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Technological Intervention for Improving Productivity & Profitability in Buffalo Husbandry

EDITORS

Dr. Jerome A

Dr. Avijit Dey

Dr. Meeti Punetha

Dr. Guraraj M

Dr. Shahaji Phand

Dr. Sushrirekha Das

Dr. T.K. Datta

ICAR-Central Institute for Research on Buffaloes (CIRB)
Hisar, Haryana
National Institute of Agricultural Extension Management (MANAGE), Hyderabad

ICAR-CIRB, Hisar & MANAGE, Hyderabad

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This e-book is a compilation of resource text obtained from various subject experts of ICAR institutes & MANAGE, Hyderabad on 'Technological Intervention for Improving Productivity and Profitability in Buffalo Husbandry'. This e-book is designed to educate extension agents, dairy farmers & entrepreneurs, students, research scholars, academicians working in the field of veterinary and animal husbandry about modern buffalo husbandry practices for enhanced milk production and its value addition. Neither the publishers nor the contributors, authors and editors assume any liability for any damage or injury to persons or property from any use of methods, instructions, or ideas contained in the e-book. No part of this publication may be reproduced or transmitted without prior permission of the publisher/editor/authors. Publishers and editors do not give warranty for any error or omissions regarding the materials in this e- book. References cited in each chapter may be obtained on request from the concerned experts.

Published for Dr. P. Chandra Shekara, Director General, National Institute of Agricultural Extension Management (MANAGE), Hyderabad, India by Dr. Srinivasacharyulu Attaluri, Program Officer, MANAGE and Dr. TK Datta, Director, ICAR-CIRB, Hisar and printed at MANAGE, Hyderabad as e-publication.

MESSAGE

National Institute of Agricultural Extension Management (MANAGE), Hyderabad is an autonomous organization under the Ministry of Agriculture & Farmers Welfare, Government of India. The policies of liberalization and globalization of the economy and the level of agricultural technology becoming more sophisticated and complex, calls for major initiatives towards reorientation and modernization of the agricultural extension system. Effective ways of managing the extension system needed to be evolved and extension organizations enabled to transform the existing set up through professional guidance and training of critical manpower. MANAGE is the response to this imperative need. Agricultural extension to be effective, demands sound technological knowledge to the extension functionaries and therefore MANAGE has focused on training program on technological aspect in collaboration with ICAR institutions and state agriculture/veterinary universities, having expertise and facilities to organize technical training program for extension functionaries of state department.

Technology is developing rapidly. In this development, the transfer of computer systems and software to the application has made an important contribution. Technological instruments made farmers easy to work in a comfortable way, which leads to increased animal production efficiency and profitability. Therefore, technologic developments are the main research area for animal productivity and sustainability. Milking, feeding, environmental control, reproductive performance constitutes everyday jobs most affected by correct management decisions. This e-book describes the valuable information on the latest challenges and key innovations affecting the animal Buffalo husbandry.

It is a pleasure to note that, ICAR-CIRB, Hisar, Haryana and MANAGE, Hyderabad, Telangana is organizing a collaborative training program on on "**Technological Intervention for Improving Productivity and Profitability in Buffalo Husbandry"** from 18-20 October, 2022 and coming up with a joint publication as e-book on "Technological Intervention for Improving Productivity and Profitability in Buffalo Husbandry" as immediate outcome of the training program.I wish the program be very purposeful and meaningful to the participants and also the e-book will be useful for stakeholders across the country. I extend my best wishes for success of the program and also I wish ICAR-CIRB, Hisar, Haryana many more glorious years in service of Indian agriculture and allied sector ultimately benefitting the farmers. I would like to compliment the efforts of Dr. Shahaji Phand, Center Head-EAAS, MANAGE, Hyderabad and the Director ICAR-CIRB, Hisar, Haryana for this valuable publication

Dr. P. Chandra Shekara
Director General, MANAGE

MESSAGE

In India, the livestock sector plays a major role for the socio-economic growth of rural households as it generates around 26% of India's overall GDP from the agriculture sector. This demand-driven growth in livestock production enables millions of poor to escape the poverty trap. In this outlay, buffalo contributes over 105 million tonnes of milk and the demand for Indian buffalo meat in international market is also increasing in recent times. Buffalo meat dominates the exports with a contribution of over 89.08% in total animal products export from India. Also, buffalo contributes high-value hides, bones, and draught power for agricultural activities. Buffaloes are well appreciated for their meat and draught as buffalo milk fetches higher price, due to its high fat content (7-7.5%), almost twice that from cows. In addition, they are efficient converters of low-quality feeds, coarse fodder and agro-industry byproducts and widely renowned for their ability to thrive on low-quality crop wastes and green forage under harsh climatic conditions.

India has been regarded as gold mine of buffalo germplasm as it harbors all the well-recognized, high producing breeds of this buffalo species. Indian dairy industry is undergoing transformational change and buffalo species is going to contribute to the overall milk production as it alone contributes to 67% of total milk in the world and around 49% of India's milk production. Buffalo farming has become a livelihood and resource generating enterprise for varied strata of our farmers as it plays a major role in alleviation of poverty and the commercial buffalo enterprises now provide employment to rural communities. Buffalo acts as an important asset of rural farmers' property, possession, and profession since it provides the greatest promise for food security and sustainable development.

Considering the pivotal role played by buffalo species in the farming systems from the time immemorial, interest in this species is expanding as never before for the growth of country's agrarian economy as well as need for entrepreneurship in buffalo farming system. In his context, I appreciate the effort of MANAGE and ICAR-CIRB to organize this collaborative training on **'Technological Intervention for Improving Productivity and Profitability in Buffalo Husbandry'** to enlighten Farmers, Faculty and students of Veterinary Universities, Veterinary Professionals of KVKs and other ICAR institutes with the modern technologies of buffalo husbandry practices. I further appreciate the organizers for choosing this important topic for training, and I am confident that this compilation of resource material from the esteemed experts will serve as a source of information and knowledge for all stakeholders.

October 2022

TK Datta
Director,
ICAR-Central Institute for Research on Buffaloes

PREFACE

This e-book is an outcome of collaborative online training program on **"Technological Intervention for Improving Productivity and Profitability in Buffalo Husbandry"**. This compendium is intended for Faculty and students of Veterinary Universities, Veterinary professionals of KVKs, ICAR institutes and state Animal Husbandry departments, play vital role in the livestock sector. The compendium is our attempt to bring together the knowledge regarding modern technologies of buffalo husbandry practices in India and to develop skills on improved technologies of buffalo production systems to field functionaries. The content of proposed training programme has been designed in such a way, so that it can provide updated information towards capacity building in the proposed area. Attempt has been made to cover topics about advances in role of buffalo in ehnacin gIndian economy, availabaility of buffalo germplasm in India, nutritional management with special reference to formulation of ration and precision feeding, alternative feed resources, technologies for fodder production, silage.

Topics like feeding management of buffaloes towards green livestock production towards efficient buffalo production system and improve milk production. Other topics include stress amelioration as well buffalo health strategies. The compendium also includes topics *viz*. management of fertility in female buffalo as well bull fertility and modern reproductive technologies such as cloning. The training will also inclide the lectures on Scope and utilization of buffalo milk as well as buffalo meat as a promising animal product. The technologies for value addition of buffalo milk and meat, and their quality assessment with reference to present FSSAI rules have been incorporated. In addition, the dissemination of technologies (mobile app./ web based) of buffalo husbandry and entrepreneurship development for profitable buffalo farming shall also be covered in this training.

The compendium has been developed with the inputs from many scientists and experts across the country specialized in their area of research and development. We sincerely thank all the contributors that have allowed us to put together this document. We hope that this collection of lectures as compendium will be useful and we shall be pleased to receive comments and suggestions for further improvement.

Best Wishes

18 October 2022

Editors

Dr. Jerome A
Dr. Avijit Dey
Dr. Meeti Punetha
Dr. Guraraj M
Dr. Shahaji Phand
Dr. Sushrirekha Das
Dr. T.K. Datta

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CHAPTER 1

Enhancing Indian Economy by Improving Buffalo Productivity

T.K. Datta*

Director

ICAR- Central Institute for Research on Buffaloes, Hisar, Haryana
*Correspondence: Dr. T.K. Datta (Director, ICAR-CIRB); director.cirb@icar.gov.in

Livestock production, in particularly buffalo, cattle and small ruminants, is an integral part of food production systems, making important contributions to the quality and diversity of the human food supply. Buffalo has been an integral part of livestock agriculture in Asia producing milk, meat, hides and draft power. With more than 90% of global buffalo population present in Asia, 77.9% buffaloes are inhabitant of south Asian countries. India is home for 57% world buffalo population and contributing nearly 50% of total milk production of the country. India is 4th largest beef exporter in the world with export of buffalo meat products to the world for the worth of Rs. 24,613.24 Cr during the year 2021-22 (APEDA, 2022). Over the last 10 years, average buffalo milk productivity increased (17%) from 4.71 kg/ d to 5.54 kg (2011 to 2021).

Buffalo milk

India is the world leader in milk production with 193.46 million cows, 109.85 million buffaloes. The total milk production of the country was increased from 198.44 million tonnes (2019-20) to 209.96 million tonnes (2020-21) with buffalo share of about 45%. The per capita availability (2020-21) of milk is about 427g/d (DAHD, 2022).

Table 1. Indian buffalo population and annual growth (20th Livestock census, 2019)

Year	Numbers (Million)	Year	Annual growth (%)
1992	84.2	-	-
1997	89.9	1992-97	1.30%
2003	97.9	1997-03	1.70%
2007	105.3	2003-07	1.50%
2012	108.7	2007-12	0.60%
2019	109.9	2012-19	1.06%

Table 2. Status of buffalo milk production (DAHD, 2022)

Production (MT)				
2001-02	2010-11	2016-17	2019-20	2020-21
45.40	62.35	80.38	97.21	94.48

Buffalo milk is rich in protein, fat, conjugated fatty acids (CLA), and minerals with low cholesterol, sodium and chloride. It has double lactoperoxidase activity, more thermally stable β- lactoglobulin, larger fat globules size (4.16-4.6 μm), higher buffering capacity (25-30% more) and more Vitamin A, E and B_{12} (4 folds more). Buffalo milk is not only sweeter in taste but also more creamier and thicker with more solids, hence preferred for preparation of several traditional sweets.

Table 3. Buffalo milk- a source of extra nutrients

Constituents	**Buffalo milk**	**Cow milk**
Water (g/L)	820	870
Total solids (g/L)	165-172	125-135
Lactose (%)	5-5.5	4.8
Protein (%)	4-5	3-4
Fat (%)	6-9.5	3.6-4
Cholesterol (mg/g)	0.65	3.14
Conj. Linoleic Acid (mg/g fat)	6.1	5.5
Calcium (mg/100g)	264.0	165.0
Phosphorus (mg/100g)	268.0	213.0
Magnesium (mg/100g)	30.0	23.0
Sodium (mg/100g)	65.0	73.0
Potassium (mg/100g)	107.0	185.0
Vitamin-E (mg/100g)	33.0	31.0
Vitamin-A (IU)	33.0	30.3
Vitamin-C (mg/100g)	6.70	1.90

Buffalo Meat (Cara- beef)

India is the fourth largest beef exporter of the world with major export destinations (2021-22) were Egypt, Vietnam, Malaysia, Indonesia, Iraq and Hong Kong (APEDA, 2022). India accounts for about 43% of the world buffalo meat production, with Uttar Pradesh producing the most, followed by Andhra Pradesh, Maharashtra and Punjab. The country has exported 1175193.02 MT of buffalo meat products to the world for the worth of Rs. 24613.24 Crores/ 3303.34 USD Millions during the year 2021-22 (APEDA, 2022).

Buffalo meat is considered a part of human diet with good vitality and favorable effect on prevention of diseases. The structure, physicochemical, nutritional, functional and sensory properties of buffalo meat are comparable with beef. Buffalo meat has been reported to have the lowest concentration of total lipids (1.37 g/100 g) among all red meats. The energy value of buffalo meat was found to be 57.22% lower than that of beef. Palmitic, stearic, oleic and linoleic acids were reported to be the major fatty acids in the phospholipids of buffalo meat. Buffalo calves have shown to produce meat with the most favorable (n-6)/(n-3) ratio (7.00) compared to bovine calves and adult buffaloes. Buffalo meat has the advantage of having less fat and cholesterol than beef and has been rated better than beef by many researchers. Buffalo meat is also reported to have a higher concentration of conjugated linoleic acid (1.83 mg/g fatty acid) than meat from zebu-type cattle (1.47 mg/g fatty acid). Buffalo meat had lower levels of atherogenicity index (0.41 ± 0.04 vs 0.57 ± 0.06) and thrombogenicity index (1.16 ± 0.13 vs 1.63 ± 0.13), compared to beef. Concentrations of vitamins B_6, B_{12} and some minerals such as iron, potassium and zinc are high in buffalo meat.

Strategies to enhance buffalo productivity:

- Genome based selection and faster multiplication of superior germplasm using assisted reproductive technique.
- Improvement in semen production technologies.
- Ensuring clean production system of meat and milk: For promoting exports and also for domestic consumers.
- Precision buffalo farming
- Economic and climate resilient buffalo feeding modules for enhanced feed efficiency.
- Enhancing value addition, processing and market linkage: Preventing wastage of products due to contamination, unfair trade practices and elimination of intermediate agencies in marketing will further enhance the profit margins.
- Strong policies and funding for solving the issues in buffalo reproduction, dissemination of superior germplasm, disease control and marketing.

References: On request

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CHAPTER 2

Buffalo Germplasm and Breeding Interventions for their Productivity Enhancement

Vikas Vohra* and Supriya Chhotaray

AGB Division
ICAR-National Dairy Research Institute, Karnal-132001, Haryana, India
*Corresponding author: Dr. Vikas Vohra (Principal Scientist), vohravikas@gmail.com

World buffalo population is represented by two major groups – African wild buffalo (*Syncerus caffer*) and Asian buffalo (*Bubalus bubalis*). The Asian buffalo consists of three sub species – the 'anoa' of Celebes, the 'tamaro' of Mindori and the 'arni' or Indian wild buffalo. Representatives Arni buffalo of which are still found in the Assam's forests in India. There are two general types of the domesticated buffalo, the river or water buffalo and the swamp buffalo. The river buffalo has 50 chromosomes and the swamp type has 48, however, the amount of genetic material is same in both. They interbreed and produce fertile progeny with 49 chromosomes. The African buffalo has not been domesticated. The *B. bubalis* is widely distributed in Asia, but it has also been introduced to European countries, South America, the former Soviet Union and the Caribbean. India is the top most buffalo producer and shares about 56.7% and 97% of total world's and Asian buffalo populations, respectively (Ahmad et al., 2022). The riverine buffalo is mostly reared as a dairy animal with several well recognized breeds, whereas the swamp buffalo are primarily used for their draught ability (Colli et al., 2018). On reviewing the literature regarding the domestication of buffalo it is revealed that there is separate origin of Riverine and Swamp buffaloes in the World.

ICAR-National Bureau of Animal Genetic Resources (NBAGR) which is the nodal agency of India having responsibility of maintain inventory of farm animals and livestock has recognized diversity of buffalo germplasm. NBAGR has till date registered and provided accession numbers to a total of 19 buffalo breeds from India (NBAGR, 2021). Majority of the milch breeds of buffalo have been originated are from North and Western parts of India, probably because the domestication of riverine buffalo in the World first started during the period of Indus Valley Civilization in India and present-day Pakistan. Presently, buffalo breeds are bred in a diverse geographical region of the country, although the distribution of Riverine buffalo breeds is not uniform in the country and very few breeds are found in North-eastern states and down Southern parts of India. However, Swamp buffaloes are present mainly in North-eastern part of India. Only one breed of swamp buffalo breed (Luit) has been recognized from Assam state. Though the recent research indicates that swamp-riverine hybrids are present in Odisha. Buffaloes have varying density of population in different states and union territories. The majority of the population (72%) is concentrated in the north and western states where most of the milch breeds of buffaloes are found comprising Haryana, Punjab, Uttar Pradesh, Rajasthan, Gujarat, and Maharashtra. Indian breeds possess adaptive characteristics to thrive in the stressful environment, resistance to various tropical diseases, and the ability of converting poor quality feed resources into meat, milk and draught ability in

the field. Hence, to enable continued and sustainable use along with genetic improvement of the buffalo germ plasm of India is necessary to uplift its contribution the agrarian economy of the country along with maintenance of genetic diversity within and between breeds. This chapter aims at making the readers accustomed to the various buffalo genetic resources of India along with methods and technologies currently available to be implemented for their improvement.

Buffalo genetic resources of India

The major buffalo breeds of India can be classified into five distinct groups according to their morphological features and breeding tract as follows: (i) North India or Murrah group: Murrah, Nili – Ravi, and Gojri (ii) Western or Gujarat group: Jaffarabadi, Mehsana, Surti, and Banni (iii) Central India group: Bhadawari, Chhattisgarhi, Nagpuri, Marathwadi, Pandharpuri, and Dharwadi (iv)Eastern India group: Chilika, Kalahandi, Manda, and Luit (v) South India group: Toda and Bargur. The detailed characters of these buffalo breeds are described below:

Murrah: Murrah is a predominant buffalo of India and the world having multi utility particularly for milk and meat. The breed can be considered as the Holstein of the buffalo world. Murrah buffaloes have native breeding tract in Rohtak, Hisar and Jind of Haryana, Nabha and Patiala districts of Punjab and Western Uttar Pradesh and NCR of Delhi. Physical characteristic appearance of Murrah buffaloes include jet-black body colour with tightly curled short and spiral horns, and tight skin. Buffaloes are kept in mixed type of housing system. Murrah is one of the predominant dairy buffalo breeds that is widely used all over India for grading up nondescript buffaloes, and is referred to as improver breed of buffaloes. Murrah buffaloes are known for its high milk yield, the buffaloes producing more than 4000kg in standard lactation is not uncommon. However, the average lactation performance varies from 1800 to 2300 kg/lactation. The buffalo milk is known for the superior milk fat and it ranges from 7 to 9%. Males are used both for breeding as well as draught purpose. This breed has even formed an important place in the livestock industry of many developing countries like Bulgaria, Philippines, Malaysia, Vietnam, Brazil and Sri Lanka. This breed has spread to almost all parts of the world and is being bred either in pure form or is being used for grading up local buffaloes. It has been exported to many developing countries and is bred there. Murrah buffalo is registered by NBAGR with accession no. INDIA_BUFFALO_0500_MURRAH_01001.

Nili Ravi: The name Nili is supposedly derived from the blue water of river Sutlej. Ravi buffaloes are mostly bred in Pakistan around the river Ravi, after which they are named. During 1960's, they were grouped into one breed as Nili-Ravi probably due to shared physical characters. These buffaloes are found in Fazilka, Ferozepur, Zira and Makhu tehsils of Ferozepur district; and Patti and Khemkaran tehsils of Amritsar district in Pujab. Nili-Ravi buffaloes are generally of black colour with walled eyes and white markings on forehead, face, muzzle, legs and tail. The most desired character of female is the possession of these white markings knowns as "Panch Kalyani". Horns are tightly curved, circular in cross section, and small in size but when compared to Murrah, Nili-Ravi buffalo has large horn size

and circumference. The forehead of Nili-Ravi is slightly convex in appearance. Production performance is similar to Murrah buffaloes. However, there is a belief that Nili-Ravi animals have superior reproduction traits when compared with Murrah buffalo. Nili-Ravi breed is registered with accession no. INDIA_BUFFALO_1600_NILIRAVI_01002.

Bhadawari: Home tract of this breed is Agra and Etawah district of Uttar Pradesh and Gwalior district of Madhya Pradesh. The body is usually light or copper coloured is a peculiarity of this breed. Eye lids are generally copper or light brown colour. Two white lines 'Chevron' are present at the lower side of the neck similar to that of Surti buffaloes. The fat content of milk varies from 6 to 12.5 per cent. This breed is an efficient converter of coarse feed into butterfat and is known for its high butter fat content in milk. In the recent past this germplasm is considered to be threatened breed of buffalo probably because the milk production is less economical and breed is facing stiff competition from the Murrah breed. Moreover, the number of animals in the native tract is low. This breed is registered under accession number of INDIA_BUFFALO_2010_BHADAWARI_01003.

Mehsana: Mehsana breed is evolved out of crossbreeding between the Surti and the Murrah and is found in Mehsana, Sabarkanda, and Banaskanta districts in Gujarat and adjoining Maharashtra state. Animals are mostly black, and black brown or brown. Horns are generally sickle shaped with the curve more upward than in the Surti and less curved than in the Murrah. Eyes are very prominent, black and bright bulging from their sockets with folds of skin on upper lids. The breed is known for its persistent milk yield and is a regular breeder. Farmers often maintain animals for commercial production unit. Grazing is practised in rainy season along road side or river beds. Animals are tied mostly at a place over day and night. The breed is registered with accession no. INDIA_BUFFALO_0400_MEHSANA_01004

Surti: Also known as Deccani, Gujarati, Talabda, Charatori and Nadiadi. The breeding tract of this breed is Kaira and Baroda district of Gujarat. The breed is named after the place of origin. Horns are flat, sickle shaped and are directed down ward and backward, and then turn upward at the tip to form a hook. Body size is Medium with two white bands below the neck. Coat colour varies from rusty brown through silver-grey to black. Skin is black or brown. The Surti buffalo is lighter in body weight, as compared to heavy breeds, consume less feed, thrives well both on stovers and on limited or no green fodder, and produce milk with high fat and SNF content. It is popular with land less, small and marginal farmers. This breed is registered by NBAGR with accession no. INDIA_BUFFALO_0400_SURTI_01005.

Jaffarabadi: Heaviest breed of buffalo due to large body size and adult body weight. The breeding tract of this breed is Gir forests, Kutch and Jamnagar districts of Gujarat. Black but some animals having white or grey tail switch. Horns are heavy and peculiar in shape and exhibit wide variation, but usually emerge out by compressing the head, go downward sideways, then upward and inward finally forming a ring like structure. Jaffarabadi has superior milk and fat production. Farmers usually mix "Chhachh" (Butter Milk) in drinking water to prevent the animals from drinking water from any other source. Animals are heavy grazers. They are maintained on natural pastures throughout the year. Jaffarabadi buffalo is registered with accession ID: INDIA_BUFFALO_0400_JAFFARABADI_01006.

Nagpuri: also called as Elitchpuri or Barari. This breed is native to the Vidarbha region of Maharashtra. The breeding tract is spread across Nagpur, Akola, and Amrawati districts of Maharashtra. These are black coloured animal with white patches on face, legs and tail. The horns are long, flat and curved, bending backward on each side of the back forming a peculiar "sword shape". This typical shape of horn provides protection to themselves from wild animals while moving through forest. Nagpuri buffaloes are adapted to hot climatic condition of Maharashtra. These buffaloes are used for heavy draught purpose. Farmers prefer this breed due to its low maintenance cost, better efficiency of feed conversion, moderate production, and better adaptation to local hot climatic conditions. This breed is registered by NBAGR with accession no. INDIA_BUFFALO_1100_NAGPURI_01007.

Pandharpuri: These buffaloes are concentrated in Pandharpur, North Solapur, South Solapur, Barshi, Akkalkot, Sangola and Mangalvedha tehsils of Solapur district; Miraj, Walwa, Jathand Tasgaon tehsils of Sangli district; and Karveer, Shirol, Panhala, Radhanagri, Hatkanangale and Gadhinglaj tehsils of Kolhapur district. Gawali and Joshi are local breeders. They maintain these buffaloes. Animals are usually housed in the open close to human dwellings. Coat colour is usually black but varies from light to deep black. White markings are found on forehead, legs and tail in few animals. Horns are very long and extend beyond shoulder blade, sometimes up to pin bones. Horns have 3 different shapes i.e., Bharkand - curving back ward and usually twisted, Toki - curving backward, upward and usually twisted outward, and Meti - flat running down. Nasal bone is very prominent, long and straight. Milking behaviour is unique. Farmer take animals to customers door and supply milk as per requirement. This breed is registered with accession no. INDIA_BUFFALO_1100_PANDHARPURI_01008.

Marathwadi: Breeding tract extends to Latur, Nanded, Beed, Parbhani, and Jalna districts of Maharashtra. Colour varies from greyish black to jet black. Some animals have white markings on forehead and lower parts of the limbs. Horns are parallel to the neck, reaching up to shoulder but never beyond shoulder blade. Horns reach up to the shoulder unlike in Pandharpuri buffaloes where these may reach up to pin bones some time. This breed is registered with accession ID of INDIA_BUFFALO_1100_ MARATHWADI _01009.

Toda: The Toda breed is known after its herdsmen, the Toda tribe of the Nilgiri Hills of south India and the management of this breed is a semi-wild. Animals are kept loose and are rarely tied. The animals are reared by Toda tribe and other tribal people of the region. These animals have religious utility. The predominate coat colours are fawn and ash-grey. Toda buffaloes thrive well in the high rainfall and humid area. Typical markings consist of a narrow band of dense hair covering the top line from the crest of neck to the point of origin of tail and two chevron markings - one just around the jowl and the other anterior to the brisket. The breed is considered to be threatened breed because of their lower numbers. This breed was registered with accession no. INDIA_BUFFALO_0018_TODA_01010.

Banni: Maldharis community rearing these buffaloes came to the banni region from Halieb in Afganishtan about 500 years ago with their animals in search of pasture and settled in India. Their animals were named after the local area of "Banni" in Kachchh district. Soil of Banni region is highly calcareous, saline and loam sandy with poor water holding ability.

Banni buffaloes thrive in this region and are adapted to the local climatic condition and high salinity. Banni buffaloes are reared in an extensive system of management with night grazing. Majority Maldharis maintained 15 to 25 animal herds and the herd size varies from 10 to 100 or 150 animals. Body colour is mainly Black, sometimes copper colour. Horns are vertical and upward in direction with inverted double/single coiling, whereas only single and tight coiling of horns is characteristic of Murrah buffalo. Compared to the Murrah buffalo these animals have loose body built and loose skin. Banni buffalo is registered under the accession no. INDIA_BUFFALO_0400_BANNI_01011.

Chilika: Name of breed is derived from its native tract, which is surrounding the Chilika lake in Odisha. Breed is distributed across Bhusandapur, Tangi, Parikuda, Maluda, Krishnaprasad, Brahmagiri, and Satapada area surrounding Chilika lake. The area mostly comprises of saline zone. These buffaloes feed on submerged weeds and aquatic vegetations in saline waters of Chilika lake. This breed is medium to small in size with compact body, strong legs and small udder. Milk production is low and animals are milked only once in the morning. The milk and curd of these buffaloes have better taste and preservation quality, and is preferred by local people. The curd from milk of Chilika buffalo can be preserved for 5-7 days at room temperature. This breed has accession no. of INDIA_BUFFALO_1500_CHILIKA_01012.

Kalahandi: Breeding tract is in Kalahandi district of Odisha. Kalahandi district in Orissa. Kalahandi buffaloes are usually left loose in the morning and allowed to graze across forest, hillocks, roadside vegetation and harvested fields throughout the day. Milking is done once in the morning before grazing. Coat colour of this buffalo is usually blackish grey, sometimes grey. Muzzle, eyelids, tail and hoofs are black. Long horns, convex head, round and medium udder with tail extending below hock are typical features of this breed. The breed is losing its relevance due to less milk production and competition faced from Murrah and its crosses. The Murrah crosses are quite common in southern part of Odisha adjoining Andhra Pradesh. This breed is registered with accession no. INDIA_BUFFALO_1500_KALAHANDI_01013.

Luit (Swamp): Also known as Assamese swamp buffalo. People of Assam have been traditionally rearing swamp buffaloes in the embankment and small islands of the mighty river Brahmaputra (Luit), hence these buffaloes are named Luit. Horns are broad at base, curved upward to form a semi-circle and taper to a narrow tip. Light white stockings up to the knee are present in both fore and hind legs. Bullocks are excellent draught animals for carting and ploughing specially in muddy field for paddy cultivation. The diploid no. of chromosome (2N) in Luit (Swamp) buffaloes is 48, out of which 23 pairs are autosomes and one pair is sex chromosome. This breed is registered by NBAGR with accession no. INDIA_BUFFALO_0212_LUIT_01014.

Bargur: also called as Malai Erumai or Malai Emmai. Named after its distribution area in Bargur hills of Tamil Nadu in Anthiyoor taluk, Erode district. Malai - means hills; Erumai/Emmai- means buffalo. Coat colour vary from black to light brown or brownish black. Greyish white stockings from carpal/tarsal joint to fetlock are present predominantly in females. The body size of the breed is small and lowest height at withers among the existing registered buffalo breeds of India. The number and distribution of these animals is limited.

Milk production is very low and milk is mainly used for house hold consumption as curd and butter milk. The breed is registered with accession no. INDIA_BUFFALO_1800_BARGUR_01015.

Chhattisgarhi: As the name suggest this breed is found in North and Central parts of Chhattisgarh State. Chhattisgarhi buffalo bullocks are preferred over cattle bullocks for ploughing the rice fields especially during monsoon. Horns are medium to large in size and directed laterally backwards and then upwards with pointing tips. Reared in semi-migratory and unique management system called "Bathaan" in which during summer buffaloes are taken away from home/village to nearby forest/hillocks/free grazing land. These buffaloes are slow maturing animals and average milkers. Peda made from milk of these buffaloes is a famous milk product in the region. This breed is registered with accession ID INDIA_BUFFALO_2600_CHHATTISGARHI_01016.

Gojri: Native tract of this breed is Pathankot, Gurdaspur, Hoshiarpur, Rupnagar, and SAS Nagar (Mohali) districts of Punjab and Kangra and Chamba districts of Himachal Pradesh. Gojri buffaloes are reared in semi-migratory/pastoral management system by Gujjar community. These buffaloes have proportionated and medium built body and are mostly brown or black in colour. Horns are medium sized; mostly curved to form a big loop. These buffaloes are well adapted to foot hills. Animals can travel long distances (seasonal migration) and can climb easily on hill tops for grazing. Used for both milk and draught power (ploughing and other agricultural operations). This breed is registered with accession no. INDIA_BUFFALO_1606_GOJRI_01017.

Dharwadi: This breed is distributed in Bagalkot, Belgaum, Dharwad, Gadag, Bellari, Bidar, Vijayapura, Chitradurga, Kalaburgi, Haveri, Koppal, Raichur, and Yadgir districts of Karnataka. Dharwadi is a medium sized black colour buffalo. Head is straight with erect ears and horns are semi-circular and almost touching to wither. Udder is medium in size with cylindrical teats. It is reared mainly for milk purpose. Its milk is used for preparation of famous Dharwad "Peda" with GI tag. The animals are well adapted to low rainfall areas. This breed is registered with accession no. INDIA_BUFFALO_0800_DHARWADI_01018.

Manda: Its native tract is in Koraput, Malkangiri, and Nawarangapur districts of Odisha. This buffalo has sturdy built, with well adaptation to hill ranges of Eastern Ghats and plateau of Koraput region of Odisha. Body colour is mostly ash grey and grey with copper-coloured hairs. Lower part of leg is lighter. Horns are broad, emerging slight laterally, extending backward and inward and making half circle. It is reared for draught, milk and manure. Both male and females are used for draught purpose and are reared mostly under extensive system. This breed has been registered with accession no. INDIA_BUFFALO_1500_MANDA_01019.

Conventional Breeding interventions for superior productivity

Water Buffalo is a key livestock species that frames the backbone of Indian dairy sector along with improving agricultural economy and supplying meat and draught power. Buffalo is a multi-utility germplasm and is often referred to as "*Black Gold*". Buffalo is well known for its ability to produce high quality milk, with superior fat (6.4–8.0% vs. 4.1–5.0%) and

protein (4.0–4.5% vs. 3.4–3.6%) contents compared to cow milk (Khedkar *et al*., 2016). Presently, there are approximately 109.8 million buffaloes contributing to 49% of the total milk produced in India (BAHS, 2018-19). This beautiful genetic resource has shown 1% increase in total population and 6% increase in milch (milk + dry) animals in 20th Livestock Census when compared to the previous Census report. These results and growth of the germplasm points towards its utility, acceptance and contribution in the livestock and dairy sector.

In India the effective selection of superior germplasm and advanced animal breeding technologies for buffaloes though has been practised over decades yet not widely adopted till today because of varied genetic resources in the organised, institutional, and unorganised farms. Several types of breeding structures are being used in India for genetic improvement of buffalo population. Almost all efforts were directed towards superior milk production. Selection and breeding are the main program for recognized breeds of buffaloes and the non-descript buffaloes are improved through grading up program with Murrah, Mehsana and Surti breeds to augment the milk production potential. Various schemes have been developed and implemented by Indian Council of Agricultural Research (ICAR) and State Agricultural Universities (SAUs) for improvement of defined breeds. Progeny-testing (PT) program was started in the Third Five-Year Plan to ensure identification of superior Murrah bulls tested on the basis of performance of their progeny rather than only the dam's yield. Network project on the buffalo improvement started in the year 1993 and its scope was extended during the 9th Five-Year plan, to undertake performance evaluation and improvement of various important breeds of buffaloes available in different parts of the country. Progeny testing is currently one of the most effective and widely adopted tool across India for superior sire selection. The tested bulls are used through artificial insemination (AI) for achieving higher genetic gain. Subsequently, field progeny testing programs for Murrah and other indigenous breeds of buffaloes supported by the Government, Cooperatives dairies, Research Institutes and NGOs have also been made to select the superior bulls for the production and supply of semen throughout India. The easiest way of multiplication and improvement of buffalo in organised farms could be done through artificial insemination technology by using available superior male germplasm. The old saying envisage that 'Bull is half of the herd' however over the years through research it has been established that through AI 'Bull is more than half of the herd' as the bull contributes more expected genetic improvement of milk production between generations than female dairy animal.

The multiplication and improvement of buffalo germplasm in organised herds could be done through AI by testing bulls and evaluating superior sires through progeny testing programme. The progeny testing may be based on single herd or multiple (Network/ Associated) herds. In Indian condition, the progeny testing in institutional herds involves testing a minimum of 12-15 bulls on about 1000 breedable buffaloes that participate in the programme. The semen of proven bulls is used for nominated mating with elite females either in single herd or multiple herds. The elite dams for particular breed should be high yielding animals to be selected based on selection criteria like defined first lactation milk yield, best lactation milk yield and breeding value for milk production for different breeds.

