BUFFALO RESERCH AND DEVELOPMENT IN INDIA: POTENTIAL AND OPPORTUNITIES

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Abstract: The overall livestock situation, production and productivity of buffalo species have been discussed. In spite of low reproductive efficiency, long generation interval, harsh climatic conditions and scarce feed and fodder supply, the buffalo number has continuously increased in India at about 1 % per annum during the last decade with 109.4m heads comprising about 56% of the world's total 193.8m population. In India, 82% of the buffalo population is concentrated in the north and western states where most of the milch breeds of buffaloes are found comprising Murrah, Nili Ravi, Jaffarabadi, Pandharpuri and Surti. Milk production in India, increased from 17 million tonnes during 1950-51 to 146.3 million tonnes during 2014 making India number one in milk production in the world and per capita availability of 332g/day. Milk constituents in buffalo milk in comparison to cow milk has been presented. Various developmental projects initiated for the improvement of buffaloes e.g. National Programme for Bovine Breeding and Dairy Development, NDP- 1 and National Livestock Mission initiated by the Govt. of India during 2014 – 15 have been highlighted and presented. Network Project on Buffalo Improvement undertaken by the CIRB since 1993 with the aim to produce progeny tested bulls for improvement of buffaloes in various parts of the country has been discussed. Seven important buffalo breeds are covered through eighteen centres, which include associated herds of Murrah (8), Pandharpuri, Surti and Jaffarabadi; field units for Murrah (3) and conservation units for Swamp, Bhadawari and Nili-Ravi breeds together with improvement through selection. Conservation and propagation of superior / champion bulls by rearing them with extra ordinary production records (3000 to 5500 kg lactation milk yield) and providing service facility either through natural mating or through semen production at Pvt. Institution like Genus ABS India and technological interventions like embryo transfer technology, buffalo cloning, sexed semen production and marker assisted selection have been discussed.

Keywords: Breed, Murrah, progeny testing, champion bulls, cloning

Introduction: It is estimated that the global human population is likely to touch 8 billion mark by 2030. India will have approximately 18% of the world population growing at about 1.3% per annum with only 7.3% of the world arable mass. India thus faces a huge challenge for its agricultural sector to meet the nutritional requirement of its huge population, as against the challenges of limited natural resources, climate change and shrinking cultivable land due to urbanisation. As a result of growing economies and per capita income, demand for animal products (dairy and meat) is growing globally and India is also witnessing enhanced production, productivity and consumption of its agricultural produce especially from livestock with annual growth rate of about 4 to 5% over the last decade. The overall growth rate in livestock sector has been quite variable for different species (Table 1) though steady growth has been observed in buffaloes over the last decade, besides poultry and ducks.

Table1. Indian Livestock Diversity and their growth pattern (*population in millions*)

Species	Livestock Census Population			% Growth Rate per annum	
	2003	2007	2012	2003 - 07	2007 -12
Cattle	185.2	199.1	190.9	1.83	-0.82
Buffalo	97.9	105.3	108.7	1.84	0.64
Sheep	61.5	70.6	65.1	3.87	-1.81
Goat	124.4	140.5	135.2	3.10	-0.77
Pigs	13.5	11.1	10.3	-4.74	-1.51
Rabbit	0.48	0.42	0.59	-11.63	7.90
Poultry	489.0	648.3	729.2	7.33	2.48
Ducks	30.0	27.6	23.5	-7.73	2.97

The ownership of the livestock is evenly distributed with landless and marginal farmers with small herd size. The reproductive efficiency is low and the generation intervals are long besides the feed and fodder scarcity and harsh climatic conditions which make dairying in India a challenging profession. In spite of all these factors, the buffalo number has continuously increased at about 1.5% per annum during the last decade and also the per head milk production increased from 4.05 kg during 2000 -01 to 4.96 kg during 2012-13 at the rate of about 2% per annum primarily due to the concerted efforts of various R&D agencies and increasing awareness of farmers to rear high producing buffaloes in almost all parts India.

