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BULLETIN OF THE FAO-ESCORENA INTER-REGIONAL COOPERATIVE RESEARCH NETWORK ON BUFFALO AND OF THE INTERNATIONAL BUFFALO FEDERATION – INCLUDES SHORT COMMUNICATIONS, RESEARCH PAPERS, TECHNICAL NOTES, ONGOING RESEARCHES

Inside this issue

In year 2022 the world is still facing with Covid-19 pandemic.

The World Buffalo Congress that should have taken place in October 2022 in Wuhan, China, has been cancelled due to the pandemic. We are now asking to all IBF members to present new candidatures.

On 15-18 November 2022 the 10Th Symposium of America and Europe will be held in Asunción, Paraguay.

In the **scientific focus** section, we are presenting two papers: the buffalo production in Paraguay, and Buffalo artificial insemination in Amazon Valley (a 40-year retrospective).
The four volumes editions book "The black gold**"** from Alina Mitat Valdes presents the buffalo production in Cuba.
The biotechnological applications in buffalo research book includes various chapters on the most advanced techniques (including omics) in buffalo production.
The patented technology for preparing buffalo milk protein concentrate that scientists Dr Sunil Kumar and Mr Kuldeep Dudi developed is presented.

In the **Reports** section are illustrated the activities of American Buffalo Breeders Associations in Argentina and Guatemala.
The IBF Training courses in Italy are still on hold but the IBF Secretariat together with CREA is organizing a series of six webinars on buffalo management, reported in the section **Upcoming events** as well as news about next 13th World Buffalo Congress and 11th Asian Buffalo Congress.

Unfortunately, Prof Fabio Napolitano, an IBF member from Italy, and Manfred Thiele, an IBF member from Germany passed away and we unknowledge their contribution to buffalo development in this number.

The IBF Secretariat worked to maintain connections and support to associates. Other requests to become members were examined, reaching 117 IBF associates, representing 39 countries. The list is enclosed, as usual, at the end of this Newsletter.

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BUFFALOES IN PARAGUAY: PAST, PRESENT AND FUTURE

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ABSTRACT

The water buffalo (Bubalus Bubalis) was introduced to Paraguay in the 1954 through imports from Brazil. Currently the buffalo population is 13.125 head distributed in the 17 departments that make up the national territory. 71% of the buffalo population is found in the eastern "Oriental" region and 29% in the western "Chaco" region. The departments with the largest buffalo population are Presidente Hayes, San Pedro and Paraguarí. There are currently 267 buffalo producers across the country. If one were to project the current Paraguayan buffalo population to have an organic annual growth similar to that of Argentina of 13.1%, from 2022 to 2032 the Paraguayan buffalo population would grow to 50.147. A National Buffalo program oriented towards the strategic objectives could triple this growth rate and increase the national buffalo herd to over 250.000 head.

Key words: buffalo, population, producers, wetlands, Paraguay

INTRODUCTION

The Republic of Paraguay is located in South America, in the south center of the South American continent, with an area of 406,752 km^2, neighboring with Bolivia and Brazil to the north; with Bolivia and Argentina to the west; with Argentina to the south; and with Argentina and Brazil to the east. Its climate is tropical to subtropical with abundant although seasonal rainfall. The country is divided in two major regions: the western "Chaco" and the eastern "Oriental". The difference between the rainy summer and the drier winter season is quite marked, especially in the Chaco. In the Oriental region, temperatures and rainfall are somewhat more uniform, but the seasons are equally marked.

Paraguay stands out as one of the most open economies in South American, and a leading world exporter in products such as of hydroelectric energy, soy, wheat and corn, mate tea, stevia and organic sugar. In 2021 it ranked as the world´s 7th largest exporter of beef products.

The country also has significant wetland areas; about 23.06%, (93,707 km^2), with 15% of this surface located in the Eastern Region. The larger wetland areas are the Pantanal Paraguay, the Lagunas del Chaco Central, the Lagunas del Chaco Húmedo, the wetlands at the foot of the Cerrados, those belonging to the Arroyos y Esteros wetlands, those of the Atlantic Forest, the Ñeembucú wetland, as well as other flooded grasslands (SEAM, 2015).

Wetlands play a key role in the development of life on earth and have been critical to the survival of human communities throughout history. These ecosystems provide important ecological services such as water supply, flood control, groundwater replenishment, shoreline stabilization, storm protection, sediment and nutrient retention and export, pollutant retention, climate change mitigation and water purification (SEAM, 2015). However, they are also areas that, without use, accumulate large amounts of senescent vegetation and dry biomass, being prone to large fires almost annually, and that without productive use often have areas of poverty and scarce human development nearby.

In this work it is proposed that the water buffalo (Bubalo Bubalis), with proper management, is an ecologically friendly alternative for sustainable social, environmental and economic development in this dynamic South American country.

PAST

The first buffalo (Bubalus bubalis) arrived in Paraguay in 1954 when Manuel Ferreira Sosa brought a herd of approximately 40 buffalo along with a larger importation of Nelore cattle from Uberaba, in the state of Minas Gerais, Brazil (Bruyn, 2017; Crudeli et al. 2021). His intention was not to approach buffalo breeding as an economic option, but rather as a curiosity to accompany cattle breeding and to give use to the lowlands on his property in Caazapa. Offspring of this initial herd, sold to different ranchers, was the beginning of the dispersion of the species throughout the different areas of the country, but always more as a curiosity than as an economic option. Another relevant source was the sporadic entry over the land border with Brazil, brought by Brazilian colonists settled in the area adjacent to the state of Mato Grosso.

The first important commercial buffalo exploitation was carried out by Eitimios Ioannidis, who in 1988 acquired 200 buffalo from the state of Mato Grosso and began raising them in Colonia Yguazu, near Ciudad del Este in Paraguay. Later, he increased his herd with small purchases in different parts of the country (Crudeli et al. 2021). In 1993, this producer acquired a herd of 50 registered Jafarabadi buffaloes, with high dairy genetics, from the San José fazenda, Franca locality, São Paulo state, Brazil. His dairy operation was considerable, reaching the milking of 600 buffaloes in various milking parlors within his establishment. Although today he is no longer engaged in dairy farming, he was, at one point, the producer with the largest number of buffalo in the country. This property, however, was sold and the herd moved to another establishment in the Santa Rosa area, where it is currently being managed by his children Karin and Thiliio.

Another buffalo pioneer is Bella Italia in the department of Misiones owned today by Jean Batista Lozio, an Italian based in Paraguay. The origin of this operation was his father's intention to install a buffalo dairy for the production of Mozzarella cheese. His premature death cut short the project that was already underway, and today the operation is currently dedicated to meat production and the breeding of Criollo horses (Crudeli et al. 2021) and is known for their yearly buffalo and Criollo horse auction.

Mention must also be made of Ruben Bruyn and his family. He is one of the first who started to mark the difference between buffalo owners and breeders, as well to manage his herd with rotational grazing and electric fencing. In 2010, Mr. Bruyn became the founding president of the Paraguayan Association of Buffalo Breeders (APACRIBU).

For many years, buffalo in Paraguay was raised in medium or large farms, mostly without management and intentionally limiting their increase in number. They were kept as something exotic and/or to cover the internal meat required by estancias. The animals were of the Jafarabadi breed or crosses of this breed with Mediterranean and/or Murrah (Zava, 2007). In extensive conditions, herds often had little human contact, without proper management, so the animals became feral and gained a reputation of being untameable (untamed). To some extent this explains the relatively small herd in spite of all its benefits and of having been introduced to the country almost 70 years ago.

PRESENT

According to official figures from the National Animal Quality and Health Service (SENACSA) in 2022, Paraguay registered a bovine herd of 13,573,375 head and a buffalo herd of 13.125 head with 267 producers distributed throughout the national territory. Buffalo is primarily in the eastern region with 71% of the herd, while the western Chaco region which registers 29%. The departments with the largest buffalo population are Presidente Hayes, San Pedro and Paraguarí. (Table 1)

Table 1: Holders of buffalo cattle, number of establishments and buffalo population by department. Year 2022.

HOLDERS OF CATTLE, ESTABLISHMENTS AND BUFFALO POPULATION BY DEPARTMENT 1ST PERIOD YEAR 2022. SENACSA - PARAGUAY				
DEPARTMENT				
CODE	DESCRIPTION	HOLDERS OF CATTLE	ESTABLISHMENTS	BUFFALO POPULATION
1	CONCEPCION	7	7	48
2	SAN PEDRO	18	17	2.704
3	CORDILLERA	12	12	535
4	GUAIRA	1	1	2
5	CAAGUAZU	33	33	698
6	CAAZAPA	34	33	693
7	ITAPUA	14	14	149
8	MISIONES	9	9	963
9	PARAGUARI	26	26	1.252
10	ALTO PARANA	13	13	496
11	CENTRAL	8	8	298
12	ÑEEMBUCU	20	18	714
13	AMAMBAY	10	9	274
14	CANINDEYU	25	23	544
REGION ORIENTAL		**230**	**223**	**9.370**
15	PRESIDENTE HAYES	20	18	3.201
16	ALTO PARAGUAY	6	7	287
17	BOQUERON	11	11	267
REGION OCCIDENTAL		**37**	**36**	**3.755**
TOTAL		**267**	**259**	**13.125**
Based on data of the first vaccinaton period in 2022 for foot and mouth disease				
Cut off date: April 22,2002				
Source: SIGOR database				
Prepared by Technical Coordination - DPE - DIGESTEC.SENACSA				

Of the 267 small, medium, and large producers dedicated to raising buffalo in the country (Figure 1), 80% are doing so mostly in marginal, difficult low and wetlands, primarily in the departments of Ñeembucú, Paraguarí, in the lower part of San Pedro and a significant amount in the Lower Chaco (Frutos, 2021).