The breed specific young test bulls should be selected from elite males (sons of proven sires) born through nominated mating in multiple organised herds, based on breed characteristics, breeding soundness evaluation, pedigree records, dams as well as paternal grand dams, dam's best lactation 305 days yield, expected breeding values, semen quality and freezability. Test bulls for the organized/institute herds may be selected, if required, from the village herds based on breed characteristics, health, free from reproductive and zoonotic diseases and performance recording such as monthly milk yield of their dams under field progeny testing programme. For selection of elite breeding bulls in the herd more importance should be given on the breed of choice under various agro-climatic regions and even within the particular region of the country so that purity of the breed is maintained. Though the present Sire selection strategy through progeny testing has invariably increased the production level of buffaloes, yet the production level is still low and evaluation relatively slow since considerable time is required to gather adequate daughter records for genetic evaluations of bulls with high accuracy.

Potential Genetic interventions in the genomic era

Here comes the role of genomic selection in the genetic improvement of dairy sires. In conventional PT programs, the generation interval of Sires and Dams of bulls are 7 and 4 years, respectively, while it is significantly reduced to approximately 2.5 years for both the Sires and Dams of bulls with the use of genomic markers in genetic evaluations (García-Ruiz et al., 2016). The generation interval refers to the average age of the Sire at which it becomes a parent, in a breeding program no Sire can't be used for breeding the cows unless its genetic merit is proven, thus, to prove a Sire i.e., by the time the daughter of a Sire performs a large time lapse thereby increasing the generation interval, but under genomic selection strategy, the time required to select the Sire is considerably low, as low as age of sexual maturity in bulls. Since genomic breeding value (GEBV) can be estimated in a dairy herd as early as the day an animal is born. Several methods for prediction of GEBVs has been developed such as SNPBLUP, GBLUP, RRBLUP, BayesA, BayesB, BayesC, and BayesCπ. The accuracy of GEBVs predicted is high, and to obtain maximum genetic gain these GEBVs should be estimated using adequate methods, adequate reference population and should be incorporated into National Evaluation Programs without further delay (Harris et al., 2008). This depends on the adaptability of National Evaluation Services and dairy industries to include genomic information for prediction of Breeding Value and selection of Sires with high genetic merit. When we consider the dairy industry then the industry-wide scale of combining pedigree, phenotype, and genomic information to predict GEBVs poses a considerable challenge. One of the major challenges lies in the limited number of animals genotyped among the population. In dairy advanced countries, genomic selection (GS) has resulted in an increased rate of genetic gains in dairy cattle and the production of genomically proven bulls. The credit for their huge success (implementation of GS) can be attributed to the fact that a well-established conventional genetic evaluation system was already in place long before the introduction of GS. On the other hand, developing countries are lagging in the implementation of GS, maybe due to a lack of proper breeding infrastructure, pedigree, systematic phenotypic recording, and the absence of computational and analytical tools, which are fundamental for the success of any conventional genetic evaluation programs.

Ribaut et al. (2010) indicated the upsurge in information and communication technology has created opportunities to counter some of the challenges specific to developing countries by establishing global virtual platforms. Next-generation sequencing (NGS) has facilitated greatly the development of methods to genotype very large numbers of molecular markers such as single nucleotide polymorphisms (SNPs). NGS technologies have improved the ability to discover genome-wide SNPs accurately and effectively and genotyping in a single step, even in species for which little or no genetic information is available (Davey *et al*., 2011). This revolution in genetic marker discovery enables the study of important questions in molecular breeding, population genetics, ecological genetics and evolution. The rapid developments in high-throughput next-generation sequencing technologies for SNP genotyping have reduced the genotyping costs. There are few consortiums working solely for genomic evaluation in dairy cattle in developing countries. Cost-effective genotyping services provided by several companies may boost the implementation of genomic evaluations, yet it is necessary to establish consortiums for genomic evaluation programs on a large scale in developing countries like India (Mrode et al., 2019).

Other interventions – improved reproduction

Reproductive bio-techniques has been seen as another significant determinant for germplasm improvement. Despite the importance of buffalo to the economic and social fabric of the region, one of the reason behind the low popularity of buffaloes as dairy bovines among farmers is their low reproductive efficiency. The low reproductive efficiency can be due to delayed puberty, higher age at calving, long postpartum anoestrus period, long calving interval, lack of overt sign of heat, and low conception rate. In addition, female buffaloes have few primordial follicles and a high rate of follicular atresia. In the developing country like India, it is essential to apply advanced animal biotechnology methods to improve animal production and to conserve the indigenous animal genetic resources. Specifically, animal reproductive biotechnologies have been proven to be useful in augmenting reproduction, implementing embryo transfer and related technologies, diagnosing diseases and controlling and improving nutrient availability. Reproductive interventions in buffaloes using new technologies helps in the distribution of elite buffalo genes and the reduction in generation interval, and provide continued genetic gain and increased production of buffalo meat and milk. Artificial insemination, super ovulation estrus synchronization, Somatic cell nuclear transfer, embryo transfer, in vitro fertilization and somatic cell cloning are some of the reproductive interventions successfully done in buffalo.

Artificial insemination (AI) uses the valuable germplasm of outstanding male animals and distribute it in the population. Use of frozen semen in straws instead of bringing the animal to spot reduces the transportation cost and increase the profit of the farmer. The major constraints in the effective utilization of AI in buffaloes include seasonal nature of reproduction, silent heat, long ovulation time, and prolonged post-partum estrus (Purohit et al., 2003). The conception rate with natural service, chilled semen and frozen semen have been reported to be more than 60, 35-60, and 25-45%, respectively (Agarwal and Shankar, 1994). Cryopreservation of buffalo embryos has been reported (Misra et al., 1992; Kasiraj et al., 1993) but pregnancy rates are very low. Recently attempts to vitrify buffalo oocytes have been made, with post vitrification recovery rate of 88-89% and morphologically normal. These oocytes could be further used for IVM and IVF (Dhali et al., 2000a, 2000b).

Likewise, in order to exploit the superior genome of a productive female animal Super ovulation technology can be implemented. Inducing super ovulation using hormones and timely harvest of ovum followed by in-vitro fertilization and embryo transfer to surrogates can increase the number of offspring from a superior female buffalo per year. This may require additional technologies like estrus synchronization, embryo transfer, embryo freezing, and in vivo maturation (IVM). Buffalo ovaries yield only a small number of quality oocytes compared to that of cattle. So, in order to improve oocyte yield *in-vitro* culture of pre-antral follicles could be done (Gupta et al., 2001). Hormones are usually added to IVM media and known to yield high maturation rates.

There are other slightly high-end technologies like the Somatic Cell Nuclear Transfer whose benefits depends on the age and type of donor cell, the stage of donor cell cycle, and nuclear programming following nuclear transfer. Cloning is yet another technique but the overall efficiency of cloning is typically lower than 10 percent, represented by the number of live offspring as a percentage of the number of nuclear transferred embryos. Reports on production of transgenic buffalo are currently not available. Gene transfer to produce a transgenic buffalo herd would be a long way with current strategies and resources.

These technologies in livestock have helped in boosting milk production, improving reproduction, faster growth of elite animals and controlling diseases, Various application of improvement, infertility treatment, breed conservation, bull fertility testing, and producing calves of desired sex, producing transgenic animals, stem cells production and therapeutic cloning. Research to make biomedical use of livestock species is in progress. India has produced first ETT calf 'PARTHAM' and several cloned buffalo calves viz., ‘GARIMA’ at National Dairy Research Institute, Karnal, India.

Conservation of buffalo germplasm

Conservation of the unique and lesser-known buffalo population at their breeding tract is essential to meet the demand of local products of various agro-climatic zones and to maintain the sufficient genetic diversity in the buffalo population. Conservation of live specimens of buffaloes consumes sizeable manpower, valuable space and costs besides demanding proper planning skills. Both in-situ and ex-situ conservation efforts are taken to conserve the buffalo breeds for sustainable utilization. The animals are maintained in their original habitats under native conditions with no interference in their mode of management, feeding and other conditions in *in-situ* conservation. However, *in situ* conservation may lead to inbreeding and genetic drift in small populations. Under the *in-situ* conservation scheme, a set of 150 buffaloes are identified on the basis of their peak yield and then the milk production is recorded at monthly interval. The owners are provided an incentive for two years so that the animals are retained and kept in good health. The tagged females are inseminated with the semen/ bull of the same breed of higher genetic merit. The farmers rear the male progeny from these females up to six months; incentives are provided to retain the calf. As these calves grow, a total of 50 unrelated males are selected as future bulls (Thiruvenkadan et al., 2013). Under the *ex-situ* conservation program, live animals are maintained at the different government institutional farms and semen samples of the superior bulls (3000 doses/animal) are collected and stored in the liquid nitrogen container for future revival (Sadana, 2010).

Summary and Conclusions

Despite having such diverse and unique buffalo germplasm, Murrah predominates the Indian buffalo population due to its with superior lactational qualities. Another reason could be extension and popularization of this breed on large scale, and the easy availability of semen from organized government farms for artificial insemination. The large-scale breeding programmes have been undertaken for the Murrah and few other buffalo breed, in India. This has led to a suppression of the need for identification and characterization of other buffalo breeds and their improvement. Monoculture of single buffalo breed may be dangerous for the milk consuming nation. Contrary to the World, where cattle are a milch animal, in India buffalo is considered as the milch animal. This may lead to situation of All or None, the case happened to European nations with the emergence of BSE in cattle. Monoculture is also detrimental to diversity. By and large, the other important utility of buffalo as meat animal has remained neglected in the country, despite of the fact that the beef eating population of the World hardly differentiate the beef from Carabeef (buffalo meat) and there is huge scope in meat export. At present there is no breed improvement programme for Carabeef trait in Indian buffalo. Every buffalo breed of India whether or not used for milk production can be bred and maintained for meat production. For example, the Chhattisgarhi breed has an excellent meat potential. Chhattisgarhi and other breeds maintained under natural pastures and extensive management must be branded for Organic milk and meat production to generate superior revenue and attach value addition to the lesser known and less utilised germplasm of buffalo in India. Chilika buffalo milk is another example where the milk and milk products, especially Dahi, has longer shelf life has largely remained unexplored. Mozerralla cheese is another example. Thus, the breeding programmes, guidelines and policies must be framed to utilising conventional and marker-based selection techniques must be initiated for productivity enhancement and sustainable utilization. Though a considerable amount of genetic improvement has been achieved through the existing conventional selection and breeding criterions, and progeny testing methods, with the advent of Next Generation Sequencing technologies, a planned action towards genomic selection and improvement of dairy buffaloes is what called for. Government along with ICAR, SAUs, Central Govt. farms should collaborate to effectively use the technologies available for implementing a large-scale genomic improvement program along with a standardized national evaluation system. State Govt. should also emphasize to conserve and breed the native breeds and should focus on the promotion of the typical products of these local breeds at national level to emphasize the importance of the breeds. Though gradually India is adopting the new technologies for genetic improvement of the buffalo germplasm yet there is a long way to go for complete expression and utilization of production potential of buffaloes. Still there are several native local buffalo populations with unique potentials whose regional demand have led to their sustenance till date, such populations need attention. However, this necessitates their identification and characterization of unique traits and understand their genetics in order to improve the genetic worth of buffaloes. Remember they are not useless but used less.

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CHAPTER 3

Round the year Fodder Management Strategies for Economic of Buffalo production

P. C. Lailer*

Division of Animal Nutrition and Feed Technology
ICAR-Central Institute for Research on Buffaloes, Hisar, Haryana
*Corresponding author: Dr. PC Lailer (Principal Scientist & Head); aglailer@gmail.com

India inhabits 15% of world livestock on 2% geographical area with 5.23% cultivated fodder area. Present availability of green fodder is 462 MT and dry fodder is 394 MT. Contribution of crop residues, cultivated fodder and grasslands is 54%, 28% and 18%, respectively. In India, cultivated fodder is limited to less than 4.5 per cent of the area under cultivation. Present area under fodder crops in India is around 8.6 million hectares. At present we are deficient by 23%, 32% and 36% in dry fodder, green fodder and concentrates, respectively.

Importance of Green Fodder

Green fodder is the primary only source of vit A for lactation which is essential for maintenance and function of the mucous membrane, directly related to vision and is essential for conception, maintenance of pregnancy and shedding of placenta. It is also essential for respiratory tract. It is essential for the urinary tract and deficiency causes stones in the kidney, ureter, bladder. During lactation 2000 I.U. of Vitamin 'A' is eliminated in every liter of milk. Green forages have cooling effect on the animal body, more palatable contain easily digestible nutrients, provide fresh effectively utilizable nutrients in natural form and laxative. Cheap source of vitamin'A', source of minerals, crude protein, total digestible nutrients and CLA. Generally leguminous fodder contains 8-12% DCP and 45-60% TDN. The phosphorus content of leguminous fodder is poor. It is advisable to supplement a ration containing a large amount of leguminous fodder with a limited quantity of wheat or rice bran, which is rich in phosphorus. The non-leguminous fodder is having 2.5% DCP and 45-60% TDN on dry matter basis. Green fodder is highly digestible (55 – 65%) mostly when harvested at a proper time.

Comparative cost of CP and TDN:

Particular	Cost of Crude Protein(Rs/kg)	Cost of TDN (Rs/kg)
Concentrate Feed	94.44	26.98
Green Fodder Legume	55.55	15.38
Net Saving (Rs)	38.89	11.60
Saving (%)	41.18	42.99

It shows that availability of nutrients from green fodder is significantly cheaper than concentrate feeds.

Major forage crops grown and their area in India

Sl No.	Crops	Botanical name	Area ('000 ha)
1.	Berseem (Egyptian clover)	*Trifolium alexandrinum*	1900
2.	Lucerne (Alfalfa)	*Medicago sativa*	1000
3.	Senji (Sweet clover)	*Melilotus indica*	5
4.	Shaftal (Persian clover)	*Trifolium resupinatum*	5
5.	Methi (Fenugreek)	*Trigonella foenum-graecum*	5
6.	Lobia (Cowpea)	*Vigna unguiculata*	300
7.	Guar (Cluster bean)	*Cyamopsis tetragonaloba*	200
8.	Rice bean	*Vigna umbellata*	20
9.	Jai (Oats)	*Avena sativa*	100
10.	Jau (Barley)	*Hordeum vulgare*	10
11.	Jowar/Chari (Sorghum)	*Sorghum bicolor*	2600
12.	Bajra (Pearl millet)	*Pennisetum glaucum*	900
13.	Makka (Maize)	*Zea mays*	900
14.	Makchari (Teosinte)	*Zea mexicana*	10
15.	Chara sarson (Chinese cabbage)	*Brassica pekinensis*	10

Fodder production technologies:

1. **Anuual fodder crops:** Various options of annual fodder crops are available to cultivate. Major annual fodder crops of India are fodder sorghum, bajra, fodder maize, lobia, guar for kharif/zaid season and berseem, oat, lucerne for rabi season.
2. **Perennial fodder crops:** Annual fodder crops are seasonal in nature and require tillage in regular manner; however, perennial fodder crops can provide green fodder for 3-5 years (Shivakumar, 2019). The major perennial fodder crops suitable for Indian climate are BN Hybrid grass, TSH grass, para grass, guinnea grass, nandi grass, anjan grass, dinanath grass and stylo (leguminous fodder). These perennial fodder crops can be grown as per the prevailing condition.
3. **Tree fodders:** Subabul, Moringa, Sesbania, Deshi babool, Anjan (*Hardwickia binata),* Khejri (*Prosopis juliflora),* Siris *(Albizia lebbeck)* are those trees which have excellent

fodder value. These trees can be grown in bund areas of field for getting supplemental fodder production.

4. **Diversified fodder production system for round the year fodder availability:** It is recommended to provide leguminous and non-leguminous fodder in balanced manner. Sole cultivation of a fodder crop provides fodder of one kind. Hence, it is advised to grow fodder crops of two or more kind. Various round the year fodder production models based on diversity of fodder crops are developed by Indian Grassland and Fodder Research Institute, Jhansi as per the rainfall availability; which are as following:

a: Round the year fodder production system (Irrigated situation): The system comprises of raising seasonal legume fodder crops, inter-planted with perennial grasses (hybrid napier / guinea grass). Hybrid Napier based cropping system (Hybrid Bajra Napier + Cowpea – Berseem) can assure round the year green fodder availability.

b: Round the year fodder production system (Rainfed situation): Subabul + Trispecific hybrid - Fodder sorghum + Pigeon pea-based fodder production system is recommended for rainfed conditions. In this system, the Pennisetum Trispecific Hybrid (TSH) is planted in paired rows at 0.75 m x 0.5 m spacing. Subabul is planted at 50 cm plant to plant spacing in between pairs of TSH. The 3 m space between such two alleys is utilized for fodder sorghum + pigeon pea cropping system in 2:1 ratio at 30 cm.

5. **Fodder on Field boundary/ Bunds/ Channels/ Non-competitive land:** Among different perennial cultivated grasses, BN hybrid grass is most suitable for bunds of irrigated areas and Tri-specific hybrid (TSH), guinea grass, anjan grass and nandi grass are suitable under rainfed conditions. Other than these perennial fodder crops, fodder cactus, subabool and fodder moringa can also be planted in bunds and non-competitive field areas.
6. **Silvo-pasture and horti-pasture based fodder production systems:** Various silvipasture models (forest trees + fodder) and horti-pasture (fruit trees+ fodder) have been recommended that produce higher forage per unit area per unit time as well as round the year than open pasture. The understory part of forest trees viz. ficus, hardvicia, morus, acacia etc. and fruit trees viz. aonla, guava, ber etc. can be utilized for growing fodder crops. Suitable fodder crop species for silvi-pasture system are cenchrus grass, marvel grass, rat-tail grass, dhaulu grass, blue panic grass, spear grass, desmodium, purple bush-bean, stylo, lablab bean etc.

Anti-quality or anti-nutritional material in Fodders

It is those substances generated in natural feed stuffs by the normal metabolism of species and by different mechanisms (for example inactivation of some nutrients, diminution of the digestive process or metabolic utilization of feed) which exerts effect contrary to optimum nutrition. Anti-nutritional factors are substances which both by themselves or through their metabolic products, interfere with feed utilization and affect the health and production of animal or which act to reduce nutrient intake, digestion, absorption and utilization and may produce other adverse effects.

Anti-Nutritional factors in forage crops

S. No.	Anti-Nutritional or Anti quality Substances	Fodder Crops
1	Nitrate	Sudan Grass, Pearl millet, Oats
2	Oxalates	Guinea Grass, Bajra and Napier Grass Hybrid, Setaria Grass, Kikyu Grass, Buffel grass
3	Saponins	Lucerne
4	Tannins	Fodder tree/Shrubs
5	Cynogens	Sorghum, Sudan grass, Jhonson grass
6	Mimosine	Subabul
7	BOAA	Lathyrus

Nitrate: Nitrate is the form of nitrogen that the plant roots take up from the soil, and is transported to the leaves. Excess nitrates accumulate in plants when they are stressed. Drought or hot dry winds put forage under water stress often resulting in nitrate accumulation. Damage caused by hail or frost impairs photosynthesis resulting in excess nitrates. Cool cloudy weather can also cause the problem. During initial growth, much of the nitrate taken up by the plant is used for root and shoot development. At this stage, the roots are able to take up more nitrate than is required and it accumulates in the stems and leaves of the plant. As the plant develops, the leaves of the plant are able to convert more nitrate into plant protein, therefore less "surplus" nitrate is found in the plant as it matures. Some of the fodder crops such as Sudan grass, pearl millet and oats can accumulate nitrate at potentially toxic levels.

Most of the nitrate accumulates in stem, followed by leaves and very little in the grains. Nitrate poisoning is better described as nitrite poisoning. When livestock consume forages, nitrate is normally converted in the rumen from nitrate - nitrite -ammonia - amino acid to protein. When forages have an unusually high concentration of nitrate, the animal cannot complete the conversion and nitrite accumulates. Nitrite is absorbed into the blood stream directly through the rumen wall and converts haemoglobin (the oxygen carrying molecule) in the blood to met-haemoglobin, which cannot carry oxygen. The blood turns to a chocolate brown colour rather than the usual bright red. An animal dying from nitrate (nitrite) poisoning actually dies from asphyxiation, or lack of oxygen. Factors affecting the severity of nitrate poisoning are the rate and quantity of consumption, type of forage, energy level or adequacy of the diet.

Level of nitrate in forage (dry matter basis) and potential effects on animals

SN	Nitrate-content (ppm)	Effect on Animals
1	0-1000	This level is considered safe to feed under all conditions.
2	1000-1500	This level should be safe to feed to non pregnant animals under all conditions. It may be best to limit its use to pregnant animas to 50 per cent of the total ration on a dry basis.
3	1500-2000	Feeds are fed safely if limited to 50 per cent of ration's total dry matter.
4	2000-3500	Feeds should be limited to 35-40 per cent of total dry matter in the ration. Feeds containing over 2000 ppm nitrate nitrogen should not be used for pregnant animals
5	3500-4000	Feeds should be limited to 25 per cent of total dry matter in ration. Do not use for pregnant animals.
6	>4000	Feeds containing over 4000 ppm are potentially toxic. Do not feed

Precaution: Nitrates are more likely to accumulate in annual forages than in perennial crops. Nitrates are a concern immediately following a period of drought or wet, dull weather. The risk of nitrate toxicity can be reduced, but not eliminated, by taking the following steps:

- Dilute the nitrate content of the total ration by feeding a combination of low and high nitrate feeds.
- Feed the ration in two or three meals per day rather than just one meal per day.
- Allow cattle to adjust to low levels of nitrate before increasing the nitrate content of the ration.
- Ensure that livestock are being fed a balanced ration for the level of production that is expected.

Most feeds that contain nitrate can be fed to cattle if managed properly with balanced diet.

Oxalates: Oxalate is an anti-quality nutrient which under normal conditions is confined to separate places. However, when it is processed and/or digested, it comes into contact with the nutrients in the gastrointestinal tract. After released, oxalic acid binds with nutrients, rendering them inaccessible to the body. If feed with excessive amounts of oxalic acid is consumed regularly, nutritional deficiencies are likely to occur, as well as severe irritation to the lining of the gut.

Strong bonds are formed between oxalic acid, and various other minerals, such as Calcium, Magnesium, Sodium, and Potassium. This chemical combination results in the formation of oxalate salts. Oxalates react with calcium to produce insoluble calcium oxalate, reducing calcium absorption. This leads to a disturbance in the absorbed calcium: phosphorus ratio, resulting in mobilization of bone mineral to alleviate the hypocalcaemia. Prolonged mobilization of bone mineral results in nutritional secondary hyperparathyroidism or osteodystrophy fibrosa. Young plants contain more oxalate than older plants. During early stages of growth, there is a rapid rise in oxalate content followed by a decline in oxalate levels as the plant matures. The distribution of oxalate in plants is uneven. Several researchers reported that oxalate content is highest in leaf tissue, followed by stem tissue.

Precaution: Dietary oxalate can be degraded by rumen microbes into CO_2 and formic acid. Ruminants adapted to diets with high oxalate content can tolerate oxalate levels that are lethal to non-adapted animals. Moreover, it has been shown that the transfer of rumen fluid from animals in Hawaii to Australian ruminants resulted in complete elimination of the toxic effects of mimosine and the bacteria involved in such effects have been identified. Evidence also exists that rumen microbes can be genetically manipulated.

Saponins: Saponins are glycosides containing a polycyclic aglycone moity of either C27 steroid or C30 triterpenoid (collectively termed as sapogenins) attached to a carbohydrate. Saponins are characterized by a bitter taste and foaming properties. The structural complexity of saponins results in a number of physical, chemical, and biological properties, which include sweetness and bitterness, foaming and emulsifying, pharmacological and medicinal, haemolytic properties, as well as anti-microbial, insecticidal activities. Saponins reduce the uptake of certain nutrients including glucose and cholesterol at the gut through intra-lumenal physicochemical interaction. Hence, it has been reported to have hypo cholesterolemic effects. Scientists Observed that 4-7 weeks of ad lib. feeding of Albizia stipulate (Siris) gave rise to toxic manifestation of sheep. The toxicity of broom breed (Gutierrezia sarothrae), a resinous shrub believed to be due to its saponin content. Symptoms include listlessness, anorexia, weight loss and gastroenteritis. In non-ruminants (chicks and pigs), retardation of growth rate, due primarily to reduction in feed intake, is probably major concern. The adverse effect of saponins can be overcome by repeated washing with water which makes the feed more palatable by reducing the bitterness associated with saponins. Saponins are among several plant compounds which have beneficial effects. Among the various biological effects of Saponins are antibacterial and anti-protozoal.

Precaution: The concentration of saponins can be overcome by repeated washing with water which makes the feed more palatable by reducing the bitterness associated with saponins [20]. Add oSamether legumes and roughes in ration along with siris leaf (which are toxic to animal).

Tannins: Tannins are water soluble phenolic compounds with molecular weight greater than 500 and hydrolysable tannins and condensed tannins are two different groups of these compounds which differ in their nutritional and toxic effects. Tannins have a property of binding to protein to form reversible and irreversible complexes due to the existence of a

number of phenolic hydroxyl groups. They occur almost in all vascular plants. Hydrolysable tannins and condensed tannins are two different groups of these compounds. Generally, tree and shrub leaves contain both types of tannins. The condensed tannins have more profound digestibility-reducing effect than hydrolysable tannins, whereas, the latter may cause varied toxic manifestations due to hydrolysis in rumen. Tannins may form a less digestible complex with dietary proteins and may bind and inhibit the endogenous protein such as digestive enzymes. The tannin protein complexes are astringent and adversely affect feed intake and all plants contains phenolic compounds but their type and concentration may cause negative animal responses. When herbivore forage on tannin rich plant, tannin protein complexes can reduce the digestion of forage protein. Tannins" directly affect digestibility of cell wall by binding with microbial enzyme in the rumen. The reduced digestibility of cell wall compounds restricts the digestible energy that animal gain from forage plants.

Tannins containing plants and effect on ruminants due to its fodder consumption

Fodder Tree/Shrub	**Common Name**	**Area of Distribution**	**Effected Animal**	**Nutritional Effect**
Acacia aneura	Mulga	All mainland states of Australia,and others	Sheep, goat	Reduction in N digestibility decreased wool yield and growth; decreased S absorption Reduced feed intake, poor growth and loss in weight.
A. cyanophylla	Wattle	Same	Sheep, goat	
A. nilotica (pods	Babul	Same	Sheep, goat	
A. sieberiana (pods),	Paper bark	Same	Sheep, goat	
Terminalia oblongata	Yellow wood	Same	Sheep, goat	
Ziziphus nummularia	Jharberi	Same	Sheep, goat	
Prosopis cineraria	Khejri	Same	Sheep, goat	
Albizia chinensis	Sirris	Asia,S E Assia	Goat	Reduced in sacco N digestibility
Leucaena leucocephala	Subabul	Asia,S E Assia	Poultry	Poor N retention, low apparent metabolisable

				energy value
Prosopis cineraria	Khejri, long tree	Australia Asia and others	Rabbit	Reduced feed intake & growth, cecotrophy increased protein digestibility

Precaution: The concentration of condensed tannins above 4 per cent has been reported to be toxic for ruminants as they are more resistant to microbial attack and are harmful to a variety of microorganisms. Physical methods like soaking and drying and heat treatment before feeding of forage can reduce the toxic level of tannin. Several studies indicate that tannin-rich leaves, in combination with concentrate rations, could be fed to animals without any adverse effect. This happens because animals consume protein in excess of their requirement from the concentrate and therefore, the anti-nutritional effects of tannins were masked. Moreover, these studies do not show the utilization of tree leaf proteins for which they are mostly fed.

Cyanogens: Cyanogens are glycosides of a sugar or sugars and cyanide containing aglycone. It can be hydrolyzed by enzymes to release HCN by enzymes that are found in the cytosol. Damage to the plant occurs when the enzymes and glycoside form HCN. The hydrolytic reaction can take place in the rumen by microbial activity. Hence, ruminants are more susceptible to CN toxicity than non- ruminants. The HCN is absorbed and is rapidly detoxified in the liver by the enzyme Rhodanese which converts CN to thiocyanate (SCN). Excess cyanide ion inhibits the cytochrome oxidase which stops ATP formation, and further tissues suffer energy deprivation and death follows rapidly. The lethal dose of HCN for cattle and sheep is 2.0-4.0 mg per kg body weight.

Prussic acid (HCN) concentration in forages

HCN Conc in ppm		**Potential Effect on Livestock**	**Remarks**
DM basis	Fresh Basis		
0-500	0-100	Forage is generally safe and should not causetoxicity.	Safe to Use
500-1000	100-200	Potentially toxic and forage should be fed at a Restricted rate in the diet.	Dangerous
>1000	>200	Very dangerous to livestock and will usuallycause death.	Toxic/ Poisonous

The lethal dose for cyanogen would be 10-20 times greater because the HCN comprises 5-10 per cent of their molecular weight. For poisoning, forage containing this amount of cyanogen would have to be consumed within a few minutes and simultaneous HCN production would have to be rapid. HCN level will be high in young seedlings rather than in matured seedlings. reported that the forage prussic acid percentage of the second cut was significantly lower than the first cut, probably due to degradation of the acid and a higher metabolic activity of the plant due to higher temperatures during growth processes which can reduce the prussic acid accumulation, these low amounts of FPAP (Forage Prussic Acid Percentage) are not toxic to animals.

Precaution: As levels of HCN is found high in younger sorghum crop which are found unwanted and out of place i.e. grown under energy stress condition and crop not get proper irrigation. Thus try to exclude fodder from such plant. Further, Post-harvest wilting of Cynogenic leaves may reduce the effect of cyanide toxicity. Sorghum, Sudan and Johnson grass must kept for drying at least six hour before its use. Fodder HCN concentration >200 ppm in fresh green fodder and >1000 ppm in dry fodder drying, ensiling or allowing the forage to mature will reduce prussic acid concentration. Retest the plant sample before feeding.

BOAA: β-N-oxalyl-L-α,β-diaminopropionic acid (β-ODAP or BOAA), a naturally occurring amino acid, possesses potent neuro-toxic activity and has been shown to be responsible for outbreaks of neurolathyrism following consumption of *Lathyrus sativus*. β-ODAP occurs naturally as two isomeric forms with the α-form being approximately 5% of the total. According to toxicological studies, this isomer is less toxic than the major, β-isomer. The level of β-ODAP in dry seed varies considerably according to genetic factors and environmental conditions. L. sativus grown in nutrient solutions that are zinc-deficient or rich in ferrous iron produced seed with elevated levels of β-ODAP. β-ODAP is biosynthesized during the ripening of the seed and is further increased during germination. The ingestion of ODAP causes neuro-lathyrism, a neuro-degenerative disease that damages upper motor neurons, causing irreversible paralysis of the lower limbs and sometimes death in humans and animals. In Ethiopia, other studies reported ODAP content in seeds varying from 5.4 to 8.9 g/kg DM or 2.0 to 4.5 g/kg DM. The green parts and the straw contain lower concentrations of ODAP: 1.9 to 3.4 and 1.3 to 2.1 g/kg DM respectively.

Precaution: Do not use in large amount and tender fodder. Water soaking or hot water soaking for few hours reduced the toxicant. It is advised to feed lathyrus to the big animals with straw and other dry feed Bhusa.

Mimosine: Mimosine, a non-protein amino acid structurally similar to tyrosine, occurs in a few species of Mimosa and all species of closely allied genus Leuceana. Concern has arisen because of importance of L. leucocephala in which the level of mimosine in the leaf is about 2-6% and varies with season and maturity. In non-ruminants animals, mimosine cause poor growth, alopecia, eye cataracts and reproductive problems. Levels of Leucaena meal above 5-10 % of the diet for swine, poultry and rabbits generally result in poor animal performance. The main symptoms of toxicity in ruminants are poor growth, loss of hair and wool, swollen

and raw coronets above the hooves, lameness, mouth and oesophageal lesions, depressed serum thyroxine level and goiter. Some of these symptoms may be due to mimosine and othe's to 3,4 dihydroxypyridine, a metabolite of mimosine in the rumen. Reduction in calving percentage due to Leucaena feeding has also been noted.

Precaution: A solution to a mimosine problem could be the development/selection of low mimosine containing cultivars. However, low mimosine types are found to be unproductive and low vigour. The approach is to feed leucaena mixed with other feeds. Use of Leucaena fodder may be restricted to 30% of green forage in the case of cattle and buffalo, and 50% for goats. The effect of Leucaena and mimosine can be reduced by heat treatment, by Supplementation with amino acids or with metal ions such as Fe, Al and Zn.

Alternate to Green Fodder: Decreasing united family and shortage of labour is requirement to find alternate to green fodder. Daily green harvesting and chaffing is a tedious work. There is need to find altenate to daily green fodder harvesting and chaffing.

Silage: In our country, the rainfall is seasonal and more than 80 % of the annual precipitation occurs in a time span of only 3-4 months. As a result plenty of grass is available just after the onset of monsoon, all of which is not properly utilized. During this season, farmers can also produce a large amount of green fodder. There- fore, a surplus stock of fodder can be accumulated in this season which can be used for cattle feeding during the lean months viz. November to December and April to June. The ideal and simple method of conserving this surplus fodder and grass is to make silage of surplus fodder and grasses

Silage is the fermented feed, resulting from the storage of high moisture crops usually green forages under anaerobic conditions. The structure in which silage is prepared is called silo. These are airtight to semi-air-tight structures designed for the storage and preservation of high moisture feeds as silage.

Silages start to spoil when exposed to air. Air stimulates the growth of spoilage yeasts that degrade lactic acid, which results in a rise in pH, heating, and degradation of nutrients in the silage mass. Because total mixed rations (TMR) contain silages, they are also prone to further spoilage in the feed bunk.

Storage life will depend on the type of plastic used. Stretch wrap plastic will give a storage life of 12 months, while plastic sheet can last for 2-3 years. Unwrapped round and square bales can be stored in pits or hillside bunkers and covered with plastic sheeting. Round bale silage should be fed within 6 months to a year from being harvested. Whereas, pit silage if properly packed and sealed can be stored for up to 2 to 3 years with minimal quality losses. Initial quality and moisture content at harvest will certainly impact length of storage.