It is well established that milk from buffaloes has low cholesterol (about 43%), High fat (about 60%), higher SNF (about 24%), Calcium (about 9%), Phosphorus (about 110%) and higher market price than cow milk. There is no religious taboo on buffalo meat production as prevalent on cattle in India besides higher capacity for pulling heavier loads which makes buffalo as the preferred triple purpose animal for sustainable livelihood. The estimated per head milk productivity of elite buffaloes is likely to increase to 2500 to 3000 kg while the national average is likely to be about 1750 kg per head by the year 2030 from the present level of about 1500 kg from over 54 million milch buffaloes in the country and about 2100 kg per head by the year 2050 while total buffalo heads are likely to be restrained to about 150 million by that year (Table 2).

Trends of Buffalo population

The world buffalo population is estimated to be approximately 194 million spread in some 45 countries of which 187.86 million (96.9%) are found in Asia, while approximately 5.96 million (3.1%) are found in rest of the world (*Fao.org/stat*, 2013). India has 109.4 million and they comprise approximately 56.4 percent of the total world buffalo population. During the last 10 years, the world buffalo population increased from 174 million during 2004 to 193.82 million during the year 2013 by 19.82 million showing annual increase of about 1.8%. The percent increase in India was about 1.0% as compared to 1.13% in Asia, 0.92% in Africa, 1.74% in Americas and overwhelmingly higher rate of increase in Europe at 5.22% indicating increasing interest of rearing water buffaloes in the temperate climate.

Table2. Buffalo population trends in India and anticipated population up to the year 2050.

year	Population (million)	% growth per annum
1982	69.78	1.84
1992	84.21	2.07
1997	89.92	1.36
2003	97.92	1.48
2007	105.34	1.89
2012	108.47	1.84
2020*	119.03	Anticipated growth rate varying from 1 to 0.75% per annum
2030*	130.93	
2040*	144.02	
2050*	150.42	

Buffaloes in India are spread over almost all parts of the country with varying density of population in different states and union territories. The majority of the population (82%) is concentrated in the north and western states (Table 3) where most of the milch breeds of buffaloes are found comprising Haryana, Punjab, Uttar Pradesh, Rajasthan, Gujarat, Madhya Pradesh and Maharashtra. During the last 20 years there has been continuous growth of this species in this region at the rate of about 1.9% per annum as against the average growth rate of approximately 1.5 % in the country.

Table3. Total number of buffaloes and annual growth rate in major buffalo producing states of India– 1992 to 2012.

Sr No	State	1992	2003	2007	2012	% Growth rate		
						92-2003	2003-07	2007-12
1	Haryana	4373	6035	5953	6085	3.80	-0.34	0.44
2	Gujarat	5268	7140	8773	10388	3.55	5.71	3.68
3	Rajasthan	7743	10416	11091	12976	3.45	1.62	3.39
4	Madhya Pradesh	7143	7575	7536	8187	0.55	-012	1.72
5	Andhra Pradesh	9154	10630	13271	10621	1.61	6.21	-3.99
6	Punjab	5238	5995	5061	5159	1.44	-3.89	0.38
7	Utttar Pradesh	20066	22914	23812	30625	1.42	0.97	5.72
8	Maharashtra	5448	6145	6072	5594	1.28	-0.30	-1.57
	Total	64433 (77.2)	76850 (78.5)	81569 (77.4)	89635 (82.4)	2.31	1.68	1.97
	Other states &UT	19066 (22.8)	21072 (21.5)	23773 (22.6)	19067 (17.6)	0.46	2.43	-3.96
	Grand Total	83499	97922	105342	108702	1.57	1.89	0.64

Source: Deptt. Animal Husbandry & Dairy, Govt of India.