The demand for buffalo meat and its derivative products has been growing steadily, but growth is limited by the supply side. Despite this, currently it can be found in delicatessens and specialty supermarkets, in various presentations of premium cuts such as rump steak, rib eye, tenderloin, sirloin, as well as hamburgers and sausages. One can also find buffalo hamburgers on menus in specialty restaurants. Dairy products such as milk, yogurt, and a variety of buffalo cheese, such as Paraguay, Mozzarella and Burrata, is produced in small quantities (Frutos, 2021).

FUTURE

In 2021, at a meeting of the American Federation of Buffalo Breeders held in Bogotá, Colombia, Paraguay was chosen as the venue for the X Symposium on Buffaloes Breeders of the Americas and Europe, to be held in November 2022. Consequently, the President of APACRIBU, Richard Moss Ferreira, became President of the American Federation of Buffalo Breeders.

The Symposium will be of fundamental importance for the growth of buffalo farming in Paraguay, since these international events are attended by producers, technicians, researchers, presidents of buffalo associations, companies, journalists, among others, from America, Europe and Asia, and will facilitate the exchange of experiences between national producers and the international delegates, and where the species and herd management techniques will be discussed, as well as the production of products derived from buffalo milk and meat.

Figure 1: Buffaloes in Paraguay (Photo Ross)

More importantly, throughout the year it will provide opportunity to bring attention to inform and educate in a manner to overcome prejudice and draw focus on the opportunities and management skills available to turn marginal lands into areas of sustainable human development, and Paraguay into a key player in the development of the regional Buffalo industry.
The opportunity is enormous; with 9.3 million hectares of marginal wetlands and estimating to allow up to 4 hectares per buffalo for sustainable buffalo farms, a population of 2.5 million head could be sustained.

Regarding the relationship between wetlands and buffaloes, researcher Gerardo Barboza Jimenez in his work in Costa Rica mentions that the water buffalo is an animal that has been and can be used successfully for grazing in wetlands since it is the herbivore with better anatomical and physiological characteristics and with the greatest potential for vegetation management in tropical and subtropical wetlands, as it can move easily in humid and muddy terrain, it can even swim, thus using areas where cows and horses will not enter, lest they get stuck . The buffalo remove the biomass of invasive plants from the wetland and exposing the water mirrors allowing the regeneration of a diversity of plants, seeds and small organism which are food sources, attracting aquatic birds, increasing biodiversity and abundance significantly, all while the buffaloes improved their productive and reproductive conditions. His studies conclude that rotational grazing with water buffaloes is an activity of great interest for the management of invasive vegetation in tropical wetlands and for buffalo production, as the animals develop well and produce milk and meat of high ecological quality while performing the function of a biological machine that cleans the biomass of the wetland, making this a relevant and interesting model for conservation and production and as a management strategy against climate change. In short, the water buffalo can have attributed the function of rehabilitating wetlands.

Also, in a work on wetlands in the islands of the Argentine delta, the authors Travaini, A.; Astrada, E., Cadoppi, A. (2019) highlight that the buffalo species is adapted to life in wetlands and that by adjusting the loads and managing the receptivity of the environment it is possible to achieve a sustainable buffalo production in an environmentally friendly manner, contributing to the maintenance of the integral ecology

of wetlands. The buffalo takes advantage of low and flooded fields, marshes, estuaries, shallow lagoons, and alluvial plains among others. Natural wetlands are put into production without the heavy modifications for water management required for other livestock species, agriculture, or urbanization. The wetlands have the advantage of having many low fields, with estuaries and embankments, where the buffalo is very well adapted. In the summer it is observed how they are constantly submerged in the estuaries, even feeding on water hyacinth (camalotes) and other aquatic plants. During the winter, with the low temperatures in the area, the buffalo generally seek higher areas, sheltered from the cold and feeding on the natural vegetation of the hills. It is also observed that they approach the estuaries to feed and drink water. For these reasons, an excellent coexistence of the buffalo with the surrounding environment is expected. The great capacity of the buffalo to convert fiber into energy is very important in this area, where in the estuaries there is an abundant supply of crude fiber, such as reeds and grasslands. These forages, which cattle does note consume, will benefit buffalo, allowing them to obtain astonishing rates of daily gain. The study concludes that the buffalo is a very interesting alternative for the wetlands of the Paraná Delta, achieving a perfect adaptation to the area, with excellent productive results. (Steverlynck, 2014).

Due to these characteristics, the buffalo can be considered a great “instrument” for regenerative and sustainable livestock farming in Paraguay. Its rusticity permits benefiting from underutilized surfaces and with rational management schemes, its trampling, grazing and dunging, combined with rest periods, stimulate the regeneration of plants and soils, and through this, favoring biodiversity, growth and carbon sequestration.

CONCLUSIONS AND SUGGESTIONS

If we take as a reference the official buffalo population according to SENACSA of 13,125 heads for the year 2022, and we consider the figure of 13.1% annual population growth of buffalo in Argentina for 10 years (Crudeli, 2011) and extrapolate it to Paraguay, a buffalo population of 50,147 heads would be reached in this country in the year 2032.

This figure is low in relation to the previously determined potential, and we propose public private cooperation to take the following relatively simple measures to foster the development of the species in Paraguay.

1. Stimulate the importation of live animals from neighboring countries,
2. Encourage the retention of 70% of heifers and
3. Educate for herd management that obtains the yearly pregnancy rates expected for this species (80% and above) and weaning rate of 70%

These relatively simple changes would contribute to creating a critical mass sufficient for Paraguay to use and develop these wetlands in an environmental, social, and economic sustainable manner

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Figure 2: Buffaloes in Paraguay (Photo Ross)

FORTY YEARS OF BUFFALO ARTIFICIAL INSEMINATION IN THE AMAZON VALLEY, BRAZIL – An HISTORICAL RETROSPECTIVE

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ABSTRACT

This review shows a forty-year retrospective and diachronic view of the AI in Amazon valley from the establishment of deep-freezing semen protocols to the application of AI on natural or synchronized estrous. The work carried out a systematic review of the existing literature on Fixed Timed Artificial Insemination (FTAI) in buffaloes at regional level, indicating new directions for future investigations. The adopted strategy included studies published since the introduction of AI at Amazon valley, which utilized more than 10 animals, and reporting the applied protocols. A careful analysis of the quality and robustness of the literature related to the use of FTAI was made through the selection of monographs, dissertations, thesis and abstracts published in national and international congresses, specialized journals, government institutions websites and through personal unpublished data obtained from specialists who work with buffalo reproduction of at Amazon valley. A total of 35 works developed in the Amazon Valley using different protocols are reported. A total of 8.573 female buffaloes, crossed between Murrah and Mediterranean breeds, were submitted to FTAI protocols within the six states of regional Amazon. Among all protocols the Ovsynch and the DIP+EB/PGF2+eCG/GnRH/FTAI were the most used.

Key words: Buffalo, Amazon valley, Breeding Technologies, Fixed Timed Artificial Insemination.

1. Introduction

The Amazon valley, Brazil, is part of the continental Amazon region which has border with seven Latin American countries. The region covers an area of 3,500,00 km^2 and has as become a self-sustainable area of agricultural and livestock farming production, where buffaloes have a great importance in food security of local humans' inhabitants and play an important socio-economic role.
The first unofficial reports on the introduction of buffaloes in Brazil was done in 1892 by the Marajó island, the largest fluvial-marine island of the world, today sheltering the largest buffalo population in the Americas continent (around 1 million of heads) mainly of Italian and Indian origin. Currently, the species is distributed throughout the national territory, where it is inserted in animal traction activities, meat and milk production.

The most common breeds in the region are: Murrah, Mediterranean, Jafarabadi and Swamp type - Carabao, (Vale, 1996; Zava, 2009; Vale et al., 2013). In general, there are four different buffalo production system in the Brazilian Amazon: 1. Using native flooded pastures on Marajó Island; 2. Native flood plain pastures in the Amapá state, Lower and Middle Amazon regions; 3. Native pastures in dryland mixed with flooded areas in the Northern of Pará state Baixada Maranhense; and 4. Cultivated pastures in dryland, in some areas previously used for cattle or for agricultural purpose.

In this scenario buffaloes represent an important alternative source of animal protein, not only as they have the intrinsic dual ability to produce healthier meat and milk, but also because they have an excellent feed conversion, making better use of native pastures of low nutritional value, in areas that are difficult

to be used by other domestic animal species. Actually, the Brazilian Amazon valley has stood out in the production of cheese and beef production, achieving national and international awards, proving that buffalo breeding is a highly advantageous economic option when compared to other domestic species. Indeed, this progress can only be achieved due to the introduction of several technological inputs such as nutrition, management, and the use of artificial insemination by semen with recognized genetic potential for milk production that provide a short-term genetic improvement and increased production and productivity for many farm herds.

Thus the aim of this paper is a retrospective and diachronic view of the AI in buffalo species raised in the Amazon valley where we sought to show the beginning of deep freezing semen process, observation on the reproductive behavior of females through the estrous cycle, constrains existent in the reproductive management and finally set up the process of AI in buffaloes at regional level, performing a systematic review of the existing written bibliography, indicating new directions and highlights for future investigations.

2.The primordial era of the AI in Latin America

In Latin America the first attempts to use AI in buffaloes were done in the Amazon valley, Brazil. In the begin of 1980, Vale and collaborators at Animal Reproduction Laboratory of the Faculty of Agricultural Sciences of Pará (FCAP), currently UFRA, and Universidade Federal do Pará (UFPA), Belém, Pará state, Brazil, with the guidance of Prof. Dr. Hans Merkt and Prof. Dr. Anne-Rose Guenzel from the Clinic for Andrology and Artificial Insemination of the Hannover Veterinary College in Germany, started a "*pilot program*" for deep freezing semen of regional buffalo bulls.

Thus, Brazil had the priority in the development and practice of AI in buffaloes in whole America's continent which began in the 80s of last century when Vale et al. (1984) using the diluters TRIS and TES reported the successful deep freezing of semen and the perform of first AI using frozen semen, obtaining indexes upon 50% of fertility based in non-return rates (NRR) at 60 days post insemination. Later, superior indexes upon to 70 per cent of calving were obtained by the same authors. The practices were spread out to other regions of Brazil and Latin America countries (Vale et al.1984; Vale, 1994b).