The fermentation process takes 10 days to 3 weeks for completion. Silages should not be fed until after this process is completed for the best milk production and feed intake. Thus, the recommendation is to wait at least 3 weeks before feeding new crop silages.

Crops Suitable for Silage Making:

Fodder crops such as maize, oat, sorghum, pearlmillets, hybrid napier rich in soluble carbohydrates are most suitable for fodder ensiling. Quality of silage can be improved with suitable additive such as molasses, urea, salt and formic acid etc.

Methods of Silage Making: The crop for silage making is generally harvested at the flowering stage when it has the maximum amount of nutrients. For maize it is about the early dent stage (well-matured stage generally harvested for seed) of maturity. Sorghum is harvested at late dough stage (stage at which seeds are soft and immature) at the earliest. Silage materials containing less than 25 % dry-matter (more than 75 % moisture) will form a very sour silage juices during storage, incurring a considerable loss of nutrients. Thus, plants for silage making should be allowed to mature till the dry matter content attains 35-40 per cent.

Silage material should be cut to a proper size in order to fit it in silo and ensure good quality of silage. The length varies from a fraction of an inch to over an inch in length. Grass silages requires to be finely chopped than maize or sorghum. Wilted and dry forages and forage with hollow stems should be chopped more finely than forage of high moisture content, thus permitting thorough packing and eliminating most of the air pockets.

Moisture content of silage material beyond 60-65 % is not desirable. In such a condition, it will be costlier to handle, susceptible to decay and loss of juices and nutrients. Due to high acidity a large amount of silage near the wall is spoiled. Fresh grass should be wilted for 3 to 4 hours on a good sunny day. If weather is not dry enough to allow wilting, mix straw (5-20%) with grass, before filling in the silos. Desirable moisture content can be ensured by combining high and low moisture crops. In some cases ground grains and dried molasses can also be used as dry preservatives.

Hay: Green fodder should be chopped in size of 5-8 cm. Spread chopped fodder on a pucca floor and dry it in the sunlight in a set of 10-15 cm thickness. Stir the drying forage every 2-3 hours during the day to speed up the drying process under exposure to sun and air.

Crops Suitable for hay Making: Legume crops such as berseem, lucern, guar, and cowpea are very good for making hay. In addition to mineral and vitamins in dried legume fodder crops, protein is rich in quantity, which is why it is important to dry the fodder used in ration. There is a special way to cut and store different fodder crops. Hay is made only from leguminous crops which are very rich in protein and minerals. The green fodder crops which are soft, are suitable for making hay, such as berseem, cowpea, lucern and ryegrass. The amount of moisture in green fodder crops is generally 80-90%, but in order to be able to store them, the moisture should be below 15%, which does not harm bacteria and fungus.

Fodder crops suitable for hay making and their time of sowing

Fodder crop (legume)	**Sowing time**	**Seed rate**

Berseem	September (24-30) to October (1-7)	8-10 kg
Lucerne	Mid October	6-8 kg
Cowpea	March to Mid July	CL 367= 12 kg Cowpea 88= 20-25 kg
Ryegrass	September (24-30) to October (1-7)	4 kg

The nutritive value (on dry matter basis) of fodders (hay) of various crops suitable for hay making

Fodder crop	**Protein (%)**	**Total digestible Nutrients (%)**
Berseem	18.0	60.5
Lucern	22.0	59.5
Cowpea	22.5	61.2
Ryegrass	16.0	63.5

Crop should be harvested at flowering stage (when flowering is initiated) because when crop matures, its lignin content increases and nutritive value decreases. As far as time is concerned, crop should be harvested early in the morning because at this time the dew has dried off.

Conclusion:

A proper plan of fodder cultivation for round the year fodder production is necessary for making availability of green fodder to animals for enhanced production and reproduction. While feeding tree fodders to animals, the presence of anti-nutritional factors should also be considered.

References: On request

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CHAPTER 4

Nutritional Technologies for Improving Reproduction and Production Performances of Buffaloes

Avijit Dey*

Division of Animal Nutrition and Feed Technology
ICAR- Central Institute for Research on Buffaloes, Hisar, Haryana

*Correspondence: Dr. A Dey (Principal Scientist, ICAR-CIRB); avijitcirb@gmail.com

Buffalo production plays a significant role in food security and poverty alleviation in Asian countries. Buffaloes, described as the ‘’Black Gold’’, are favourite multipurpose animals of farmers and are in fact the ‘’bank on hooves’’ with huge potential for social and economic changes for the agrarian community. Buffalo has been an integral part of livestock agriculture in Asia for over 5000 years producing milk, meat, hides and draft power. With more than 90% of global buffalo population present in Asia, 77.9% buffaloes are inhabitant of south Asian countries and India is home for 57% world buffalo population.

With 20 per cent share of world’s bovine population, India is one of the largest producers and exporters of buffalo meat. India has exported 1.175 million tonnes of buffalo meat products to the world for the worth of Rs. 24613.24 Crores/ 3303.34 USD Millions during the year of 2021-22 (APEDA, 2022). During the last 70 years, buffalo contribution of nearly 50 per cent in milk pool elevated India to the No. 1 pedestal in total milk production, while buffalo meat export earned India another distinction of being the 4th largest beef exporting country in the world.

Status and contribution of buffalo milk

Livestock sector is growing faster than any other agricultural sub-sector. While percentage contribution of agriculture and allied sector at constant prices (2011-12) in total gross value added (GVA) decreased from 18.5 to 14.8 per cent from 2011-12 to 2019-20; the share of livestock to total GVA increased from 4.0 to 4.4 per cent. The GVA of livestock sector was about Rs. 9,62,682 crores at current prices during FY 2019-20, which was about 28.36% Agricultural and allied sector GVA and 5.21% of total GVA. Buffalo is prominent in UP, Rajasthan, Gujarat, MP, Bihar, AP, Maharashtra, Haryana, Telangana and Punjab, where it contributes between 54-85 percent to total milk produced and is important contributor to rural household incomes.

The total milk production of the country was increased from 198.44 million tonnes (2019- 20) to 209.96 million tonnes (2020- 21) with buffalo share of about 45% (DAHD, 2022). The productivity of buffalo is highest in Haryana followed by Punjab. Uttar Pradesh, having about 26% of total in-milk female buffaloes, is having per animal productivity of 4.44 litres per day (about 50% of the best producing state). There is scope for further improvement in buffalo productivity through improved germplasm dissemination, nutrient availability and health care.

Buffalo meat production

Meat production from buffaloes contributes immensely to Indian economy and plays a pivotal role in sustainable buffalo husbandry through improvement in productivity, remunerative price for the culled/ unproductive stocks, prevention of degradation of soil and water resources and reduction in the greenhouse gas effect. Resultantly, there are no stray buffaloes on the streets. The total meat production of the country was 8.6 million tonnes in 2019-20 with a steady increase from 7.4 million tonnes since 2016-17. Buffalo contribute 22% of total meat production of the country (2019-20) and UP, Maharashtra, AP and Telangana are the largest producers of buffalo meat (2018-19).

India is the leader in buffalo meat production with an export of 1.175 million tonnes of buffalo meat (APEDA, 2022; https://agriexchange.apeda.gov.in/indexp/exportstatement.aspx) worth of Rs. 24613.24 crore (2021-22). According to the All India Meat and Livestock Exporters Association (AIMLEA) export abattoirs-cum-meat processing plants in India registered with the export regulatory authority (APEDA) are employing74,000 workforce directly and 1,50,000 indirectly. Slaughter restrictions on utilization of male buffalo calves and other unproductive buffaloes need to be relooked for increasing revenues from buffalo meat. FMD control programme needs to be implemented effectively for control / eradication of this important economic disease, which will enhance the market potential of both milk and meat across the globe for attracting better prices.

Nutritional factors associated with low productivity

Buffaloes are better converter of poor-quality fibrous feeds to milk and meat. Because of larger rumen, slower rumen movement, passage rate and higher microbial activity in the rumen, buffaloes have better digestive ability of feeds. Better degradability of protein was also reported in buffaloes (Terramoccia et al., 2000). Despite of these merits, buffalo have relatively poor reproductive efficiency. Buffalo exhibit many reproductive anarchies including delayed onset of puberty, poor estrus expression, longer postpartum ovarian quiescence, and most importantly lowered conception rates. However, higher fertility could be achieved through better feeding and management (Paul and Lal, 2010).

The majority of the farmers in developing countries use low input production system of animal rearing, resulting low productivity. The greatest scope for improving milk production is through a strategy which targets improvement of reproductive performance. To reduce the cost of rearing and be economical to the farmers, the buffalo must grow rapidly, achieve puberty as soon as possible, conceive readily, produce a healthy calf and continue producing calves and milk until productive life. Buffaloes exhibit a distinct seasonal change in displaying oestrus, conception rate and calving rate which could be the cause of the prolonged inter-calving period since buffalo calving during the unfavourable season may not resume their ovarian activity until the following favourable season, decreasing their reproductive efficiency.

Nutrition plays a pivotal role in maintaining the body condition, production and reproductive efficiency of dairy animals. Nutrition consists of different nutrients, but mainly

includes protein, fat, carbohydrates and micro-elements. Carbohydrates and proteins provide substrates for rumen fermentation, which results in the production of volatile fatty acids (VFA). Animals utilize these VFAs as their main source of energy for maintenance, milk production and reproductive performance (Ibtisham et al., 2018).

1. *Energy*

The most important nutritional factor affecting production and reproduction in dairy animals is the energy intake. Inadequate energy intake in heifers will lead to delay in sexual maturity. Negative energy balance (NEB) is a common feature in high yielders during early lactation because of inadequate intake of energy due to reduced feed intake and mobilization of body fat reserves to meet its energy demand for high levels of milk production. Energy stores in body tissues are mobilized and weight losses occur, resulting delayed oestrus and conception (Nishany et al., 2013).

The mobilisation of body fat due to low levels of blood glucose causes increased blood levels of non-esterified fatty acids (NEFA) and ketone bodies like acetoacetate, acetone and β-hydroxybutyrate (BHBA). At the same time, blood levels of growth hormone (GH) are increased and that of insulin and IGF-1 are decreased (Boisclair et al., 2006). These metabolic changes are negatively associated with fertility. They include disturbances in LH pulse frequency, growth rate and diameter of the dominant follicle, weight of the corpus luteum, and progesterone and oestradiol concentrations (Van Knegsel et al., 2005). Increased NEFA concentrations in blood have been linked to greater incidences of ketosis, displaced abomasum, retained placenta and altered blood metabolites and hormone profiles. Proper feeding management can help to prevent the animal from NEB, which is regarded to improve the production and reproduction of the animals.

Fatty acids and cholesterol act as the substrates for reproductive hormone synthesis. Increasing fat in the diet may increase levels of reproductive hormones. Potential improvements in fertility with supplemental fat have generally been associated with increased dominant follicle diameter, improved oocyte and embryo quality (Sauza et al., 2017). The level of total dietary fat in ration should not exceed 6-7% of diet. Mixture of cereal grain and forages usually contain about 3 % fat, so up to 3 or 4 % of dietary DM can come from supplemented fat (Paul and Lal, 2010). A number of studies have revealed that dietary supplementation of bypass fat in crossbred cows and Murrah buffaloes improved the milk yield and fat content (Naik et al., 2009; Ramteke et al., 2014). In buffaloes, about 100-150 g bypass fat per day could be supplemented to increase milk production performances. Supplementation of bypass fat at 2.5% of DMI reported an increase in birth weights of the calves, while time taken for expulsion of foetal membranes, involution of uterus, onset of cyclicity, the service period and number of inseminations per conception were reduced ($P < 0.05$) in supplemented group (Tyagi et al., 2010). Increase in milk fat and yield was also reported in buffalo while supplementation of 15g bypass fat per kg milk yield, however, there was no effect on cyclicity and pregnancy rate (CIRB, 2017).

However, overfeeding of energy is wastage and costly; and may produce adverse effects on production and reproduction. Energy intake should be in adequate levels, excess intake during late lactation and the dry period can cause “fat cow” problems which lower

reproductive efficiency in the next lactation. Excessive energy intake inhibits development of milk secretary tissue in mammary gland, which reduces lifetime milk production ability. Over-conditioned animals after calving have a higher incidence of retained placenta, more uterine infections and more cystic ovaries. They also have a higher incidence of metabolic disorders and have a greater tendency to go off feed. All of these problems can result in poor reproductive performance (Dunn and Moss, 1992)

2. *Protein*

Protein is vital to the maintenance, reproduction, growth, and lactation of animals. Low level of dietary protein severally affects rumen microbial growth and fermentation resulting in increased retention time of nutrients, decreased capacity to digest organic matter and depressed feed intake which affect animal performance. When fed in excess, protein is utilized as source of energy especially in cases of energy shortage, since deposition of protein in reserve tissues of ruminants is limited to the extent of 8-22 % of total body protein (Huber, 1976). Inefficiencies arise from elimination of surplus urea, which in turn increases energy requirement and may affect health and reproduction of the animals.

Table 2. Energy and protein requirement of buffaloes

Stages of life	**Maintenance (g/kg W $^{0.75}$)**	**Growth (g/g ADG)**	**Lactation (g/kg 6% FCM)**	**Reference**
Energy requirement (TDN)				
Growing	35.4- 39.3	1.4-2.23	-	Paul and Patil, 2007; Paul et al., 2004
Lactating	35.3	1.97	406	Paul et al., 2002
Protein requirement (CP)				
Growing	5.98- 6.74	0.44-0.51		Udeybir and Mandal, 2001
Lactating	5.43	0.33	90	Paul et al., 2002

Quality of protein in term of its ruminal degradability is also important for ruminants. When bypass protein is fed to ruminants, protein remains mostly undegraded in rumen thus lowering production of ammonia in rumen, which otherwise is produced in large quantities and excess is absorbed and converted to urea in liver and is mostly excreted through urine. By feeding bypass protein, at least 30% of the dietary amino acids are saved from getting excreted as urea. The excess amino acids absorbed from the lower tract are converted to

glucose in liver and serve as precursor for lactose synthesis in mammary gland, which regulates osmotic pressure of milk and resulting in larger volume of milk produced.

Heifers fed a diet deficient in protein may show a delayed onset of puberty while buffaloes with protein deficient diets may have an increase in the number of days open. Adequate protein is necessary for the proper functioning of the reproductive organs and normal development of the foetus. In lactating animals, protein deficiency may result in low milk production, decreased appetite and hence a low body condition. Protein deficiency results in reduced forage intake and poor digestibility. Rumen microbial activity for efficient forage utilization is hampered when diet contains less than 7% crude protein. However, reproductive performance may be impaired if a protein is fed in excess amount of the requirement. Negative association between high dietary CP and fertility parameters was reported (Funston, 2014). Excessive feeding of protein during the breeding season and early gestation, particularly when the rumen receives an inadequate supply of energy, may be associated with reduced fertility. This decrease in fertility may result from decreased uterine pH during the luteal phase of the oestrous cycle in animal fed high levels of degradable protein (Cheng et al., 2015). Animals fed 10-15% excess protein above the requirement involve more services per conception and had longer calving intervals (Titi et al., 2013). Supplementation of 15-19% excess CP resulted in lowered conception rate from 65 to 53% (Thatcher et al., 2001). The negative effects of protein supplementation are associated with an increase in blood urea-N, which affects ovarian follicular and embryo development. Successful embryo development depends upon the nature of the uterine environment, while increased urea level can decrease the uterine pH that would negatively affect the implantation and development of embryos, mostly at the cleavage and blastocyst formation stage (Humer et al., 2016). Higher serum urea levels because of excessive CP intake and poor body condition due to lower energy intake were the key factors for inferior reproductive efficiency in *Nili-ravi* buffaloes (Qureshi et al., 2002). Therefore, over feeding of protein should be discouraged not only due to impede in the reproductive health, but also in the preview of environmental pollution and economy of animal production.

3. *Minerals*

The complex interactions of minerals and other nutrients are important to all physiological processes, including reproduction. Forages low in protein content are usually low in phosphorus. Mature forages are mostly deficient in phosphorus, and its deficiencies have been associated with reproductive problems. Overfeeding of phosphorus should be avoided as it is expensive and there is no added benefit to reproductive performance. Calcium (Ca) is usually adequate in most forage-based diets. It is also included in commercial mineral mixtures as many phosphorus sources contain calcium. Other major minerals are sodium (Na), magnesium (Mg), potassium (K), chlorine (Cl) and sulphur (S). Their deficiency or excess may adversely affect production and reproduction. The level at which the particular minerals is present in the feed will determine the need for its supplementation. The same is true for microminerals, including copper (Cu), cobalt (Co), iodine (I), iron (Fe), manganese (Mn) and zinc (Zn). It is also advisable to analyse water as it could be a significant source of

minerals that affect reproductive health. Several macro and micro minerals are found to be in lower levels in various types of anoestrus buffaloes (Abou-Zeina et al., 2009).

3.1. *Macrominerals*

Calcium deficiency is common during parturition or within few days following parturition. The Ca:P ratio imbalance may affect ovarian function resulting in prolongation of first oestrus and ovulation, delayed uterine involution, increased incidence of dystocia, retained placenta and prolapse of uterus (Kumar, 2003). Low calcium level in blood is associated with anoestrus and excess of calcium may affect reproductive health of animal by impairing absorption of phosphorus, manganese, zinc, copper and other elements. Ca:P ratio between 1.5:1 and 2.5:1 for lactating animal should be maintained. Dry animals should be provided with optimum levels of calcium and phosphorus to decrease the incidence of milk fever which is important for maximizing reproductive efficiency. Calcium present in the seminal plasma of buffalo bulls plays an important role in preserving spermatozoa motility and viability, as well as antioxidant status by protecting the sperm cells oxidative damage (Eghbali et al., 2010).

Phosphorus deficiency is associated with reduced production performance, abnormal sexual behaviour resulting in decreased fertility rate, feed intake, milk production, decreased ovarian activity, delayed sexual maturity and low conception rates in dairy animals. Delayed attainment of puberty, silent or irregular estrus in heifers, failure of estrus, long inter-calving period, still birth or weak calves or even embryonic death due to lack of uterine muscle tone are reported to be some of important clinical manifestation exhibited by the animals on phosphorus deficient diet and the excess of phosphorus renders the endometrium susceptible for infection (Chaudhary and Singh, 2004). Phosphorus is also needed for the maintenance of glycolysis and motility. The phosphorus concentration detected in the seminal plasma of bulls was positively correlated with quantity and quality parameters of semen (Machal et al., 2002).

Sodium deficiency may adversely affect the normal reproductive physiology by preventing the utilization of protein and energy; and potassium deficiency causes muscular weakness and affect the musculature of female genital tract causing impairment in the normal reproductive process. Sodium deficiency is also associated with general infertility and embryonic mortality in farm animals (Dittman, 2008). Sodium and potassium are responsible for maintaining osmolarity and activity of spermatozoa and regulate sperm motility and the acrosome reaction (Barfield et al., 2005). However, feeding of high levels of potassium may delay the onset of puberty, delay ovulation, impair corpus luteum development and increase incidence of anoestrus in heifer causing poor fertility (Smith and Chase, 2010).

Magnesium is required for capacitation, hyperactivation and acrosome reaction of spermatozoa (Semczuk and Kurpisz, 2006) in male reproduction. Mg level in the seminal plasma increases with sperm concentration but has no significant bearing on sperm motility (Wong et al., 2001), however, positive effects of Mg on the motility, morphology and concentration of spermatozoa were reported by Marzec-Wróblewska et al. (2012). Mg content in the seminal plasma was positively associated with the total antioxidant status of

semen (Eghbali et al., 2010). Sulphur containing compounds increased the mobility and survivability of the cryopreserved spermatozoa (Tumenbaevish *et al*., 2012).

3.2. *Microminerals*

Manganese deficiency results in delayed estrus, reduced conception rate and deformed calves (Hansen et al., 2006). Copper deficiency in ruminants is associated with delayed or depressed estrus, long post-partum return to estrus period; anoestrus, silent heat, abortion and fetal resorption. Copper rich diets enhance spermatozoa motility (Cheah and Yang, 2011) and it may also act on the pituitary receptors which control the release of luteinizing hormones (LH). Cu deficient or excess levels may affect spermatocytogenesis with regard to sperm production, maturation and fertilizing capacity (Wong *et al*., 2001; Cheah and Yang, 2011). Excess feeding of Mo resulted in decreased libido, reduced spermatogenesis and sterility in males and delayed puberty, reduced conception rate and anoestrus in females. Cobalt deficiency causes reduced fertility and increased early calf mortality. Depletion of cobalt at parturition results in decreased milk production. Chromium potentiates the effect of insulin by increasing the uptake of glucose and amino acids by the cells in the body which in turn improves the energy balance leading to improved reproduction in early lactation. Chromium also exerts a significant influence on follicular maturation and luteinizing hormone release (Chaudhary and Singh, 2004). Chromium supplementation resulted in increased pregnancy rate and reduced age of puberty (Stahlhut et al., 2006).

Semen contains a certain amount of Fe, required for a normal spermatozoa production and functions. The total Fe content of the buffalo seminal plasma was highly associated with sperm motility and viability. Fe content within the seminal plasma is important for the preservation of sperm motility and viability after ejaculation, and its presence will help spermatozoa to maintain their functions (Eghbali et al., 2010). Increased Fe concentration can affect negatively the morphology and DNA integrity of spermatozoa (Perrera et al., 2002). Zinc is essential for sexual maturity and early attainment of puberty. Zn supplementation in bulls resulted in higher semen volume, sperm concentration, percentage of live sperm and motility (Kumar et al., 2006). Zinc deficiency in females has been associated with abortion, fetal mummification, lower birth weights and prolonged labour. Adequate zinc levels are vital for repair of the uterine lining following calving, return to normal estrus cycles and maintenance of the uterine lining necessary for implantation of embryos. Zn is a vital component of enzymes involved in steroidogenesis and has shown to act indirectly through the pituitary to regulate the gonadotropic hormones. Seminal Zn has an important role in the physiologic functions of the sperm cell and that its reduced levels result in low seminal quality and subsequent chances of fertilization (Colagar et al., 2009).

Selenium deficiency can result in impaired fertility, silent heat, cystic ovaries and the birth of unthrifty calves with poor immunity, increased incidence of retained placenta, and uterine infections following calving and high embryonic mortality. In males, Se deficiency has resulted in reduced sperm viability and reduced testicular growth. Iodine deficiency in breeding female results in reduced conception rate and ovarian activity, suppressed estrus, abortion, still birth, increased frequency of retained placenta and extended gestation period, weak calf and may be hairless. Excess of iodine intake is associated with abortion and

decreased resistance to infection and disease. Therefore, animal feed should be supplemented with minerals particularly with those which are deficient in feeds and fodders of that particular area.

The major cations present in feeds and the charge they carry are sodium (+1), potassium (+1), calcium (+2) and magnesium (+2). The major anions and their charges found in feeds are chloride (-1), sulfate (-2) and phosphate (-3). The difference between the number of cation and anion particles absorbed from the diet determines the pH of the blood and has significant role in feed intake, milk production, occurrence of metabolic diseases and reproduction of animals. The dietary cation-anion difference (DCAD) value of the diet can be calculated by finding the difference between cations and anions present in the diet. Calculation of DCAD is normally expressed using milli equivalents of the major cations and anions as follows: (sodium + potassium) – (chloride + sulfur). A negative DCAD diet contains more equivalents of anions than cations, a zero DCAD diet contains equal equivalents. By reducing subclinical hypocalcaemia, it may be possible for dairy animals to increase the dry matter intake faster in early lactation, resulting in increased milk production and decreased disease incidence. The incidence of retained fetal membranes (RPM) was significantly reduced by feeding anionic salts (Goff and Horst, 1997). The literature on the effect of anionic diets on reproductive performance indicated that such diets reduced the incidence of parturient hypocalcaemia and RPM and resulted in an increase in conception rate of cows.

Nestor et al. *(*1988) observed that severity of udder edema increased when heifers were fed NaCI (23 or 136 g/d) or $KHCO_3$ (0 or 272 g/d) in the diet but not when both salts were added together. Alteration of dietary cation-anion difference by addition of Cl may effectively reduce incidence and severity of parturient hypocalcaemia (Gaynor et al., 1989). Patel (2018) in a study with periparturient buffaloes observed that out of 10 control group of animals one animal suffered from prolapse and two animal showed the symptom of milk fever. But, out of 10 treatment group of animals only one animal showed the symptom of milk fever.

Senthil Kumar and Kaur (2005) also reported that then numbers of postpartum metritis cases were 71.43% in Cationic group compared to no incidence in Anionic group. One cow out of seven cows (1/7) under Cationic group was affected with mastitis, milk fever and death. There was 50% reduction in the incidence of retained placenta in anionic group.

4. *Vitamins*

Vitamins C, D, E and B-complex are either synthesized by rumen microorganisms, by the animal body or available in most common feedstuffs. Vitamin A is deficient in mature forages, crop residues, and other poor-quality forages. The deficiency of which definitely plays a role in embryo development and its supplementation before and after calving may increase conception rates. Vitamin A deficiency in dairy animals results in delayed sexual maturity, abortion, birth of dead or weak calves, retained placenta and metritis. Dairy animals and heifers consuming diets deficient in β -carotene suffered from delayed uterine involution, first estrus after calving, or ovulation and increased incidence of cystic ovaries. Buffaloes require vitamin A or its precursor beta- carotene in its diet. Daily feeding of 2-3 kg green

generally meets the requirement of vitamin A. When no green fodder is available, milch buffaloes should be supplemented with vitamin A (20000 to 45000 IU/d) and growing buffaloes should be fed 2000 to 8000 IU/d. Generally, there is no need for supplementing other vitamins to adult buffaloes. Vitamin E supplementation at 1,000 IU from 30 to 60 days postpartum decreased postpartum estrus interval, days open and services per conception suggesting that the supplemental dose might be reduced from 1,500 IU to 1,000 IU from 30-60 days postpartum in buffaloes (Panda et al., 2006).

Feeding strategies to improve reproduction and production performances of buffaloes

Meeting requirements of critical plane of nutrition at each stage of life cycle has comportment on performance at subsequent stages and thus on lifetime production. Hence, proper scientific feeding should be followed at all stages of life cycle for sustaining productivity. Feeding of pregnant and lactating buffaloes should be taken care judiciously which would affect milk production and subsequent reproduction.

Feeding of buffalo calves

Buffalo calves should be fed colostrum, the first milk of the mother secreted immediately after the birth of a new born as early as possible and positively within 30 minutes of birth and should be continued up to 4 days of age @ 3-4 litre/ day. The colostrum is rich in protein, fat, vitamin A and minerals. It also contains antibodies, which provide passive immune protection to the calf from many diseases and has laxative properties which help in expelling muconium. After 4 days when colostrum feeding is over, calf should be continued to feed whole milk @1/10th of body weight i.e. around 2.5 kg per day upto the age of 7 days. From 8th day, calf should be allowed to take calf starter and green grass along with the whole milk to boost up the rumen development in the following manner.

Age (days)	Whole milk (kg)	Calf starter* (kg)	Green grass (kg)
1-7	2.50	-	-
8-14	2.50	0.050	0.250
15-21	3.00	0.100	0.350
22-30	3.50	0.300	0.500
30-45	3.00	0.600	0.600
46-60	2.00	0.900	0.900
61-75	1.25	1.250	1.200
75-90	0.75	1.500	1.800

*Calf starter should be prepared with good quality, easily digestible feed ingredients (crushed maize, 38 kg; crushed barley, 9 kg; groundnut cake, 30 kg; wheat bran, 20 kg;

mineral mixture, 2 kg; salt, 1 kg; Vit A, B2, D3; 10 g per quintal so that it contains 22% CP and 70% TDN.

At the age of 3 months, calves attain about 60 kg body weight and microbial digestion in rumen become functional. Calves are meagrely allowed to suck milk from their mother and fed a complete diet (7.5 kg non- leguminous fodder, 2.5 kg leg. fodder, 1 kg concentrate mixture and wheat straw *ad lib.*) which contains 13% CP and 62% TDN to support 500- 600 g average daily weight gain.

Feeding of buffalo heifers

Calves from 6 months of age to adult can be reared on roughage-based diet with minimum amount of concentrates. Generally ration containing 12% CP and 60% TDN (10 kg green fodder, *ad lib* straw and standard concentrate mixture containing 20% CP and 70% TDN (maize, 30 kg; GNC, 30 kg; wheat bran, 38 kg; mineral mix, 2 kg; salt, 1 kg per 100 kg) @ 1.5, 2.0, 2.5 and 3.0 kg per day for 100, 150, 200, 250 kg BW will provide an average daily growth of 500g. Scientific feeding is required for optimum growth, as buffalo heifers attain puberty when threshold body weight is about 60% of mature weight. With proper feeding, buffalo heifers attain puberty at the age of 17-21 months with a body weight of 270-300 kg. However, very high plane of energy nutrition inhibits development of milk secretary tissue in mammary gland, which in turn reduces lifetime milk production ability. Generally, a growth rate of 500-600 g/d between 100 and 300 kg body weight is considered optimum growth rate for Indian buffalo heifers. Underfeeding that reduces growth rates during the rearing phase to 50% of the animal's potential delays puberty.

Feeding of pregnant buffalo

In general, until the last one third of gestation, nutrient requirements for intra-uterine growth are very small relative to mother's maintenance. In early pregnancy, buffaloes should be fed on maintenance level or restricted level of feeding to increase conception rate. Only about one-third of the total products of conceptus are produced during the first 7 months of gestation period. Subsequently, there is rapid acceleration in foetal development during the last 3 months of gestation period. Generally, buffaloes should be fed to support 750-900 g average daily weight gain during last 2 month of pregnancy and about 700g average daily weight gain during the last 3 month of pregnancy. In pregnancy of adult buffaloes, CP requirement increases by 3, 8.4, 16, 26, 43 and 64% of maintenance requirement on 5th 6th, 7th, 8th, 9th and 10th month of pregnancy, respectively. The corresponding increases in TDN requirements are 4.3, 7.2, 18.8, 22.2, 39.0 and 67.4 % of maintenance requirement, respectively. During pregnancy substantial extra uterine growth in the mother takes place especially during early pregnancy. This is necessary in immature animals that are still growing i.e. those in first and second pregnancy and hence 20 and 10% of maintenance requirement of energy and protein should be additionally fed to the heifers in first and second pregnancy, respectively. In case of adult buffaloes, maternal growth is considered as non-essential for pregnancy. However, in high yielding buffaloes, additional allowance during early pregnancy may be given to facilitate building up of extra body reserve which can be utilized to meet out energy deficiency in early lactation when animals are invariably in negative energy balance due to limited DM intake capacity. Pregnant buffaloes should be

dried at least 2 month before expected date of calving. In pregnancy, DM intake is low (about 1.7- 2.0% of BW). Pregnant dry buffaloes (at > 5 month of pregnancy) should be fed with 30 kg green fodder and 2 kg concentrate mixture (20% CP & 70% TDN) and *ad libitum* wheat straw. With decrease in availability of green fodder 1 kg concentrate mixture should be additionally fed to replace every 10 kg green fodder. This ration will meet protein requirement for entire pregnancy and energy requirement upto 9.5 month of pregnancy but will fall short of energy requirement on the last 2 weeks of pregnancy when additionally, 1-1.5 kg grain has to be fed. For pregnant immature buffaloes in first pregnancy, additional 1 kg grain or 5.5 kg cereal fodder or 7.5 kg legume fodder should be fed to support 300-350 g average daily maternal growth. Similarly, buffaloes in their 2nd pregnancy should be fed additional 0.5 kg grain or 2.7 kg cereal fodder or 3.7 kg legume fodder to support 120-200 g average daily maternal growth. Challenge feeding of buffaloes with good quality fodder and concentrate mixture during last three weeks of pregnancy helps in priming the rumen for increased concentrate feeding in early lactation and build up body reserve for lactation.

Feeding of lactating buffalo

Dietary energy is the most limiting factor in milk production. Lactating buffalo should be fed sufficient nutrients for their milk production and maintenance. Milk production increases gradually, reaches peak at 42- 56 days after calving, and the peak is maintained for next 70 days. It declines gradually thereafter from 126 to 305 days. Inadequate energy intake in early lactation leads to loss of body weight and delay in initiation of post calving oestrous cycle. Generally, ovarian cycle ceases when buffaloes loose 15 to 24% of body weight. Thus utmost care should be taken so that they are not underfed during early part of their lactation. The lactating buffaloes in their first and second lactation continue to grow and thus additional 20 and 10% of maintenance requirement should also be provided in first and second lactation, respectively. For a buffalo of 450 kg producing 10 kg milk; 5.0 kg concentrate mixture, 7 kg straw and 20 kg legume fodder/40 kg cereal fodder per day should be fed depending on the availability. For every 50 kg increase or decrease in BW, 350 g grain+ 1 kg straw+ 3 kg berseem + 2.5 kg cereal fodder has to be added or reduced, accordingly. For buffaloes in their 1st or 2nd lactation, should be fed additional 0.5- 1.0 kg grain to support the maternal growth. High yielding buffaloes (> 15 kg milk/d) may be fed bypass fat (cracked un decorticated cotton seed, soybean seeds and mustard seeds @ 1-3 kg per buffalo per day depending on milk production), starch (rice polishings), protein or amino acids (fish meal @ 1-2% of DM, methionine hydroxyl analogue @ 20-30 g per day per animal), probiotics (yeast culture, 10 g) or vitamin B complex (5-10 g niacin) to support high rate of production.