Breeds and their performance characteristics

The number of pure bred animals of specified breeds is expected to be about 25 to 30% of total population. The rest of the buffaloes are nondescript in type and have extremely variable composition being either nondescript, or crosses among various breeds and cannot be categorized in any other well established breed. The number of purebred animals is further diminishing due to migration and interbreed crossing. The well-distinguished and the location-specific breeds available in the country are discussed below

India possesses the best milch buffalo breeds of the world namely Murrah, Nili Ravi, Surti, and Jaffarabadi, which originated in the north-western states of India. These breeds have high potential for milk and fat production besides being used for work and surplus stock used for meat production. Besides these relatively well-known breeds, there are several other breeds in India which have regional importance and add to economic value of the farming community. Some of these breeds are Bhadawari and Tarai in Uttar Pradesh, Nagpuri and Pandharpuri in Maharashtra; Parlakhemundi, Manda, Jerangi, Kalahandi, Sambalpur in Orissa and Andhra Pradesh, Toda in Tamil Nadu and South Kanara in Karnataka and Kerala. Mehsana breed has been developed from grading up of Surti buffaloes with Murrah in Mehsana District of Gujarat. Similarly continued grading up of local nondescript buffaloes with Murrah breed in Krishna and Godawari District of Andhra Pradesh has produced Godavari.

Table 4. Production and reproduction characteristics of different breeds of buffaloes

Breed	Av. Age at 1st calving (months)	Av. Service period (days)	Av. Dry period (days)	Av. 305 day or less milk yield (kg)	Av. Calving interval (days)
Murrah*	44.6±1 (154)	147±11 (333)	148±10 (260)	2275±54 (404)	450±11 (260)
Nili Ravi	39.7±17 (69)	172±11 (70)	172±11 (70)	1979±67 (108)	489±16 (70)
Surti	51.5±1.4 (13)	96±7 (20)	169±16 (20)	1566±46 (34)	481±18 (20)
Bhadawari	51.2±1.3 (6)	148±26 (15)	206±32 (15)	1434 (31)	499±33(15)
Pandharpuri	43.28 (02)	85±5 (14)	100±7 (13)	1841 (19)	425±11 (14)
Swamp	49.33 (1)	134±9 (11)	177.±17 (11)	412 (6)	440± 8(11)

* Weighted averages from the participating centres of Murrah breed under Network Project (2013) Figures in paranthesis are number of observations

Trends of Buffalo Milk Production

India, for the past about two decades is the world's highest milk-producing country. Milk production in India during the period 1950-51 to 2014-15, increased from 17 million tonnes to 146.3 million tonnes which accounted for about 16% of the world's total milk production of 789 million tonnes during 2014. Milk production in India remained more or less stagnant

from 1950 to 1970. Thereafter, it increased rapidly, reaching 80.6 million tons in 2001 and 146.3 million tons during 2014-15 with average growth rate ranging between 1 to 2% up to the year 1974 and 3 to 5% thereafter. The per capita availability of milk increased from 112 gm per day in 1973-74 to about 226 gm per day in 2001–02 and 322g per day (Table 5) during 2014–15 surpassing the world average of 285g per day per head.

This represents sustained growth in the availability of milk and milk products for our growing population. Dairying has become an important secondary source of income for millions of rural families and has assumed the most important role in providing employment and income generating opportunities particularly for marginal and women farmers. Most of the milk is produced by animals reared by small, marginal farmers and landless labourers.

