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Enhancing Reproductive Performance in Dairy Buffalo

Review Article

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Abstract

The aim of the review is to report on some of the factors affecting fertility in buffalo and the techniques that allow an improvement of the reproductive performance.

In Italy and in Latin America in farms in which mating is natural and bulls are always present in the herd the inter-calving interval is approximately 400 days and the culling rate is lower than 15%.

This species shows a reproductive activity tendentially seasonal, that is favoured by the decrease of daily light hours, but the ovarian activity stops if conception does not occur within 3-5 ovarian cycles. Hence it is important an appropriate management of the transitional period, particularly concerning the hygienic conditions of uterus.

In tropical Countries located North of the equator feed deficiencies and heat stress are considered the main factors of summer hypo fertility. In Pakistan, for example, the increasing body condition score during the autumn was associated with the commencement of breeding season in buffaloes. However, anoestrus is observed also in Italy where the average daily temperature in the same period is 23.5-13.5°C and feeding is constant throughout the year.

The only common element between the two areas is the progressive increase of daily light hours between April and June and the daily length higher than 12 h up to September.

In Italy in herds where out of season breeding strategy is applied an improvement of fertility, as percentage of corpora lutea corresponding to subsequent pregnancy, is observed when water pools are present in the farm, demonstrating that the improvement of environmental conditions reduces the incidence of embryonic mortality and/or abnormal cycles. To summarize, in the absence of serious nutritional problems, the improvement of the environmental conditions increases fertility.

Introduction

Buffalo represents a fundamental and irreplaceable resource for tropical Countries. Between 1961 and 2008 the world population has increased by 43% and 105% and between 1995 and 2007 has increased by 2% and 13%, respectively in bovine and buffalo species (Faostat.fao.org/faostat). The FAO data are referred to the population present in 43 of 143 Countries that currently breed buffalo [1].

The majority of the authors believe that the main reproductive characteristics of buffalo are the delayed puberty, the prolonged post-partum ovarian inactivity, the long inter-calving periods [2] and the tendency toward seasonality.

The reproductive problems are different depending on the area of the breeding. In tropical countries North of the equator (TCNE), for example, the majority of the authors assert that the summer anoestrus is due to heat stress and forage scarcity. However, in Italy anoestrus is observed in the same period of the year of TCNE, as demonstrated by the calving distribution shown in Figure 1, although the diet is constant throughout the year and the temperatures are milder.

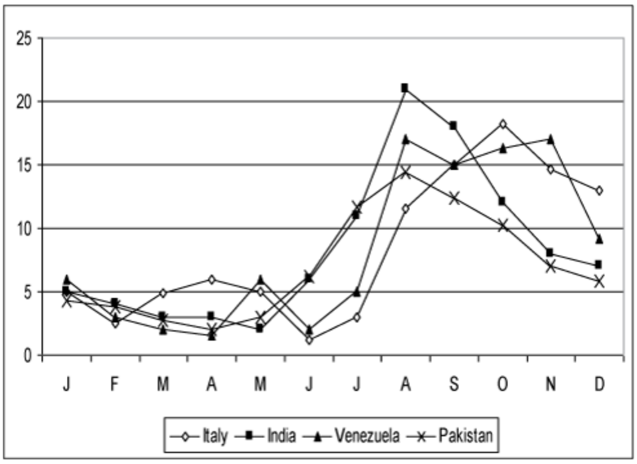

Figure 1: Monthly calving percentage: Italy (Zicarelli et al., 1977), India (Singh, 1988), Venezuele : Guanota : farm, Apure State (Zicarelli, 1993), Pakistan (Hassan et al., 2007).

The aim of the review is to examine the main factors affecting fertility in buffalo and the strategies that may be adopted to enhance the reproductive performances.

Reproductive seasonality

The place of origin and the duration of gestation have undoubtedly influenced the way in which reproductive seasonality occurs. The natural necessity to coincide calving and weaning with the most suitable parts of year in order to satisfy the nutritive requirements of the offspring through a period in which etiologic agents (infectious and parasites) are less aggressive and/or present [3] represents one of the causes of this 'adaptation' process. The subjects born under the most favourable conditions, have brought about natural selection of subjects endowed with a more ideal reproductive seasonality towards the survival of the species [4].

Spring calving (March-May), which guarantees the offspring good availability of forage in temperate zones north of the Equator, occurs when reproduction takes place in autumn (September-November) in the case of five month gestation (sheep and goats) or in the previous spring in the case of 11 and 12 months gestation (horses and donkeys). Therefore, the same calving period is conditioned by the neuroendocrine system sensitivity, for the re-activation of the reproductive cycle, short day breeder (negative photoperiod) or long breeder (positive photoperiod) as regards the length of gestation.