Feeding of breeding buffalo bulls

Breeding bulls should be fed good quality balanced ration for proper development of testicular tissue and improved semen quality. They should attain about 400 kg BW at 30 months of age. They should also be fed good plane of nutrition as low protein diet could delay puberty by 5-6 months with poor testicular development and small semen volume. Experiments conducted at CIRB revealed that about 40 to 60% restriction of energy and protein during growing phase causes retardation of testicular growth and the effect persists throughout the life. On the other hand feeding high concentrate diets (80% concentrate in

DM) to growing bulls reduced testicular sperm reserve and also reduced semen quality as compared to total roughage diet. Maintenance level feeding of bulls without energy allowance resulted in reduction in sperm numbers by 32 and 41% as compared to those receiving twice and thrice maintenance requirements. Therefore, breeding bulls should be fed 100% higher CP and 20% higher energy than maintenance requirement of mature female buffaloes (Paul et al., 2005). Caution should be taken to avoid overfeeding as fatness lead to reduced libido and reproductive performance. For a 700 Kg buffalo bull following feeding schedule can be followed: a) 40 kg cereal fodder + 0.8 kg deoiled ground nut cake/ deoiled soyabean cake or b) 10 kg berseem + 10 kg straw + 1.2 kg deoiled ground nut cake/deoiled soyabean cake or c) 8 kg straw + 2.0 Kg concentrate mixture + 2-3 kg green fodder + 1 kg deoiled ground nut cake/deoiled soyabean cake. For every 50 kg increase or decrease in body weight from 700 kg, a) 0.6 kg straw +100 g deoiled groundnut cake/deoiled soyabean cake or b) 3 kg green berseem or c) 3 kg green cereal fodder should be added/deducted from the ration suggested.

Feeding for buffalo broiler production

Oversight of male buffalo calves in scientific feeding and management reduces the growth rate, resulting wastage of precious resource, which could otherwise be utilized for buffalo veal production. Experiments conducted at CIRB Hisar to develop scientific modules and package of practices for economic rearing of buffalo calves for veal production. In the experiment, eighteen male calves (4-5 months age and 80 kg body weight) were divided into three groups of six each and fed as per following schedule for a period of 12 months. Group - I: All forage diet comprising of green and dry fodder *ad lib*, group -II: roughage: concentrate (70:30) and group -III: roughage: concentrate (30:70). The roughage: conc. ratio changed to 30:70 in group I and II in last four months of rearing. The feed intake in three different groups was 2.56, 2.60 and 2.63 per cent of body weight, respectively. The growth rate was 318.50, 435.69 and 647.21, g/d in G-I, G-II and G-III, for the first eight months and 692.37, 704.95 and 595.17, g/d during last four months, respectively. The overall growth rate in different groups was 442.88, 528.02 and 628.25, g/d in G-I, G-II and G-III, respectively. The economics of production was calculated taking the prevailing cost of the feed ingredients and the values for cost of feeding per animal per day were Rs 42.10, 56.10 and 85.92 in Group-1, II and III, respectively. The cost per kg body weight gain in corresponding groups were Rs. 95.05, 106.24 and 123.32, respectively. The study indicated that buffalo calves for veal production could be reared economically on all forage diets initially followed by a finisher ration for the last four months of rearing.

Conclusions

Feeding of buffaloes with low quality feeds results loss of body weight and body condition, delays the onset of puberty, increases the post-partum interval to conception, interferes with normal ovarian cyclicity and increases infertility besides low production performances. The main factors to low income of the farmers are delayed puberty, long calving intervals, short productive life and high calf mortality which ultimately affects the lifetime productivity of a buffalo. The level of the excess, deficiency or imbalance of nutrients affecting production and reproduction is not very lucid. Therefore, ideal feeding as

per requirements of energy, protein, vitamins and minerals is necessary for enhancing production performances in buffaloes.

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CHAPTER 5

Towards Green Livestock Production: Feeding Strategies for Abatement of Enteric Methane Emission

Avijit Dey*

Division of Animal Nutrition and Feed Technology
ICAR- Central Institute for Research on Buffaloes, Hisar, Haryana

*Correspondence: Dr. A Dey (Principal Scientist); avijitcirb@gmail.com

Methane (CH_4), one of the important greenhouse gases, produced normally during the enteric fermentation of feeds in ruminants mainly, has global warming potential of 21 times than of carbon dioxide. Although there is intense debate on contribution of domesticated ruminants on global methane emission, it is thought to be responsible for about 15% of total methane emission due to anaerobic enteric fermentation of feeds. Besides, ruminant animals lose a substantial (8-12% of gross energy) amount of feed energy through methane emission, which otherwise could be converted to metabolizable energy for productive purposes. Therefore, reduction in methanogenesis is of interest of ruminant nutritionists since long for efficient utilization of feed and more recently from the perspective of global warming. Sustainable strategies for mitigation of methane from ruminant livestock are necessary for cleaner environment and diverting the energy for productive purposes.

Feeding Strategies for Enteric Methane Abatement

Multi-dimensional strategies (genetic improvement strategies viz. breeding for increased animal productivity, and specific traits related to reduced methane emissions, selection for better feed and nitrogen utilization efficient animals; biotechnological strategies and nutritional strategies) for mitigation and adaptation to climate change are to be developed. Proper nutritional management is the most developed and ready to be applied strategy in the field for reducing methane emission from livestock.

Direct inhibition of Methanogens (Methane analogue)

Halogenated methane analogue and related compounds have been used for reducing methanogenesis due to their specific inhibitory effect on rumen archaea. Bromo-chloromethane (BCM), 2-bromo-ethanesulfonate (BES), chloroform and cyclodextrin are the compounds used successfully for reducing enteric methane from live animals. These CH_4 inhibitors reduced methane production by up to 50 percent *in vivo* in cattle and small ruminants (Lila *et al.,* 2004; Knight *et al.,* 2011) and majority of them reported no effect on feed intake, and productivity of ruminants. As these compounds are banned for animal use because of their known carcinogenic effect, they cannot be recommended as methane mitigating agents, however, research in these directions are necessary to develop natural and synthetic compounds that directly inhibit rumen methanogenesis.

Ionophores and Organic acids

Ionophore antibiotics viz. monensin and lasalocid have been used successfully for a long time as feed additives to improve efficiency of animal production and decrease methane emission (Beauchemin *et al.,* 2008). Interaction of ionophores with bacterial cell membrane decrease trans-membrane ionic gradient, resulting lysis or lowered cell growth. The modulation of particular group of microbial community results a shift of fermentation towards propionogenesis, but recent feed regulation does not allow use of these antibiotics into the livestock ration.

Organic acids (malate, fumarate) have been assayed as feed additives, but *in vivo* results are inconsistent. It has been suggested by Martin (1998) that the high malate content in fresh forages at early growth stage, especially lucerne, could lead to significant changes in rumen fermentation. McCaughey *et al.* (1999) observed a decrease in methane production by 10% when replacing grasses by a mixture of lucerne and grasses (70:30). An increase in dietary malate of 3%, could explain the decrease in methane. However, other factors may be involved such as the high intake and a high rate of passage out of the rumen for lucerne, and presence of saponins. Further research is needed in these directions.

Nitrates and Sulphates

Nitrate is known to compete effectively with methanogenesis as a sink for hydrogen generated during fermentation. It is energetically more favourable to reduce nitrate than CO_2, outcompeting methanogens for hydrogen. Recent research (van Zijderveld *et al.,* 2010; Hulshof *et al.*, 2012) has shown hopeful results of feeding with nitrates in decreasing enteric CH_4 production by up to 50 percent. Therefore, feeding of nitrates may be promising enteric methane mitigation agents, particularly in low-protein diets, where rumen bacteria may benefit from non-protein source. The potential use of nitrate to decrease rumen methanogenesis has been hindered by the toxicity of the intermediate product nitrite. Rumen microbes rapidly reduce the nitrate into nitrite, but the rate of reduction of nitrite into ammonia is slower, which can cause nitrite accumulation in the rumen. Therefore, to avoid nitrite toxicity, it is important to consider suitable dose and proper adaptation to animals.

Addition of sulphate in the diet increases the sulphate reducing bacteria (SRB) in the rumen (Paul *et al*., 2011), which can act as alternate hydrogen sink to compete with methanogens. However, possibility of sulphide (H_2S) toxicity due to reduction of sulphate could be a major impediment for its acceptance as methane inhibitor. A synergistic effect has been reported when both sulphate and nitrate are used. Many SRB appear to have dual roles, that is, they reduce inorganic and organic sulphur, and the majority of them can reduce nitrite to ammonia. Their ability to reduce nitrite was explored to avoid the potential toxicity problem encountered when supplementing with nitrate alone. Moreover, an additive effect was observed on decrease in CH4 emission *in vivo* when sulphate (2.6% of DM) and nitrate (2.6% of DM) were used together (van Zijderveld *et al*., 2010).

Fat and Oils Supplementation

Dietary fats and oils seem a promising nutritional alternative to reduce ruminal methanogenesis without decreasing ruminal pH. Fat reduces methanogenesis in three ways viz. direct effect on methanogens, reducing fibre digestibility and bio-hydrogenation of fatty

acids. Medium-chain fatty acids (FAs) are known to affect methanogen numbers (Machmüller *et al.*, 2003) but not long-chain FAs such as linolenic acid. Polyunsaturated FAs also contribute to methane decrease through a toxic effect on cellulolytic bacteria (Nagaraja *et al.*, 1997) and protozoa (Doreau and Ferlay, 1995). This effect, observed with all long-chain FAs, is probably through an action on the cell membrane particularly of Gram-positive bacteria. Linolenic acid was found toxic to cellulolytic bacteria *F. succinogenes, R. albus* and *R. flavefaciens* (Maia *et al.*, 2007). These microbial changes favour a shift of ruminal fermentation towards propionate, and thus to an increase in hydrogen utilisation. *In vitro* studies with supplementation of mustard and sesame oil resulted reduction in methane production with concomitant lowered fibre degradation, however propionate production was increased (Dey *et al*., 2015). The multiple actions of fats and oils may impair digestion if the number and activity of primary microbial fermenters is too affected or if the negative effect on methanogens leads to an accumulation of hydrogen in the rumen. Bio-hydrogenation of polyunsaturated FAs results in an uptake of hydrogen. As adaptation of the rumen microflora to FA supplementation over the long term may be possible, the sustainability of methane suppression by FA supply should be thoroughly tested (Grainger *et al.*, 2008). It has been shown that FAs from linseeds may decrease methane production in growing lambs (Machmüller *et al*., 2000) or in dairy cows (Martin *et al*., 2007) without altering animal performances. Supplemental linolenic acid also contributes to improve the quality of fatty acids of ruminant products and this may compensate for the additional cost of lipid supply.

Elimination of protozoa (Defaunation)

Although methanogens are present as an individual entity, physical association between protozoan cells and methanogens exist in the rumen ecosystem. Methanogens associated extra and intra-cellularly to ciliate protozoa have been estimated to contribute between 9 to 37% of the rumen methanogenesis (Newbold *et al.*, 1995). The removal of protozoa from the rumen has been shown to reduce CH_4 production by up to 50% depending on the diet (Hegarty, 1999). The decrease in methane production of 26% per kg DM intake in protozoa-free lambs was related to a decrease in the proportion of methanogens in the total bacterial population of the whole ruminal content (McAllister and Newbold, 2008). In another study, where CH_4 production decreased by 20% in protozoa-free sheep (Morgavi *et al.*, 2008), the quantity of methanogens estimated was not different between faunated and defaunated animals suggesting that the decreased methanogenesis might be due to a reduction in the amount of hydrogen substrate. Therefore, reduction of rumen protozoa by feeding of diets rich in saponins and others could be another promising approach for decreasing enteric methane emission from ruminants.

Exogenous enzymes

The potential of exogenous enzymes in reducing enteric methane production have been extensively studied (Grainger and Beauchemin, 2011). There is no evidence of direct effect of these preparations on CH_4 production, but they appear to improve diet digestibility and animal production in some studies. Thus, there might be an opportunity to increase fibre digestion in ruminants, which would help to improve feed efficiency of forage-based diets.

Improved feed digestibility may decrease fermentable organic matter in (stored) manure, thus reducing overall CH_4 emissions from ruminant production systems.

Precision feeding and Ration balancing

Matching animal requirements closely to dietary nutrient supply, is important for maximizing feed utilization, stabilizing rumen fermentation, improving rumen and animal health, and minimizing nutrient excretion in manure. These effects of precision feeding are expected to decrease enteric and manure GHG emissions. Formulation of feed with ingredients having low methane production potential could reduce the methane emission. Dietary supplementation of grains induces a decrease in ruminal pH and modification in microbial population. A shift in volatile fatty acid production from acetate to propionate results in less hydrogen production. The relationship between concentrate proportion in the diet and methane production is curvilinear with 56% decrease in methane production observed when diet contained 45 % starch (Sauvant and Giger-Reverdin, 2007). However, practical implications need to be considered due to soaring cost of concentrates.

Feed processing such as reduction in particle size, pelleting, steam flaking increases the intake and digestibility of nutrients, thus reduce methane production per unit animal product. This mitigation practice may not be economically feasible in low-input production systems of developing countries. Adoption of science-based feeding systems with subsistence animal agriculture will have economic benefits for the farmer and will also help to maximize production, feed utilization, and consequently reduce GHG livestock emissions.

Supplementation of locally available tree leaves, grasses, hays and agro-industrial by products to straw based feeding system increases the utilization of basal diet by favouring rumen environment, therefore, reduces methane emission (Dey *et al*., 2014). Tree leaves are rich source of proteins, minerals, vitamins and some growth promoting factors which can alleviate the deficiencies and stimulates specific group of microbes resulting increased digestibility of feeds (Dey *et al*., 2006). In developing countries, where animals are reared on poor quality fibrous feeds, strategic supplementation could improve the feed digestibility, therefore, less methane production per unit product.

Feeding of Direct-fed Microbials (DFM)

The use of direct-fed microbials for the stimulation of rumen microbial populations capable to decrease methane emissions potentially remains an interesting approach. Direct-fed microbials have been defined as a 'source of live, naturally occurring microorganisms' (Krehbiel *et al*., 2003) and, they have been successfully used in ruminant production to increase productivity, to prevent digestive disorders like acidosis and to decrease pathogenic load in young animals. However, to date there is little evidence to suggest the efficacy of DFM to control the production of methane in ruminants.

Yeast (*Saccharomyces cerevisiae*) has been extensively used as DFM for calves for improvement in rumen maturity by favouring microbial establishment, stabilizing rumen pH and increasing fibre degradation. Although feeding of live yeast has been reported to increase growth and hydrogen utilization by acetogenic bacteria (Chaucherus-Durand et al., 1995), their effects on rumen methanogenesis remained inconsistent. Increase in propionate production, a hydrogen utilization pathway decreases methane production. Lactate utilizing,

Megaspheara elsdeni is the major rumen bacteria producing propionate from lactate via acrylate pathway. *Prevotella ruminicola* also produce propionate from lactate, but it is not significant in rumen. *M. elsdeni, Propionibacterium sp.* and *lactobacillus sp.* have been used in increasing animal productivity and to prevent rumen acidosis, but their effect on ruminal methane production was not reported. Decrease in CH4 emission was reported in lactating dairy cows receiving a mixed culture of DFM (*Propionibacterium* sp. and *Lactobacillus* sp.) showing the potential of this approach to mitigate rumen CH4 emission (Lettat *et al*., 2012).

Providing nitrite-reducing bacteria as DFM along with nitrate may improve the nitrate reduction process with abatement of methane production and avoid nitrite toxicity. The major nitrate-reducing bacteria present in the rumen are *Wolinella succinogenes, S. ruminantium* and *Veillonella parvula*. However, to compete with methanogens it may be advantageous to increase the number and/or the activity of nitrate- and/or nitrite reducing bacteria in the rumen if nitrate is going to be a strategy to decrease methanogenesis. Addition of nitrate increases the number of nitrate reducing bacteria but, this increase may not be sufficient to compete with methanogenesis. *In vitro* addition of nitrate-reducing-bacteria *W. succinogenes, S. ruminantium* or *V. parvula* to mixed methanogens in the presence of nitrate (5 mM) was reported to decrease (>70%) methanogenesis (Iwamoto *et al*., 2002). However, there is still scarce *in vivo* data on their ability to decrease CH_4 production and/or avoid nitrite toxicity particularly when nitrate is used as feed additive.

In rumen, competitive and co-operative relationship exists between methanogens and sulphate reducing bacteria (SRB). As energetic value of sulphate reduction is slightly higher than methanogenesis, introduction of SRB should decrease methane production theoretically. The major SRB present in the rumen are *Desulfovibrio, Desulfotomaculum* and *Fusobacterium.* However, as SRB are present in rumen in lower number, introduction from external source and/ or increase in existing SRB population along with sulphur sources could be an approach to reduce methanogenesis. A decrease in CH_4 emission was observed in an *in vitro* study using the newly identified SRB, *Fusobacterium* sp., as a DFM with a high sulphate diet. The CH_4 production at 72 h was decreased from 2.66 to 1.64 mmol/g digested dry matter (DM) without H_2S accumulation (Paul *et al.,* 2011).

Diverting hydrogen from methanogenesis towards acetogenesis has been assayed by several authors. The final product of the reaction, acetate, has the additional advantage of being a source of energy for the animal. However, in the rumen environment, acetogens are less efficient than methanogens in the competition for reducing equivalents and attempts to boost their activity had been so far not successful. The recent isolation of new acetogen species, which can grow at low threshold levels of hydrogen could offer a better alternative than previously tested acetogens (Boccazzi and Patterson, 2011).

Therefore, strategy to reduce methane production from ruminants through DFM needs to be examined thoroughly through animal experimentation ad could be a promising technology for enteric methane abatement.

Plant Bio-active Compounds (PBAC)

There is a growing interest in the use of plant bio-active compounds (condensed tannins, saponins, essential oils) as a CH_4 mitigation strategy because of their natural origin in opposition to chemicals additives. For tannin-containing plants, the anti methanogenic

activity has been attributed mainly to condense tannins (Dey *et al*., 2008), a direct effect on ruminal methanogens and an indirect effect on hydrogen production due to lower feed degradation (Dey, 2016). The mode of action of saponins seems to be clearly related to their anti-protozoal effect (Newbold *et al*., 1997; Dey *et al*., 2015a). However, the effect of saponins on protozoa may be transient (Koenig *et al.*, 2007).

Essential oils (EOs) are gaining importance in ruminant nutrition for reducing enteric methanogenesis and improvement in health and productivity of animals. The antimicrobial effect of EOs suggests their relevance as antimethanogenic feed additive but challenge is to maintain feed digestibility. It is indicated that essential oil reduces methane production either by inhibiting methanogenic archaea, changes in the phylogenetic distribution of archeal population or activity of methane producing genes (Ohene-Adjei *et al*., 2008). Macheboeuf *et al.* (2008) reported an inhibition of methane (0.93- 89%) production on incubation of corn-soybean and hay based feed with graded (1-5mM) doses of cinnamaldehyde, however, feed degradability was also reduced at higher doses. Singh *et al.* (2017) observed a linear decrease in methane production with increasing doses of eucalyptus oils (ECO) with a reduction in methane production (82.60% to 85.30%) with a dose of ECO at 2-3ml/l. Similarly, a dose dependent reduction in methane production (58%, 1.66 ml/l, Kumar *et al*. (2009); 90.3%, 2ml/l, Sallam *et al*. (2009) were reported with supplementation of eucalyptus oil to *in vitro* fermentation medium. A reduction in methane production was reported by Kumar (2017), on incubation of oats hay with petroleum ether extracts (1 ml/ 30 ml) of eucalyptus leaves, poplar leaves or clove buds. The blends of these extracts were observed to have more pronounced effect on methane reduction at lower doses

Garlic oils contain various bio-active components like allicin, diallyl sulfide, diallyl disulfide, allyl marcaptan which have direct antimicrobial effects to a group of organisms (Busquet *et al*., 2005). In a study with graded doses of garlic oil (GOL) supplementation (Dey *et al*., 2016), total gas production was increased with supplementation of GOL-1 (33.3µl/l) however, it remains unaffected ($p>0.05$) with higher doses (GOL-2 and GOL-3, 83.33 and 166.66 µl/l, respectively) of supplementation. Methane concentration in the head space gas was less and thus, total methane production was reduced ($p<0.001$) with the increasing doses of garlic oil. The organosulfur compounds present in the garlic oil has direct inhibitory effects on methanogenic archaea by inhibiting HMG-CoA reductase enzyme, which is required for the synthesis of membrane lipid of the archaeal community (Busquet *et al*., 2005). Wanapat *et al*. (2008) supplemented lemongrass powder at 100 g/d to beef cattle and found improved digestibilities of nutrients, rumen microbial population, and microbial protein synthesis efficiency. A linear decrease in methane production was reported by Singh *et al*. (2016) with *in vitro* incubation of oat hay or mixed feed with graded doses (10- 120 µl/40 ml) lemongrass oil (*Cymbopogon citrates*). However, feed digestibility and volatile fatty acids production were reduced at higher doses, suggesting general inhibitory effect on rumen microbes. The essential oil components (citral, limonene, citronellal, geraniol) present in lemongrass oil are the key factors for inhibiting methanogenesis. Therefore, essential oils could be used as antimethanogenic feed additive to reduce enteric methane emission from livestock, however, long term *in vivo* experiments with different feeding regimes need to be conducted before their use in animal ration.

Therefore, PBAC have the potential to improve nutrient utilization efficiency and reduce the impact of livestock production on environment. Essential oils and their components, especially could reduce entric methane and ammonia emission by modulating rumen microbial community structure and could be used as phytogenic feed additive. However, their effects need to be studied by long term *in vivo* experimentation, as adaptation of rumen microbes could lead to ineffectiveness. Optimum dose of bioactive components and their appropriate combinations needs to be standardized in relation to dietary composition of animals to achieve consistent benefit of their feeding for enteric methane mitigation and improvement in livestock health and production.

Conclusions

Multi-dimensional strategies for enteric methane mitigation are to be developed through genetic selection of animals for desirable traits as well as feeding management for efficient conversion of fibrous feeds into animal products. Development of new products/delivery systems for anti-methanogenic compounds or alternate hydrogen sinks in the rumen is the most promising area of current research for reducing methanogenesis. However, their consequence on animal performances, safety for the ruminant and the consumer, and economic viability is an important issue to consider before their application in ruminant production systems.

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CHAPTER 6

Efficient Buffalo Production System

Sanjay Kumar* and A. Bharadwaj
Division of Animal Genetics and Breeding
ICAR-Central Institute for Research on Buffaloes, Hisar
*Corresponding author: Dr. Sanjay Kumar (Senior Scientist), sanjayivri@gmail.com

Buffalo farming is a very important occupation for many people in the Indian subcontinent especially in India and also has good potential for employment generation. Buffaloes have several advantages over crossbred cows as farm animal. The buffaloes are known to be highly valuable for the purposes of milk and meat efficiency. India is home to great biodiversity of buffalo germplasm, including the world famous Murrah buffaloes- renowned for high milk production potential. Total buffalo population in India is over 109.85 million headcounts constituting 20.45 per cent to total livestock population (20th livestock census, 2019), representing approx. 57 percent of the world buffalo population. People in India and some other Asian countries place more emphasis on buffalo rearing than any other domestic animal.

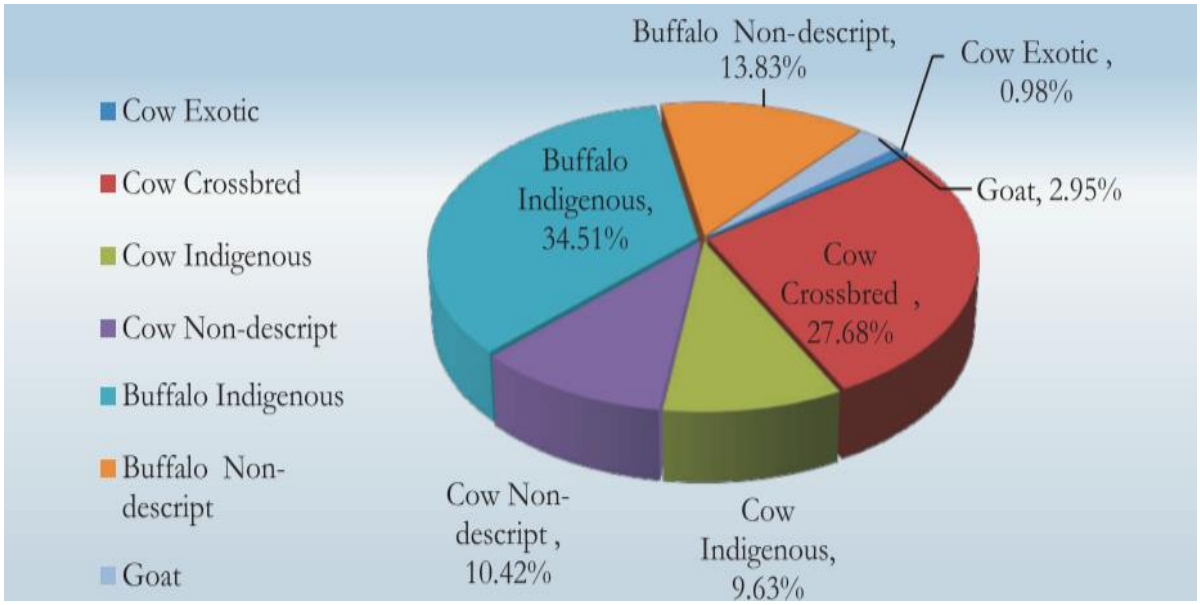

Fig 1:Species-wise milk contribution across the country (BAHS, 2020)

Buffalo milk, its dairy derivatives, meat and hide are the major bi-products of buffalo farming. Indian buffaloes are required for necessary supply of milk nowadays and approx. half of the total milk produced (48.34%) in the country is contributed by the 55.00 million milch buffaloes. There are around ten native customary breeds of buffalo viz. Murrah, Nili Ravi, Surti, Jaffrabadi, Bhadawari, Mehsana, Nagpuri, Toda, Godavari, Pandharpuri which are well known for their milking qualities.

Buffaloes contribute 18.43 percent of total meat production of India and having major part (89.08%) in meat export from the country.

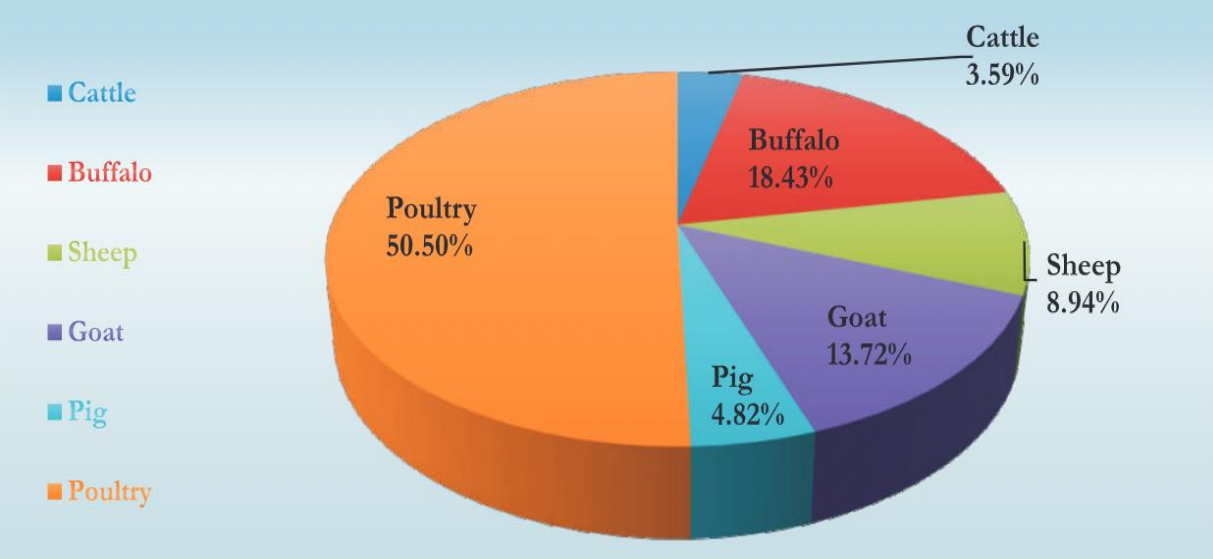

Fig 2:Species-wise meat contribution across the country (BAHS, 2020)

Buffalo milk is better in quality as compared to any other domestic animal due to high content of fat, protein and SNF. The average content of fat 7%, protein 4.6%, SNF 10%, lactose 4.5% and Total solids is 17% is available in buffalo milk. Due to presence of more fat, solids not fat and total solids, yield of products prepared from buffalo milk such as cream, butter, cheese or condensed milk will be always higher. The inherent properties of buffalo milk like high total solids content, superior whiteness and viscosity render it eminently suitable for the manufacture of traditional (indigenous) milk products like khoa, dahi, paneer, kheer, malai, kulfi and ghee.

Buffalo farming/rearing system depends on the purpose for which they are bred and maintained. Most often the buffaloes are raised by the farmers who have small land holding. In Indian scenario adoption of efficient buffalo production system is the need of the day for successful buffalo farming.

Following steps need to be adapted to make buffalo farming a profitable business by implementing efficient buffalo production system.

1) Selection of suitable area
2) Selection of good quality breed of buffaloes
3) Housing
4) Feeding
5) Breeding
6) Care of buffalo and calf after calving
7) Disease control

1) Selection of suitable area: An area with all required facilities will be good for successful buffalo farming. Following facilities are required in buffalo farming area.

(a) Source of water: Plenty of clean water is needed for farm operations like washing, fodder cultivation, processing of milk and byproducts and for drinking. Hence a water source that provides continuous water supply is essential.

(b) Land for animal farm, cultivating fodder and feed: There should be vast area to construct all building and should give way to future expansion of farm. Adequate land is required to cultivate the fodder for livestock.
(c) Drainage facility: Proper drainage of rain and subsoil water should be provided to keep healthy environment and to protect the building from dampness.
(d) Availability of electricity and all required equipment: Proper electricity supply should be available at the site. All required equipment such as milking machine, bulking tanks, strainers, teat cups, milk pails, milk pans, weighing balance etc. should be there.
(e) Availability of labour: Availability of cheap and trend labour should be arranged. Animal farm should be designed is such a fashion so that labour cost can be reduced.
(f) Availability of Market facility: The farm should be away from the city but at the same time it should be nearer to city thereby the products produced from the farm could be marketed easily.
(g) Good transportation service: A vital location issue surrounding infrastructure of approach roads, nearness to main roads and straightforward accessibility need to be considered. Road facility is prerequisite to reach the nearest commercial hub for day to day milk supplies. Along with the need of transportation facility for milk, a large number of associates such as vendors, technicians, consultants and even customers would need a facility to reach the site.

2) Selection of good quality breed of buffaloes:
The basis of increase buffalo productivity is the "only one way to select and to keep the best and cull the poorest". This is a two-stage operation in which the superior individuals are first identified / selected and then used as parents / breeding stock for future generations. Selection of dairy animals is based upon individual performance, pedigree records and progeny performance as discussed below:

1. Selection based on breed characters and own performance
2. Selection based on pedigree records
3. Selection based on progeny performance
4. Show ring selection

Before considering the above basis of selection, some important points are considered for general selection of dairy animals (male and female) as:
i) Birth weight (should be above breed average)
ii) Animal should be free from any genetic abnormalities and
iii) Growth rate (growth rate should be optimum @ 500 to 600 gm /day in buffalo)
Selection of good breed is required for production purpose as some breeds are good for producing milk and some are good for meat purpose. Some well-known and popular buffalo breeds for milk purpose are Murrah, Nili-Ravi, Surti, Jafarabadi, Pandharpuri, Nagpuri, Bhadawari, Banni and Marathwadi.

3) Housing: Proper ventilated and clean housing is prerequisite for optimum buffalo production. Buffalo being an animal well adapted to temperate environment, has special housing needs. Buffalo housing should be well ventilated, protected from cold and hot weather and with ample supply of fresh and clean drinking water. This section deals in detail with buffalo housing for optimum production.

Housing Management

Loose housing is the most economical system of keeping animals. Animals are kept loose in an open paddock throughout the day and night except at the time of milking. The open paddock is provided with shelter along one side under which the animals can take feed and do rest when it is very hot or cold. A common watering tank and feeding manager is provided to make management more effective. Loose housing system has an advantage over the conventional housing system as cost of construction is significantly lower and further expansion is possible according to requirement. In loose house animals get optimum exercise and it also facilitate easy heat detection of animals. Experiences in the Indian subcontinent suggest that loose housing should be preferred over conventional animal sheds.

Construction of Buffalo Sheds

During construction of a house for buffaloes, care should be taken to provide comfortable accommodation with proper sanitation, durability and arrangement for the production of clean milk under convenient and economic conditions. Climate of the region is also important and should be considered before construction of animal housing facilities so as to protect them from extreme hot or cold seasons.

For construction of buffalo shed following points should be undertaken:

- Entire shed should be surrounded by wall of 5 feet height from three sides and the manger from one side. The feeding manger should be designed in such a way that quick and proper distribution of feed and fodder is possible.
- The height of the inner wall of the manger from the ground should be 50 cm for adult buffaloes and 20-25 cm for young calves.
- Depth of feeding manger should not exceed 40 and 20 cm for adult buffalo and young calves, respectively.
- Near the manger under the roofed house 5 feet wide, non-slippery easy to clean floor having a little slope should be provided.
- The floor of the covered area should be a little above the ground level of the open area.
- The drains should be located near the junction of covered and open area. Beyond that there would be an open unpaved or paved area. The covered and open area per animal should be 30- 40 and 800- 100 sq ft for adult buffaloes and 20-25 and 50 -60 sq ft for calves, respectively.
- The feeding space should be provided with 2½ - 3 ft of manger space per buffalo and 1½ ft for calves in the covered area.
- The roofing of shelter is done with slight slope by asbestos sheets on pipes and angle iron. The thatch roofing may also be considered to reduce the initial cost and provide comfort to animals as compared to asbestos or tin sheets.
- A common water tank is also provided in open area so that every animal can have access to fresh and clean water.

Floor Space Requirements for Different Categories of Buffaloes

Category	Floor Space/animal (sq. ft.)		Manger Length /animal(ft.)
	covered	**Open**	
Buffalo	30 - 40	80 - 100	2.5
Calves	20 - 25	50 - 60	1.5
Pregnant buffalo	100 - 120	180 - 200	2.5
Bull	120 - 140	200 - 250	2.5

4) Feeding: Adequate green fodder is mandatory for raising buffaloes because green fodder is essential for intensive milk production and also for fattening purpose. Fodder can be preserved as hay and silage for future use. Proper quantity of concentrate ration and mineral mixture should also be provided to various age groups of buffaloes.