Table 5. Trend of annual total and buffalo milk production and growth rate since 1960-61

Year	Total milk (million tons)	Buffalo milk (million tons)	Per capita availability (gm/day)
1960-61	20.37 (1.64)	11.08 (2.30)	124
1973-74	23.20 (1.15)	12.76 (1.51)	112
1980-81	31.60 (4.51)	18.86 (5.97)	128
1990-91	54.06 (5.68)	28.67 (5.20)	176
2000-01	80.64 (4.21)	45.40 (5.83)	220
2006-07	100.0 (3.53)	56.20 (3.40)	251
2010-11	121.8 (5.45)	62.35 (1.54)	281
2011-12	127.9 (5.01)	65.35 (3.65)	290
2012- 13	132.4 (3.52)	67.68 (3.56)	299
2013-14	137.7 (5.51)	70.44 (4.07)	307
2014-15	146.3 (6.24)	~ 75.0	322

( ): Percent growth per annum. Source: Deptt. of A H & D Deptt. Statistics

About 51 to 55% of the total milk produced in the country is contributed by buffaloes since beginning in spite of the fact that number of adult buffaloes being only 60% to the total milch cattle population including crossbred cows producing on an average about 7 kg per day as against 4.96 kg from buffaloes. Buffaloes have significantly higher production potential as compared to indigenous cattle and much higher fat percentage in milk than crossbreds as well as indigenous cow's milk. The average milk production potential of all buffaloes in India is approximately 1500 kg (Range: 500 to 5500 kg) as compared to about 900 kg in cattle including crossbreds (Range: indigenous 400 to 3500 and Crossbreds 1200 to 6000 kg).

Buffalo milk - high nutritional value: It is well established that buffalo milk is more nutritional than cow milk and it is also considered to be better for manufacture of milk products. The compositional difference between the buffalo and cow milk is presented in Table 6. In addition to the significant cholesterol and calcium benefits, buffalo milk is also rich in iron, phosphorus, vitamin A and protein and the natural antioxidant tocopherol. The lactoferrin content of buffalo milk is 32mg/100 ml compared to only 15 mg/100 ml in cow milk. The protein efficiency ratio (PER) value of buffalo milk proteins are 2.74 and that of cow milk are 2.49. Buffalo milk fat contains higher proportion of high melting triglycerides (9 – 12%) than cow milk (5 – 6%) which gives more solidness in nature (Srivastava and Kumaresan, 2014).

Table 6. Composition of buffalo milk compared to cow milk

Sr No	Constituents	Buffalo Milk	Cow Milk
1	Water (g/L)	820	870
2	Total Solids (g/L)	172	125
3	Lactose (%)	5 – 5.5	4.8
4	Protein (%)	4 - 5	3 – 4
5	Fat (%)	6 – 9.5	3.6
6	Cholesterol (mg/g)	0.65	3.14
7	Cong. Linolinic Acid (mg/g fat)	6.1	5.5
8	Polar lipid (mg/L)	189	140

Source: Srivastava and Kumaresan, 2014

Development / Improvement Programmes

National Programme for Bovine Breeding and Dairy Development: Government of India launched the National Programme for Bovine Breeding and Dairy Development (NPBBDD) by merging four ongoing schemes i.e. Intensive Dairy Development Programme (IDDP), Strengthening Infrastructure for Quality & Clean Milk Production (SIQ&CMP), Assistance to Cooperatives and National Project for Cattle & Buffalo Breeding with the budget provision of Rs.1800 crores (~US$ 300 million) for implementation during 12th Plan.

The National Dairy Plan Phase-I (NDP-I) was launched in February, 2012 with a total investment of about Rs.2242 crore (~US$ 350 million) to be implemented from 2011-12 to 2016-17, with the objective to improve milch animal productivity and increase milk production to meet the growing demand for milk in the country. NDP-I will help to meet the projected national demand of 150 million tonnes of milk by 2016-17 from domestic production through productivity enhancement, strengthening and expanding village level infrastructure for milk procurement and provide producers with greater access to markets. The strategy involves improving genetic potential of bovines, producing required number of quality bulls, and superior quality frozen semen and adopting adequate bio-security measures etc. The scheme is implemented by NDDB through implementing agencies like state Dairy Cooperative Federations/Unions/Milk Producers Companies. NDP-I focuses on 15 major milk producing States - Uttar Pradesh, Punjab, Haryana, Gujarat, Rajasthan, Madhya Pradesh, Bihar, West Bengal, Maharashtra, Karnataka, Tamil Nadu, Andhra Pradesh, Telangana, Orissa, Uttarakhand, Chhattisgarh, Jharkhand and Kerala which account for over 90% of the country's milk production.