In tropical zones where the domestic buffalo developed, between 31°N to the north and 2°S, forage availability is usually found after the rainy season which begins normally in July and ends in September. Therefore, in species like the domestic buffalo, whose pregnancy lasts 310-316 days, it seems that the sensitivity to decreasing of light stimulus and the reproductive season take place between September and January.

Around the equatorial belt, where light/dark ratio varies little throughout the year, the reproductive season is highly conditioned by forage availability [5-7]. It may be predicted that there is a tendency towards seasonality as far away is the buffalo species from the equator. Transferring the species to other breeding areas has not modified hypophyseal hypothalamic axis sensitivity to a decreasing light/dark ratio.

In Italy the seasonality makes necessary to change the calving calendar in order to meet the milk market demand; this is accomplished by the technique of out-of-breeding-mating-strategy (OBMS) that entails the interruption of natural mating or the use of AI between October and late January in adult females (between September and late March in heifers), which is the most favorable period for the reproductive activity [8,9].

In a research carried out in Italy [10], it was verified that buffaloes (observed for a year) which have a tendency towards seasonality, showed high melatonin blood plasma levels two hours after the sunset even when they were moved to another farm where other group of females showed low melatonin blood plasma levels and less sensitivity towards light stimulation [11]. The blood plasma levels of melatonin had a repeatability value of 0.733 [12]. If the heredity of this character turns out to be high, as we expect on the basis of high repeatability, such phenomenon will be involved in genetic selection programmes of this species [13]. To confirm this theory, we report that ovine Romanov 58°N, Karakul 41°N and White-faced 51°N [14] showed a continuous cyclic activity throughout the year even if living at latitudes where other genotypes are sensitive to light/dark ratio.

In heifers and adult female buffaloes [15] the difference in March (begin of spring) between night/day values of blood plasma melatonin were less in heifers (5,02 times) compared to adult buffaloes (28,3 times). Finally, both the buffaloes [16] that calve in spring (more adaptable to out-of-breeding-mating-strategy) and the heifers [17], which, as is known, are less sensitive to the photoperiod, show the same behavioral pattern.

In the heifers the fertility is not penalized by the season [17-19]; during the summer and when daylight hours are more than those of darkness, there is an increase in prolactin levels in the blood but, contrary to assertions by Madan [20], subjects regularly conceive. We believe that hyperprolactinemia is a consequence of the hypothyroidism during the warm months, which exerts a positive feedback on TSH and hence on TRH that notoriously promotes an increase of haematic levels of prolactin [21-23].

The majority of authors attribute reproductive seasonality to nutritional factors like in areas where 97% of the buffalo population are bred and the breeding period takes place in the months of greater forage availability [24-30]. This condition is found during the months (July –November) characterized by decreasing daylight length in TCNE.

Recently Hassan et al., [31] reported the different seasonality of Nili-Ravi Buffaloes, purebred Sahiwal and cross-bred cattle in Pakistan. In this study, the difference between Nili-Ravi Buffaloes and purebred Sahiwal characterized respectively by negative or positive photoperiod has been highlighted. Data similar to those recorded in buffaloes have been observed in cross-bred cattle (Holstein or Jersey x Sahiwal). However, it cannot be ruled out that the high temperatures (41C° - 35C°) and milder (from November to February) temperatures (28C°-22C°), may influence respectively, negatively and positively the ovarian activity. This finding suggests, moreover, that the crossbred with Bos Taurus may modify the seasonality of native bovines, which show the higher incidence of deliveries during the first 5 months of the year and, hence, conceive in the hottest months (April-August).

If heat stress was the main cause of the anoestrus it should be admitted that it adversely affects the reproductive activity of buffalo compared to Bos indicus. In Italy exactly the opposite pattern is observed because in out-of-season mated buffaloes the percentage of conception increases between July and September, a period during which the Holstein shows a low rate of conception.

In Italy in herds where the OBMS technique is not used as well as 30 – 40 years ago in the majority of farms [32], the resumption of the reproductive cycle (R.R.C.) took place (Figure 1) from September (decreasing light period) until January (light increasing period predominantly dark hours).

Sensitivity to the negative photoperiod is also found on farms where a constant balanced diet is provided around year. This type of seasonality, where reproductive events are not synchronized with forage availability, indicate that the buffalo bred in Italy are not autochthonous in the sense that they are sometimes apt to calve in periods of forage scarceness and low temperature which in turn hinder survival of the calf.

Italian findings should be sufficient to define the buffalo as a short day breeder animal. Indeed, a similar seasonality to that found in Italy and Asian tropical areas is found also (Figure 1) in Venezuela [33,34] and Egypt [35].