A balanced ration should provide all the essential nutrients (carbohydrates, protein, fat, vitamin, minerals and water) in such a proportion and quantities that is required for nourishment of the particular type of animals for a period of 24 h and also improves FCR, ruminal nutrients flow, enhances immunity and suppresses parasitic load and methane inhibition. Ration balancing is the process to balance the level of various nutrients in ration, from the available feed resources, for optimum utilization of feeds to meet nutrient requirements for a particular physiological stage of animals (maintenance, production and reproduction). It should also be cost effective with minimal impact on environment and have positive effects on general health and well-beings of animals.

Requirement for balanced ration for:

- Increases efficiency of utilization of available feed resources
- Improvement in production performance viz. milk production, growth rate etc.
- Improvement in product quality viz. milk quality, meat quality
- Enhancing of net profit of animal rearing by reducing the feed cost
- Improves reproductive performance
- Improvement of overall health and well-being of animals by improving antioxidant and immunity status of animals
- Reduces environmental pollution by lowering enteric methane and ammonia emissions and excretion of nitrogen through faeces and urine by efficient protein utilization.

Feeding of buffaloes should be done with the consideration of following points:

- Standard mineral mixture (BIS type II or area specific) and salt may be added in the concentrate mixture or fed separately @ 0.6% and 0.3% of total DMI, respectively. Thus, it comes around 20 g mineral mixture for young calves and 60-80 g for lactating and adult buffaloes depending on body weight and milk production. Mineral mixtures of good companies only bearing BIS or ISI mark should be purchased.

• Daily feeding of minimum 2-3 kg green fodder is necessary to meet the requirement of Vitamin A. Milch buffaloes should be supplemented with 20,000-40,000 IU Vitamin-A per day, when no green fodder is available.
• High yielding lactating buffaloes should be fed at regular interval (4 times daily) to maintain continuous fermentation in rumen. Forage should be chaffed and may be mixed with required quantity of concentrate mixture to make total mixed ration for better utilization of feeds.

5) Breeding: Reproductive efficiency of the buffaloes shows wide variation throughout the year. The females exhibit a distinct seasonal change in displaying oestrus, conception rate and calving rate. The age at first oestrus of heifers varies between breeds from 24-36 months depending on the breed. Gestation of the buffaloes generally lasts from 281-334 days.
Reproductive problems in buffalo are associated with poor heat (estrus) detection by the owner. In buffaloes, heat signs are very minute and owner is not able to identify them. As a result buffalo owner complains about delayed puberty, silent estrus, summer anestrus, post-partum anestrus, repeat breeding and long calving interval. These problems lead to huge economic losses to the farmers and lengthen the interval between successive calvings, reduce the life time production and net calf crop and are responsible for high culling rate.
Buffalo come into heat mostly during night hours and may not be very active in hot weather, and remain in heat only for a short period (roughly 6–12 hours), making it difficult to observe. Two to three visual observations per day will increase the chances of identifying animals in heat. Different strategies are required to be employed for detection of heat in buffaloes.
a. Observe carefully for heat signs: Buffalo does not express all signs of heat with similar intensity. Transparent mucus discharge hanging from vulva while sitting, bellowing and frequent urination are commonly observed estrus signs in majority of buffaloes.
b. Observe at least three times in a day: Unless declared as pregnant, each buffalo should be considered as non-pregnant. In a day each buffalo should be checked at least thrice to take care of short duration of estrus.
c. Record every suspicious heat and make heat expectancy chart: Each suspected heat must be recorded properly on a register or on a calendar. This enables the farmer to expect next heat after ~21 days. Whenever, buffalo is suspected for heat, it should be presented either to bull for natural service or to a veterinarian for AI and confirmation of estrus signs.
d. Get Buffalo checked by a trend veterinarian: If a buffalo does not show signs of heat even 90 days after calving, it must be get checked by a veterinarian for proper hormonal treatment. Hormone treatments are useful only when animal is healthy and receiving balanced diet.
e. Provide extra care during summer: During summer months estrus symptoms are very weak, of shorter duration and expressed particularly at night therefore extra care for heat detection should be done during summer.

Major infertility problems, possible causes and suggestions for their prevention and control

A. Anestrus:

Absence of periodic manifestation of estrus, with absence of palpable follicular or luteal structure (true anestrus), or absence of normal physiological signs of estrus (subestrus) is defined as Anestrus. It is caused due to persistent CL is mostly associated with uterine pathology (pyometra, fetal resorption, and mummification), early embryonic motility, pregnancy, subestrum and unobserved estrum. Broadly speaking, anestrus may either follow parturition (post-partum anestrus) or following service (post-service anestrus).

I. Silent estrus with normal ovarian activity may result from:

a. Inadequate estrus detection at an appropriate time.

b. Inadequate animal identification and / or inadequate records

c. Lack of a sufficient secretion of estradiol from the mature and secondary follicles or due to need for a higher threshold of estrogen in central nervous system to produce nervous symptoms characteristic of estrus and acceptance of the bull.

II. True anestrus (lack of ovarian activity) is caused by:

a. Failure of follicular growth through lack of endocrine stimulus and/ or failure through nonregression of corpus luteum.

b. Inadequate LH pulse frequency due to negative energy balance, malnutrition, environmental stress, lameness, endogenous opioid peptide, lactational stress, suckling and lower insulin concentration.

c. A lesser number of primordial follicles, higher rate of atresia and lower levels of circulating gonadotropins are some of the inherent causes of ovarian inactivity in buffaloes.

Control suggestions:

1. Silent estrus/ subestrus

(a) Closely observe animals for estrus at least twice and preferably 3 times a day for at least 20 to 30 minutes at each detection time. (b) Provide non-slippery floor. (c) Adopt procedures to tackle heat stress. (d) Improve management practices.

2. True anestrus

(a) Check problem animals for parasites and treat accordingly. (b) Examine blood for macro and micro-mineral status. In case of deficiencies provide mineral mixture supplementation. (c) Submit forage samples for standard and mineral tests. (d) Examine animals at least once between 15 and 45 days after calving to ensure proper uterine health, cystic ovaries and, for resumption of ovarian activity. (e) Provide round the year access to fresh forage. (f) Animals should be in a good body condition at the time of breeding. (g) Keep hormonal therapy as last resort in case herbal therapy and supplementation of mineral mixture fail to yield results.

B. Repeat breeding

Repeat breeder animal is usually defined as sub-fertile animal which mated three or more times during the proper period and does not become pregnant and continually return to service in the absence of any obvious pathological disorder in the genital tract and normal estrous cycles. Possible causes:

(1) Genetic factors. (2) Anatomical defects like congenital abnormalities of uterus, cervix and vagina are infantilism, segmental aplasia, uterus unicornis, double cervixes, vaginal constriction and septum. (3) Hormonal imbalances such as abnormal endocrine status during

folliculogenesis and ovulation. Repeat breeding has been associated with ovulatory disturbances like delayed ovulation, anovulation and follicular cysts. (4) Managemental factors including improper heat stress, poor nutrition, and improper insemination techniques. (5) Infectious causes including trichomoniasis, vibriosis, and endometritis due to *E. coli*, *Archanobacterium pyogenes* and *Staphylococcus aureus*.

Control procedures:

(a) Regular checkup of old bulls and newly purchased bulls for vibriosis and trichomoniasis. (b) Inseminate cows 12 hours after initial observation. In case of long estrus cows (more than 24 hours), a second insemination 12 hours later should be performed. (c) Examine repeat breeder cows for presence of endometritis, delayed ovulation or other abnormalities and treat according. (d) Use high fertility bulls. (e) Adopt fixed time insemination (FTAI) and embryo transfer procedures in large herds.

C. Retained fetal membranes

When a cow fails to expel afterbirth (fetal membranes) within 12 hours after calving, the condition is known as retained fetal membranes (RFM).

Possible causes:

(a) Specific infections associated with abortion like abortion, leptospira, infectious bovine rhinotracheitis (IBR) etc. (b) Twin births and abnormal deliveries, including prolonged or difficult ones that require manual handling or caesarean section. (c) Deficiency of selenium, vitamin A or E. (d) Deficiency of collagenase enzyme. (e) Inadequate rise of estradiol during last stage of gestation due to oxidative stress. (f) Over conditioning of dry cows due to excess energy intake.

Control suggestion:

(a) Periodic tests against specific infections. (b) Keep calving area clean and well bedded. (c) Breed heifers to bulls of appropriate size and with a record of calving ease. (d) Perform manual removal after 96 hours of fetal birth and avoid manual removal in case of fever. (e) After correction, advise for anti-microbial and anti-inflammatory therapy. (f) Give calcium borogluconate etc. to treat uterine inertia. (g) Provide supplemental selenium in deficient areas.

D. Metritis

Infection of uterus that extends into deeper layers of uterine wall and is associated with serious systemic signs, discharge of sero-sanguinous and foul smelling fluids from the uterus. Metritis generally occurs within days to weeks of parturition as hence termed as puerperal metritis.

Possible causes:

(a) Prolonged dystocia that requires manual handling resulting in contamination of uterus. (b) Retained fetal membranes and injury to reproductive tract due to difficult calving or use of excessive force during handling. (c) Selenium and vitamin E deficiency. (d) Over conditioning at the time of calving or during early lactation.

Control:

(a) Allow animals to calve in a clean place. Follow maximum antiseptic procedures during handling of dystocia. (b) Avoid over-conditioning during late lactation and dry period, while maintaining adequate, balanced vitamin and mineral intake. (c) Treat RFM cows

judiciously. (d) Stabilize animal using intravenous fluids and also by drenching. (e) Give anti-inflammatory drugs like Flunixin meglumine that have additional anti-endotoxic effect. (f) Give broad spectrum antibiotics that provide sufficient MIC inside uterus via systemic route. (g) Treat animals for secondary hypocalcaemia.

E. Cystic Ovarian disease

Ovarian cysts are structures, usually greater than one inch in diameter, which persist on one or both ovaries for 10 days or more in absence of CL. Major Categories of cysts include follicular cysts, luteal cysts and cystic corpora lutea. Follicular cysts result from failure of ovulation and luteinization. Follicular cysts are blister-like structures, flaccid to the touch. Luteinized cysts apparently fail to ovulate, but some luteinization occurs. Because of the varying degree of luteinisation, luteinized cysts are firmer to the touch than follicular cysts though not as solid as CL. Cystic CL is CL with a fluid filled centre and is considered non-pathological. Ovarian cysts seriously affect the herd fertility by increasing days to first service; prolonging calving interval and reducing conception rates at first service.

Possible causes:

(a) Neuro-endocrine imbalance. (b) Excessive calcium intake or wide calcium phosphorus ratio. (c) Genetic predisposition. (d) Stressful conditions at calving or early postpartum. (e) Ingestion of estrogenic fodders or some mould toxins.

Control suggestions:

(a) Remove predisposing causes. (b) Use progesterone releasing devices to reset HPG axis. (c) Use of luteinizing agents like hCG and GnRH at the time of breeding. (d) Give single dose of hCG or GnRH 12-14 days postpartum. (e) Give prostaglandins in case of luteal cyst. (f) Fixed time insemination using GPG programme.

F. Abortions

It is the expulsion of a living fetus or more specifically of a dead fetus of recognizable size at any stage of gestation prior to completion of term. Causes of abortion can be either infectious or non-infectious.

Possible causes:

(a) Specific infections like brucellosis, leptospirosis, IBR, BVD, listeriosis, vibriosis, trichomoniasis, fungal infections etc. (b) Genetic defects resulting in severely abnormal embryo or fetus. (c) Multiple-fetus pregnancies like twins, triplets etc. (d) Toxicities due to nitrate, cyanide, chlorinated naphthalenes or some weeds. (e) Drug-induced abortions due to Dexamethasone, Prostaglandins etc. (f) Injuries like accidental removal of CL, excessive manipulation of reproductive tract and violent injuries during late pregnancy.

Control procedures:

(a) Avoid injuries and use extreme care during handling of reproductive tract of possibly pregnant animals. Provide non-slippery floor. (b) Submit suspected feed and water samples for laboratory analysis. (c) When abortion occurs, submit required samples for serological and microbiological testing. (d) Isolate aborting animals from rest of the herd. (e) Follow strict quarantine procedures. (f) Vaccinate animals for specific infectious diseases at proper time intervals.

6) Care of buffalo and calf after calving:

The success of any dairy farm depends upon fast growth of calves to a breedable age with a minimum mortality, so as to have replacement stock. To keep the calves healthy it is important for every farmer to implement best calf management practices.

The buffalo calf may have some innate systematic problems which make it more vulnerable to the physical, nutritional and disease environment. After birth there are some problems which may arise such as: (i) Infection of calf during and just after birth (ii) Catching cold just after birth (iii) Improper maneuvering of navel cord (iv) Too late or insufficient supply of colostrum, (v) Diarrhoea in calf after birth.

Proper expulsion of placenta after calving is highly essential for prevention of uterine infection in buffaloes. Colostrum feeding to new born calves for few days increases the immunity in their body. Calves should also be prevented from cold, heat and diseases like diarrhoea.

Infectious and non-infectious causes are the main reason of buffalo calf mortality. Various infectious agents are capable of causing diarrhoea and pneumonia in the neonatal calf which includes bacteria (*E. coli*, *Klebsiella* spp., Salmonella spp., *Pasteurella multocida*, *Mycoplasma bovis*), Virus (Rotavirus, coronavirus), Protozoa (Cryptosporidium, Coccidia) and Helminthes (*Toxocara vitulorum*, *Moniezia* spp.). The major non-infectious causes are dystocia, improper feeding of colostrum, low birth weight, and poor management practices.

During the 1st few hours of birth the calves should be protected from source of infection since natural immunity at that time is not yet achieved and besides, the calf has to overcome the discomfort and stress due to change in environment and physical surroundings. The new born calf gets infected mainly by germs penetrating into the body through mouth, nasal passage and through navel cord. Therefore, it is essential to wash the vulva and hind quarter of the animal with disinfectant before and during parturition and help out the calf at the time of calving with clean hands or disinfected gloves. Calves are frequently exposed to disease from the dam at birth; therefore minimum contamination of calving area should be kept. In the calving pens where the birth has to take place, the calves should be placed on the floor and putting sufficient straw on the floor and removing the dam's faeces if any.

Immediately after birth, the mucous or phlegm should be cleared from the nostril and mouth of calf. It becomes necessary to provide artificial respiration, if calf does not start respiring after birth. Allow the mother to lick her calf dry, if necessary the calf should be cleaned and dried with gunny bags or wheat straw. It is most important to disinfect the navel cord soon after birth. The simplest way is to cut the navel cord, if it is longer than 3 inches, by the sterilized scissors and apply tincture iodine on the cord by the cotton swab or the cord itself is dipped in tincture iodine.

Placenta of the buffaloes effectively separates the blood of the fetus from that of the dam and prevents any transfer of protective immunity while in the uterus, therefore, the antibodies cannot pass from dam to offspring through placental membranes. As a result, calves are born with no circulating antibodies to combat infection and the calf is born completely dependent on the absorption of maternal antibodies from colostrum after birth. Total amount of 4-5 litres colostrum should be ensured to fed the calves during the first 24 hours of calving.

Colostrum of another animal in the herd should be fed, if colostrum is not available from the calf's mother. High nutritive value and laxative action to remove the meconium (first faeces) are other important function of colostrum feeding. Buffalo calves seem to have some problem with absorption of immunoglobulin. This may one of the reasons for high mortality rates in buffalo calves during early calf hood.

At the stage upto 3 months, calves require high plane of nutrition and good quality easily digestible feed. However, to speed up development of rumen and early initiation of microbial fermentation the calves should be offered calf starter and green grass from second week of life. As intake of calf starter and green grass increases milk has to be reduced gradually.

7) Disease control

Diseases and other health problems are generally less in buffaloes as compared to cattle. Vaccination against harmful diseases such as FMD, HS, BQ and brucellosis and deworming against parasitic diseases of different age group animals need to be done at proper time. Control of mastitis is one of the major issues in milking buffaloes.

Vaccination is the easiest and cheapest way disease control therefore vaccination of buffalo should be done as given schedule:

Name of Disease	**Age at first dose**	**Booster dose**	**Subsequent dose**
Foot and Mouth Disease (FMD)	3- 4 months	6-8 months	Six monthly
Hemorrhagic septicemia (HS)	6 months and above	-	Annually
Black Quarter (BQ)	6 months and above	-	Annually
Brucellosis	4-8 months of age (Only female calves)	-	-
Anthrax	4 months and above		Annually only in endemic areas

(Bivalent (FMD, HS), Trivalent (FMD, HS, BQ) forms of vaccine are available in the trade name of Rakshabiovac and Rakshatriavac)

Deworming against parasitic diseases

- Deworming should be started from the 2nd week of buffalo calf.
- A single oral dose of 10 g piperazine adipate is recommended preferably in the second week of life to control neonatal ascariasis in buffalo calves. Repeat at 15 days interval
- Different types of dewormers like albendazole, fenbendazole, praziquental, Ivermectin (ecto & endo paracites) etc should be done on rotation basis to avoid resistance.
- The choice of dewormers will depend upon the parasites prevalent in a particular area.
- Deworming every month for first 6 months, thereafter as and when required.

Vitamin A @ 10,000 IU should be given to buffalo calf as anti-infective agent once a week and a total dose of 500,000 IU (usually 1 cc) of vitamin A is required early in life. Special care is necessary to prevent tick infestation in buffalo calves.

Control of mastitis in buffaloes

Mastitis is inflammation of the mammary gland. It is usually caused by microorganisms which have entered the teat canal and moved to the udder. The microorganisms multiply and cause a mastitis infection which results in an inflamed udder.

Significance of bovine mastitis

Economic loss: Mastitis causes severe economic losses to the dairy farmers throughout the world. Mastitis cause annual losses of about USD 153 million (Rs.72 billion) in India as per 2009 estimates. The losses include reduced milk production, discarded milk, replacement cost, extra labour, treatment and veterinary services.

Culling: Mastitis is the major caused of culling of milking animals.

Health concerns of mastitis

Animal health: Loss of functional quarter, Lowered milk production, Death of animals

Human health: Poor quality milk, antibiotic residues in milk

Types of mastitis

Subclinical Mastitis and Clinical Mastitis

Diagnosis of mastitis

a. Clinical symptoms: hot inflamed udder, pain in udder, changes in milk, off colour milk, presence of pus and clots.

b. Presence of high proportion of somatic cells: The SCC is quantified as the number of cells per ml of milk.

- SCC of 100,000 or less indicates an 'uninfected' cow.
- SCC of 100,000 - 200,000 indicates normal cow and requires observation
- SCC of 200,000 - 300,000 indicates cows susceptible to mastitis
- SCC of 200,000 - 300,000 indicates subclinical mastitis
- SCC of >400,000 indicates clinical mastitis and milk unfit for human consumption

c. California Mastitis Test (CMT): The CMT reagent reacts with somatic cells present in milk to form a gel. A plastic paddle having four shallow cups marked A, B, C and D for easy identification of the individual quarter. Approximately 1/2 teaspoon (2 cc) of milk is poured. An equal amount of the CMT reagent is added to the milk. A circular rotating to thoroughly mix the contents. Score in approximately ten seconds while still rotating. Read the test quickly as the reaction tends to disintegrate after about 20 seconds.

Mastitis control Programme

A. Monitoring udder health status: Implementing and effective system of monitoring udder health involves monitoring at herd and individual level. Use of animal side diagnostic test like California mastitis test, somatic cell count and milk bacteriological culturing are important for udder health and milk quality.

B. Therapeutic management: Identify the causative bacteria by culture and treat with antimicrobials after antibiotic sensitivity test. Intramammary antibiotics should be preferred.

C. Prevention of new infection: Premilking and postmilking teat disinfection are the most effective mastitis control practice in lactating animals. Premilking teat disinfection with chlorhexidine in association with post milking teat disinfection reduces new intramammary infection. Post milking teat disinfection is regarded as the single most effective control practice in lactating animals.

D. Concentrate feeding post milking: In order to keep animal in standing position for some time until the teat sphincters close, concentrate should be fed immediately after milking.
E. Cull chronic cases: Culling infected (particularly older) buffalo is a key strategy in mastitis control. Chronically infected buffaloes are likely to be a source of bacteria for other animals. Culling buffaloes with chronic infections helps protect the healthy, young buffaloes which are the future of the herd.
F. Milker hygiene: Absolute cleanliness of personnel is required specifically - Milking should be carried out under good personal hygiene of the milker. He should not be suffering from diseases, especially cough and cold, should wear clean clothes, wash hands properly and cut nails periodically before milking. Milker should be free from contagious diseases like cholera, typhoid, diphtheria and tuberculosis and should be monitored for these diseases rigorously on regular basis. Milker should avoid the wrong milking practice like knuckling and incomplete milking, which leads to multiplication of organisms in the left over milk.

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CHAPTER 7

Stress and its Ameliorating Strategies in Buffaloes

A.K. Balhara and Sajjan Singh*

Animal Physiology and Reproduction Division
ICAR-Central Institute for Research on Buffaloes, Hisar, Haryana
*Corresponding author: Dr. Sajjan Singh (Principal Scientist); sajjansingh@mail.com

Stress in biology refer to an external stimulus that places a 'strain' on the system. Most commonly, in livestock a stress disturbs the nutrient-energy balance in the animal body meaning the animal has to spend some body energy reserve to overcome it. The energy loss results in compromised production status. Physiologic stress can result from deviations in normal physiology especially related to endocrine functions and can be induced by mineral deficiencies, chemical stimulants or binders or changed environmental conditions. Some stressors can be prevented by adopting alternate management practices or through nutria-pharmaceutical interventions. Environmental stress especially heat is often difficult to avert, and leading to heavy losses in animal production systems. Physiological adaptation is defined by biologists as the physiological processes involved in adjustments by the body to various internal and external stimuli for survival and performance. Body's water content and temperature are critical components in the adaptation processes.

Stress type	Indicator	Implications
Physical	Disturbed body condition	Multiple e.g. wound
Environmental	Elevated/reduced body temperature	Thermoregulatory mechanisms disturbed
Disease	Multiple	High lymphocytic activity
Social	Behavioural changes	Low production, dull and low activity
Production	Abnormal body fluids	Disturbed metabolism

Among different weathers and climatic environments, it is the hot weather that ubiquitously affects the performance of flora and fauna. The Indian subcontinent, home to > 90% of worlds' buffaloes, experience extreme climates with temperature reaching up to 48°C in summer and as low as 2°C in winter. The very presence of such large population in such climatic environment indicates that buffaloes are well adapted to such type of ecosystem. Despite that there are sufficient studies to prove that buffaloes are very sensitive to heat stress. Main reasons suggested include:

- Black Colour: absorbs more solar radiations
- Sparse hairs: less insulation from environmental heat

- Fewer number of sweat glands situated deep in skin: lesser evaporative losses from skin

All the factors enumerated above contribute to increase in internal body heat. In fact, these peculiar morphological and anatomical characteristics make buffaloes poor thermoregulators.

Common terms associated with Heat Stress

(a) Heat wave: prolonged period of excessive heat, often with high relative humidity.
(b) Heat Cramps: muscular pains and spasms due to heavy exertion in high heat.
(c) Heat Exhaustion: excessive loss of body fluids (usually through sweat) occurs.
(d) Heat stroke/Sun Stroke: thermoregulatory system of body breaks down; body's internal temperature rises, no sweating occurs and death may occur if not treated immediately

Adaptive changes in Buffaloes in response to heat

Numerous physiologic changes occur in the animal system such as acid-base chemistry, and blood hormones during hot weather; some in response to reduced nutrient intake, but many changes occur as a result of strain in the animal. Neurons that are temperature sensitive are located throughout the animal's body and send information to the hypothalamus, which invokes numerous physiological, anatomical or behavioural changes in the attempt to maintain heat balance. During heat stress buffaloes exhibit reduced feed intake, decreased activity, seek shade and wind, increase respiratory rate, and both peripheral blood flow and sweating. Table 1 lists different physiological adjustments in the animal body in response to heat.

Table 1. Physiological effects of Heat on Buffaloes

S.No.	**Effect**	**Implication**
1	Hemodynamic effects	Increased blood flow to skin and peripheral tissue resulting in :- - increased hydrostatic pressure, - increased capillary permeability, - leucocytic and antibody infiltration, - analgesia
2	Neuromuscular effects	Increased nerve conduction velocity, decreased firing rate of motor neurons resulting in muscle relaxation and increased pain threshold
3	Metabolic effects	Stimulation of Hypothalamus resulting in increased metabolic rate, oxygen uptake and accelerated healing
4	Soft tissue extensibility	Increased collagen extensibility for maintaining greater length after stretching for: - decreased elasticity - less force required to increase length - risk of tissue tearing decreased

These responses have a deleterious effect on both production and reproductive status of the animal. Fig. 1 illustrates different physiological adjustments in buffalo body in response to heat.

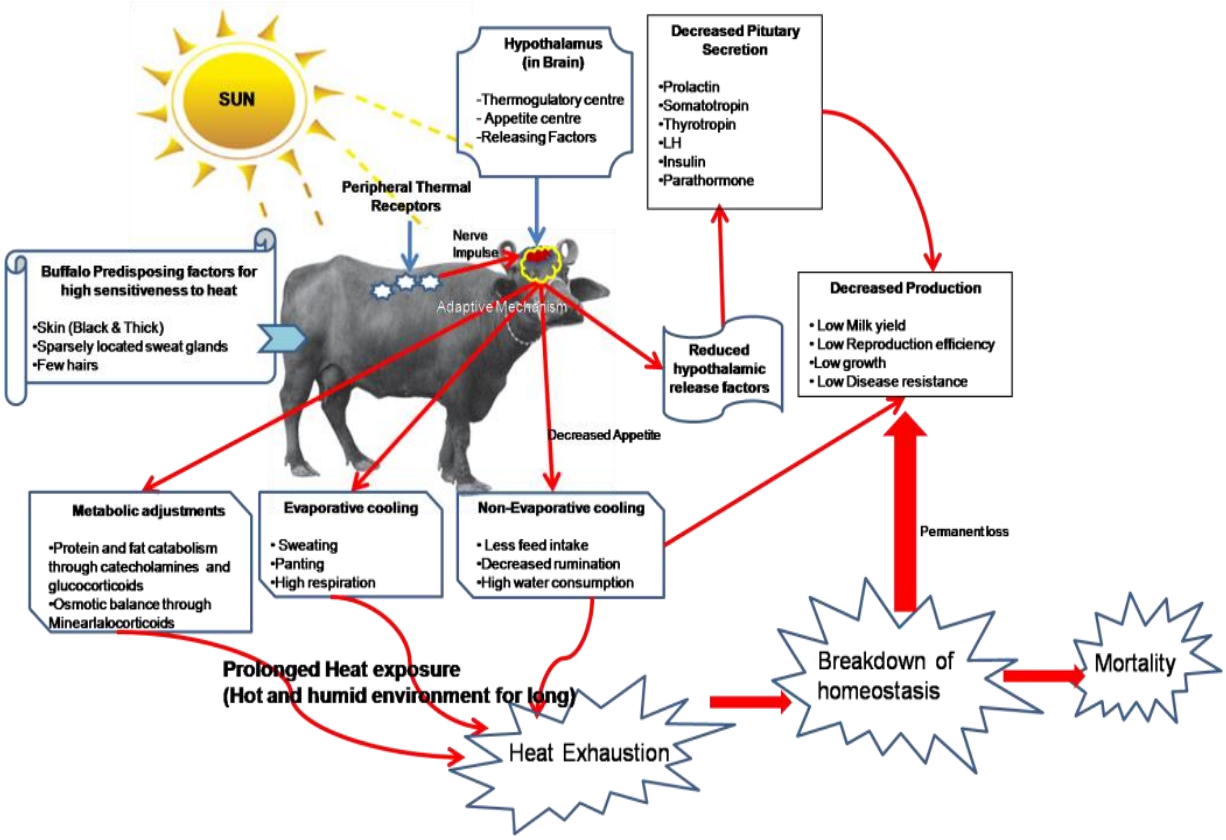

Fig. 1 Physiological mechanisms involved during Heat stress in Buffaloes

The negative effect of heat stress on milk production is due to the decreased nutrient intake and decreased nutrient uptake by the portal drained viscera of the buffalo. Blood flow shifted to peripheral tissues for cooling purposes alters nutrient metabolism and contribute to lower milk yield during hot weather. Hormonal alterations also occur with heat stress such as plasma somatotropin and thyroxine concentration tend to decline with heat stress. Reduced concentrations of these key metabolic hormones with heat stress shows the animal attempt to reduce metabolic heat and milk production. Heat stressed buffaloes generally exhibit altered blood acid-base chemistry as a result of the shift in cooling from conductive, convective, and radiation to evaporative cooling. Heat-stressed animals have elevated rectal temperature and respiratory rate accompanied with a diurnal variation in blood pH and blood bicarbonate levels. Their acid-base chemistry exhibits wide swings from alkalosis to a compensated acidosis resulting in a metabolic acidosis during the cooler evening hours. Reduced concentrations of blood bicarbonate compromise the buffering capability associated with the bicarbonate system, which may be critical during summer. During hot seasons the dry matter intake of buffaloes is decreased moreover the ratio of forage to concentrates intake is also decreased. In other words, the decline in milk yield at higher temperature is more marked in buffaloes that produce more milk. There is a significant relationship between the level of

milk yield and the decline in milk yield with each increase in daily mean environment temperature.

Changes in gene expression levels associated with Heat Stress

The central dogma of life (DNA-RNA-Protein) suggests that changes do occur at the gene levels which percolate as changes in proteins in the body. The heat shock proteins group (HSP) is one of the fundamental groups of molecules which have been used by scientists to study stress at molecular level. The study of these proteins both at gene level and protein level in different tissues including muscles is a useful indicator of the damage at molecular level caused by stress. Study of antioxidant enzyme systems especially in erythrocytes has also been used for long for the study of oxidative changes in the animal body.

How to recognize Heat Stress

- Changes in consciousness: rapid, weak pulse; rapid, shallow breathing
- Abnormal vital parameters: high Heart Rate, Respiration Rate, Rectal Temperature
- Unusual Salivation: Capillary refill is very fast
- Increase of Heat stroke, very high body temperature – sometimes as high 106 – 108°F. Heat stroke is life threatening so immediately call for veterinary aid. Move the animal to cooler place, give bath with cold water or wrap in wet sheets and fan it.
- Signs of heat exhaustion
  - Animal shows sign of dizziness/ unconsciousness
  - Skin becomes dull and may be cold also.

Treating Heat Stress

Buffaloes are well adapted to hot climatic conditions due to special anatomical, behavioural and morphological advantages: -

- Buffaloes have sparsely distributed hair on the characteristics black skin containing numerous melanin granules. Melanin provides protection against UV rays component of sun light, which are abnormally light in typical hot climates like that of tropical regions
- Buffalo dermis layer have well developed sebaceous glands. The oily secretion from these glands makes skin slippery for water and mud. This way extra thick epidermis along with sebum prevents water and solutes from entering the skin. This makes buffaloes defend against harmful ingredients present in mud, water while wallowing. Further this oil secretion from skin makes it more lustrous during summer to reflect solar radiations more effectively.
- Buffaloes are comparatively more susceptible to heat than indigenous cattle breed due to obvious reasons. In fact, buffalo skin has much less ($1/6^{th}$ of cattle) number of sweat glands in the skin. This makes evaporative cooling by sweating less efficient in buffaloes as compared to cattle.

Other Stressors

Physically, animals are able to survive the harsh conditions of cold areas because with their fur and fat they are able to blend in and combat harsh cold weather. Behaviourally, they live a generally solitary life. There are certain adaptations listed below:

a) Low surface area to volume ratio (anatomical) – here animals have small extremities, proportionally shorter legs and a stockier build. This reduces the surface area to lose heat from the body. This is a common characteristic amongst animals that live in cold environments that enables them to retain

their heat and conversely means they often can't cope with warm temperatures as they easily overheat in warmer temperatures.

b) Thick layer of body fat / blubber (anatomical / physiological) - This can be up 10cm (4 inches) thick, it is used both for insulation and also for food storage to help survive when food supply may be intermittent especially in the summer months when animals often go hungry for long periods due to not being able to hunt their preferred food.
c) Processing body fat to gain metabolic water (physiological) - They store a lot of fat which they use for energy in the process combining it with oxygen to release carbon dioxide and metabolic water. They live in a polar desert with little access to fresh water especially in the colder months (which are in the majority) in this way they can get extra fresh water - camels do a similar thing with their stored fat.
d) Pregnant females go into a kind of hibernation in their maternity den during the winter in which they give birth. (behavioural / physiological) - Males and non-pregnant females don't make dens and don't hibernate.
e) They eat a very high fat diet and have adaptations to allow them to process this food (physiological).
f) One of the most important genes in the speciation of polar bears has been one known as APOB, this makes a protein which is part of small molecule called Low Density Lipoprotein (LDL) and a large molecule made of a protein and lipid (fat) commonly known as "bad" cholesterol. The APOB gene protein is important to the process of transporting lipids (fats) from the blood and in the uptake by body cells and in the formation of fatty plaques in arteries that can lead to cardiovascular disease, blood clots and heart attacks. The AOB gene helps to remove LDL from the bloodstream dumping it into fat cells. In a sense polar bears have an anti-junk-food gene which allows them to survive on a diet that would kill most mammals.

Vasoconstriction commonly occurs in order to reduce blood supply to under-ventilated areas of the lung and maintain homeostasis in other respects. At high altitude such vasoconstriction would not be a good long-term response to permanently hypoxic conditions. Because the whole lung becomes a low-oxygenated area at high altitudes, the vasoconstriction reflex would be very damaging, as it would then affect the whole lung.

Female yaks at pasture at an altitude of 3450 m have respiration rate was between 20 and 30 per minute when the air temperature was below 13^{o}C, but above that temperature the respiration rate increase rapidly. Respiration rate was significantly higher in the evening than in the morning. The capacity to take in sufficient air by virtue of anatomical features, respiration rate and physiological response is clearly an important aspect of yak adaptation to life at high altitudes. It is also important that absorption and retention of oxygen from the air should be adequate for the need.

Adaptation Strategies for stress amelioration

It is the process of appropriate adjustment and modification of livestock microclimate to reduce the consequences of climate change. The modification must fulfil the need i.e., survivability, reproduction and production. It is necessary to build or improve the adaptive capacity of animal through implementation of adaptation strategies. It can be either modification of animals' genetics to the changed environment. The important step towards adaptation to climate change is reducing vulnerability and exposure to climatic extremes.