Therefore, the practice of AI in Latin America has not been so widely adopted for buffaloes as for some other species of farm animals, especially for bovine. The reasons for this are several, probably the most important is related to the lack of support by the local governments assessing the performance of existing AI programs for small-holders and medium farmers. Thus, it is necessary to identify constraints and formulate assistance to implementation of remedial measures including appropriate strategies, establishing sustainable routine of deep-freezing semen programs and supporting services to farmers and harmonizing Veterinarian managerial and field practices and sharing of expertise within the region.

3. Basic necessary requirements for the success of AI in buffaloes

After the begin of this century, buffaloes have steadily gained acceptance as an alternative to cattle, and indeed, throughout South America buffalo perform favorably in both meat and dairy production. Ranchers were impressed by their resistance to common bovine diseases, superior weight gain than cattle, high quality in dairy and meat products, and the ability to fatten on a wide range of grasses. These characteristics and reputation are part of the reason that buffalo herds in the Brazilian Amazon have increased at a rate of nearly 13% per year.

Before programming any reproductive events in buffalo, it is essential to know the specific reproductive characteristics of this species. Buffaloes reach puberty and sexual maturity a little bit later than cattle, however with good management practices buffalo heifers of Murrah, and Mediterranean breeds can reach the puberty between 13-15 months of age, and it is recommended to inseminate heifers at this age if they have more than 320 kg of body weight (Vale et al. 1997; Vale, 2000).

Although buffaloes are considered a short-day seasonal polyestrous breeder animal, like equine, sheep and goats, in the equatorial zone like Amazon valley, buffaloes present themselves as continuous polyestrous breed, since the reproductive activity is subjected to nutrition and management. Differently, in the central-south regions of Brazil, where there is an annual variation in the duration of light hours, depending on the season of the year, a greater concentration of estrous manifestations in female buffaloes is observed in the autumn and at the beginning of winter period (Vale et al. 1990; Baruselli, 1993; Vale, 1994a; Vale & Ribeiro, 2005). The buffalo estrous cycle has a length that varies between 18 and 32 days, with an average of 21 days. The duration of the estrus varies between 5 and 27 hours, with an average of 20 hours, with ovulation between 24 and 48 hours after the start of the heat (Vale et al,1984).

4. Management and nutrition

Normally, one of the important points in buffalo production system for novice farmers that start to use AI, is provide a sustainable pasture production system linked with forage production of nutritive value, stocking rates, stocking methods, and opportunities to match forages according to animal requirements for reproduction and production. However, in the majority of buffalo farming systems in Brazil, the native grass production system is nearly favorable for the time of calving buffaloes as usually occur in the floodplains areas of Amazon River and its tributaries. When the flood occurs, it deposits a layer of fine soil, rich mineral salts incorporated in silt sediment which is spread across a wide area of the riverside banks. These sediments make the soil very much fertile and lead to the formation of a very flat fertile flood plain area. Grasses of the family *Poaceae* sp. are the main members of the vegetation stratum herbaceous from alluvial soil native pastures. It is in this stratum that cattle herds and buffalo graze with greater intensity when the water level allows. Most are hydrophyte species (“amphibians”) that vegetate floating or even submerged in water, during river floods, for more than four months; these species belong to the genera *Echinochloa sp, Oryza sp, Leersia sp, Luziola sp and Hymenachne sp.* (Camarão et al. 1997).
Recently many farms are paying attention to the production and reserve food for animals used in FTAI programs. Notwithstanding we have been promoting the importance of feeding on the fertility of herds and many farmers are producing food, such as the *Pennisetum purpureum* varieties Elephant and BR Cupiaçú grass, sugar cane *Saccharum officinarum*, cassava *Manhiot sculenta*, *Sorghum sp* and corn *Zea mays*. However, higher levels of productivity are achieved with food supplementation of by-products with macro and micro minerals, according to local deficiencies.
Female buffaloes, without adequate food, does not come into heat and, when it does, has a reduced conception rate due the impossibility to carry on a normal pregnancy (Vale, 1996). Moreover, due to the erroneous view that most of the breeders have about the rusticity of the species, meat, milk and fertility really decrease when the herd have precarious offer of food (Vale et al. 1990).

The number of animals per hectare varies depending on the location of the farm, soil fertility, quality and type of pasture, property management, etc. Likewise, overcrowding of pastures has a negative effect on fertility. It is common to find properties with a greater number of animals than their stocking rate capacity. The rainy season normally offer to buffalo and cattle herds a richer food (Vale, 2007; Rolim Filho et al. 2009).
Indian researchers (Bhalaru et al., 1987) observed that the body status of buffaloes interfered with the onset of heat and the fertility indexes obtained with AI. Buffaloes that were thin at the time of birth took a long time to manifest heat and had reduced fertility when compared to others in good body score condition (BSC). Animals that lose a lot of weight after giving birth also have a drop in fertility. Consequently, the good nutritional status of the herd is fundamental for the success of the reproductive management in buffalo.

5. Body Condition Scoring (BCS) at calving and at insemination.

The Body Condition Scoring (BCS) at calving is pivotal for the selection and fertility success of females used in FTAI programs because it influences the subsequent interval from calving to first estrus (ICFE) and conception rate (CR). Farmers must aim to have cows in an optimal BCS between 2.5 and 3.5 (based on a scale of 1-5) and to minimize loss of score between calving and insemination (Vale, 2007). Therefore, it seems obvious that buffalo cows that are too fat at calving are likely to have calving difficulties and are more prone to early fetal absorption or death as well as, females that are too thin, especially if they are losing body condition, will have delayed estrus and poor CR (Figure 1).

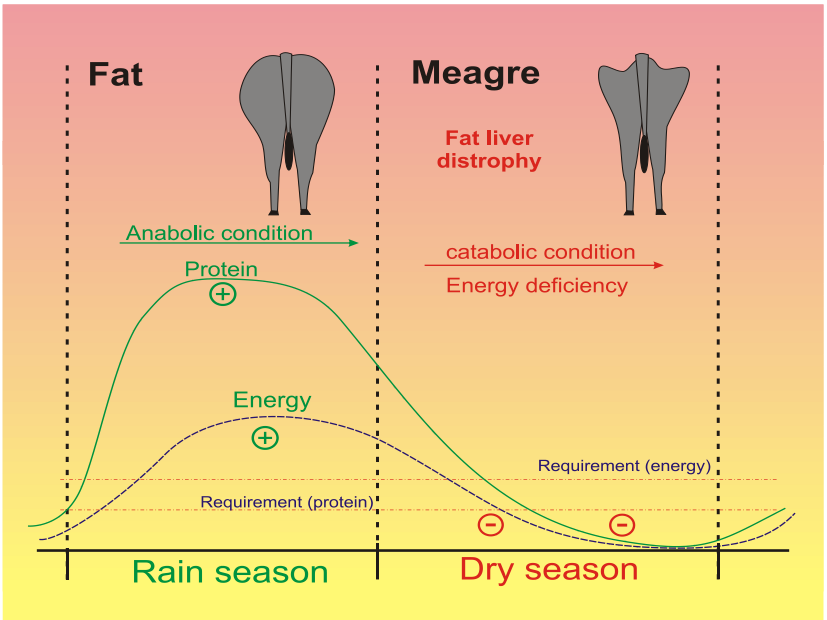

Figure 1 - Presentation of nutritional *status* and body score condition (BSC) of buffaloes raised in the Amazon Valey. The differences in the rain season – anabolic condition due to larger offer of fodder and in dry season – catabolic condition due to shortage of fodder- are shown. Vale (2007).

Other factors to be considered before FTAI, should be the total uterus involution. Buffalo female usually have uterus involution between at least 20–30 days if the parturition was normal without trauma. Then, after calving, a clinical examination to check uterine condition is an important tool, before they are served again. For high yielding buffaloes, if the BCS is lesser than 2.5, a longer period may be necessary to obtain a good CR and to reduce embryo and early fetal losses (Vale et al. 1986). On this concern buffalo cow should be in good health, specifically, she should be free of any evidence of infection of the reproductive tract.

Particular attention should be paid to females that have had abnormal calving (e.g., dystocia, retained placenta and prolapse of cervix or uterus), as they may require a longer period after calving for involution of the uterus and to return to normal fertility. The case that natural estrous is used, AI technicians must be sure that the buffalo is genuinely in estrous and that she is not pregnant confirmed by a veterinarian. In addition to the aspects of disease control that affect buffalo females, very important are the pathologies that often affect the genital tract like plague, endometritis and metritis which should be treated clinically (Vale et al. 1988).

6. Data analysis

36 buffalo farms localized in six states of Amazon region were used as a base to present the results of this paper in Parà state (15 farms), Amapá state (10 farms), the others in Amazon and Maranhão state and in Rondônia and Roraima state (Figure 2)

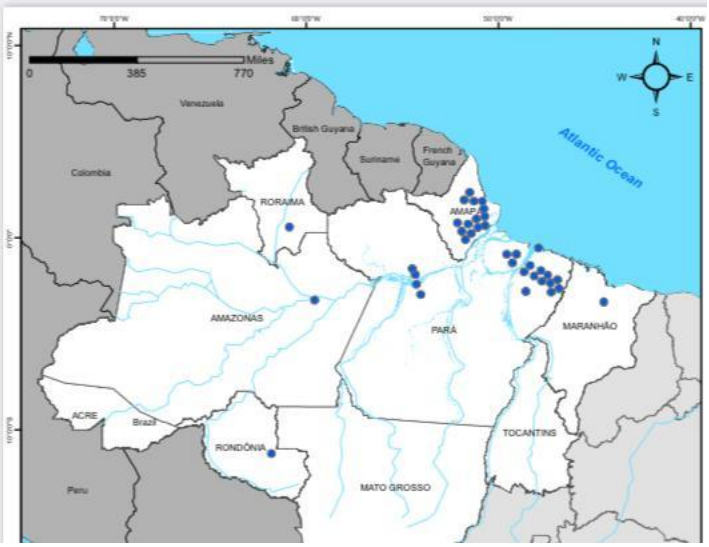

Figure 2 - Presentation of the map of the Amazon region with the location of buffalo farms where AI programs were used. In some farms the practice of AI has been used more than twice and have already become routine.