National Livestock Mission: The National Livestock Mission (NLM) has commenced from 2014-15. The Mission is designed to cover all the activities required to ensure quantitative and qualitative improvement in livestock production systems and capacity building of all stakeholders. This Mission is formulated with the objective of sustainable development of livestock sector, focusing on improving availability of quality feed and fodder. NLM is implemented in all States. NLM has 4 sub-missions as follows:

Sub-Mission on Fodder and Feed Development will address the problems of scarcity of animal feed resources, in order to give a push to the livestock sector making it a competitive enterprise for India, and also to harness its export potential. The major objective is to reduce the deficit to nil.

Sub-Mission on Livestock Development This sub mission has provisions for productivity enhancement, entrepreneurship development and employment generation, strengthening of infrastructure of state farms with respect to modernization, automation and bio-security, conservation of threatened breeds, minor livestock development, rural slaughter houses, fallen animals and livestock insurance.

Sub-Mission on Pig Development in North-Eastern Region: This sub mission would support the State Piggery Farms and importation of germplasm so that the masses get the benefit of livelihood and contributes in providing protein-rich food.

Sub-Mission on Skill Development, Technology Transfer and Extension: This sub-mission will enable a wider outreach to the farmers for a sustainable livestock development.

Network Project on Buffalo Improvement: The Network Project on Buffalo Improvement was initiated in 1993 with the aim to produce progeny tested bulls for improvement of buffaloes in various parts of the country. Seven important buffalo breeds are covered through eighteen centres, which include associated herds of Murrah (8), Pandharpuri, Surti and Jaffarabadi; field units for Murrah (3) and conservation units for Swamp, Bhadawari and Nili-Ravi breeds together with improvement through selection. A bull certification laboratory for disease status testing is established under the project at CIRB, Hisar.

As per technical program for Murrah breed, a set of up to 15 pedigreed bulls are selected in each set and used for AI in the associated herds (comprising of approx. 1000 breedable buffaloes) and field buffaloes (approx. 16000 AIs per annum) for test mating over 18 months duration. So far, 203 superior breeding bulls have been test mated in 15 sets comprising 10 to 16 bulls in each set. The first 10 sets of bulls have been progeny tested and the percent superiority of the top ranking bulls ranged from 24.89% to 9.37% in the various sets (Sire Directory- 2007, Network Project CIRB) which have been used for nominated mating.

Table 6. Top ranking progeny tested bulls, % superiority of bulls selected for elite mating under Network Project on Buffalo Improvement

Set No.	Bull No.	Location	Dam No.	Sire No.	Dam's best lact. 305 day yield (kg)	Daughter's first lact 305 day or less av. yield (kg)	No. of daug-hters	Sire index	% superiority over contempor ary daughters
I	392	CIRB	238	PQ1	2594	2074	13	2118	22.80
II	761	CIRB	474	366	2878	1960	15	1987	9.37
III	1354	PAU	762	989	3088	2072	6	1975	13.11
IV	1506	PAU	-	988	3018	2065	12	2089	18.81
V	4393	NDRI	2762	1908	3898	2143	13	2187	22.29
VI	1153	HAU	618	759	2675	2022.8	21	2121	13.31
VII	4915	NDRI	3521	2921	3437	2038	17	2116	17.26
VIII	1875	GADVAS U	1669	558	2714	2357	8	2300	24.89
IX	1994	GADVAS U	1884	392	2938	2432	18	2487	11.73
X	1693	LUVAS	1050	392	3194	-	-	2320	1.23*

Field progeny testing under Network Project on Buffalo Improvement to strengthen the ongoing sire evaluation program of Murrah breed was initiated by involving performance recording on farmers animals in the field using the semen of bulls selected under the Network Project. Details of the AI services and daughters born and recorded are shown in Table 7.