I. Tolerant breeds

Thermal tolerant varies between and within the breeds. It also depends upon the productivity of the animals. The high yielding animals are more susceptible to heat stress (Naqvi and Sejian, 2011). Local breeds are already adapted to their harsh conditions. Adaptation include not only their tolerance to heat, but also their ability to survive, grow and reproduce in conditions of poor nutrition, parasites and diseases (USDA, 2013). Zebu cattle are capable of utilizing poor quality feeds and are more resistant to local parasites and diseases. To increase productivity suitable breed should be selected to the changed climatic conditions through (a) introduction of breeds more adapted to the environment (b) breeding for improved adaptation to heat, disease and harsh conditions using tropically adapted breeds/genes or insert specific genes via strategic crossbreeding or biotechnology, and (c) use of adaptive traits of indigenous animal genetic resources.

II. Shelter management

Good housing management not only comfortable to animals and workers but also good economic output. A higher conception rate of around 80% was obtained in animals given showers in addition to wallowing facilities. Over-crowding of cattle or buffaloes in the shed should be avoided, with a maximum 25 animals in a floor space of 25 ft x 50 ft. Design, orientation and height of the shelters, choice of roofing material, open space ventilation and provision of adequate space per animal are some of the important aspects for cooler microenvironment of the animal. During the period of high temperatures, water can be used to bring down the micro-environmental temperature within the animal shelters. Use of air-cooling systems is also an efficient method. It has been observed that wallowing groups of buffalo showed higher milk yield than the group under water showers.

III. Nutritional Management

There are a number of nutritional technologies for improvement in rumen efficiency like, diet manipulation, direct inhibitors, feed additives, propionate enhancers, methane oxidisers, probiotics, defaunation and hormones (Eckard et al., 2010). Composition of diet has the effect on the rumen microbial ecosystem so any manipulation in the diet by means of forage, concentrate and their components results in change in the microbial community and may decrease or inhibit activity of methanogenic bacteria. Dietary manipulation through increased green fodder decreased methane production by 5.7% (Singh, 2010). Low fibre digestible diet is beneficial during summer season and digestibility of poor forage can be improved by energy supplementation like molasses. In addition, supplementation of fat and providing cold water is helpful. Supplementation of antioxidant is beneficial to combat the oxidative stress. Supplementation of micromolecules, oil and modification of micro-climate may help to alleviate the heat stress. The effect of niacin, yeast, edible oil provided with curtains, additional ceiling fans, and mist were beneficial as ameliorative measures against heat stress. Administration of Vit E helped in reducing superoxide dismutase activity to about 15-20 % and catalase about 5-10 %. Thermal stress on high producing cows and buffaloes may be reduced by addition of Vit E (800-1000 IU/day/animal). Supplementation of vitamin C @10g/animal/day improved the immune status and reduced the oxidative stress in primiparous Murrah buffaloes during thermal stress. The concentration of superoxide

dismutase and catalase were reduced in zinc supplemented group as compared to non-supplemented groups at 42 °C.

IV. Special measures needed during droughts

Drought is normally associated with crop failure and water scarcity. Drought is a regular affair in India at present. There will be decline in growth, milk production due to feed and water scarcity during drought (Thornton et al., 2009). Prolonged drought conditions may lead to reduced reproductive efficiency. Unlike crops, impact of drought on livestock is slow as they are more resilient to drought. During such conditions, focus can be made on efficient utilization of existing feed and water resources. Early warning system can help the people to prepare and act appropriately in sufficient time to reduce the possibility of harm or loss due to drought. Diversification of livestock and crop varieties increases drought and heat wave tolerance, and also livestock production when animals are exposed to temperature and precipitation stresses (IFAD, 2010). Agroforestry (establishing trees alongside crops and pastures in a mix) as a land management approach help to maintain the balance between agricultural production, environmental protection and carbon sequestration to offset emissions from the sector (Smith et al., 2012). Agroforestry also increases productivity and improve quality of air, soil, and water, biodiversity, pests and diseases, and improves nutrient cycling.

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CHAPTER 8

Buffalo Health: Respiratory Disease Complex in Buffalo

Sushila Maan*, Anju Sehrawat, Kanisht Batra and Aman Kumar

College of Veterinary Sciences, LUVAS, Hisar, Haryana, India
*Corresponding author: Dr. Sushila Maan, Professor and Head, Department of Animal Biotechnology, COVS, LUVAS, Hisar (sushilamaan105@googlemail.com)

India has retained the status of highest milk producer in the world, annual milk production being 187.7million tonnes, with a per capita availability of 374 gm/day (NDDB, 2018-19). Out of different states of India, Haryana possesses 2.5 per cent of country's total bovine population with growing annual milk production of 83.81 lakh tonnes in 2018-19 (DAHD, 2018-19). Haryana is among the top ten milk producing states of India, ranking second in terms of per capita per diem availability of milk (835 grams) against the national average of 374 gm (Department of Animal Husbandryand Fishery, 2018-19). Haryana is famous for Murrah breed of buffalo popularly named as 'Black gold. This species alone contributes more than 80% of total milk production in the state. In addition, they are a source of quality lean meat and valuable draught power. Murrah buffaloes are heavy milk producers with high fat content in addition to being efficient feed converters even when fed poor quality roughages. However, it is suffering from many health problems and reproductive problems which limit its production efficiency. Among different diseases of buffalo, Respiratory Disease Complex (RDC) is the most prevalent and damaging diseases which adversely affects production Respiratory disease in calves causes great economic losses for the dairy and beef industry worldwide with a mortality rate in the range 1.5–4.2% (Ames, 1997) (Snowder *et al*., 2007).

RDC is a term used to describe severe respiratory disease and is sometimes referred to as shipping fever due to the increased risk of infection and transmission during transportation (Urban-Chmiel and Grooms, 2012). RDC is a multifactorial process, infectious agents including viruses, bacteria and mycoplasma. Viral pathogens that cause primarily respiratory lesions are Bovine and bubaline herpesvirus 1 (BoHV-1/BuHV-1), Bovine respiratory syncitial virus (BRSV) and bovine parainfluenza Virus type 3 (BPI3V) (Lazić *et al*., 2009; Pardon *et al.,* 2011). Unique among the bovine respiratory viral agents is bovine viral diarrhoea (BVDV), because intrauterine infection can lead in persistently infected (PI) cattle, chronically ill or dying in feedlots (Loneragan *et al.,* 2005; Kurćubić *et al.,* 2011). Infection with BVDV lead to immunosuppression which causing progression of BRDC because facilitate invasion with opportunistic secondary pathogens such as *Mannheimia haemolytica* (16 serotypes), *Pasteurella multocida*, *Haemophilus somni* and a number of mycoplasma species such as *M. bovis* and *M. dispar* (Ellis, 2001; Hodgson *et al.,* 2005; Fulton, 2009; Pardon *et al.,* 2011).

1. Different entities of RDC

RDC consists of at least three clinical entities, as well as several additional diseases which affect the respiratory tract secondarily or as part of a more generalized disease (Anon, 1968).

The three clinical entities are: 1) Enzootic pneumonia of calves 2) "Shipping fever" complex and 3) Atypical interstitial pneumonia.

1) Enzootic pneumonia of calves:

Enzootic pneumonia of calves is an entity from which numerous viral and bacterial agents have been isolated, including *Parainfluenza 3 virus* (PI-3), *Adenovirus, Chlamydia* agents, *Rhinovirus, Reovirus, Enterovirus, Herpesvirus, Mycoplasma* spp (Jubb *et al.,* 1970; López and Martinson, 2017; Baghezza *et al.,* 2021), and a variety of more conventional bacteria, usually *Pasteurella* spp. These diseases tend to occur in the first six months of life often in enclosed crowded conditions where ventilation and humidity are inadequate. The sequelae of consolidation, bronchiectasis, purulent or obstructive bronchiolitis, secondary bacterial infections, and lung abscessation appear to be common to many of these agents and predispose to ill thrift and further lung diseases in later life.

2) "Shipping fever" complex

The "shipping fever" complexes of diseases are the acute diseases of adult life in dairy, feedlot or cow-calf operations. The principal constituents of this group are infectious bovine rhinotracheitis (IBR) and pneumonic pasteurellosis. It is with this group of diseases subsequent discussion will be restricted to.

3) Atypical interstitial pneumonia.

The third group of diseases consists of the atypical or hypersensitivity pneumonias. This group of diseases has becoming more prevalent and a significant cause of death, particularly in beef cattle. Some forms of interstitial pneumonia, such as ‘fog fever’, ‘bovine farmer’s lung’ or hypersensitivity reactions against massive *Dictyocaulus viviparous* infections, have previously been classified as atypical interstitial pneumonia.

3. Viral pathogens involved in RDC:

Viral pathogens can cause primary infections, usually leading to mild clinical symptoms. The viruses which are most frequently associated with BRDC include infectious bovine rhinotracheitis virus (IBRV), bovine viral diarrhea virus (BVDV), bovine herpesvirus type 1 (BHV1), bovine respiratory syncytial virus (BRSV), and parainfluenza virus type 3 (PI3). Some viruses which may have role in respiratory infection but remain underestimated like bovine adenovirus (BAV) and bovine coronavirus (BCV). Moreover, several viruses are occasionally implicated by serological evidence but no consistent association was revealed like bovine calicivirus, bovine parvovirus, BHV4, bovine reovirus, bovine enterovirus, bovine rhinovirus, and malignant catarrhal fever virus.

a) Bovine Viral Diarrhea Virus: BVDV is a member of the genus *Flaviviridae*. It is a single-stranded negative-stranded RNA. According to the 5′UTR, Npro, and E2 gene sequences, BVDV can be divided into 2 genotypes: BVDV-1 and BVDV-2. Furthermore, BVDV-1 can be divided into 17 subtypes between 1a-1q, and BVDV-2 is divided into 4 subtypes between 2a-2d (Flores *et al.,* 2002; Xue *et al.,* 2010). In 2004, a third genotype named D32 / 00_HoBi was isolated in Brazil. After that, countries such as Canada, Mexico, and Australia have also isolated BVDV-3 (Gao *et al.,* 2016). Besides, BVDV can be divided into two biotypes according to their ability to cause cytopathic effects in cells, cytopathic and non-cytopathic (Nettleton and Entrican, 1995).

b) **Bovine Herpesvirus**-1: BoHV-1, a member of the subfamily *Alphaherpesvirinae*, can lead to upper respiratory tract disorders, conjunctivitis, genital disorders, and immune suppression (Jones and Chowdhury, 2007). BHV-1 can be divided into 3 subtypes: BHV-1.1, BHV-1.2a, and BHV-1.2b. Majority of the isolates from respiratory infections and abortion in cattle are BHV-1.1, while BHV-1.2a is mostly isolated from cattle with genital infections, and BHV-1.2b has no relationship with abortion (Muylkens *et al.,* 2007). As BHV-1 has caused huge economic losses to the cattle industry, many countries have launched its eradication programs.

c) **Bovine Parainfluenza virus type 3 (PI3):** It is an enveloped non-segmented negative-strand virus, belongs to the family *Paramyxoviridae*, genus *Respirovirus* (Fauquet *et al*., 2005). It is always associated with bovine respiratory disease (BRD), which can damage tissues and cause immune suppression and secondary bacterial infections (Haanes *et al*., 1997). Previously, the complete genome analysis of the representative BPIV3 isolates indicated two genotypes, BPIV3a and BPIV3b. SD0835 strain that was isolated in China in 2010 was classified as a new genotype: BPIV3c (Zhu *et al.,* 2011).

d) **Bovine Respiratory Syncytial Virus (BRSV):** BRSV is a non-segmented, negative-stranded, enveloped RNA virus that belongs to the genus *Pneumovirus* of the family *Paramyxoviridae* (Yunus *et al*., 2001). It shares common epidemiological, clinical, and pathological characteristics with the human respiratory syncytial virus (Valarcher *et al*., 2000). BRSV causes lower respiratory tract disease in cattle, which may be very severe, and sheep can also become infected (Masot *et al*., 2000). In addition, a BRSV outbreak is related to geographical region, seasons, temperature, age of the host, management of the pasture, and health conditions. A good knowledge of the BRDC epidemic situation could help us to take effective measures to prevent economic losses, such as strengthening feeding management, and the development of multiple vaccines.

4. Bacterial pathogens involved in RDC:

Immune suppression caused by viral infection is also an important cause of secondary bacterial infection. Studies have shown that viral infection can enhance the adhesion of bacteria to cells (Galdiero *et al.,* 2002; Solís-Calderón *et al.,* 2007; Grissett *et al.,* 2015). Secondary bacterial pneumonia is typically attributed to members of the family *Pasteurellaceae*, including *Mannheimia haemolytica* (formerly *Pasteurella haemolytica*), *Pasteurella multocida*, and *Haemophilus somnus*. Other bacteria that have been isolated from pneumonic lungs of cattle with BRD, with some frequency are mycoplasmas, especially *Ureaplasma diversum*, *Mycoplasma dispar*, *Mycoplasma bovis*, *Mycoplasma bovirhinis* and *Chlamydia* spp.

a) *Pasteurellaceae* family members : These are responsible for severe pneumonic damage characterized by pulmonary invasion of *Mannheimia haemolytica* and along with the production of virulence factors which are responsible for colonization of the lower respiratory tract (Confer *et al.,* 1990). It is responsible for lung injury characterized by vascular damage, excess fibrin effusion, and neutrophil infiltration results from the host's response to LPS produced by this Gram-negative organism (Whiteley *et al.,* 1991; Whiteley *et al.,* 1992). Other pathogens often associated with bacterial pneumonia include *Pasteurella multocida* and *Haemophilus somnus*. Like *M. haemolytica*, these are also normal inhabitants of the

upper respiratory tract of buffalo. *P. multocida* is considered to be less virulent than *M. haemolytica*, and in experimental challenge studies, more organisms are required to produce primary pneumonia (Ames *et al.,* 1985). The *P. multocida* is responsible for causing acute fibrinous bronchopneumonia along with chronic suppurative bronchopneumonia. The pneumonia attributed to *H. somnus* is more subacute or chronic than that caused by infection with either *M. haemolytica* or *P. multocida*. Lesions associated with *H. somnus* infection include necrotizing bronchiolitis and alveolitis. It is unclear if necrotizing airway lesions are due to colonization by this organism or a predisposing infection with respiratory viruses such as BRSV (Bryson *et al.,* 1990).

b) Mycoplasma and Ureaplasma Species

Mycoplasma mycoides subsp. mycoides (small colony type [SC]), *M. bovis, M. dispar* and *U. diversum*are associated with respiratory tract disease in buffalo (Rosendal, 1993). *M. mycoides* subsp. *mycoides* SC causes contagious bovine pleuropneumonia, an enzootic disease in cattle in Africa, Asia, and parts of Europe with major economic impact. *M. bovis, M. dispar* and *U. diversum* contribute to respiratory disease in housed calves and feedlot cattle. *M. bovirhinis* has been isolated from the lower respiratory tract of calves but evidence of a role in lung disease remains elusive (Otto *et al*., 1996).

M. dispar mainly affects tracheal epithelial cells ranging from ciliostasis to degenerative changes and death. Experimental infection of calves with *M. dispar* leads to decreased clearance of *Serratia marcescens* (Almeida and Rosenbusch, 1994), suggesting that *M. dispar* infection facilitates infection by other bacterial species. *M. bovis* infection, in contrast, has less effect on the function of ciliated epithelium but invades deeper into lung parenchyma, incites a stronger cellular response, and induces more lung damage.

Epidemiology and economic impact:

BRDC accounts for approximately 75 percent of feedlot morbidity and 50-70 percent of all feedlot mortality (Edwards, 2010; Loneragan *et al*., 2001). The percent of morbidity and mortality depends on the management system in place, prevention program and the kind of pathogens involved. For example, according to Duff and Galyean (2007), higher morbidity rates but fewer fatalities are typically observed when viral pathogens are primarily involved. In calves with bacterial infections only – there is sporadic morbidity, but higher mortality. The highest number of animals affected and higher mortality are observed in case of mixed viral and bacterial infections. Annual losses to the US cattle industry are estimated to approach US$1 billion, whereas preventative and treatment costs are over US$3 billion annually (Griffin, 2006; Snowder *et al*., 2007). The average cost of a single treatment was estimated at US$15.60. This cost is amplified to US$92.30 when indirect costs are also considered such as reduction in average daily gain (ADG) and feed efficiency, and decreased carcass value (Schneider *et al*., 2009). In calves, the overall prevalence rate of BRD is 2.07% (Joshi *et al.,* 2016). The highest prevalence was observed (11.58%) in agegroup of 0–1 months due to more susceptibility to infections as immune system is in developing phase. The highest prevalence (3.61%) was seen during winters in calves due to environmental stress and poor immunity in early partof life. Sex-wise prevalence in male calves is higher (3.08%) (Joshi *et al*., 2015).

5. Diagnosis

Diagnosis of RDC can be done by different methods.

1. **Classical methods**: These are based on physical examination and observation. Often observations are quantified in the form of a clinical score. The following clinical scoring system has been suggested by Perino and Apley (1998):

1. noticeable depression without apparent signs of weakness;
2. marked depression with moderate signs of weakness without significantly altered gait;
3. severe depression with signs of weakness such as significantly altered gait
4. moribund and unable to rise

According to this scoring scale, calves with a rectal temperature higher than 40^{o}C and clinical score ≥1 need therapeutic treatment.

2. **Laboratory methods:** The second diagnostic method is to use laboratory tests to identify microbial agents involved. Several samples can be taken to identify pathogens involved in BRDC. These include blood, nasal or nasopharyngeal swabs, tracheobronchial lavage and tissue samples at necropsy. A firm understanding of the disease pathogenesis is necessary to select the correct sample and interpret the results. There are many useful laboratory methods available for identifying both viral and bacterial pathogens including culture, immunohistochemistry (IHC), antigen capture ELISA, culture and PCR assays (Duff and Galyean, 2007; Thonur *et al*., 2012). One useful method for identification of calves with predisposition to BRDC after transportation is to identify changes in acute-phase proteins, including haptoglobin and fibrinogen. Changes in acute-phase proteins, such as haptoglobin, could be useful as a support in diagnostic of high risk calves after transportation (Arthington *et al*., 2003, Svensson *et al*., 2007). Molecular tests allow the assessment of development trends of microorganisms, a retrospective analysis of their geographical distribution and development of a database.

6. Prevention and control

Implementing RDC prophylaxis and control systems benefit producers by reducing economic loss. It also benefits consumers because of the reduced disease incidence which has both food safety and quality implications. As with any disease control program, BRDC control can best be accomplished by following two procedures:

(a) **Increase in disease resistance:**

- It can be done by integrated vaccination programs against both viral and bacterial pathogens involved in RDC.
- Arranging of sufficient nutrition program with good quality diet especially for calves exposed to stress conditions. The nutrition should be rich with energy, should contains high protein concentration and proper micro nutrient concentrations including minerals such as Zn, Cu, Fe and Se and vitamins E, B complex and C).
- Reducing stress brought on my management practices.

(b) **Decrease in exposure to RDC pathogens**: It can be done by

- Reducing the risk of infected animals being introduced into a herd.
- Bringing in only animals from uninfected herds.
- Bringing in only animals from herds with a known effective vaccination program.

- Avoiding the purchase of animals from sales barns.
- Testing new animals for persistent infections.
- Isolating new animals for 30 days before allowing contact with animals on-farm.
- Sanitation to reduce pathogen build-up.
- Isolation of sick animals.

7. Vaccination

Vaccination against important pathogens involved in BRDC is a useful tool to help reduce the risk of BRDC occurring. In the United States, vaccines against the viral pathogens IBR, BVD, PI-3, BRSV and the bacterial pathogens *Mannheimia haemolytica*, *Pasteurella multocida* and *Histophilus somnus* are readily available. They are available in different combinations and with respect to the viral pathogens, they can be found in both killed and attenuated forms. Research has demonstrated that appropriate use of these vaccines can reduce the risk of BRDC. Optimal vaccine response requires providing an efficacious vaccine to an immunocompetent animal. Immunity takes 1 to 3 weeks to develop, and may require multiple doses of vaccine to elicit protective immunity. Upon entering the feedlot, timing of the initial vaccination may vary if cattle experience transit time of more than 12 hours, it may be beneficial to allow 1 hour of rest for every 1 hour of transit before administering the vaccination and processing protocols (Edwards, 1996). A study conducted comparing on arrival versus delayed (14 days) vaccination with a multivalent modified-live virus (MLV) vaccine showed improvement in daily body weight gain at day 0 to14 and day 0 to 42 in the delayed procedure (Richeson *et al*., 2008). Ideally, calves entering the feedlot would be vaccinated prior to entry. This is best accomplished as part of a preconditioning program. These value added calf programs addresses vaccination and management strategies that provide the calf an opportunity to build immunity during a time when stress and disease challenge is minimal.

8. Future Perspectives

Increasing beef and dairy production in all part of the world will benefit from improved prevention and control of BRD. In North America, the cattle industry uses the largest amount of animal health products, consuming 37% of the global supply. The animal health market for cattle products is second only to companion animal, with over $1 billion U.S. attributed to vaccines (Wilkinson, 2009). The cost of all losses due to BRD approaches US$1 billion per year and the average treatment cost is US$15.00 per cattle (Schneider *et al*., 2009). Because of these significant economic losses, it is necessary to develop strategies which will reduce the incidence and impact of BRD both in dairy and feedlot cattle. In the future, logical changes will include the development of better management and nutrition technology, and improved genetics. According to McVey (2009) very important information will be learned by continuing to investigate the pathogenesis of BRD in cattle. Areas of research that will help to improve the prevention and control of BRD include:

i. Improved vaccines, vaccine formulation and administration strategies.
ii. Understanding the genetic basis of disease resistance.
iii. Understanding the role of inflammation in the pathogenesis of disease.

Most importantly, further enhancement of BRD control will be accomplished by:

a) Use of epidemiological concepts to manage BRD, especially as it relates to understanding the relationships between pathogens and predisposing factors.
b) Developing strategies to manage or reduce stress, especially associated with transportation, handling, feeding, and mixing of cattle.
c) Developing new antimicrobial therapies and therapeutic strategies that not only provide clinical cures but also minimize selection of resistant organisms.
d) Implementation of effective disease prevention strategies such as metaphylaxis and vaccination programs.
e) Development of diagnostic tools and methods necessary for early intervention.
f) Improving research to production system technical knowledge transfer.

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CHAPTER 9

Management of Fertility in Female Buffalo

R. K. Sharma*, S.K. Phulia, A.K. Balhara, Jerome A and MH Jan

ICAR-Central Institute for Research on Buffaloes, Hisar, Haryana
*Correspondence: Dr. RK Sharma (Principal Scientist); rksharmascientist@gmail.com

Fertility management is very important aspect in buffaloes as buffalo have long intercalving interval, longer age at first calving, silent oestrus, summer anoestrus, post-partum anoestrus and repeat breeding problem. Most of these problems are associated with poor nutrition and ignorance of oestrus signs. These problems lead to huge economic losses to the farmers and lengthen the interval between successive calvings, reduce the life time production and net calf crop and are responsible for high culling rate. The oestrus signs are not detected either due to less intensity of heat, due to short duration of heat, occurance of heat during night hours or ignorance of signs as farmers often compare them with cow. The oestrus signs in buffaloes are very week and assuming that buffalo is not in perfect heat, we do not present buffalo for breeding. Therefore, oestrus signs should be observed very minutely atleast thrice in a day particularly when buffalo is sitting for discharge and any suspicion should be looked into by a veterinarian. Buffalo come into heat mostly during night hours and may not be very prominent during hot weather, and may remain only for a short period (roughly 6–12 hours), making it difficult to observe. It is not necessary that all listed signs below will appear in a buffalo. It is also not necessary that signs appeared last time or in last lactation will be same all the time. Farmer always should suspect for signs of heat if any one sign appear and get them confirmed by a veterinarian.

The important heat signs in buffaloes are:

- Frequent bellowing
- Mounting on other animals
- Clear mucous discharge hanging from vagina while mounting or during rest
- Chin resting and rubbing
- Swollen red vulva
- Frequent urination
- Tail raising
- Muddy flanks and ruffled tail head
- Restlessness, sniffing behaviour
- Decreased milk production and off feed
- Temporary teat engorgement in buffaloes about 3 days prior to impending estrus

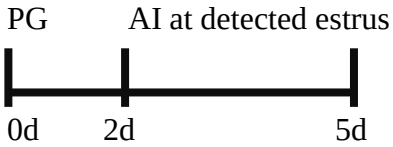
Different strategies may be employed for heat detection in buffaloes.

1. **Carefully observe heat signs:** As buffalo does not express all signs of heat with similar intensity, observe all signs very carefully. Bellowing, transparent mucus discharge hanging from vulva while sitting, and frequent urination are most commonly observed heat signs in majority of buffaloes.
2. ***Increase daily observations:*** Each buffalo should be oserved atleast thrice a day to take care of short duration of estrus. Observations should be made especially when buffalo is taking rest after taking food to see any clear vaginal discharge.

3. ***Every suspicious heat should be noted down on house calendar or note book:*** Whenever, buffalo is suspected for heat, it should be noted down on calendar and presented either to bull for natural service or to a veterinarian for AI and confirmation of estrus signs. If not mated, it should be carefully observed 21 days after suspected heat for recurrence of estrus signs on the expected date.
4. ***Non-pregnant buffalo must be checked by a Vet*:** If a buffalo does not come into heat even after 90 days of calving, it must be examined by a veterinarian for proper treatment. Buffaloes inseminated 50-60 days earlier should also be examined for pregnancy diagnosis. Buffaloes showing purulent discharge after calving should also be treated at the earliest.
5. **Provide balance diet and extra care during summer:** Balance diet is very important as poor diet firstly impair the reproductive function. While during summer months estrus symptoms are very weak, of shorter duration and expressed particularly at night. Provide ameliorative measures like wallowing tank, coolers, fogging system, tree shed, exhaust fan, and bathing etc during summer. Fresh drinking water should be available at all times. Avoid overcrowding. Buffalo should be kept indoor during day time or under tree while during night hours it should be allowed to rest in open paddocks. Green fodder availability should be ensured and it should be offered when environment temperature is low i.e. late evening and early morning.
6. **Hormonal treatment for anovulatory and silent oestrus conditions:** Anoestrus buffaloes should be suitably treated with hormones to induce the estrus at desired time. Following hormonal protocols may be used suitably

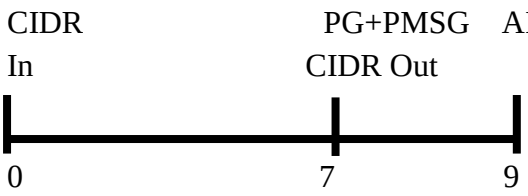
Prostaglandin (PG) injection for silent oestrus buffaloes

All animals having a detectable CL are injected one injection of PG and observed for estrus for five days. Most of the buffaloes are in heat 72h after PG injection. Animals found in heat are identified and inseminated.



Those animals not found in heat within 2-5 days are again injected one more injection of PG 11-14 days later and insemination is made again at observed estrus.

Progesterone implant (CIDR) for anovular buffaloes: In anovular buffaloes CIDR implant is inserted for 7-9 days and thereafter pulled out. An injection of PG as well as PMSG (400 IU) is also administered preferably at the time of implant removal. It is always good to squeeze the vagina after CIDR removal by back racking to clear the vagina for any pus type discharge at heat. Animals are found in heat within 48 of CIDR removal.



How to select oestrus synchronization protocol

There are several protocols available for oestrus induction and synchronization. It is very difficult to choose the protocol among different available protocol. However, it should be kept in mind that these protocols are not a replacement of poor heat detection system but help in reducing days open in those animals which are not detected in estrus at proper time. Veterinarian should ensure that animal is having minimum BCS of 3 and on balance diet. He should only choose protocol that works best in the given situation and it is simple with minimum use of hormone that is absolutely required to reduce the cost with minimum handling of animal and minimum visits.

First breeding in buffalo heifers:

Buffalo heifers express their first heat when they attain a body weight around 300 to 350 kg which is usually at an age of 24-30 months provided nutrition is proper. However, if heifers are not coming into heat even after attaining this weight may be checked by a veterinarian. To enhance the puberty at an early age interventions like balance diet, mineral mixture supplementation, feeding of green fodder, regular deworming and reducing heat stress during summer may be instituted.

First breeding in buffaloes after Calving:

In order to get ready for the next pregnancy, uterus return to its normal non-pregnant state by day 35 post-calving. By this time, nearly 40-50% buffaloes also show first heat if they are fed a balance diet. Despite providing balanced diet, nearly 20% buffaloes do not resume cyclicity by day 90 post-partum. First breeding of buffaloes should not be carried out before 50 days of calving. If the vaginal discharge in first heat is not good then one heat can be missed or some intrauterine treatment may be given. In a few animals the first heat cycle is short and next heat recurs within 7-10 days. In that case, repeat insemination should be carried out after examination by a veterinarian. However, if animal is not fed properly, the first heat delayed for >90 days resulting in long duration of anoestrus condition. In such cases buffalo should be examined by a veterinarian for suitable treatment.

Getting calf a year:

All efforts should be made to get a calf every year from females. For this calving interval is a very useful marker which is interval between two successive calving's. The ideal calving interval buffaloes is 400 days. This means that the buffalo must conceive by day 100 postpartum. If buffalo conceived by 200 days it will have next calving in 500 days and if conceived after 300 days it will have next calving in 600 days. The calving to conception interval is influenced by three factors: how soon after calving buffaloes come in heat, how accurately heat is identified and how readily they become pregnant following breeding. Early treatment of uterine infection, balanced diet, accurate estrus detection, comfortable housing and managemental factors are very important that has to be looked into.

Early diagnosis of pregnancy:

Early pregnancy diagnosis is very important so that non-pregnant animals can be detected and rebred at the earliest. Ultrasound scanning has emerged as a very powerful tool to diagnose pregnancy by day 30 post AI. The animals found nonpregnant on day 30, can be given prostaglandin injection to induce the estrus and subsequent rebreeding 3 days later.

Comfortable housing and goal setting:

Better management and comfortable housing is very essential for higher productive and reproductive performance. The shed should be designed in such a way that it protects the buffalo from direct sun rays, well ventilated and there is no overcrowding. Electric appliances viz ceiling fan, exhaust fan or cooler should be fixed in the shed to improve ventilation and decrease shed temperature. A wallowing tank may be constructed as buffalo enjoy wallowing in the pond. Practice of taking buffalo to nearby river or canal may be followed. Two to three times sprinkling of cold water also helps in lowering body temperature. During day time buffalo may be kept indoor or under a tree while during night hours it should be allowed to rest in the open paddocks. Feeding times can be arranged in such a way that buffalo consume fodder when environment temperature is low i.e. late evening and early morning.For improved reproductive performance of the herd, targets should be fixed and efforts should be made to achieve these targets. Any deviation while achieving these targets may be discussed with the veterinarian or subject experts.

S.No.	**Parameters**	**Targets**
1	Age at First heat	24-30 Months
2	Minimum body weight at first breeding	300-350 kg
3	Age at First Calving	<42 Months
4	Calving Interval	<14 Months
5	Voluntary Waiting Period for AI	45-60 days
6	Calving to Conception Interval (Service period)	<100 Days
7	AI per conception	<2
8	First Service Conception Rate	>50%
9	Minimum Dry Period	60 days

References: On request

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CHAPTER 10

Animal Cloning: Application and Status

Prem Singh Yadav* and D. Kumar

Animal Physiology and Reproduction Division
ICAR-Central Institute for Research on Buffaloes, Hisar, Haryana
*Corresponding author: Dr. PS Yadav (Principal Scientist); psycirb@gmail.com

Animal cloning is an asexual mean of reproduction, in which whole stretch of genetic material is identical between parent and offspring. With advantage of cloning, animals can be multiplied like primitive type of printing machine results in similar progenies with identical genetic value. Major scientific breakthrough in animal cloning was the birth of 'Dolly', first cloned animal produced by the somatic cell nuclear transfer (SCNT) technique. After this, many animal species have been produced through this technique. In this process, genetic material of donor cell (nucleus) is transferred into female gamete whose nucleus has been removed. The reconstructed egg containing foreign DNA is treated with chemicals or electric current in order to stimulate cell division. Once the cloned embryo reaches a suitable stage (morula or blastocyst), it is transferred to the uterus of a surrogate female where it continues to develop until birth and finally produce cloned animal. This approach is extensively used in farm, lab and wild animals as a tool for multiplication and conservation of genetic resource. In addition, SCNT is also an important method for studying various basic research investigations of cell biology and nuclear reprogramming of the mammalian genome.

Facts about the animal cloning

Several myths and negative perceptions are mostly based on the regretfully early death of the Dolly, the cloned sheep. Later, many clones were born that are normal, healthy and fertile, including buffalo clones. There are some facts about the animal cloning.

1. Cloning is an assisted reproductive technology (ART) like artificial insemination, embryo transfer, in vitro fertilization and sexed semen.
2. Cloning allows creating a genetic copy of an existing animal, which is in principle similar to an identical twin.
3. Aging is normal in clone animals (Study published by Scientists who created Dolly, https://www.nature.com/articles/ncomms12359)
4. Clones are not genetically engineered/modified animals.
5. Offspring's of clones that produce through conventional breeding techniques (AI or ET) are not clones.
6. In January 2008, the U.S. Food and Drug Administration (FDA) stated that clones and their offspring's products (milk and meat products) are safe for human consumption and it is not different from food products of natural or artificial bred animals (Risk assessment study conducted in worldwide)

7. As per FDA guidelines, products from animal clones will not require any kind of labelling. Therefore, it impossible for the EU to monitor the import of cloned animal products (Embryos, Semen).
8. Many countries, except the European Union, use animal cloning technology to improve the quality of animals for food production. Many commercial companies are producing and supplying cloned animals worldwide.