6.1 Location

The survey was performed at 36 farms located in different Amazon states, Brazil, and it was a representative buffalo commercial farm model for the region. The climate is characterized by a humid tropical, with a temperature oscillation from 20 to 38 C° and an air relative humidity of 80-95 per cent and rain fall average of 2.200-4.000 mm per year. Usually there are two main seasons, a rainy one when Amazon River and its tributaries are high flooding large areas of the Amazon basin and the dry or go out season time one, promoting the formation of large floodplain areas covered with abundant and rich native pasture used for buffalo graze.

6.2 Animals selection and management

Female buffaloes mixed graded of Murrah x Mediterranean breeds, pluriparous, from 4 to 12 years old, lactating and calved for more than 30 days postpartum (DPP) and the body condition score (BCS) equal or >3.0 were used. BCS was recorded on the day of enrolment and 45 and 60 dpp (days postpartum) according to Edmonson et al., (1989) where 1.0 was thin and 5.0 obese. Animals were subjected to a gynecological examination (rectal palpation of the ovaries for follicles, corpus luteum, uterine contraction and presence of uterine fluid) and some herds underwent sequential trans-rectal ultrasonography using ultrasound equipment from different origins and brands to assess the presence and morphology of follicles and corpus luteum (CL) in the ovaries.

The management semi-intensive native pasture in the floodplain areas between July to February and between March to June in artificial pasture of *Brachiaria humidicola* and *Brachiaria decumbens* grass under semi-intensive grazing system corresponding to the flooding season of the Amazon River. Access to water was done free in artificial tanks, lakes and natural lagoons for drinking and bath. At 4:00 PM the animals were brought to a yard when received a free stall supplement *ad libitum* of chopped Elephant grass (*Penisetum purpureum*) mixed with a soyabean bran 16 per cent of crude protein/dry matter composing 10 per cent of the whole supplement. At 5:00 AM animals where milked and then release for the pasture. Between 4:00 PM and 5:00 AM, if necessary, animals were subjected to successive gynaecological examination and submitted to intensive observation for oestrus detection by teaser bulls with a chin-ball device.

All groups have free access to mineral supplement performed throughout the whole year, consisted in a commercial mixture with normal Ca:P relationship and macro and micro minerals which was offered *ad libitum* in artificial trough.

Animals were subjected to the health program of the farm consisting in vaccination against Food and Mouth Diseases, Brucellosis and Rabies in areas subject to the presence of hematophagous bats and annually tested against Tuberculosis (TBC), Food and Mouth Diseases (FMD) and dewormed.

7. From conventional AI to FTAI

7.1 Phase I

Conventional AI had its first test in the Amazon Valley in the early 1980s, through a research work carried out by the Animal Reproduction Laboratory of the Faculty of Agricultural Sciences of Pará (FCAP), currently UFRA, Belém do Pará, in partnership with the Hannover School of Veterinary Medicine – Federal Republic of Germany – and the breeders Mr. Liberato de Castro (owner of the Itaqui and São Joaquim Farms) and Mr. João de Almeida Moreira (owner of Fazenda São Marçal) who ceded the infrastructure, leaving their animals available for carrying out the experiments.

Many experiments were carried out until a successful field semen cryopreservation of the Murrah bull called “Rajastan II” was achieved on the Itaqui Farm, located 70 km from Belém. Using an egg yolk-based extender and TES/TRIS as buffer substance, researcher team of FCAP, obtained satisfactory results in field tests that resulted in the first buffalo calves generated by mechanical deposition of semen in the uterine body, in 1982, at Fazenda São Joaquim, located in the central region of Marajó. This project is considered a pioneer in Brazil and Latin America and had repercussions throughout the world. At São Marçal farm, some calves were also born generated by AI, using semen of the Carabao breed, donated by the FAO (Food and Agriculture Organization of the United Nations) and imported from Germany through the Ministry of Livestock Agriculture and Supply (MAPA).

In the early 1980s, Vale et al. (1984), carried out a study at Fazenda São Joaquim, in Santa Cruz do Arari – Marajó. 25 lactating buffalo females, crossbred between the Murrah and Mediterranean breeds, maintained in extensive regime with native pasture in a floodplain area, underwent a complete gynaecological evaluation, 15 were detected with uterine infection problems being discarded, and 10 were selected to be artificially inseminated. In these selected buffaloes, 2ml of PGF2α (D-cloprostenol) was applied on the day of evaluation, with estrous observed at 6:00, 12:00 and 18:00 hours, for three days. The 10 selected females were observed until the manifestation of estrous signs, inseminated soon after. All were inseminated until 72 hours after the application of PGF2α, with frozen semen processed at FCAP´s Animal Reproduction Laboratory. The greatest manifestation of estrous with uterine contraction, characteristic mucus discharge, and vulva swollen occurred between 48 and 72 hours after IM (Intramuscular) administration of PGF2α. The pregnancy rate was 60% (06/10), diagnosed by radioimmunoassay (RIA) at the Hannover Veterinary College-Germany. However, in the following year four calves were born, resulting in a calving rate of 40%. It was supposed that there was embryonic loss or abortion in some buffaloes which were detected pregnant.

7.2 Phase II

Using semen collected from a Murrah sire of good genetic history, which was processed in regional laboratory installed previously on the FCAP campus, in 1986 our research group started to work at Jari Enterprise.

Jari Enterprise was a mega farming project owned by an American entrepreneur Keith Daniel Ludwing where there were 15,000 buffalo heads, bred by selecting female buffalo for the program of AI. There female buffaloes were selected among the best existing dairy buffaloes in the different farms alongside the banks of the Jari River called locally “*retiros*”, whose milk produced was already sent to human consumption within the project area. In addition to the daily milk production performance, the buffalo females were subjected to general clinical examinations, with an emphasis on the genital tract followed by an examination of brucellosis and tuberculosis.

The existence of pasture cultivated with different types of *Brachiaria* sp., including *Brachiaria humidicola*, and rich native pasture in the floodplain as well as an additional supplementation of chooped Elephant grass (*Penicetum purpureum)* and a commercial mineral mixture offered *ad libitum* in multiply tough, allowed to increase the quality of the food offered to the herd.

A group of female buffaloes selected through the phenotype and milk production record, and free of brucellosis and tuberculosis, were submitted to the trial. Estrus was detected by visual observation and with the use of teaser bull using a chin-ball device with ink for identification when it mounts the female and was checked two times for day (06:00 and 18:00), for periods of one hour observation; started 48 hours after the use of PGF2α and was recalled and ended up to the moment when AI was performed. The pregnancy diagnosis was made by transrectal palpation 60 d after the AI. The results were tabulated in the table 1.

Table 1 - Conception rates obtained in an artificial insemination program in buffaloes in the Jari Project farms, State of Pará.

Year	Number of buffalo females	Number of AI performed				Doses per conception	Conception rate (%)	
		1.	2.	3.	Total		n.	%
1986-87	42	42	27	10	79	1.88	25	59.0
1987-88	64	64	47	37	148	2.31	29	43.3
1988-89	70	70	21	6	97	1.38	47	67.1
1990-91	95	95	44	15	154	2.44	64	67.3
1991-92	105	105	58	21	184	1.44	73	69.3
1992-93	98	98	24	11	133	1.92	69	70.4
1993-94	94	94	35	07	136	1.97	69	73.4
1994-95	156	156	78	24	258	2.03	127	81.4
Total	724	724	334	131	1189	1.92	503	64.4

Of the total of the buffaloes inseminated n=724 in seven years, conception rates vary between 43,3 to 81,4 per cent with a total average of 64,4 per cent (table 1). It seems that these variations were related to the management, feeding, insemination technician skill in handling the semen and even semen quality.

The nutrition was of course a factor that has caused some problems mainly in the years 1987-88, due to a great flood of the Amazon River and its tributaries, which caused some shortage of grass in the areas of pasture. After this gap it seems that the nutrition problems were overcome when supplemented fodder with the addition of rice and corn bran and better-quality artificial pasture were used. Furthermore, during the AI season there were always a veterinarian and students following the process.

The overall results were considered very good and after this experience it was concluded that AI was a feasible technique to be used as an important tool in the genetic improvement of buffaloes in the Amazon region.

7.3 Phase III

This phase was carried out at Fazenda Carabao, belonging to the Baraúna Pinheiro family, located in the district of Novo Remanso, municipality of Itacoatiara, State of Amazonas. The farm is located on the banks of the Amazon River, with an immense area of lowland and floodplain rich in grasses and legumes, which pulses between the months of September and March, as well as areas of artificial pastures of *Brachiaria humidicola* and *B. byzantha* in normal areas of dry land.

During the period 2004-2006, 426 crossbred female buffaloes between the Murrah and Mediterranean breeds, aged between 4 and 12 years, were selected. These animals were submitted to tuberculosis and brucellosis tests and became part of three distinct batches according to the description in Table 2. Furthermore, in the table 3 it can be observed the experimental groups, number of animals used, and treatments performed in the different experiments.

Table 2 - Experimental groups, number of animals and treatments Performed in the different groups.

Groups	Number of females	Treatments
G-1	**121**	Ovsynch (D_0) GnRH+(D_7) $PGF_{2\alpha}$ (D9) GnRH (D9)+(D10) FTAI
G-2	**121**	CL-synch (D_0) $PGF_{2\alpha}$ +(D_2) GnRH+(D_3) EB +FTAI (D4)
G3 Control	**184**	AI 12 hours after heat detection
Total	**426**	

In the G-1 group, independent of the day of oestrus cycle, every female received the Ovsynch protocol [Ovsynch (D_0) + GnRH+(D_7)[1] + $PGF_{2\alpha}$ (D_9)[2] + GnRH+(D_{10}) + FTAI] consisted of an intra muscle (IM) of the first dose of GnRH, (Lecirelina) (1ml/25µg), at the day seven, after a IM dose of $PGF_{2\alpha}$ (D+cloprostenol), (2ml/150µg) and GnRH, (Lecirelina) (1ml/25µg), in the ninth day followed by a FTAI at 10 days.