In South of Brazil [36,37] and Argentina, the wet season, and consequently, pasture availability, starts around October-November and continues until March - April, while the pasture scarcity goes from May - June until October-November. The buffalo calving period under these conditions is mainly concentrated from February to May (Figure 2), the breeding period from April to July, and the weaning calf period, under free range and suckling calf conditions, from September to December. These events permit the coincidence of forage availability within the first 2- 4 months of lactation and most of the dry milk period (October-April). The breeding period, however, is mainly concentrated during the pasture scarcity period (May-July).

Baruselli et al., [38] who elaborated the data of Brazilian Breeders Association observed that the seasonality is

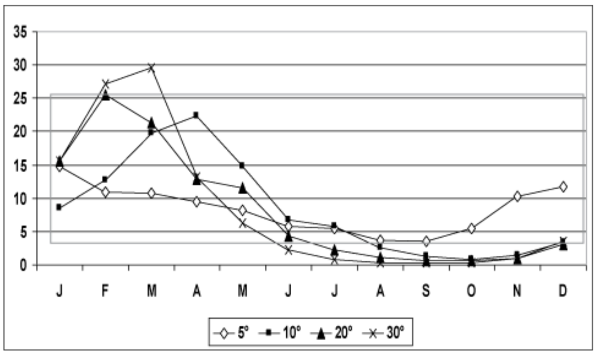

Figure 2: Monthly calving percentage in Brazil in function of latitude (°).

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more accentuated from the North (0-8 latitude degrees) to the South (24-32 latitude degrees) of Brazil (Figure 2) and hence the calving season is influenced by latitude.

From the findings shown here, it can be unequivocally stated that, although the domestic buffalo shows reproductive activity throughout the year, there is greater tendency to concentrate reproductive activity in months of decreasing daylight or if increasing, when dark hours prevail.

Reproductive efficiency

Many scientists affirm that the buffalo shows the delayed puberty and a long intercalving period, which is affected by several factors, such as the year of calving, the season of delivery, the genotype and the heat stress [39].

The delayed puberty and the consequent older age at first calving are referred by many authors. In a study performed in 86 farms (30,735 primiparous buffaloes, which calved between 1975 and 2005), it was observed that the mean age at first calving lowered by 1 month every 5 years (44.7 ± 6.6 and 35.33 ± 6.46 respectively in 1975 and 2005).

The hormonal pattern in cyclic buffalo is similar to that described in a cyclic cow [40]. The main difference between the two species is the rate of cyclic subjects in the different seasons.

The different reproductive efficiency of buffalo species compared to the cow is due above all to these features:

a) in the buffalo is present a lower number of primordial [41] and antral follicles, as well as a lower weight (4 vs. 8,5 g) and volume (mean length: 2.5 vs. 3.7 cm) of the ovary [42]. The number of oocyte in a buffalo calf is 1/5 of that recorded in a bovine calf [43];

b) need to modify or not the calving calendar in order to meet the market demand.

a) In our opinion, after calving, due to the low number of follicles and waves, few cycles occur and hence, if conception does not take place, an anoestrus of variable length begins.

With regard to this topic, the transition period and the post-partum have a major importance for fertility in buffalo species compared to the cow. It is worth to point out that in the farms adopting semi-free housing, the presence of mycotoxins in the roughages, of Clostridia, of Coxiella burnetii, the incorrect input of Ca, P and crude proteins in the last two months of pregnancy often lead to the occurrence of vaginal or uterine prolapse which impairs the R.R. [44].

In my opinion, it is not clear whether the seasonality of the species depends on the reduced follicular population or if this latter is the cause of buffalo seasonality.

b) It has to be underlined that if the calving calendar is not modified, the delayed R.R.C. after calving can be due to the lack of the "bull effect" and/or to poor nutritional conditions. In TCNE, in the small farms where the bull is not present, the dry period of the animals (March-June) coincides with the scarcity of forage, while in the South of the equator over the 20th parallel (November-March) it coincides with the abundance of forage and the bull is always present in the herd. In the first case prolonged inter-calving intervals are observed whereas in the second case inter-calving intervals less than 400 days are recorded.

These observations suggest that in those areas, the covering of nutritional requirements and the absence/presence of the bull are the most important factors, especially taken into account that the protein content of the pastures is very low (6 - 10% on DM) and leguminosae are found in irrigated areas or during the rainy season. In Pakistan, Qureshi et al., [45] refer that the seasonality is influenced by nutritional and non-nutritional factors.

However, unless the feed deficiencies are serious [46-48], nutritional factors in general do not play a significant role because the species cannot yet be considered exactly as having a "lactiferus habitus" and therefore a "catabolicus habitus" as the highly producing milk bovine cow, which uses its reserves to compensate energy and protein deficiency during the first phases of lactation. As matter of fact, within reasonable limits, the buffalo uses its reserves in its aim to reproduce and so to the detriment of his own production [49].