Status of the buffalo cloning in India

The first buffalo cloning was attempted in buffalo in the late 1990s by researchers of ICAR-National Dairy Research Institute (NDRI), Karnal (Singla et al. 1997). Cells from in vitro fertilized or in vivo produced embryos (called blastomeres) were injected in to enucleated oocytes to generate cloned embryos. The method used during that time was similar to Dolly's method having use of micromanipulator instruments but could not produce blastocyst-stage embryos. Later, in the early 2000s, NDRI researchers have acquired the economical, safe, and efficient handmade cloning (HMC) method. In 2009, the NDRI has created history in the field of animal cloning by producing the world's first cloned riverine buffalo calf, named "Samrupa." Over 25 buffaloes have been cloned using different donor cell types such as embryonic stem cells, urine-derived epithelial cells, semen-derived epithelial cells, etc. In 2015, researchers of the ICAR-Central Institute for Research on Buffaloes (CIRB), Hisar produced a clone of superior bull and CIRB becomes India's second institute that produced cloned buffalo. At present a total of 17 clones of superior breeding bulls are available at both CIRB and NDRI centres. Out of that, 13 clones are produced from 3 superior breeding bulls indicating multiple clones from a superior breeding bull (seven copies of a superior breeding bull M-29 (Dam's SLMY 4600 Kg) and three copies of bull no 4354 (Dam's SLMY 3605 kg). The buffalo clones growth, blood hematology, plasma biochemistries, and reproductive organs is found normal. The cloned buffalo bulls semen was evaluated using CASA (Computer Assisted Semen Analysis) variables and in vitro fertilizing ability of sperms and found similar to non-cloned bulls, including the donor bull (Selokar et al., 2019, Scientific Reports. 9(1):11366). Successfully cryopreserved semen of the cloned animals and some of the cryopreserved semen doses were used for artificial insemination in farm animals or farmer's animals on prior consent and a total of 62 progenies were produced and they are growing healthy and normal. Two male progenies had born using semen of cloned bull available at CIRB, Hisar which now donating semen and qualifying minimum standard for semen cryopreservation. Three female progenies born using semen of cloned bull semen are now pregnant, indicating the normalcy of the cloned animals.

Problems associated with animal cloning

There is no doubt that AC is a powerful tool providing many opportunities for farm animal production, conservation of endangered breeds / species, and human medicine. However, AC is suffered from several challenges such as (1) poor success rates (only 5 –10% of embryos transferred into recipient animals could result to healthy surviving clones, whereas in other ARTs (AI and IVF), 30–40% results following embryos transferred / semen insemination), (2) abnormal placental development, (3) loss of clones throughout gestation, at time of

calving, and post-natal period (generally within 6–8 months of age), (4) prolonged gestation period in cloned pregnancies, and the birth weight of clones may be 20–30% more than normal, (5) born clones require special care and management since they may succumb to infection and stress (Keefer 2015). Extensive research on genomic and epigenomic showed that these abnormalities are probably due to incorrect /improper genetic and epigenetic reprogramming of a differentiated donor cell by a recipient oocyte that leads to abnormal gene expression throughout the development of clones (embryo to calf). Not all clones are abnormal; studies showed that once clones mature, they grow and reproduce similar to non-cloned animals.

Prospective of animal cloning

Commercialization of the cloning technology

Cloning technology is already being used commercially in the livestock industry in different parts of the world for the replication of elite breeding animals (Table given below). It has been widely reported in the media that products from the offspring of cloned animals have already allowed in the human food chain in the United States and elsewhere (Weiss, 2008; Plume, 2009). Following the decision by the US Food and Drug Administration (FDA, 2008) that products from cloned animals are safe, food from clones and their offspring can freely enter the marketplace in the US and there is no requirement for these products to be labeled. A number of companies in the US offer cloning services to the livestock breeding industry, primarily for cattle and also for pigs (ViaGen, 2009; Trans Ova Genetics, 2009; Cyagra, 2009).

List of companies engaged in animal cloning worldwide

Sr. No.	Company Name	Country	Animals
1	Trans Ova Genetics	USA	Bovine, Sheep, Goat, Pig
2.	Tianzin Animal cloning Centre	China	Cattle, Dog, Horses, Endangered & extinct species
3.	Advanced cell technology	USA	Holstein Cattle
4.	Bio Sidus	Argentina	Pampa, Cattle
5	Boyalife	Chinese	Cattle, Dogs
6.	CRC for innovative dairy products	Australia	Cattle, bulls
7.	ViaGen	USA	Dog, Cat, Equine
8.	Sinogene	USA	Dog, Cat, Horse
9.	Sinogene	Beizing, China	Dog
10.	Sooam Biotech	South Korea	Dog
11.	Bio Art International	USA	Dog
12.	Genetic savings and clone	USA	Cat
13.	Camel Reproductuve centre	Dubai	Camel
14.	Geneticas	CA, USA	Cat

Examples from the Asia are not so many but it is likely that products from the offspring of cloned animals will enter the food chain in at least some Asian countries. In 2002, calves cloned from an elite Holstein dairy bull were sold to China by Australian-based Company, Clone International (BBC, 2002). Cloning of livestock is also being undertaken within China by Yangling Keyuan Cloning (People's Daily, 2001). While Bovance (2009) considers that "cloning will remain a technology suited exclusively for the most elite tier of genetics, and cloned individuals will represent only a fraction of a 25 percentage of tomorrow's cattle breeding foundation", some authors have suggested that there will be a transition from the commercial use of semen and offspring of clones to the production of food products from cloned animals themselves over the next few years (Suk et al, 2007).

Application of animal cloning

Rapid multiplication of desired livestock

Cloning could enable the rapid dissemination of superior genotypes from nucleus breeding flocks and herds, directly to commercial farmers. Genotypes could be provided that are ideally suited for specific product characteristics, disease resistance, or environmental conditions. Cloning could be extremely useful in multiplying outstanding F1 crossbred animals, or composite breeds, to maximize the benefits of both heterosis and potential uniformity within the clonal family. These genetic gains could be achieved through the controlled release of selected lines of elite live animals or cloned embryos. More appropriately, given that cloning is not particularly efficient at present; a niche opportunity exists in the production of small numbers of cloned animals with superior genetics for breeding. These could be clones of performance tested animals, especially sires. This would be particularly relevant in the sheep and beef industries, where cloned sires could be used in widespread natural mating to provide an effective means of disseminating their superior genetics. This could be used as a substitute for artificial insemination, which in these more extensive industries is often expensive and inconvenient.

Animal conservation

Cloning can be used along with other forms of assisted reproduction to help preserve indigenous breeds of livestock, which have production traits and adaptability to local environments that should not be lost from the global gene pool. In some situations, inter-species NT and embryo transfer may be used to aid the conservation of some exotic species. At the very least, it is appropriate to consider the cryopreservation of somatic cells from these endangered animals as insurance against further losses in diversity.

Research models

Sets of cloned livestock animals could be effectively used to reduce genetic variability and reduce the numbers of animals needed for some experimental studies. This could be conducted on a larger scale than is currently possible with naturally occurring genetically identical twins. Lambs cloned from sheep selected either for resistance or susceptibility to

nematode worms will be useful in studies aimed at discovering novel genes and regulatory pathways in immunology.

Human cell-based therapies

There are also direct applications of nuclear transfer technology in human medicine; principally therapeutic cloning as opposed to human reproductive cloning. Patients with particular diseases or disorders in tissues that neither repair nor replace themselves effectively (as occurs, for example, in insulin-dependent diabetes, muscular dystrophy, spinal cord injury, certain cancers and various neurological disorders, including Parkinson's disease) could potentially generate their own immunologically compatible cells for transplantation, which would offer lifelong treatment without tissue rejection. Initially, this approach could employ human nuclear transfer and embryonic stem cells, the use of which is controversial. In the longer term, however, fundamental understanding of reprogramming will enable one cell type to be directly trans-differentiated into another cell type specifically required for cell-based therapy.

Cloning for transgenic applications

A significant application of nuclear transfer is to clone animals from cells that have been genetically modified in order to produce transgenic livestock. Even acknowledging the current problems with nuclear transfer, the cloning route is more efficient than conventional pronuclear injection of DNA, where typically less than 1% of injected zygotes develop into transgenic animals.

References: On request

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CHAPTER 11

Bull Fertility and Related Technological Interventions for Improving Productivity and Profitability of Buffalo Husbandry

Shivanagouda Patil, Arsha Shaji and Arumugam Kumaresan*

Theriogenology Laboratory, Veterinary Gynaecology and Obstetrics, Southern Regional Station of ICAR - National Dairy Research Institute, Bengaluru, Karnataka
*Correspondence: Dr. A Kumaresan (Pr. Scientist, ICAR-NDRI);
A.Kumaresan@icar.gov.in, ogkumaresan@gmail.com

Reproduction is a "luxurious process" and it demands high energy (in the form of glucose, protein, and enough fat), suitable climatic conditions, and disease-free body irrespective of the species that support the birth of a young one followed by its survival. As animal productivity generally follows reproduction, dairy animal reproduction is highly important and it is influenced by several factors; genetic, nutritional, hormonal, physio-pathological conditions, and management practices. Maintenance of male animals for breeding purposes is not economical for a farmer and invariably they are dependent on Artificial insemination (AI) in which semen from high genetic merit bulls with good production records is used. Here, the bulls have more importance than females because one low-fertile or sub-fertile bull can render thousands of breedable females infertile and make them repeat breeder or increase the number of inseminations per conception thus affecting both production and economy of production. Therefore, male-related subfertility or infertility will impair the reproduction ability of counterpart females by increasing the calving to conception period and extending the calving interval, which risks the aim of obtaining a calf per cow a year.

The motto of getting a calf per cow per year in dairy animals will be difficult without assisted reproductive technologies (ARTs). The development of rigorous bull selection protocols with promising tests/fertility markers would be helpful in yielding high-quality semen and on the other hand effective bull management is mandated. Though the society of theriogenology has given the standards for the selection of breeding bulls, buffaloes don't have a well-established breeding bull scoring system and are being selected majorly based on the dam's lactation yield (Table 1) and few sperm phenotypic parameters (Table 2).

Table 1: Dam's lactation yield for selection of breeding bull

Breed	Dam's Lactation Yield (Kg)		
	First	Best	Fat %
Murrah	2400	3000	7.0
Mehsana	2400	3000	7.0
Nili Ravi	2400	3000	7.0
Jaffrabadi	2800	3500	8.0

Banni	2400	3000	7.0
Surti	1600	2000	7.0
Bhadawari	1300	1600	8.0
Pandharpuri	1300	1600	7.0

Table 2: Minimum quality control parameters for frozen semen

Sr. No.	**QC Parameters**	**Cut-off Values**
1	Bacterial load/Colony forming units (CFUs)	<5000 per ml of processed semen
2	Hypo osmotic swelling test (HOST)	≥ 40%
3	Incubation Test	standard drop in motility by 10% after every 30 minutes
4	Acrosome Integrity	≥ 70%
5	Percent Intact Acrosome	≥ 65 %
6	Sperm Concentration	20 million spermatozoa per dose (0.25 ml Mini straw)

It has been more than centuries, and researchers are still working to find out tests or combinations of tests to arrive at promising bull fertility markers which can aid in the bull selection and utilization of bulls in breeding programs (Saacke et al., 1994). Previously semen quality was assessed only by direct assessments (volume, colour, consistency, density, and pH), and gradually that got advanced towards the microscopic evaluation where they started assessing motility, morphology, viability, abnormalities, etc. Regardless of the evaluation type utilized, outcomes are usually influenced by subjectivity and have a lower degree of repeatability because of the evaluator's abilities. As the research, advanced, high throughput data generation with more specificity and sensitivity is possible with flow cytometry in the present days; now it is possible to assess several thousands of sperm in a few seconds for a variety of molecular/structure-related studies (Elango et al., 2022).

Buffalo bulls and their semen

India is leading in buffalo population with 109.85 million buffaloes contributing 49% of the total Indian milk production (DAHD annual report, 2020-21). Buffalo bulls require shorter AV so that the ejaculate is properly collected because their penis is thinner and shorter than those of cattle bulls. Buffalo semen composition is different from cattle semen in terms of chemical and biochemical characteristics as well as the morphology of sperm. Buffalo semen has low buffering capacity and less electrolyte tolerance. Significantly decreased fructose content and fructolytic index values are recorded in buffalo semen. Buffalo semen has lower levels of ascorbic acid, citric acid, and sialic acid but contains

greater levels of calcium, inorganic phosphorus, acid-soluble phosphorus, and total phosphorus. The scrotal circumference is less, and the volume of ejaculate and sperm concentration is also less in buffalo bulls. In addition, the buffalo bull's ejaculatory thrust, mount, and service conduct differ from cattle bulls. Buffalo bulls are sensitive to thermal stress, high humidity, high ambient temperature, and strong radiation effect in the summer thus altering the scrotal thermoregulation mechanism and affecting the semen quality and sex drive. The ejaculate rejection rate is higher from June to August. Due to low post-thaw motility, a high sperm abnormality count, and a significant number of primary abnormalities indicative of disruptions in spermatogenesis, the majority of the frozen semen doses are rejected at this time (Bhosrekar, 2014). Inadequate sex drive and poor semen quality were the main contributing factors for bull disposal in cattle whereas poor semen freezability was most frequently observed in buffalo bulls (Bhosrekar, 2014).

Scrotal circumference is highly correlated with the semen production potential of a bull. For mature (>600 Kg BW) Murrah buffalo bulls (n=86), mean SC values were found to be 35.23 cm, with an S.D. of 3.00. It is concluded that that SC had a positive relationship with initial semen quality, but a combination of SC, TL, TW, TV, and TM may yield superior results for standardizing the breeding soundness evaluation protocol for the selection of crossbred bulls (Vijetha et al., 2014). Under the field progeny testing program semen of test bulls are used for artificial insemination in the field, followed by pregnancy diagnosis, calving records, and follow-up of progenies until the completion of the first lactation for milk records on the basis of monthly test day recording. Then the value of the bull is evaluated.

Notable factors that affect buffalo semen quality and quantity

Nutrition

For successful reproduction, nutrition is very much crucial. Bulls typically do not receive the proper attention when it comes to feeding, which delays puberty and the collecting of the first sperm. Balanced nutrition during pre-weaning and post-weaning stages of male calves has a significant impact on the hypothalamus in turn testicular steroidogenesis and gonadotropins releasing hormone. A low plane of nutrition will delay the puberty. Low protein feeding may delay the puberty of bulls by 3-5 months followed by poor testicular development and less ejaculate volume. The nutritional status of a bull is one of the main aspects, which determine the semen quantity and quality of semen. Moreover, undernutrition, malnutrition, or nutritional imbalance leads to reduced androgen secretion and poor semen quality in adult bulls. Balanced nutrition during pre-weaning and post-weaning stages of male calves has a significant impact on testicular steroidogenesis as it affects the Leydig cell functions.

Energy and protein

The amount of energy in a feed indirectly affects testicular activity. Additional dietary energy has been shown to accelerate the beginning of puberty in male calves, particularly through improved testicular function as indicated by elevated levels of serum testosterone, testicular testosterone, Leydig cell size, and sperm generation (Nolan et al., 1990).

Prepubertal development is accelerated by dietary energy up to a certain amount. Bulls don't need extra nutrition as compared to producing female buffaloes; e.g. 3% CP and 40% TDN + 17% oil cake + 25% molasses mixed with 4% bran, 1% salt, 2% mineral mixture, and 1% urea is maintenance ration for adult animals.

Puberty and sexual maturity

Puberty in male buffalos could be defined as the first ejaculation with >10% sperm motility and the onset of puberty is observed at the mean age of 25 months (Korejo et al., 2018). The onset of puberty is earlier than sexual maturity where it depends on the attainment of 2/3rd of the adult body weight at a particular age. Laterally development of testicles along with increasing scrotal circumference is a good indication. According to the society of theriogenology bull should have a scrotal circumference of 30 cm at <1.25 years, 31 cm at >1.25 to 1.5 years, 32 cm at >1.5 to 1.75 years, 33 cm at >1.75 to 2.0 years and 34 cm at >2 years as an optimum indicator of good breeding bull. Bull nutrition should be aimed to attain desired body weight and scrotal circumference at the earliest possible age. Mandal et al. (2015) found that 484 Frieswal bull calves attained an average daily gain (ADG) of 512.82 g/d in the growth phase of 13-18 months and showed appropriate sexual behaviors and had normal semen quality. Sperm ejaculates from sexually mature males should have at least 50 million spermatozoa and at least 10% motile sperm (Wolf et al., 1965). Further, sexual interest and mounting were observed at the mean age of 19.1 ± 0.72 and 21.1 ± 0.89 months respectively in buffalo bulls.

Low energy and protein intake result into delayed onset of puberty (Soliman, 2014), poor testicular development (Tegegne et al., 1994), decreased thickness and diameter of seminiferous tubules (Barth et al., 2008) impaired sperm production (VanDemark and Mauger, 1964) and reduced fertility (Lindsay et al., 1984). Regardless of nutrition during the peripubertal period, it has been reported that restricted feeding during calf hood causes impaired sexual development because of negative effects on the hypothalamus-pituitary-gonadal axis, such as inhibition of the hypothalamic GnRH pulse generator that reduced pituitary response to GnRH and altered or suppressed release of gonadotrophins. Higher mass motility and progressive motility of spermatozoa, increased semen volume and sperm concentration has been reported following supplementation of high energy/protein diet (Perkovic et al., 2001). Feeding 1.5 kg/day of concentrate to Zebu bulls in a sub-humid environment for 50 weeks reduced the aberrant sperm count (11.40% Vs 8.3%) both in comparison to the control group and within the same group after supplementation (Kebede et al., 2007). Maintenance of bull may cost is given below (Table 3) (Bisla et al., 2018).

Table 3: Cost of Rearing of male calf up to breeding age

Particulars	**Parameters**
Purchase cost of 6-month-old calf	Rs 10,000/- per calf (at prescribed weight gain)
Feeding cost	Rs 50/- calf/day

Labour cost	Rs 10/- calf per day
Medicines	Rs 300/- per calf per year
Utilities	Rs 5/- per calf per day
Salaries	1 supervisor; Rs 10,000/- month per supervisor

Effect of vitamins and minerals

Minerals constitute about 2 to 4 % of the animal tissue and play a vital role in the nutrition of buffaloes. Among these, Calcium, phosphorus, sodium, potassium, magnesium, sulphur, and chloride are required in fairly larger quantities as they are macro-minerals. However, copper, zinc, cobalt, manganese, iodine, and iron are required in less quantity as they are trace minerals. Minerals are essential for bone development, body ionic equilibrium, vital enzyme systems, growth, milk production, and reproduction in buffaloes; hence, they must be present in enough amounts in the diet. Any mineral imbalance or excess in the diet is also detrimental.

For healthy body metabolism, development, milk production, and reproduction, vitamins are necessary. Vitamins C, K, D, and B complex are examples of those that may be produced by the body or by rumen bacteria in buffaloes, whilst others must be obtained through food (Vitamin A and E). Inhibition of spermatogenesis, a decrease in testicular size, and a decrease in testicular steroidogenesis are all typical signs of vitamin A insufficiency in male rats (Ganguly et al., 1980). Depending on how severe the insufficiency is, vitamin A deficiency in men leads to testicular germinal epithelium degradation, which either slows down or stops spermatogenesis. Bulls which had vitamin A deficient feed showed delayed puberty, reduced libido and reduced spermatogenesis. Vitamin-E deficiency reported to have deleterious effect on germ cell proliferation. Male rats showed degeneration of the germinal epithelium in vitamin E deficiency (Scott, 1978) and Se deficiency results in an inhibition of spermatogenesis (Wu et al., 1973). Area-specific mineral mixtures fortified with vitamins based on a geographical survey of minerals would be helpful.

Supplementation of fermented yeast culture (FYC) 12 g/animal/day during the growing period and 24g/animal/day during the prepubertal period. The supplemented Murrah male calves attained puberty by 25 months of age, whereas the control animals attained puberty by 27 months of age (Sehgal et al., 2016)

Summer Management

Temperature humidity index >72 will affect the quality of semen (Armstrong, 1994). As buffaloes are highly susceptible to heat stress supportive management practices like feeding Niacin @ 6 gms/day/animal, yeast at 10 gms/day/animal, and mustard oil at 150 gms/day/animal; are reported to enhance milk production of lactating buffaloes by reducing thermal stress. Further, provision of cold drinking water ad. lib., ceiling fans, fogger, molasses, partially mixed wet feed but avoid > 50% moisture in the total ration have been shown to reduce the effect of heat stress. Succulent fodder and increased frequency of

feeding, feeding the majority part of the ration in the early morning; 6-8 AM and night; 8-10 PM would be helpful. Infrared thermography (IRT) is being successfully used as a non-invasive tool for monitoring scrotal temperature in breeding bulls (Silva et al., 2016).

Cryopreservation

Buffalo semen is reported to be a poor cryotolerant compare to cattle bulls, but species- specific extenders have improved post thaw motility. Glycerolization at room temperature at the initial stage of semen dilution decreases or abolishes backward motility of post thaw buffalo sperm (Kumar, 2020). Cryopreservation media like low density lipoprotein (LDL) extender found best suitable for buffalo semen, followed by liposome based and egg yolk-based medias (Patil et al., 2020). The presence of high-density lipoproteins, calcium, and steroids hormones in extenders affect the sperm quality. The issue of cryocapacitation and oxidative damage during the cryopreservation process can be mitigated by deoxygenation of buffalo semen by Oxyrase to enhance post-thaw sperm quality (Dalal et al., 2020). BUFFASOL – a novel buffalo semen extender (tris-citrate-fructose-glycerol-egg-yolk based extender) developed by NIANP, Bengaluru, is found to improve progressive motility of the cryopreserved buffalo sperm. Semen additives at various concentrations are found to improve the post thaw quality of sperm but no semen station is following fortification of extenders with additives. Inyawilert et al. (2021) concluded that, the addition of melatonin at 1 mM to the semen extender exert the better protection against sperm damage in swamp buffalo bulls during cryopreservation.

Sperm dose with respect to the fertility of bull

Recent studies reported that buffalo semen dilution from 20 million sperm per 0.25 ml straw to 16 million, 12 million and 2 million found that field fertility did not vary significantly up to the sperm concentration of 12 million, however large-scale insemination is required to proceed further (Patil et al., 2020). The sperm load in semen dosages has to be adjustable to allow lower loads for bulls with high genetic merit and demonstrated high fertility and greater loads for bulls with relatively lower fertility. This would optimise the use of bulls with high genetic quality and provide better benefits for dairy farmers. The conception rates of 60.34% (20 million/straw), 61.97% (16 million /straw) and 60.12% (12 million /straw) was obtained under field conditions (Singh et al., 2020).

Breed and bull Variation

It is generally known that breeds differ significantly in how they react to frozen semen processing procedures, as well as within breeds, and that the uniform processing strategy for semen processing results in a significant amount of rejection in post-thaw discards. The equipment that is now available does not allow for breed- or bull-level modification of setup with short lead times. Additionally, no equipment suppliers cater to the unique requirements of various semen stations based on their scale of operation. Because of this, each and every semen station must ultimately incur significant capital expenditures for the equipment, and most semen stations continue to have capacity utilization issues with their built-up infrastructure.

Advanced technologies for bull sperm quality assessment and improvement

Flow cytometry

Flow cytometry is a technology that measures the physical and or chemical properties of cells while moving in a fluid stream through a laser which works by the principle of hydrodynamic focusing. By using, flow cytometry it is possible to analyze a large number of spermatozoa in a short time and it has higher repeatability than other routine traditional quality control tests (Kumaresan, 2014). It can gather a precise understanding of sperm physiology by assessing a variety of sperm parameters that can be rapidly measured on a cell-by-cell basis such as sperm count, viability (CFDA and Propidium iodide), plasma membrane integrity (SYBR 14 or CFDA along with Propidium iodide), acrosomal integrity (FITC-PSA or FITC-PNA), membrane scrambling (Merocyanine 540) mitochondrial membrane potential (JC-1 or JC-9), apoptosis (Annexin V), intracellular Ca level (Fluo 4), reactive oxygen species (BODIPY, MitoSOX) and DNA integrity [Chromomycin A3 (CMA3)], etc. These peculiarities make flow cytometry an aid for quality control tests for spermatozoa which may be able to predict the functionality of the spermatozoa so as to speculate fertilizing ability more accurately.

Nano purification of semen: picking a superior sperm population

Although enrichment or removal of a specific subpopulation to increase fertility has been tried little, several studies have also shown that there are subpopulations in an ejaculate, and some subpopulations are superior in terms of quality and fertilizing capacity. Sperm with inferior quality removal is the main aim and the technique of cell separation using nanoparticles-based magnetic activation gained momentum in recent years. The idea is to coat the iron nanoparticles with the right antibodies or lectins so that desired or undesirable cell populations are selectively captured and eliminated (Bisla et al., 2020; Paul et al., 2022). High-quality spermatozoa concentration may be enhanced using this method, according to several research.

Possible markers reported through various techniques

Talluri et al. (2022) found that low-fertility bulls had considerably lower levels of metabolites related to the metabolism of taurine and hypotaurine, suggesting that molecules regulating sperm metabolism potentially have an impact on bull fertility. Several areas are being explored reportedly random amplified polymorphic DNA (RAPDs), microsatellites, and traits governing specific genes such as growth hormone, seminal plasma protein gene-specific primer-based buffalo genome characterization for bull performance. Numerous studies have been conducted on the sexual behaviour of Murrah buffalo bulls and how peripheral hormones (kisspeptin and testosterone) interact with it (Bhardwaj et al., 2022). CYP11B2 was highly abundant in high-fertile spermatozoa and MT1A was highly abundant in low-fertile spermatozoa (Aslam et al., 2019). Though a number of studies are available, there are no reliable markers, which are used on a commercial/broad scale bases in routine practice.

Omics in animal reproduction: Era of biomarkers

Routine semen analysis only indicates the phenomes of sperm but not the molecular health of the sperm, which is highly important for a sperm to fertilize the oocyte and establish the pregnancy. Sperm proteins like inhibition of ODF2 adversely affect sperm motility, and downregulation of ATP5A1 resulted in reduced sperm motility (Huang et al., 2015) metabolites like taurin, tetradecanoyl-CoA were found in subfertile bulls (DasGupta et al., 2021) and transcripts are also found to be relatively promising markers as they are involved at the molecular level. Sperm transcripts code for the functions starting from the testicles (spermatogenesis) to early embryo development in the uterus. Paul et al. (2021) reported that buffalo sperm transcript ORAI 3 gene was higher and the expression of YBX1 and TFAP2C genes were lower in low-fertile bulls when compared to high-fertile bulls. There are possibilities for the identification of promising markers for bull sperm health and its fertility potential by exploring more in this area. Genomic selection has created substantial changes in the dairy cow market as accurate breeding values, which double the reliability of the pedigree index, may be collected considerably earlier in an animal's life. Therefore, compared to genetic gains gained using conventional methods, genetic gains from correctly planned genomic programmes are significantly higher. Within weeks of birth, long before they reach puberty or are even capable of producing spermatozoa, genomic selection has made it possible to identify potential elite sires (Kenny and Byrne, 2018). Characterization of the SPAG11 gene may serve as a base for future research to explore the association of the SNPs in the SPAG11 gene with buffalo reproduction and fitness traits, which could be an addition to the existing knowledge for better estimation of genetic breeding values in the genomic selection (Deshmukh et al., 2022). For the purpose of genotyping cattle and buffalo, respectively, NDDB has created the INDUSCHIP and BUFFCHIP genotyping chips. These chips have been used to genotype more than 20,000 animals. Genomic breeding values, a measure of an animal's genetic potential, are computed using both genotype information and performance records. Gir, HF crossbred, Jersey and Murrah breed bulls undergo genetic selection before distribution to semen stations to improve the breeding standards of the animals and thereby realize a faster gain and growth rate in milk production of the country.

Sperm oviduct epithelium binding assay

Although it is a well-known fact that bulls that have crossed BSC are varying by 20-25 % in their fertility potential, these *In vitro* oviduct explants models could be great in assessing the bull semen quality *in vitro*. Nag et al. (2022) reported that the membrane integrity, acrosome intactness, and mitochondrial membrane potential of spermatozoa from bulls with low DFI% were considerably greater ($P<0.05$). Further, the binding index is reported to be positively correlated with conception rate ($r = .703$), intact sperm membrane ($r = .631$) and mitochondrial membrane potential ($r = .609$). It is concluded that breeding bulls with high sperm DFI% have impaired sperm morphological traits and oviduct binding capacity, which may explain why these bulls have poor conception rates.

Epilogue

Although it is not wise to compare buffalos with cattle, often, when talking about reproduction, buffaloes are compared with cattle and are said to have comparatively higher sub-fertility problems. Both male and female contribute to infertility, however, the role of males is amplified because one bull is used to artificially bred several thousands of females. Buffalo spermatozoa is unique in terms of structural, compositional and functional attributes, and thus the fertility prediction methods/tools used in cattle may not hold good for this species. During the last one or two decades, there have been a quantum of information generated on buffalo bull reproductive physiology and fertility. Incorporation of the promising findings in buffalo bull selection and production could help in expanding the potential benefits of buffalo rearing to the unreached population.

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CHAPTER 12

Scope and Utilization of Buffalo Milk

Kaushik Khamrui* and Writdhama Prasad
Dairy Technology Division
ICAR-National Dairy Research Institute, Karnal, Haryana
*Corresponding author: Dr. Kaushik Khamrui (Principal Scientist); kkhamrui@gmail.com

Buffalo rearing has been an integral component of agriculture in several regions of the world. Buffalo milk contributes a major portion of the total milk production in several countries of Asia, Africa, Europe and Latin America. In tropical and subtropical regions of the world, water buffaloes contribute significantly towards milk production, because of their ability to thrive under hot and humid climatic conditions. In terms of the percentage of world milk production from buffaloes, India ranks first (69.69%), followed by Pakistan (23.91%), China (3%), Egypt (2.56%) and Nepal (1.67%). Differences in various compositional and functional properties render buffalo milk eminently suitable for the manufacture of dairy products *viz*., paneer, *khoa*, dairy whiteners, UHT cream, ice cream mix powder, edible casein and caseinates. Again, due to several species related biochemical differences between buffalo and cow milk it is often considered not ideal for the manufacture of several types of cheeses, milk powders, evaporated and condensed milk and infant formulae. It should be realized that buffalo milk is distinct in its physicochemical and processing characteristics and deserve a position of importance by virtue of its unique processability and utility for producing many better quality milk products. It is often falsely presumed that technological information generated on cow milk processing can be directly extrapolated for buffaloes. Over the years, a wealth of information has been generated about the chemical, functional characteristics and processing technology of buffalo milk. This paper delineates the uniqueness and scope of buffalo milk from a techno-functional point of view.

Composition of Buffalo Milk

Richness and creaminess are two characteristics sensory attributes of buffalo milk. It has higher content of fat, proteins, lactose and minerals as compared to cow milk. In general, buffalo milk fat content varies between 7.20-12.60%, proteins between 3.60-6.04%, lactose between 3.70-5.48% and minerals between 0.71-0.86%. A comparative summary of the gross chemical composition of buffalo milk, milk from Indian cow (*Bos indicus*) and Western cow (*Bos taurus*) is delineated in Table 1. Concentration of protein constituents in buffalo and cow milk is presented in Table 2.

Constituents	**Concentration in g/100 ml of milk**			
	Indian buffalo [*Bubalus bubalis*]	Egyptian Buffalo [*Bubalus bubalis*]	Zebu [*Bos indicus*]	Western cattle [*Bos taurus*]
Water	83.18	82.75	85.28	87.2
Total solids	16.32	17.25	13.82	12.8
Solid-Not-Fat	10.01	10.11	9.11	9.1
Fat	6.71	7.14	4.64	3.7
Protein	4.52	4.33	3.3	3.5
Lactose	4.45	4.99	4.44	4.9
Total ash	0.8	0.79	0.73	0.7

Table 1. Chemical composition of buffalo milk and cow milk

Polypeptide	Concentration in g/100 ml of milk		
	Buffalo [*Bubalus bubalis*]	Zebu [*Bos indicus*]	Western cattle [*Bos taurus*]
α_{s1}-Casein	1.44-1.8	1.04-1.3	1.2-1.5
α_{s2}-casein	0.22-0.28	0.26-0.34	0.3-0.4
β-Casein	1.26-1.58	0.94	0.9-1.1
γ-casein	0.16	0.15	0.1-0.2
k-casein	0.43-0.54	0.3-0.37	0.3-0.4
β-lactoglobulins	0.39	0.31-0.38	0.2-0.4
α-lactalbumins	0.14	0.01-0.1	0.1-0.15
Protease peptone	0.33	0.24	0.6-1.8
Serum albumin	0.029	~	0.1-0.4
Lactoferrin	0.032	0.005	~

Sahai (1999)

Table 2. Concentration of protein constituents in buffalo and cow milk

Advantages of Buffalo Milk in terms of its Processing Characteristics

The typical chemical and functional attributes of buffalo milk which render it unique for processing is described under:

Buffalo milk protein is A2 type hence considered to be more healthful and beneficial than A1 type. Buffalo milk contains higher total solids as compared to cow milk, which provides higher yields of dairy products *e.g.*, milk powders, *paneer*, *khoa* etc. Due to higher fat content gives better yields of cream, butter and *ghee*. Higher protein concentrations enhance

the yields of casein and caseinates. Larger average size of fat globules in buffalo milk, results in better separation of cream and easier churning of butter resulting in more yield and lesser fat loss in skim milk and butter milk during manufacture of these products as compared to cow milk. Higher fat content coupled with better emulsifying capacity of buffalo milk due to a higher proportion (50%) of butyric acid containing triglycerides via-a-vis only 37% in cow milk. This results in better body and texture attributes in *khoa* and sweets made therefrom and superior absorption of fat in the human body. Buffalo milk is better suited for the manufacture of *dahi* and yoghurt because of its higher total solids content. The need for prior concentration of milk or addition of milk powder is eliminated. The product exhibited a superior body texture and a creamier mouthfeel. Buffalo milk fat contains less cholesterol and more tocopherol, which is a natural antioxidant. There were significantly lower concentrations of total and free cholesterol in buffalo milk (275 and 212 mg respectively/ 100 g fat) when compared to 330 mg and 280 mg respectively /100 g of cow milk fat. The higher calcium content in buffalo milk produces better quality paneer in terms of its body and texture attributes. The higher levels of taurine and lactoferrin in buffalo milk, also made it a superior feed for infants.