Table 7. Total AI, pregnancies, conception rate, Calving, Conception and daughter's milk recording in Field Units up to March 2012

Duration	Total AI	Pregnancies	CR%	Calving	Females born	Daughtres recorded	Av. AFC (mts)	Av. Milk Yield (kg/day)
Unit - I	20931	10469	50.02	6201	3022	205	41.97	7.73
Unit - II	40550	14448	35.60	8854	4138	121	47.05	8.10
Unit - III	21123	9509	45.01	6366	3005	151	46.18	6.88
Total	82604	34426	41.68	21421	10165	477	45.22	7.57

Average CR from artificial insemination in the field was 41.68% based on 82604 inseminations spread over 3 locations. It is estimated that at least 8 to 10 AI are needed for each daughter born in the field while number of AI for each daughter recorded is likely to be very high (approx.25 -30) due to frequent movement of animals in the breeding zone as well as outside the catchment area. The large and fast increasing buffalo population of the country requires a large number of breeding bulls for natural mating, but scarcity of superior breeding buffalo bulls may slow down the desired rate of genetic improvement of non-descript low-producing buffaloes. The only method through which we can make the best use of the limited

availability of superior buffalo bulls is through the use of frozen semen technology with artificial insemination.

BUFFALO FOR MEAT PRODUCTION

In India, meat production from buffaloes is largely a by-product of livestock production system utilizing spent animals at the end of their productive life and surplus males. Buffalo meat production accounts for about 31% of the total 6.5 million tons meat production in the country and their contribution is next to milk as major source of livestock economy and comes to about 16% of the total output of the livestock sector. During the last 25 years meat production has increased indicating average growth rate of about 3 to 4% per annum (Table 6) indicating increasing trend and interest of buffalo producers in buffalo meat production.

Increased exports are primarily due to the large number of spent un-productive buffaloes and surplus male calves being diverted for meat production with lower cost of buffalo meat production. Buffalo meat is generally produced from spent buffaloes and emaciated young male calves. Dressing percentage in such animals is low and varies between 40 to 45% with average carcass weight as low as 138 kg. However, dressing percentage in these animals can be substantially increased by proper feeding prior to slaughter and in such animals growth rate as high as 1000 gm per day has been reported under feed lot system (Ranjhan, 2007). Dry matter intake of such animals is generally more than 3% of body weight and dressing percentage as high as 50 to 55% can be achieved. A strong need has been felt to establish a production base around each modern abattoir to produce quality disease free animals as per the sanitary and phytosanitary (SPS) requirement of OIE standards (Ranjhan, 2013). In experimental trials on daily body weight gain up to 541 gm per day has been achieved under conventional concentrate feeding system. Weight gain during summer months is higher than in hot humid months. (Bharadwaj and Sethi, 1994).

Farmers initiative for production of neo champion breeding bulls

Buffalo breeders maintaining superior quality buffaloes and breeding bulls have initiated the exercise of conservation and propagation of new generation superior bulls by rearing them with extra ordinary production records and providing service facility either through natural mating or through semen production and dissemination for breeding of farmers buffaloes in the field. These animals are reared under high protein high energy rations so as to achieve high growth rate and early maturity. The average body weight of such breeding bulls varies from 800 to 1200 kg at maturity with average growth rate of about 700 to 800g per day. The average peak yield of bull mother dams of these bulls varies between 23 to 28 kg and the lactation milk yield of such animals is estimated between 4000 to 5500 kg.