The G-2 was assigned with the Cl-synch protocol [CL-Synch (D_0) + $PGF_{2\alpha}$ (D_2) + GnRH (D_3) + FTAI consisted of an IM dose of $PGF_2\alpha$ (D+cloprostenol), (2ml/150µg) at the day two, an IM dose of GnRH (1ml/25µg) at day three, followed EB (Estradiol Benzoate) and FTAI after 24 hours.

For Group 3, it was subjected to artificial insemination after 12 hours of a natural estrous affected by a teaser buffalo bull with a chin-ball device and cowboys' intensive visual observation with a second AI performed if the female came in estrous after a negative pregnancy diagnose. Data on the presence of vaginal mucus discharge and tonus (uterine contractility), presence of oestrus symptoms at the moment of the FTAI as well as the relationship between BCS and pregnancy rates were collected for the G1, G2 and G3 for pregnancy diagnose.

Results

On 426 buffaloes used in the last trial, 311 (73,0%) became pregnant. Therefore, the pregnancy rate for the 242 female buffaloes subjected to synchronization in the two groups was 167 (68,9%), for the Group I-Ovsynch, 79 (65.2) and Group II-Clsynch 88 (72.7%) whereas for Group III inseminated in natural oestrus with one or second artificial insemination were 105 (57,0%) and 39 (52,0), respectively (Table 3).

The service period rates in the groups submitted to synchronization were 100 per cent as a consequence of the adequate feasibility of the management proportioned by the use of this assisted reproductive technologies which took only ten days to perform the whole work meanwhile in the control group with natural service and AI took 120 days. The overall results here obtained 73,0 per cent of pregnancy rates, can EB considered very good when compared with the literature which show a variation between 30 to 75 per cent at Ovsynch and its similar synchronized protocols, with or without the utilization of steroids.

Table 3 - Conception rate in an AI program according to the protocol used.

Groups	Number	Conception rate (%)
Protocol I- Ovsynch	121	79 (65.2)
Protocol II- CLsynch	121	88 (72.2)
Sub-total	242	167 (73.1)
Protocol I- Ovsynch - 2nd AI	39	29 (74.3)
Protocol II- CLsynch - 2nd AI	32	26 (81.2)
Sub-total	313	55 (77.4)
Group III: 1st AI	184	105 (57.0)
2nd AI	75	39 (52.0)
Sub-total	259	144 (55.5)
Sub-total for the overall AI	572	366 (63.9)
Total	663	452 (68.1)

7.4 FTAI trials in 3 states of Amazon region and in Marajo Island

Recently, Rolim Filho et al (data not published) found conception rates between 44,4 to 54,1 per cent though the use of FTAI in different herds raised in extensive farming management, in three different states of Amazon region. The same protocols were used and based in the association of five types of commercial hormones. Also, it demonstrated the efficacy of GnRH on the fertility of buffalo females subjected to FTAI, managed in range condition in different areas of Amazon region (Table 4).

Lima (2021) evaluate the influence of the dominant follicle (DF) and the use of GnRH at the time of insemination and pregnancy rate through FTAI. A total of 384 multiparous buffalo cows with 30 to 60 days postpartum, predominantly Murrah breed with body score 3.4±0.6, were used.

The synchronization protocol consisted of D0 intravaginal device 1g of P4, application of 2.0 mg of Estradiol Benzoate (EB) and administration of 10 ml of injectable mineral supplementation. On D9, also at 4 pm, the device was removed, application of 0.265 mg of PGF2α and 300 IU of eCG was held. On D11 at 4 pm, 0.1 mg of GnRH was administered. On D12, 16 hours after the application of GnRH (at 8 am) artificial insemination was performed and the 4 groups were separated according to the diameter of the DF.

Group 1: Buffaloes whose dominant follicle was ≥ 13mm with application of 0.1mg of GnRH.
Group 2: Buffaloes whose DF was ≥ 13mm without application of GnRH.
Group 3: Buffaloes whose dominant follicle was < 13mm with application of GnRH.
Group 4: Buffaloes whose dominant follicle was < 13mm without application of GnRH.

The overall pregnancy rate was 53.9 per cent (207/384, table 5). Female buffaloes with DF greater than 13 mm had a higher pregnancy rate ($P<0.05$) than those with follicular diameter below 13 mm; overall, there was no significant difference in the group that received GnRH at the time of insemination compared to the group that did not receive it; considering only the animals that had a follicle <13mm, the treatment with GnRH at the time of insemination had a higher rate than the untreated group ($P<0.05\%$). Therefore, it can be concluded that the use of GnRH at the time of artificial insemination can be a tool to improve the results of the FTAI protocol in buffaloes when applied in specific situations.

Table 4 – Conception rate of buffaloes through FTAI raised in different states in the Northern Region of Brazil.

Local	Year	Protocol	Female number	Conception rate (%)
Amapá state	2019	Progesterone + EB + eCG + PGF + GnRH	85	48,2% (41/85)
Amapá state	2020	Progesterone + EB + eCG + PGF + GnRH	125	54% (65/125)
Marajó island / Pará state	2020	Progesterone + EB + eCG + PGF + GnRH	384	53,9% (207/384)
Maranhão state	2020	Progesterone + EB + eCG + PGF + GnRH	108	44,4% (48/108)
Marajó island / Pará state	2021	Progesterone + EB + eCG + PGF + GnRH	132	51,5% (68/132)
Santa Maria County Pará state	2020	Progesterone + EB + eCG + PGF + GnRH	90	45,5% (41/90)
Santa Maria County Pará state	2021	Progesterone + EB + eCG + PGF + GnRH	85	54,1% (46/85)

Table. 5 -. Conception rate and GnRH effect at the time of the FTAI of buffaloes raised on Marajó Island

GnRH effect on pregnancy	Pregnant	Open
With GnRH	56,5% (78)	43,4% (60)
Without GnRH	52,4% (129)	47,5% (117)
Total pregnancy	53,9% (207)	46,1% (177)

P=0,44

Table 6 - Influence of the dominant follicle (DF) diameter on the conception rates of buffaloes submitted to Fixed Timed Artificial Insemination (FTAI) at the Marajó Island.

Effect of folicular diameter	Pregnant	Open
>13 mm	59,6% (111)[a]	40,3% (75)
<13 mm	48,4% (96)[b]	51,5% (102)

P=0,0279

8. Conclusions

After 40 years, since the pioneering work of AI began in the State of Pará, Brazil, we can say that many positive and progressive results have been made. These achieved progresses were only possible due to the saga of a group of veterinarians, most of them still students, who allied themselves and who, despite the difficulties, were able to overcome natural and human obstacles that got in the way of this victorious march. Thus, basic studies were started on the morpho physiology of the female genital tract, collected from slaughterhouses, studies of the causes of infertility in buffalo herds, especially in animals from the Marajó island, where the feeding requirements are not often satisfied.

On the contrary, in herd for which reasonable pasture were available all year and mineral supplements were provided, calving occurred throughout the year, but were more frequently between November and

March. Furthermore, studies on the puberty of female and male in buffaloes, showed the important aspect on the parameters never studied before at regional level.

Following these first steps, everything was prepared to start studies on deep freezing the semen of buffalo bulls at a regional level and for this we have great support from the Hannover Veterinary School, in Germany, through the person of Prof. Dr. Hans Merkt and Prof. Dr. Anne-Rose Guezel, who left us vast knowledge, which provided the possibility of freezing buffalo semen, with the state of Pará being the pioneer in this technology in all the Americas. However, none of this could have been done without the strength of the team and here we want to refer to the person of two pioneers, Prof. Otávio Mitio Ohashi and José Silva de Sousa, who left us early, but who will forever be revered as an important piece of this historical epic. Now, the new generation of Veterinarians not only in Amazon region, but in all over the Brazil and Latin America are using these new technologies provided by veterinarians native from Pará state, Amazon region.

In memory of Ohashi and Sousa, without a doubt, the great merit of this victory belongs to you...
Floreat bubalus !!!