Challenges in Buffalo Milk Processing

Due to higher content of fat, proteins, lactose and calcium as compared to cow milk, necessitates buffalo milk to be standardized to fat and solids not fat (SNF) ratio by either cream separation or addition of SNF an additional step during its processing. The ripening or fermentation process is often slower in buffalo milk. The rennet clotting time of buffalo milk is much lower due to high level of calcium. This factor also leads to harder body and texture attributes in *chhana* produced from buffalo milk making it less suitable for preparation of *chhana* based sweets. The curd obtained had a higher curd tension when compared with cow milk. Due to higher levels of milk proteins, high calcium content and micellar differences in milk proteins the challenges that noticed during the production of powders from buffalo milk are low heat stability, solubility and faster browning.

Heat processing of buffalo milk

During heat processing the compositional attributes of buffalo milk influenced the thermal destruction of microorganisms. It was reported that spores of *Bacillus subtilis* were more heat resistant in buffalo milk than in cow milk. The D value for the thermal destruction of *Bacillus subtilis* in buffalo milk with 6% fat on heating to 100°C was calculated to be 53 minutes. It was reported that *E. coli* can survive pasteurization temperatures in buffalo milk. The Z values for the destruction of three strains of *E. coli* in buffalo milk were higher than in cow milk.

Various parameters such as kinetics of bacterial spore inactivation, Maillard browning, whey protein denaturation and storage stability of UHT buffalo milk have been extensively investigated. Decimal reduction time value (D) *i.e.,* the time required to reduce microbial population by one log cycle at 130, 135, 140 and 145°C were 18, 6, 2 and 0.8 seconds

respectively. The death rate constant (k) determined at the above temperatures was 0.12, 0.38, 0.87 and 2.87/second.

Buffalo UHT milk, was considerably whiter in appearance than raw buffalo milk. Buffalo UHT, was sensorially comparable to cow UHT milk. Organoleptic scores ranging between 83 and 92.5 were obtained for buffalo UHT milk on a 100-point composite scoring scale. Scores in respect of cow milk were only marginally higher and varied from 84 to 94, when the two milks were evaluated by a trained taste panel. However decimal reduction times for organoleptic scores during UHT processing of buffalo milk, were higher than those for cow milk.

Buffalo UHT milk showed faster deterioration when compared with similarly processed cow milk. Maillard browning reaction results in severe depletion of nutritional value of milk. High levels of proteins and lactose in buffalo milk, create favourable conditions for rapid progress of the Maillard reaction during heat processing of buffalo milk.

Milk Powders from Buffalo Milk

The physical characteristics of skim milk powder from buffalo milk were similar to that of cow milk product. The insolubility index of buffalo milk powder was slightly higher as compared to powders made from cow milk. The level of preconcentration prior to spray drying of buffalo milk powder was slightly higher particularly for whole milk also effected powder characteristics. It was observed that with increased total solids in the milk concentrate; the moisture content, solubility index, wettability, sinkability, dispersibility, particle size, bulk density of powder also increased. Near instant buffalo skim milk powder was prepared by a single pass method using buffalo skim milk concentrate having 50% total solids.

Buffalo Milk Cheeses

Buffalo milk is conventionally used for the preparation of some traditional cheeses *e.g*., Mozzarella, Feta, Domiati, Queso Blanco etc. However, due to relentless efforts of many researchers protocol for the production of cheeses conventionally prepared for cow milk e.g., Cheddar, Gouda etc. has been perfected using buffalo milk.

Mozzarella is a well-known variety of Italian cheese which has gained popularity throughout the world. Mozzarella is used on pizza toppings, which is undoubtedly a very popular product now in India. In Italy, the name 'Mozzarella' is exclusively used for the cheese prepared from buffalo milk without the admixture of milk from other species. The term 'Mozzarella di bufala' in Italy now enjoys a legal protection as a product that is made strictly from buffalo milk. Buffalo Ricotta is prepared from the whey left after production of Mozzarella cheese from buffalo milk. Pure white in colour, buffalo Ricotta has a fine, slightly moist texture and a bland sweetish flavour. Like fresh, unripened cheeses, Ricotta does not have a long life, and must be consumed within a short time.

Fat Rich products

Various functional properties such as thermal behaviour, emulsification properties, crystallization and fractional behaviour of buffalo milk fat is different that fat of other species. Due to inherently high fat content and larger size of fat globules, the churning attributes of buffalo cream were superior to that of cow milk cream. Loss of fat in butter milk was lower, and the fat mass obtained was firmer. The churning ability of buffalo cream, improved considerably with increasing fat content, acidity and pre-cooling. Buffalo milk fat was rich in high melting triglycerides and saturated fatty acids particularly the palmitic and stearic acids. The crystallization of triglycerides of buffalo milk fat was relatively easier than that of cow milk triglycerides. As the amount of crystal formation in the fat of cream before it is churned into butter greatly influences the rheological properties of butter, the fat in cream was tempered for a shorter duration than cow milk cream. The ripening temperature was also higher than that for cow milk cream.

Ghee is a complex mixture of lipids such as triglycerides, esterified fatty acids, phospholipids, sterols, sterol esters, fat soluble vitamins, carbonyls, lactones, other hydrocarbons together with traces of charred casein and moisture. Due to the absence of carotenoids in buffalo milk, the ghee produced from it did not exhibit the typical golden yellow colour observed in cow milk ghee. A slight greenish blue tinge in buffalo ghee is owing to the presence of bilirubin and biliverdin pigment.

Conjugated linoleic acids (CLAs), which are shown to possess anti-carcinogenic properties, were reported in buffalo ghee. Presence of these acids has generated much interest about the physiological role of ghee in human nutrition. Ghee prepared by the traditional process contains more CLAs (1.2-2.9%) as compared to ghee and prepared by the creamery butter (0.6-1.5%) or direct cream (0.65-0.80%) method. The levels of CLAs in cow ghee were lower than in buffalo ghee when prepared by traditional and direct cream method.

Traditional Indian Dairy Products from Buffalo Milk

Traditional dairy products are the products of indigenous origin and it is presumed that they were developed to preserve the nutritional quality of milk constituents for longer duration in the tropical climate. There are many technological interventions available for preparation of heat desiccated, heat and acid coagulated, and fermented traditional products using buffalo milk.

Buffalo milk is generally preferred for *khoa* preparation primarily because of three reasons *viz*., higher total solids (which corresponds to increased yield), pleasant sweet taste (which is slightly salty in case of cow milk) and white coloured appearance (slightly yellow in case of cow milk). Traditionally, *khoa* is prepared by the boiling milk in an open pan with continuous stirring and scrapping till a mass of the desired consistency is obtained. This is also accompanied with ‘pat’ formation, which occurs as a result of fat de-emulsification and the contents starts leaving the heating surface. This is followed by transferring the mass (*khoa*) on a greased tray and allowing it to cool. A number of equipment was developed to mechanize the process of khoa preparation. This includes thin film scrapped surface heat

exchanger (TSSHE), inclined SSHE (ISSHE), conical process vat, etc. All these equipment are based upon the principle of rapid concentration of milk solids along with constant scrapping the contents to prevent over desiccation and browning. This has resulted into increased capacity of production, lower manpower requirement, improved hygienic and microbiological quality of the product.

Heat and Acid Coagulated Products

Paneer is a heat and acid coagulated product preferably prepared using buffalo milk because of higher calcium and casein content. Paneer comprises of an aggregated mass of casein micelles and fat globules, in which lactose and other soluble components (including whey proteins) of milk are entrapped. Composition of paneer prepared from cow and buffalo milk is provided in Table 3. Fore-warming (above 90°C) prior to acid coagulation results into denaturation and interaction of whey proteins with casein in the form of a porous structure. This increases the paneer yield and imparts spongy texture into it. During paneer preparation addition of coagulant results into decrease in the pH and leads to solubilisation of calcium, which was previously attached to casein micelles as calcium hydrogen caseinate and tri-calcium phosphate.

Constituent (% by weight)	**Buffalo milk (5.8% fat in milk)**	**Cow milk (4% fat in milk)**
Moisture	50.8	52.6
Total Solids	49.2	47.4
Fat	27.5	23.5
Protein	18.2	20.4

Table 3: Gross Composition of buffalo and cow milk paneer

Method for paneer preparation from Buffalo Milk

Buffalo milk (containing fat: SNF ratio of 1:1.65) is heated to 95ºC (no hold) and subsequently cooled to 70ºC for acid coagulation. At this temperature, acid solution (1 % citric acid), maintained at 70°C is added into the milk along with very gently stirring till the greenish whey appears. This is followed by drainage of whey and pressing the coagulum to shape the product. The curd (*paneer*) is then cooled by dipping in chilled water (at about 5ºC) for 4 hours (preferably overnight). Chilling results into proper shaping and decrease in the temperature of the product. The chilled *paneer* is then packaged in suitable packages (before or after slicing into pieces) and marketed.

Chhana

Chhana is also categorized as a heat and acid coagulated traditional dairy product. Because of low calcium content, cow milk is generally preferred for preparation of *chhana*, however, technology was perfected for *chhana* preparation using buffalo milk. Chemical composition of *chhana* depends upon the quality and type of milk used and the processing conditions maintained during the product preparation. Gross chemical composition of *chhana* prepared using cow and buffalo milk is provided in Table 4.

Constituent	Buffalo Milk	Cow Milk
Fat	29.7	24.4
Protein	14.7	17.8
Lactose	2.3	2.3
Ash	2.0	2.2
Moisture	51.7	53.4

Table 4: Gross Composition of buffalo and cow milk *chhana*

Good quality *chhana* possess soft, smooth body and slightly adhesive texture. *Chhana* prepared using buffalo milk without any change in the processing parameters exhibits higher chewiness, hardness and lesser cohesiveness. This necessitates the need to modify the processing conditions for *chhana* preparation using buffalo milk, *viz*., delayed whey draining (results into higher amount of whey retention in the coagulum and thus higher amount of moisture and decreased hardness), mixing of 25 % cow milk with buffalo milk and maintaining a coagulation pH at around 5.7.

Sandesh

Sandesh is a *chhana* based product. When prepared from buffalo milk, *sandesh* exhibit slightly less cohesive, springy, gummy and chewy body as compared to the product prepared using cow milk. In order to improve the sensory quality, buffalo milk is adjusted to 4 % fat. Also, stabilizing salts, *viz*., sodium phosphate is added at 0.05% level in the standardized milk. Product obtained from buffalo milk using these modifications has sensory quality similar to that of cow milk *sandesh*. Also, an admixture of buffalo and cow milk in the ratio 3:1 could also be used for acceptable quality *sandesh* preparation. Rheological properties of *sandesh* (*narampak* variety) are provided in Table 5.

Characteristic	Cow milk	Buffalo milk
Hardness (kg)	3.50 kg	3.59 kg
Adhesivenes (kg)	0.215 kg	0.212 kg

Cohesiveness	0.311	0.254
Springiness	0.337	0.311
Gumminess (kg)	1.025 kg	0.814 kg
Chewiness (kg-cm)	0.344 kg-cm	0.251 kg-cm

Table 5. Rheological and textural attributes of *Narampak sandesh* of cow and buffalo milk.

Dahi

Dahi is a fermented traditional milk product obtained by lactic starter fermentation of milk. During its preparation, the milk is first heat treated to high temperature (80-95°C) for 5-15 minutes. This extent of heat treatment results into inactivation of microorganisms present in the milk and also results into denaturation of whey proteins and their interaction with caseins. The free sulfhydryl group exposed by this heat treatment aids in maintaining reducing environment by decreasing the activity of oxygen. The heat-treated milk is then cooled to incubation temperature and once the temperature is reached, it is inoculated with starter culture and incubated for acidity development and product preparation. As soon as the desired acidity is achieved, the temperature is cooled rapidly to refrigeration temperature in order to limit the activity of starter culture and maintain the sensory quality of *dahi*. Composition of *dahi* is similar to the milk from which it has been prepared. Gross chemical composition of *dahi* obtained using cow and buffalo milk is provided in Table 6.

Constituents	**Buffalo milk**	**Cow milk**
Moisture (%)	83-86	84-87
Fat (%)	6.1-8.4	3.5-4.5
Protein (%)	3.5-4.0	3.0-3.5
Lactose (%)	4.6-5.2	3.8-4.5
Ash (%)	0.7-0.72	0.64-0.66
Lactic acid (%)	0.5-1.1	0.5-1.0

Table 6. Gross composition of *dahi* from buffalo and cow milk

Misti doi or *payodhi* is a fermented sweetened traditional dairy product prepared from partially concentrated and sweetened buffalo milk. The product has firm body, light brown colour, cooked flavour and a sweet caramelized taste. It contains about 30-32% solids, 10-12% sucrose and has 0.95-1.0 % titratable acidity.

Shrikhand is also a sweetened fermented product popular in western part of India. *Shrikhand* of superior quality is obtained using buffalo milk of 6.0 % fat and 9.0 % SNF. Its preparation involves separation of whey from the set *dahi* by suspending it by a cloth bag or by pressing

to squeeze out the whey to yield chakka or *maska*. *Chakka* obtained from buffalo milk has smooth body, while skim milk *chakka* has rough texture and dry appearance. About equal amount of sugar is added to *chakka* and the mixture is thoroughly kneaded to form a homogenous mass. Flavourings, nuts and edible colours are added during this stage. This is followed by cooling and packaging the product for sale.

Kheer

Kheer is a milk, cereal and sugar containing traditional milk product which possesses nutritional goodness of both cereal and milk. For *kheer* preparation, rice is first soaked in water and added to buffalo milk at about 3% of milk and concentrated in an open pan till the contents decreases to half of the initial volume. At this stage, sugar is added (5-6 % of the initial quantity of milk) and cooked for about 5-10 minutes to melt and uniformly mix the sugar. Chopped nuts and cardamom are generally added after this stage and just prior to ending the heating process. This is followed by cooling the contents to room or refrigerated temperature. Similar to heat desiccated products, yield of *kheer* depends upon the initial total solid concentration and hence buffalo milk is preferred.

Other Value-added Products from Buffalo Milk

Increasing share of value-added products in overall dairy products market is expanding the market growth. Some of the value-added products from buffalo milk having great market value are discussed below.

Curcumin *Ghee* from Buffalo Milk

Curcumin, the biologically active yellow pigment found in Indian spice turmeric, possesses numerous functional attributes *e.g.*, antioxidant, antibacterial, antifungal, antiviral, anti-inflammatory, anti-Alzheimeric, anticarcinogenic, antidiabetic, etc. Moreover, oxidative deterioration is one of the major factors that limit the shelf life of ghee. Synthetic antioxidants are often used in ghee to prevent oxidative deterioration that may cause liver damage and cancer. For the same reason natural anti-oxidants derived from plants has gained focus as an alternative to the synthetic ones, besides of increasing the product's functionality. To develop curcumin fortified *ghee* (CFG) from buffalo milk creamery butter method was found to be the best. Response Surface Methodology (RSM) used to optimize the level of process variables *viz*., heating time and temperature. Conjugated diene, TBA, FFA content were lower and DPPH radical scavenging activity was significantly higher in case CFG as compared to BHA added ghee. Storage studies of CFG were carried out in HDPE bottles and metallized PE pouches and it was found that peroxide value, TBA, FFA, conjugated diene value of all samples increased significantly with progress of storage period and DPPH radical scavenging activity was decreased. Consumer acceptance study revealed that 98% of respondents preferred CFG, among them 40% rated excellent, 36% rated very good, 17% rated good while 7% rated fair. Ninety percent of respondents, who had liked the CFG, was interested to purchase knowing that it contained functional ingredient despite of higher cost.

Haldi Lassi

Lassi is a widely consumed fermented milk beverage of India. *Lassi* is a highly perishable commodity and has a shelflife of around 8-12 days at refrigeration temperature. Spoilage of *lassi* is primarily caused by excessive fermentation, enzymatic breakdown or contamination with undesirable microorganisms. A technology was developed for production of functional *lassi* using buffalo milk by incorporation of curcumin and its shelflife was ascertained with respect to organoleptic, chemical and microbiological changes in poly ethylene terephthalate (PET) bottles. The developed product had a shelf life of 20 days at 4 ± 1°C and 90-95% RH when packed in low density polyethylene (LDPE) pouches or in PET bottles, whereas the control *lassi* had storage life of 12 days. No loss of curcumin was observed during the storage of the product in both the packaging materials. The developed product may possess great potential as a functional food with increased shelf life through natural preservation.

Reduced Fat Dairy Spread

Due to rapid increase in life style related disorders and the consequential rise in health consciousness of general populace, high fat and energy rich foods products are finding less acceptances among the consumers. Reduced fat dairy spreads could be a better alternative to their full fat counterparts because of their low fat and high protein content, low energy density and increased convenience as they are spreadable at refrigerated temperature. A functional reduced fat dairy spread by using relatively cheaper ingredients *viz*., chhana, skimmed milk powder (SMP) etc. has been developed. The optimization command of Design Expert Software for Response Surface Methodology was used to in order to achieve the best combination of the ingredients. The chemical composition of the optimized product was observed to be 62.5% moisture, 10% fat, 16.8% protein, 4.75% total carbohydrates 3.6% ash and 2.35% salt. The product contained 30% less calorific value as compared to the full fat market samples. The reduced fat *chhana* based dairy spread added with curcumin @ 200 and 300 ppm could be satisfactorily stored for more than 60days under refrigerated temperature without adversely effect on sensory, physic-chemical and microbiological qualities.

Conclusion

Buffalo milk contribute almost half of the milk business in India. Distinct physicochemical, functional and processing characteristics of buffalo milk makes it better suited for producing many milk products whose sensorial, textural and nutritional quality is better as compared to the products prepared from milk of other species. Over the years, due to numerous efforts of many academic researchers and industrial workers technology has been developed to prepare milk products which were considered cannot be produced from buffalo milk. In recent years many value added functional milk products have also been developed using buffalo milk. Venturing into production of these value-added products can help the dairy industry to become more viable and definitely open up new markets for buffalo milk.

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CHAPTER 13

Buffalo Meat as a Promising Animal Product

M. Muthukumar*, Rituparna Banerjee and B.M. Naveena
ICAR - National Research Centre on Meat, Hyderabad
*Corresponding author: Dr. M. Muthukumar (Principal Scientist);
muthukumar55@rediffmail.com

Livestock Sector has continuously been growing at compound annual growth rate (CAGR) of 7.93% (at constant price) from 2014-15 to 2020-21, which is higher to CAGR of Agriculture (2.05%) and even manufacturing (4.93%) and Services (4.82%) sector. Further, livestock sector contributed 4.90% (at constant prices) of total GVA in 2020-21(DAHD, 2022). Among the livestock, buffaloes constitute about 20.45 per cent of total livestock population and have been recognised as multipurpose animals (milk, meat, leather and draught utility). The meat function of buffaloes became more evident over the last few decades due to the development of organized nature of the buffalo meat export industry.In terms of value, buffalo meat became the leading agricultural product exported from India and fetches foreign revenue of about US $ 4 billion.

Emergence of buffaloes as quadruple purpose animal

The contribution of buffaloes to country's current milk production of more than 209 million metric tonnes is very significant as about 45% of total milk is produced by buffaloes. Though buffaloes were not utilized for meat production in the past years, over the last few decades, the meat function of buffaloes has increased due to surging exports. The average live weight of buffaloes used for meat production in India is reported to be around 300 kg. The average carcass weight is around 150 kg and yield ranges between 45 and 55% for buffaloes (Muthukumar et al. 2017). Buffaloes contribute about 98 percent of total meat exports from India. Therefore, now buffaloes have been recognised as multipurpose animals having milk, meat, leather and draught utility.

Quality attributes of buffalo meat

The most noteworthy feature is that buffalo meat did not possess any religious taboo against its consumption, and emerged as the healthiest meat among red meats in many parts of the world. Buffalo carcass has higher proportion of muscle and lower proportion of bone and fat than beef carcass (Joksimovic and Ognjanovic, 1977). Buffalo meat is low in fat and cholesterol content and high in lean and protein contents (Lazar, 2001). In terms of quality and organoleptic properties, buffalo meat is nearly identical to beef, with lower contents of fat, cholesterol, and calories. Buffalo calves have shown to produce meat with the most favorable (n-6)/(n-3) ratio (7.00) compared with the bovine calves and the buffalo cows (Dimov et al., 2012). Among all of the red meats, buffalo meat has been reported to have the lowest concentration of total lipids (1.37 g/100 g). Water buffalo meat was also reported to contain a greater concentration of conjugated linoleic acid (1.83 mg/g fatty acid methyl esters) compared with meat from zebu-type cattle (1.47 mg/g fatty acid methyl esters; de Mendoza et al., 2005). The colour of buffalo meat is brighter, more uniform than beef (Cosentino et al. 1982; Matassino et al. 1976). Myoglobin content of fresh buffalo meat

varied from 2.7 to 9.4 mg/g depending on the type of the muscle and animal age, and meat becomes darker with increasing age (Naveena and Kiran, 2014). Buffalo meat contains proteins of high biological value in terms of their essential amino acid content (Ziauddin et al., 1994). Buffalo meat has higher units of essential amino acids, biological value and iron content (Anjaneyulu et al. 1990). Buffaloes produce tender meat by virtue of their docile habits.

Buffalo meat production

During the year 2020-21, buffalo meat production was around 1.59 million tonnes (DAHD, 2021). Though small numbers of buffaloes are slaughtered in the local body slaughterhouses for the domestic consumption, majority of buffalo meat production is carried out in the export oriented integrated facilities. Currently, there are about 80 plants registered with Agricultural and Processed Food Products Development Authority (APEDA). It is mandatory that all these meat export plants should have adequate facilities for meat animal handling, ante-mortem and post mortem inspections, hygienic slaughter, dressing and meat processing, by-products collection and processing, rendering, effluent treatment and disposalto get approval and recognizing the facility for registration as meat export plant by APEDA. Further, in addition to monitoring the pollutants level and environmental pollution by the State Pollution Control Boards (PCB), many of the meat export establishments are certified with relevant standards for quality control and assurance (HACCP and ISO standards) and environment management (ISO 14001: 2015 and ISO 14004:2016).

Buffalo meat export

India attained number one position in bovine meat export category from nowhere position about 50 years back. The increased popularity of buffalo meat in several south-eastern and middle-eastern Asian countries is due to its reduced fat, reduced cholesterol, and low calories. At the beginning, meat export was carried out utilizing carcasses sourcing from the domestic slaughterhouses and processing in the general-purpose cold stores. During the year 1945-49, meagre quantity of beef, mutton and goat flesh and pork were shipped from major port towns like Bombay, Madras and Calcutta for use on their voyages. After 1950s, export ofmeat was nearly ceased with ban on cattle slaughter in several states and ban on beef export. However, export of buffalo meat has resumed in the 1970s with the establishment of Deonar abattoir at Mumbai and the higher price realization in the international market.In the year 1973-74, about 2000 MT of buffalo meat at a value of Rs 6.84 crores was exported and the export of buffalo meat was increased to about 60,000 MT by 1987-88 valued at Rs.88 crores. During the decade 1990-2000, buffalo meat export was increased by 164% with an average annual growth rate of 16%. In value terms, buffalo meat export increased by 562% and realisation per kg of buffalo meat has increased by 150% in this decade. The number of countries to which Indian meat is exported has increased from about 30 in 1994-95 to 50 in 1999-2000. Between 2009 and 2014, India's buffalo meat exports more than tripled from around 0.6 million tonnes to around 2 million tonnes in carcass weight equivalent terms. In value terms, shipments more than quadrupled from $ 1,163.54 million in 2009-10 to $ 4,781.18 million in 2014-15. Currently buffalo meat is exported to more than 70 countries of the World and has becomes India's No. 3agri-export item ($ 3.30 billion), after non-basmati ($ 6.12 billion) and basmati rice ($ 3.54 billion) (APEDA, 2022).

Table 1. Details of buffalo meat production

Year	Total meat production (Million metric tones)	Buffalo meat production (Million metric tones)
2007-08	4.0	0.557
2008-09	4.2	0.623
2009-10	4.5	0.670
2010-11	4.9	0.805
2011-12	5.5	0.975
2012-13	5.9	1.104
2013-14	6.2	1.164
2014-15	6.7	-
2015-16	7.0	1.611
2016-17	7.4	1.451
2017-18	7.7	-
2018-19	8.1	1.19
2019-20	8.6	1.22
2020-21	8.8	1.59

Data source: BAHS 2020; DAHD 2021; APEDA 2022

India reached number one status in 2011-12 surpassing the three majors- Australia, Brazil and US. Brazil and India have been competing for the top spot in world beef exports over the past several years and India and Brazil tied on top — with both countries accounting for just under 20% each of the world's total beef exports. These four countries exported more than 1 million metric tons of beef in 2016. During the year of 2020-21, India has exported 1.17 million MT of buffalo meat products for the worth of 3.30 USD Billions.

The top 10 destinations for buffalo meat by value in 2021-22 were in Asia and Africa countries. There are several reasons for the popularity of buffalo meat in the meat market. Comparatively lower cost of buffalo meat, desirable compositional characteristics (lean meat of higher quality protein, lower fat and cholesterol, desirable fatty acids, minerals and vitamins etc.), ethical and traditional considerations, the risk free status, etc. have patronized Indian buffalo meat in various importing countriesfor about five decades. As the buffaloes in India are salvaged for meat production only after completing their productive life, the buffalo meat has a competitive price advantage over beef. The average free-on-board price of buffalo meat exported from India is around $ 3,500- 4,000 per tonne. Continuous increase in the

prices of buffalo meat in the export market has in turn contributed for better farm economics together with demand driven growth in dairy sector leading to sustained buffalo production.

Table 2. Top 10 Indian buffalo meat importing countries

Importing Countries	2021-22	
	Qty (Metric Tonne)	Rs. Crore
Egypt	28,86,109	5,509
Vietnam	1,49,379	3,637
Malaysia	1,53,690	3,318
Indonesia	98,056	2,288
Iraq	91,374	1,663
Hong Kong	67,045	1,580
Saudi Arab	51,631	1,178
Philippines	43,317	947
United Arab Emirates	45,315	888
Jordan	21,796	499

The proximity to key meat consuming countries viz., Asia, Middle East (80 % of the total) and Africa (15%) has reduced the transit cost and pass on a huge competitive edge to Indian buffalo meat exports. Further, the Indian buffalo meat finds acceptability in the Muslim countries as the buffalo are slaughtered and processed, as defined in the Islamic religious text Koran.

Contribution of buffaloes to National economy

The economic contribution of buffaloes is both direct by way of contribution in terms of milk, draught, meat and leather and indirect on livelihood, employment, natural resource utilization and value addition function. A precise estimate of contributions to National economy is complex but it can be stated with certainty that they are considerable. Buffalo milk production at 94 million tonnes contributes about Rs 3,29,000 crores (at Rs 35 per kg) and earnings from dairy exports is at Rs. 2,928 crores (largely from buffalo products).

During the year 2021-22, buffalo meat production was around 1.61 million tonnes. About 98 percent of total meat exports from India are contributed by buffaloes. Export of buffalo meat fetches about US$4 billion. Buffaloes also contribute for foreign exchange earnings from growing modern leather sector by way of contributing quality hide and skins from buffaloes slaughtered for meat export. Buffalo hide makes excellent, thick, tough leather much valued for making shoe soles and other leather articles. Tasty buffalo chips are made from buffalo hides in parts of Thailand, Nepal and Indonesia. Leather sector is known for its consistency in high export earnings and it is among the top ten foreign exchange earners for the country. With an annual turnover of over US$ 12 billion, the export of leather and leather products increased manifold over the past decades and touched US$ 6.5 billion during 2014-15, recording a cumulative annual growth rate of about 13.10% (5 years). Share of buffalo leather exports is at Rs 15000 crores (about 38 percent of total leather exports at Rs. 37,835 crores excluding non-leather foot wear).

In addition to meat, there is a huge export market potential exists for all co-products and by-products generated during meat production. By-products such as tripe, heart and tongue after processing are exported in good quantity. It may be noted that share of edible

offal in the total meat and offal export has been growing significantly and is around 6 percent amounting to an export value of Rs. 2000 crores. Inedible offal have been promoted successfully as pharmaceutical raw material and raw material for the pet treat sector (Rs. 500 crores). Other waste meat, tissues, rejected organs and bones are processed through modern rendering plants to produce meat cum bone meal, tallow and gel bones that have markets for poultry feed, soap, lubricants, sizing in textile industry and gelatin manufacture. Massive horns of buffalo are used in wide variety of fancy and decorative horn article manufacturing. Organic material wastes such as rumen, intestinal contents and dung are utilized for methane production or used as fuel resource for boilers.Recognizing the importance of this species as back bone of Indian dairy industry as well as Indian meat export industry, buffaloes have been described 'the Black Gold'. The important role in food security and livelihood of farmers has been recognized as the important aspects of buffalo production.

Value addition of buffalo meat

As majority of buffalo meat produced in India comes from aged or spent animals at the end of their productive or working life, the meat has low consumer appeal and eating quality attributes. However, as the demand for lean red meat has been increasing consistently world-wide, buffalo meat is expected to get consumer preference due to its leanness and lower cost (Naveena et al., 2022). Functional properties like pH, water holding capacity, emulsifying capacity, and emulsion stability of buffalo meat play a major role in processing of meat products. Expansion in buffalo meat consumption is related to its nutritional advantages compared to beef. Researchers suggested that water buffalo meat consumption could be associated with several beneficial effects on cardiovascular risk profile, including lower carotid atherosclerotic burden and susceptibility to oxidative stress. The processing of buffalo meat is a profitable market, with customers potentially preferring it because of its lipid content, nutritional properties, and excellent palatability (Giordano et al., 2010). Due to higher proportion lean, buffalo meat blends well with other ingredients, hence, it is more suitable for production of various value-added processed products and canned food industry (Anjaneyulu and Muthukumar, 2010). Hence, the development of value-added meat products is one of the best strategies to enhance consumer appreciation, open up new markets and improve net profitability. Comminuted buffalo meat products viz., sausages (Sachindra et al., 2005), loaves (Devatkal et al., 2004), burgers (Modi et al., 2003), patties (Suman and Sharma, 2003), nuggets (Thomas et al., 2006; Prince et al., 2010) haleem (Muthukumar et al., 2005) with better sensory attributes have been developed. The development of value addition and further processing of buffalo meat sector has larger prospects as it boost the market demand for buffalo meat promotes per capita consumption, ensures nutritional security, stabilizes the prices of buffaloes and their produce, thereby support sustainable growth of buffalo husbandry benefiting farmers, processors and consumers.

Way forward

As the world meat markets for both the raw and processed meat is expanding at a greater pace, the opportunities for Indian buffalo meat export are very promising. The USDA anticipates a rise of total world beef exports. Increase in the purchasing power and changing taste buds in these growing economies are few of the growth drivers. It has been well recognized in developed countries that the prospects for growth in beef and veal production will depend heavily on the future growth and structure of the dairy industry, by-products of

which are the major source of cattle slaughtered for meat. With buffalo emerging as the major dairy animal in Indian situation and the slaughter buffaloes as by-products of the dairy industry, the production system of Indian buffalo became more sustainable and could be able to compete successfully in the World in the post WTO liberalized trade scenario.

Improvement in productivity and increase meat production are very significant for expansion of meat export.About 1.5 million MT of buffalo meat could be produced, if the available 15 million male buffalo calves are grown to higher weights and salvaged for meat production. Buffalo veal is having high market potential and majority of males born can be effectively used for veal production. Due to lack of demand for draught purpose and ban or restrictions on slaughter and meat production, farmers do not take adequate care of health and management of male buffalo calves. Farmers grossly neglect milk feeding as it is uneconomical. Hence early age mortality of male buffalo calves is abnormally high (more than 50 percent) in India. When slaughter policy permits male buffalo calf rearing and utilization, large number of young male calves would be reared upto optimum age/weight (200-300kg), which would not only increase meat and leather production, but also provide opportunity to choose them for selection of breeding bulls and draught buffaloes (Kondaiah, 2014).

Increase in the slaughter rate of buffaloes also will boost meat export. Current slaughter rate is about 10 % in India. Pakistan with similar type river buffaloes showed an increase in its population by 183.19 % with a slaughter rate of 19 % during1980 to 2012. Therefore, with good management and feeding practices slaughter rates upto 20 percent could easily be sustained without any adverse effect on population sustainability with relevant slaughter policy and economic utilization of buffaloes.

In additions to the existing markets, China and Russia are going to be potential markets in the coming years. One of the next ends of the market to look at is the organic or green label segment of the meat market. Increasing awareness among consumers about the health issues resulting from the consumption of meat from those animals raised in industrial farms has resulted renewed demand for organic and "green" label foods especially in developed countries. It is yet almost non-existent in India but has a huge potential of growth. As most of the animals are raised very naturally, meat harvested from those animals could be labelled 'green' or 'organic'. It is possible in the near future to create the awareness about organic animal production among the stakeholders especially farmers and labelling the animal products in conjunction with regulatory authorities. It is one of the possible ways of diversification for Indian meat exports, targeting this high end market, to increase its margins.

Though India is free from economically important bovine diseases, viz., rinderpest, contagious bovine pleuropneumonia, lumpy skin disease, the outbreaks of foot and mouth disease is reported sporadically. Department for Animal Husbandry, Dairy & Fisheries, Government of India has taken serious control measures through 100% vaccination and creation of FMD-free zones across the country where there is scientific proof of the virus being absent is in progress. Recognition of these zones by OIE will for pave way for the entry of animal products of India into the developed counties markets (DADF, 2018).

Conclusion

The transition from agrarian subsistence to more diversified market economies hastens the demand for livestock products and the livestock sector will increase its share in

agricultural value added market. The production potential of food animals has to be enhanced to meet the fast growing domestic and export market demand and also making animal production activities more viable and sustainable. India is witnessing change in the livestock production and utilization pattern. The integrated system of poultry farming led to achieving fifth place in the broiler meat production. Buffalo meat export has attained new heights in the recent years. Further progress could be possible through bring desirable changes in the slaughter regulations pertaining to salvaging the male buffalo calves and unproductive animals. Establishment of disease free zone for Foot and mouth disease (FMD) and implementation of livestock traceability system will open up many markets to Indian livestock products.

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