Genus ABS India, in Joint Venture collaboration with B G Chitale, Bhilwadi, have opened their new bull stud facility BRAHMA located in village Brahmanand Nagar, District Sangli. Genus ABS claims that their Buffalo bulls are selected through a rigorous selection protocol for higher milk yield and high fat content. It is also said that all bull mothers have been recorded for the entire lactation twice a day. Production data is supported by data on fat %, further adding on to the reliability. ABS buffalo bulls have been classified under the categories of ROYALE (Dam's yield >5000), ELITE (dam's yield >4000), PLATINUM (Dam's yield 3000 to 4000) & GOLD as per the milk production and fat % of their respective dams. The names of bulls listed in the various categories include Redhu, Khalli, Bheem,

Dara, Gabbar, Angad, Vijay, Mahabali and some more belonging to farmers in different parts of the country and placed at their respective locations (*genusabsindia.com*).

The number of such superior breeding bulls is increasing with the increasing demand of elite germplasm for higher milk production in the country but there is need to coordinate and make appropriate use of the rapidly increasing gene pool with proper IPR and compensation to the owners. After having the pedigree, breed characteristics, health examination and semen quality check the semen from such bulls can be collected and examined for its freezability at various semen freezing laboratories and disseminated in an integrated manner with appropriate data recording system for achieving the desired levels of genetic gain.

Research Interventions for technology development

Nutritional technologies to enhance production: Area Specific Mineral Mixture (ASMM), Urea Molasses Mineral Blocks, and complete feed blocks may be popularized amongst the farmers and industries to scale up such technologies. Supplementation of critical nutrients (nitrogen, soluble carbohydrates, minerals and preformed proteins) through urea molasses mineral block (UMMB) @1-1.5 kg/d for adult buffaloes can increase efficiency of utilization of nutrients in straw and crop residue based diets. Urea treatment of straw can increase digestibility and intake of straw, enriches straw with additional nitrogen.

Scientific feeding practices like feeding ration components as TMR (total mixed ration) / complete feed blocks, feeding chaffed fodder and straw, feeding at regular interval, preparation of balanced concentrate mixture, feeding of bypass protein and fat, use of unconventional feeds in ration at recommended rate, supplementation of area specific mineral mixtures (ASMM) improves performance of buffaloes significantly with reduction in cost of milk and meat. These have been popularized amongst the farmers and dairy owners.

Other technologies like silage making from surplus fodder for use in lean season and dissemination of information on feeding as per nutrient need of the animals is becoming more popular in states like Punjab for efficient land use planning and increased productivity.

Embryo transfer technology: The technology implies multiple ovulation and oestrus synchronization for accelerating genetic gain through increased selection intensity in females. Protocol for embryo transfer technology has been standardized for cattle and buffalo at NDRI, CIRB, GADVASU and many other institutions. The world first in vitro fertilized buffalo calf was born at NDRI. Since then more buffalo calves using this technology have been produced. Number of calves has been produced through ETT at these institutions. Though the technology has been standardised in buffaloes as well, however the success of ETT is not encouraging at many of these institutions primarily due to the low response in super ovulation and ovum collection technique.

The OPU allows repeated pick-up of immature ova directly from the ovary without any impact on the donor female and the use of these ova in IVM/IVF. The technique can be used for rapid multiplication of elite germplasm, however concerted efforts are needed for focused research both on basic issues regulating the super ovulation and applied aspects to obtain the maximum benefits.

Cloning in buffaloes for production of superior germplasm: Somatic cell nuclear transfer technique enables production of infinite copies of an existing animal of proven genetic merit

and result into dramatic improvement in genetic merit of animals in just one generation. The work on buffalo cloning in India was initiated about two decades ago using the traditional micromanipulation-based approach which resulted in few transferable quality blastocysts, however in 2005 a simplified cloning technique called handmade cloning (HMC) was standardised which resulted in the world's first cloned buffalo calf through the hand guided cloning technique and India entered an era of buffalo cloning, through a simplified technique. After the birth of the first cloned buffalo, the scientists at NDRI further worked on this approach and succeeded to produce cloned calves using ear skin-derived somatic cells , embryonic stem cells , semen-derived somatic cells and urine-derived somatic cells as nuclear donors, or produced from a vitrified–warmed cloned embryo. Recently, they have been able to produce superior quality male cloned calf which is the clone of a progeny tested bull with genetic superiority of more than 22% over contemporary daughters.