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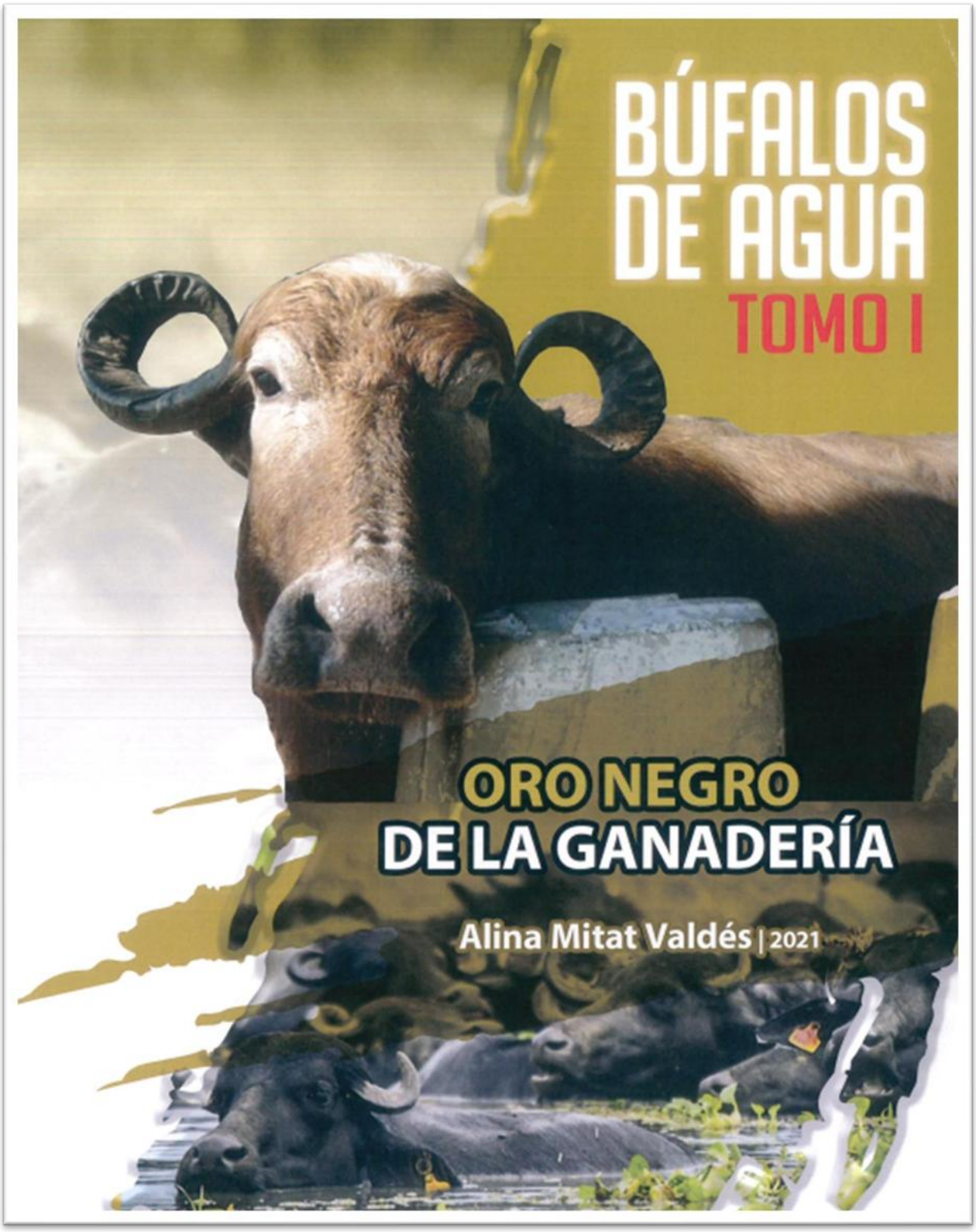
FIGURE 3 BUFFALOES BRED ALONG AMAZON RIVER, CLOSE TO MANAUS (BORGHESE FOTO, 2015)

THE BLACK GOLD

“El Bufalo de Agua. Oro negro de la ganaderia” book.

Editors: Alina Mitat Valdes



This four-volumes book summarizes the story of the experience with water buffalos in Cuba, and it also brings precious information about nutrition, health, animal breeding and reproduction.
An essential book for scientists, students and everyone who wants to know the adaptation of buffalos to the American continent.

Written by Alina Mitat Valdés, the title also shows the experience of those who dedicated most of their professional life to the study of buffalos and their adaptation to Cuban conditions. Admired for her perseverance in defending the species, Alina is an American icon in the development of the buffalo species.

The book is divided into 4 volumes. In the first volume, the author narrates the “The introduction of the Buffalos in Cuba”, held in the 80's of the last century and offers us all the information about the beginning of the animal´s importation and their subsequent adaptation to the productive system in Cuba, including absorption between the species.
In the second volume, entitled “Water Buffalos” Alina Mitat Valdés describes the general characteristics of the species, worldwide distribution, genetic differences between the river species and swamp species, as well as various aspects of reproduction. Buffalo breeds are also described, including the Buffalypso.
The third volume brings the different breeding systems, stages of animal’s growth and aspects of reproduction. The book offers us comparative tables of physiological states and productive purpose, bringing extremely useful and relevant graphics.

In the fourth volume, Alina Mitat Valdés evaluates one of the most important aspects of buffalo breeding: management and feeding. In other words, the role of the breeder for the success of these animals in different situations.

An important American contribution to the bibliography of this spectacular species. Those who know the work of Alina Mitat Valdés and know its importance to the buffalo development in the Americas, especially in Cuba, understand this book as a summary of her experience and an important contribution to all who love this species.

Alina Mitat Valdès
Asesora del Centro de Investigacion para el Mejoramiento Animal de la Ganaderia Tropical
Former Professor at Faculty of Veterinary Medicine – Agraria University
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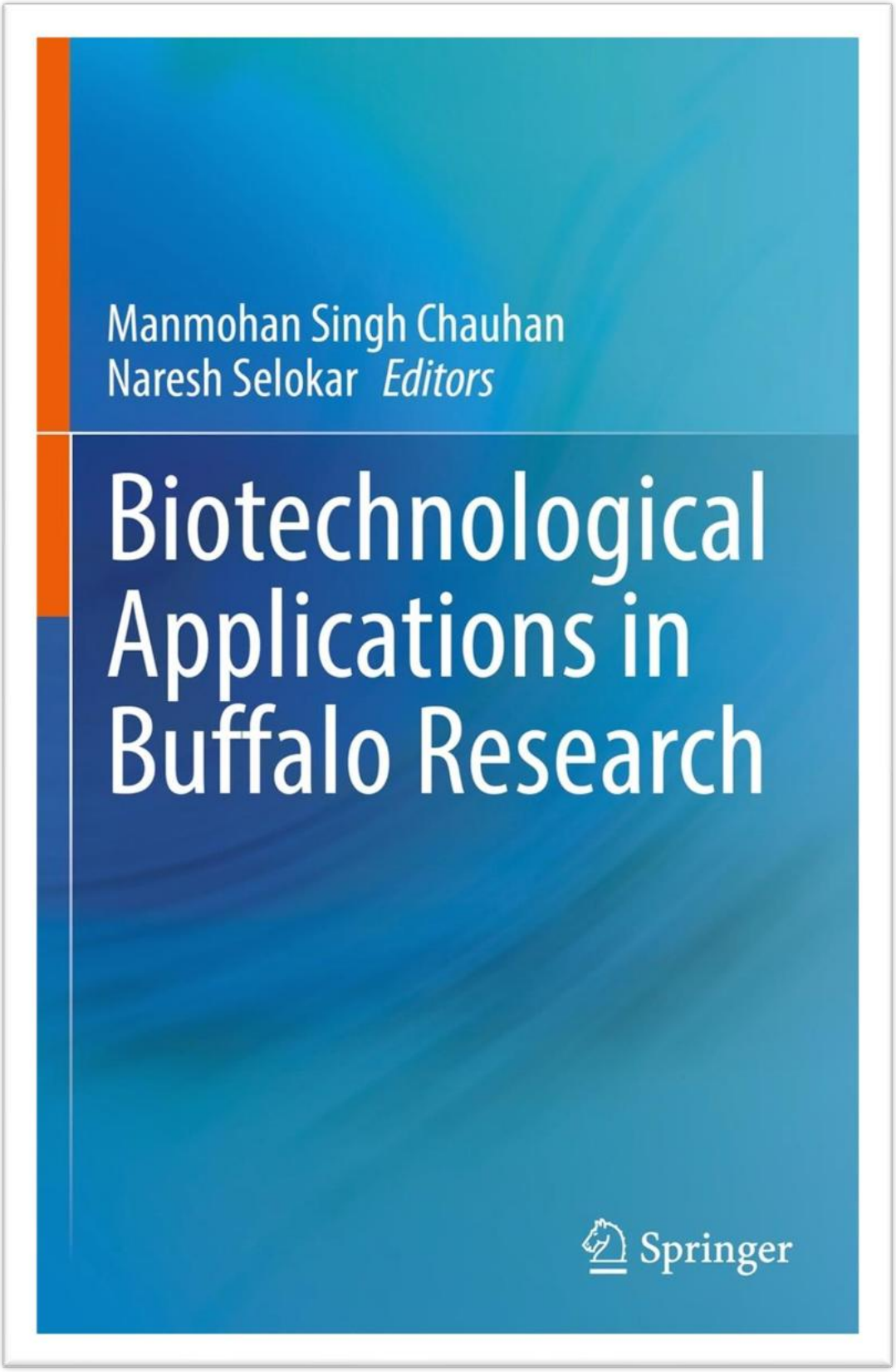
BIOTECHNOLOGICAL APPLICATIONS IN BUFFALO RESEARCH

Editors: Chauhan, Manmohan Singh, and Naresh Selokar



- Highlights recent developments in buffalo husbandry practices
- Summarizes traditional and advanced reproductive technologies in buffalo
- Reviews genome-wide association studies for improved productivity of buffalo

(https://link.springer.com/book/10.1007/978-981-16-7531-7)

Buffalo, A Black Beauty in Livestock Farming

1 **Buffalo in the World: Situation and Perspectives**

- Antonio Borghese, Antonella Chiariotti, Vittoria Lucia Barile

2 **Water Buffalo Genomic Diversity**

- Licia Colli, John L. Williams, Paolo Ajmone-Marsan

3 **Advances in Buffalo Breeding: A Journey from Classical Breeding to Genomic Selection**

- G. R. Gowane, Vikas Vohr

Reproductive Management of Dairy Buffaloes

- Nasim Ahmad, Mubbashar Hassan, Usman Arshad

4 **Behavior and Welfare of Dairy Buffaloes: Calving, Milking, and Weaning**

- Daniel Mota-Rojas, Fabio Napolitano, Agustín Orihuela, Francesco Serrapica, Adriana Olmos-Hernández, Julio Martínez-Burnes et al.

