for the disease resistance ability of Mali pig. Further studies may be carried out to explore its better adaptive capability due to ambient stress experienced in tropical climates.

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