Garima, the world's first cloned calf was inseminated with frozen thawed semen of a progeny tested bull of NDRI on February 27, 2014, which resulted in conception and calf born maintained under standard scientific management system during its gestation. Earlier, a female calf 'Mahima' was born to 'Garima' at NDRI on January 25, 2013, which was the first calf born to a cloned buffalo produced through 'hand guided cloning technique. The list of Clones that were born at the National Dairy Research Institute, Karnal are Sammrupa (6 February 2009); Garima (6 June 2009); Garima-II (22 August 2010); Shrestha (26 August 2010); Swaran (18 March 2013); Purnima (6 September 2013); Lalima (2 May 2014); Rajat (23 July 2014); Wild buffalo Deepasha (12 December 2014) and Apurva(5 February, 2015) (Singla, 2015).

Scientists at Central Institute for Research on Buffaloes (CIRB) have also successfully produced a cloned buffalo offspring 'Hisar Gaurav' under the project entitled "Cloning for conservation and multiplication of superior buffalo germplasm". This cloned buffalo calf is distinct from the earlier clones produced in India, as this is produced from cells of ventral side of tail of superior bull, which is least exposed to sunlight and may have less mutation rate, and might result into a good choice for isolation of donor cells to produce healthy clones in future.

Development of diagnostics for enhancement of reproductive and productive efficiency: Reproductive efficiency is one of the most important factors affecting economics in production system. Prolonged postpartum anestrus, estrus detection and early pregnancy diagnosis are factors that limit reproductive efficiency in buffalo.

Marker assisted selection: Whole genome selection has revolutionized animal selection. In the absence of parental information, one can still predict the genetic merit of an individual. However, performing whole genome selection is much easier said than done.

India has sequenced buffalo genome in Network mode at National Bureau of Animal Genetic Resources, Karnal and Central Institute for Research on Buffaloes, Hisar with the main objective to improve breeding of buffaloes having higher genotypic and phenotypic potentials. The buffalo assembly represents 91 to 95% coverage with cattle assembly (B-taurus) 4.0 as reference. ***BuffSatDb*** (**Buff**alo Micro**Sat**ellite **D**ata**b**ase) was developed using PHP and MySQL during 2013 (*http://www.biomedcentral.com/1471-2164/14/43/abstract*). It is simple and systematic web based search for customised retrieval of chromosome wise and genome-wide microsatellites. The BuffSatDb considers all the chromosomes (1–24, X, M, U)

sequenced at NBAGR, India for the development of microsatellite marker database. Copy of data used in BuffSatDB can be obtained from the URL (*http://210.93.84/bbu_2.0alpha/*). (Tantia et al, 2011).

Allelic variation ranging from 9 to 36% was observed by RAPD primers in high and low yielding buffaloes. Sequence, showing the highest polymorphism in buffaloes associated with high and low lactation milk yield was identified. Similarly, genotypic differentiation for buffaloes with varying AFC and Service period has also been demonstrated Six grand sire families have been identified and phenotypic data have been compiled for undertaking linkage studies (Sethi and Sikka, 2004).

Sexed semen production: The requirement for sex pre-selection in cattle and buffaloes is of paramount importance to India where the livestock population is the highest in the world. Semen sexing technique can help to quickly address the requirement of superior germplasm through programmed birth of male calves to elite females so that these can be used extensively in AI programme to cover the vast population. Progeny testing can also be accelerated through controlled births of required number of daughters within short period. Recent development of modern cellular methodologies has led to the development of a flow-cytometric system capable of differentiating and separating living X- and Y-chromosome with about 90% accuracy. The technology is yet to be standardised under Indian conditions either by procuring the available technology abroad or developing indigenously by undertaking research on alternative methods.

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