5 **Buffalo Milk and Its Products: Composition, Nutrition and Benefits**

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6 **Welfare of Buffaloes at Slaughter: Signs of the Return of Sensibility**

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2. **Omics Approaches to Understand Buffalo's Genome, Physiology, and Reproduction**

1 **Molecular Evolution and Genome Architecture of Water Buffalo (*Bubalus Bubalis*), the "Living Bank" for Marginal Farmers in Developing Countries**

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2 **Advances in Buffalo Bull Fertility Prediction**

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3 **Being Sweet Is Being Smart: Lessons Learnt from Buffalo Spermatozoa**

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6 **Domesticated Buffalo: A Model for Human Biomedical Research**

- Vijay Pal Singh, Sujoy Khanna
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3. **Reproductive Biotechnologies**

1 **Advances in Embryo Production in Buffaloes: In Vivo Versus In Vitro Procedures**

- Otávio Mitio Ohashi, Satish Kumar, Sebastião T. Rolim Filho, Haroldo F. L. Ribeiro, Vicente José de Figueirêdo Freitas, William Gomes Vale

2 **Application of Fixed-Time Artificial Insemination in Water Buffaloes**

- Juan Carlos Gutiérrez-Añez, Adriana Camacho de Gutiérrez, Héctor Nava-Trujillo

3 **Semen Sexing in the Buffalo (*Bubalus bubalis*)**

- Giorgio A. Presicce

SCIENTISTS DR SUNIL KUMAR AND MR. KULDEEP DUDI DEVELOP PATENTED TECHNOLOGY FOR PREPARING BUFFALO MILK PROTEIN CONCENTRATE

Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), India

Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana has added another feather to its cap by obtaining a patent for the technology of preparing Buffalo (*Bubalus bubalis*) Milk Protein Concentrate.

The inventors, Dr Sunil Kumar and Mr. Kuldeep Dudi, from the College of Dairy Science & Technology, asserted that currently, there were serious shortcomings in the technology for the concentration and drying of buffalo milk protein. Maintenance of high solubility and functionality of final powder were important issues hampering its acceptability. On the other hand, there were well-established methodologies for the preparation of cow milk-based milk protein concentrate but, no such process was available for buffalo milk because of the more complex ionic and protein environment of buffalo milk.

Buffalo milk has unique quality attributes like significantly higher total solids, fat, energy, protein (both casein and whey proteins), larger casein micelles, availability of only A2 variant of β-casein, better buffering capacity, higher minerals (calcium, magnesium, and phosphates) etc. Due to these qualities of buffalo milk, its demand is boosting across the globe. While buffalo milk is preferred for preparation of several ethnic sweets in Indian subcontinent, but it presents technical challenges in preparing other milk products, especially milk protein concentrate or high protein powders.

This challenge has been addressed through the novel technology developed by the GADVASU scientists by modifying the ionic and protein environment of buffalo milk for the production of highly soluble and functional buffalo milk protein concentrate.

Dr Sunil Kumar (co-inventor).

Dr. Inderjeet Singh, Vice-Chancellor of the university and past President of the Asian Buffalo Association, congratulated inventors of the technology and emphasised that the developed technology would open

new avenues for dairy industry while potentially increasing the opportunities of value addition of buffalo milk in the production of quality protein powders and products. Hence, it will boost the demand of buffalo milk worldwide and by adopting the GADVASU technology, the issues pertaining to concentration, drying and solubilization of buffalo milk protein can easily be resolved while keeping the protein's functionality intact. He further stressed that the technology would also help in the enhancement of income and the livelihood of our farmers & producers and is now available for commercialisation to interested speciality milk food industries.

Mr RS Sodhi, Managing Director of India's largest Gujarat Cooperative Milk Federation (GCMF), popularly known by its tradename - Amul, twitted it to be a breakthrough technology developed by GADVASU, reminiscing of the 1950s technology of making milk powder from buffalo milk by Dr. Kurien and Mr Dalaya, which was otherwise negated and challenged by the western experts.

Dr. J.P.S. Gill, Director Research of the university, claimed that the current invention overcomes the drawbacks of prior arts in terms of being simpler and efficient. He also added that the superior functionality concentrated protein is from buffalo milk thus, it would be free from any type of protein allergy as seen in most of the other protein concentrates available in the market.

The developed buffalo milk based highly soluble and functional protein concentrate contains both the proteins of milk, i.e. casein and whey protein, thus, considered a potent source of quality protein for the consumers. In addition, it also comprises significantly depleted level of calcium, phosphorous, potassium, and magnesium which ranges from 85-88%, 87-90%, 80-97%, and 87-91%, respectively. Developed high protein concentrate can also be used for the development of functional food products, processed cheese, ice-cream, beverages, high protein food products, nutritional bars, sports foods, supplements, etc. This is expected to make importance of buffalo milk more pronounced as a superior output.

Dr. Inderjeet Singh
Vice Chancellor, Guru Angad Dev Veterinary and Animal Sciences University
Ludhiana, Punjab.

BUFFALO IN ARGENTINA
FOLLOW THE GROWTH OF THE BUFFALO

FIGURE 1 MEDITERRANEAN PREGNANT BUFFALO HEIFERS

The XI Expo Buffalo 2022 showed that the buffalo is a productive node in continuous growth, in the context of the country's economic crisis and in a region hit by drought, heat and fires.

The Argentine Buffalo Breeders Association convened more than 200 people and enclosed almost 600 buffaloes in a two-day event, including conferences, judging, selection of champions and auction.

The Expo Buffalo was held on May 5 and 6 at the Rural Society of Corrientes, in Riachuelo, with sales by Colombo and Magliano S.A. Very good quality and body condition in all the lots presented. A very dynamic auction: 540 heads were sold in just two hours, with face-to-face and streaming buyers, including traditional breeders from Corrientes and Formosa and new breeders (including some from Santiago del Estero) who had a great role in the auction. Prices improved approximately 20% compared to the previous auction, in November 2021. Buffalo bulls (about 27 months old) averaged US$ 1.705, with highs of US$ 2.350. Pregnant buffalo heifers sold for an average of US$ 695, and buffalo heifers for service sold for US$ 525. It is necessary to highlight the good prices (there were disputed batches) and the new breeders.

The conferences presented by breeders and technicians from Brazil, Venezuela and Argentina were of a high level. Data on greater productive efficiency and greater profitability were provided in an important livestock company in the north of Corrientes. An example of intensive production on excellent quality soils in southern Brazil was presented, with gross margins higher than cattle (500 heads per year are fattened on 50 hectares with fertilized pasture and under irrigation).

FIGURE 2, FIRST BUTCH OF THE AUCTION: MEDITERRANEAN BUFFALO STEERS AT 319 KG LIVE WEIGHT AND 14 MONTHS OF AGE

The buffalo is more productive also with high quality forages. Another presentation referred to buffalo farming in the different regions of Brazil: buffalo is the only livestock that adapts efficiently to the different ecosystems of the Brazilian subcontinent.

The milk production of a farm in South of Lake Maracaibo, Venezuela, was also presented: 22 liters per day, which is equivalent to 55 liters of cow milk (due to its yield in cheese and other dairy products). A researcher from the UNNE Corrientes presented novel results of parasitological studies carried out with buffaloes. International and local juries did a job of excellence (Renato Amaral and Regis Goncalves from Brazil, and Federico Goicoechea from Argentina).

The new generation of members of the Association had a leading role in the impeccable organization of this event. Mainly Marcelo Breard and Federico Goicoechea. Both are breeders in Corrientes, where Breard is also Delegate of the AACB. This event included buffalo burgers as part of the attention to those present during the two days.

In short, everything indicates that we are on the right track.

Ing. P.A. Marco A. Zava
Buffalo breeder and member of the Board of Directors of the Argentine Buffalo Breeders Association

ASOBUFALOS

ASOBUFALOS stands for Guatemala Association of Water Buffalo Breeders and it embraces the following essential objectives:
To promote the development of buffalo breeds in the country;
To implement genealogical records;
To carry out scientific studies, conferences, publications and others for encouraging the promotion of this animal species;
To carry out exhibitions, performance tests, awards and other activities aiming to stimulate the development of the species and to encourage its exploitation;
To providing technical assistance to its partners or third parties interested in the exploitation of the species;
To advocate for any other subjects related to the previous objective already approved by the Association bodies.

Within its objectives, **ASOBUFALOS** Guatemala organized an exchange of best practices in a conference meeting held in YouTube on 26 April 2022. The recording is available in the following link:
https://youtu.be/0VIJHsiNyRE

Presenters highlighted key issues in a round of lectures followed by questions and answers, ending in fruitful discussions.

Lic. Rodolfo Estrada Nicol

Buffalo breeder and Strani product developer.

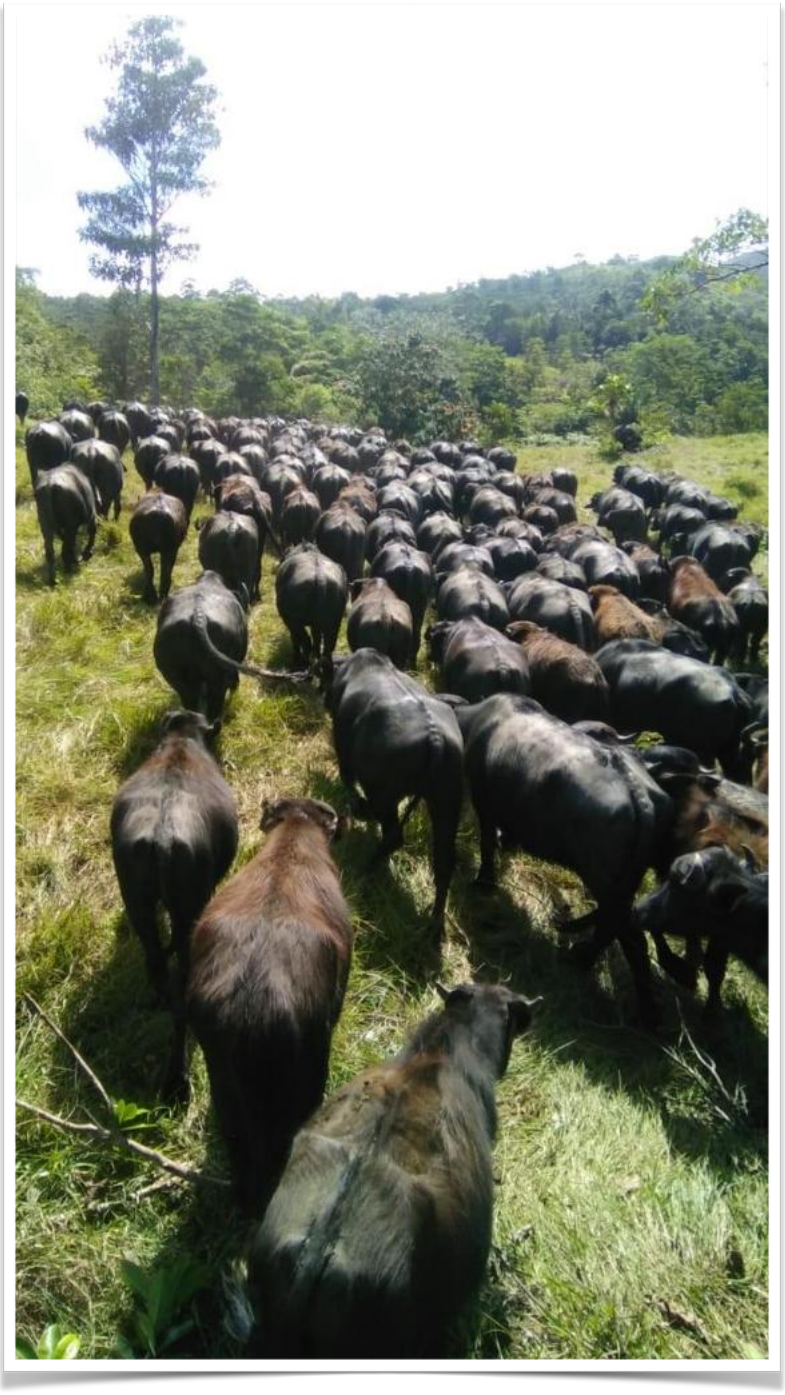
World-class organicity and sustainability certifications.
Presentation: Strani Buffalo Products:

- Meat (Hamburger patties, ground beef and beef)
- Mozzarella cheese
- Burrata cheese
- Ghee cheese

Dr. Julio Adolfo Kopp Goméz

Veterinarian graduated from the San Carlos University of Guatemala, specialized in Parasitology in Hannover, Germany.

Presentation:
Parasitology: The economic impact of parasitic diseases in a buffalo herd.
Parasitic diseases cause millions in losses within livestock.
It is necessary an in-depth knowledge of parasitic diseases and their consequences on production.
Paradigm, doing something without knowing why, is the way we intend to control parasites.



Lic. Leonel Sandoval

Buffalo breeder and producer of buffalo of triple purpose: meat, milk and work at Hacienda La Reserva.

Presentation: The Buffalo as a Working Animal
Hacienda La Reserva is a company dedicated to the production of palm fruit oil and buffaloes for work.

UPCOMING EVENTS

10TH AMERICAN AND EUROPEAN BUFFALO SYMPOSIUM

Asunciòn, Paraguay 15-19, November 2022

APACRIBU, the Buffalo breeders association of Paraguay has achieved important milestones in the process of preparing for the X Symposium
of Buffalo Breeders of the Americas and Europe, which will take place Nov 15-19, 2022, in the capital city of Asunciòn, with headquarters in the 20 ha campus of the Asociacion Rural del Paraguay, (Paraguayan Rural Association), the prestigious private sector organization that amalgamates and represents Paraguayan cattle producers throughout the country.

En route to the Symposium, APACRIBU has worked hand in hand with the Ministry of Agriculture, with the support of the GIZ, to establish a multisector working group to promote the competitiveness of the Buffalo in Paraguay, including various ministries, universities and other public and private sector organizations. The National Senate has also given its support by declaring the X Symposium as an event of National Interest.

The event will have two phases: Conferences and a Buffalo Tour. The first two days are primarily academic conferences focusing on subjects related to the management of a buffalo herd, as well as those related to sustainable and regenerative ranching and the role that buffalo can play in the development of Paraguay and in the region. The conferences will have simultaneous translation between English and Spanish.

The rest of the event will be a buffalo tour with the objective of sharing the country's scenic beauty, culture and history, as well as showcasing leading ranching establishments and visiting buffalo producers.

Do join us!

Richard Moss
President of the Paraguayan Association of Buffalo Breeders (APACRIBU)

Details can be seen, and inscriptions made on the APACRIBU website: www.bufalodeagua.com

13th WORLD BUFFALO CONGRESS

The World Buffalo Congress that should have taken place in October 2022 in Wuhan, China, has been cancelled due to the pandemic.

Prof. Liguo is the elected President for the period 2019-2022 and has agreed to ask other countries to host the 13th World Buffalo Congress in 2023.

The new Venue and date will be communicated.

11TH ASIAN BUFFALO CONGRESS

The elected President of the Asian Buffalo Association Prof Omar Md Faruque is organizing the next Asian Buffalo Congress in Bangladesh in 2024.

IBF-CREA course 2022

"BUFFALO FARMING ON AIR: FROM FARM TO FORK"

Due to Covid-19 pandemic the IBF Training courses in Italy are still on hold. The IBF secretariat together with CREA has decided to organize in 2022, a series of six webinars covering the main aspect of buffalo production chain: milk production process, meat production process, and sustainability.

The two-hour webinars already on air are WEB 1. *Buffalo calf management: From birth to weaning*; WEB 2. *Buffalo heifer & cow management* and WEB 3. *Reproductive management*

Info on the webinars are available at www.internationalbuffalofed.org website.

Video recording of the webinars are available at the International Buffalo Federation YouTube channel: shorturl.at/ikyN2

IBF Survey

Dear Members,
many years have passed since last survey in the nineties, it is time to carry out a new one to update the **state of the art of buffalo farming in IBF member countries**. For this reason, the IBF will develop together with ARB (an innovation broker company with twenty years of work experience devoted to innovation in agriculture and to the development of rural areas) a questionnaire containing about 20-30 questions (closed and open) focused **on the main aspects of buffalo farming.**

The questionnaire could be filled online (offline compilation will be provided where necessary) and will include four sections: i) Country overview; ii) Farming systems; iii) value chain supply; iv) Impact of buffalo farming on the development of territories. The IBF secretariat together with the executive officers will coordinate the organization, the data collection, the activities as well as the support to the compilation. A final report will be published. We hope this initiative will be welcomed by all IBF members.

MEMORIES

Fabio Napolitano

(1963-2022)

Fabio Napolitano was full Professor at the School of Agriculture, Food, Forestry and Environmental Sciences (University of Basilicata, Italy) since December 2021 and Coordinator of the PhD course in Agricultural, Forest and Food Sciences. At the same University, Fabio started his academic career as researcher in 1995. He was involved in several projects concerning animal behavior (regional and national level) and animal welfare (national and international level), and in particular on Mediterranean Italian buffaloes.

He has been nominated member of the scientific committee of external reviewers by the European Food Safety Authority (EFSA) for the period 2009-2011 and contributed to draft the external reviews of the quality of the scientific outputs of EFSA. He has been nominated member of the working group on sheep welfare by EFSA in 2013 and contributed to draft and publish a “Scientific Opinion on the welfare risks related to the farming of sheep for wool, meat and milk production” and a Technical Report titled: “Outcome of a public consultation on the Draft Scientific Opinion of the EFSA Panel on Animal Health and Welfare on the welfare risks related to the farming of sheep for wool, meat and milk production”.

He has been nominated member of the working group on Animal-based measures (ABMs) gap analysis and contributed to draft and publish a Technical Report titled: “The use of animal-based measures to assess animal welfare in EU - state of the art of 10 years of activities and analysis of gaps.
He has been nominated expert evaluator for the calls of the Societal Challenge of Horizon 2020.

He was member of the editorial board of the journals “Animals” (MDPI), “Journal of Food Quality” (Hindawi), and “International Journal of Food Studies” (ISEKI_Food Association), and editor in chief of the “Journal of Buffalo Science” (LiveScience Global).

In 2021 he was included in the "career" and "single year" categories of the "Updated science-wide author databases of standardized citation indicators", published by Stanford University. He was author of 150 indexed scientific articles, with 3,150 citations and an h-index of 33.
Fabio was a recognized pioneer of the study of buffalo behavior and welfare. He had the gift of conversing to everyone as an equal, Fabio was not a man who put on airs or thought he was superior to others. We will miss his smiles, cheerful advice, availability and scientific expertise. His way of approaching life will be impressed in our minds forever.

Ada Braghieri
Corrado Pacelli
Giuseppe de Rosa
University of Basilicata, Italy

Manfred Thiele
(1950-2022)

It is with deep sadness that we say goodbye to Manfred Thiele, President of the German Buffalo Association (DBV).

Manfred Thiele was born on March 02, 1950, and passed away on May 11, 2022, after long illness that was endured with great patience.
He grew up on a farm in the Ore Mountains. After vocational training in animal husbandry, he began studying at the Karl Marx University in Leipzig, where he graduated as an agricultural engineer.

Professionally, he spent the following years in various leading positions in animal husbandry.
In 1991, together with other partners, he founded a farm where, in addition to arable farming, about 500 dairy cattle including offspring were kept.
In 1999 the idea of buffalo breeding was born and the first three animals were brought from Italy to gain first experiences. The herd then grew steadily to around 250 buffalo at times.
Manfred Thiele made himself deeply familiar with buffalo husbandry. Through his intensive research in theory and practice, he acquired extensive knowledge.
In the early 2000s, he was elected President of the German Buffalo Association, which was founded in 1999 in Wardenburg in Northern Germany, and the Association's headquarters were simultaneously moved to Chursdorf in Saxony.
He established multiple contacts with domestic and foreign scientists and practitioners in the field of buffalo husbandry, as well as politicians. Through his perseverance, he succeeded in getting the buffalo included in the German animal breeding law.
One of his great merits was the organization of the German Buffalo Congresses on a regular basis near Chemnitz with international participation.
With his irrepressible energy he moved a lot in the field of buffalo husbandry and left his mark forever.
We have lost a true pioneer and unique personality in German buffalo economy, a great thinker and doer, and a wonderful human being.
We bow deeply to his life's work.

Andreas Hoeflich
(DBV and IBF)

Institute for Genome Biology
Institute for Farm Animal Biology
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