The effects of the seasonal reproductive trend of the species are particularly evident and studies conducted in Italy may be useful for Countries wishing to make more consistent their production of buffalo milk in the year. In the calving calendar of the farms that are adopting the OBMS technique, when the primiparous are excluded from the survey (heifers are less sensible to the photoperiod), a decrease in calving rate between March and June (Figure 3) is observed in Italy, both if OBMS is applied or not, confirming that the increase of daily light hours (April-

June) or a number of daily light hours >12 h, negatively interfere with reproductive activity.

The subjects that undergo OBMS can show anoestrus, and this phenomenon is emphasized by unfavorable climate conditions.

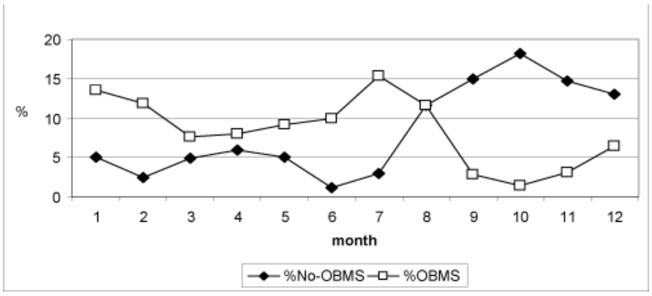

Figure 3: Calving percentage in farms that utilize (%OBMS) or not (%No:OBMS) the OBMS technique

It is possible to recognize a "temporary anoestrus" (<150 days) and a "deep anoestrus" (>150 days) according to the number of days open. It is possible to distinguish between subjects that come into oestrus within 70 days from calving, and conceive within or after 90 days, from those which come into oestrus after 70 days and conceive within or after 150 days. Around 70% of buffaloes, that deliver out of the breeding season, showed R.R.C. within 70 days from calving. Fifty-three percent of these are able to conceive within 3 months (around 75% of the animals that resumed the ovarian activity within 70 days) whereas 17.3% later than 3 months. The remaining 30% of animals show the R.R.C. after 70 days from calving and 10.6% and 18.8% conceive within and after 5 months, respectively.

Anoestrus can be identified as a non R.R.C. after parturition or as an interruption of cyclic activity for varying reasons. Environmental conditions that are responsible for the anoestrus are accompanied by serum variations of hormones [50] like prolactin [51], follicle stimulating hormone [52], luteinizing hormone [53,54], progesterone [55,56] as well as 17β-estradiol [57,58] T3, T4 [59,60], and corticosteroids [2] that represent the endocrine response to stressful factors rather than the cause of the arrest of reproductive activity.

Hence, the subjects that deliver out of the breeding season conceive very soon or need several months. During this period, they are not always acyclic, but are able to conceive and undergone embryonic mortality, in particular between April and May, months characterized by progressive increasing daylight length. During the year the embryonic mortality rate is 10% of the pregnancies diagnosed at 40 days and becomes 22% for conceptions that take place in the month of April. The incidence of this event is considerably affected by the "farm" factor and ranges between 10% and 45% if calculated on the pregnancies after AI diagnosed by ultrasonography at day 26 [61].

After the observation of a corpus luteum, at the following clinical examination the pregnancy is not always detected. In these cases, the buffaloes show an anoestrus condition. Therefore, it can be supposed the occurrence of an early embryonic mortality or an anoestrus condition preceded by an ovarian cycle which forms a corpus luteum responsible of an inadequate luteinic secretion due to short luteal phase or normal luteal phase but with low progesterone production. This phenomenon has been found upon the onset of anoestrus season in sheep, upon first ovulation post-partum in bovine and during the pre-puberal phase in both species [62].

Some recent papers [63] report an incidence of double ovulations of 15.5% in dairy cows and similar value has been previously reported in buffalo. In the latter, however, only 0,06% of double ovulation lead to a pregnancy twin. The double ovulations reduce the efficiency of the AI in case of spontaneous heats, but not in the case of induced ones.

In Italy, the OBMS technique penalizes fertility. In fact, when the OBMS technique was not applied, values of 400 – 445 days [64,65] have been recorded in Italy.

Recently a mean inter-calving period of 487±133 days was registered (6,052 inter-calving periods throughout an interval of 5-10 years) in 5 farms, in which a culling rate lower than 10% for year was present, the OBMS technique was adopted and a constant rationing was given throughout the year. It was demonstrated that the shortest inter-calving periods were recorded in buffaloes that delivery in the periods between April and June, and that, consequently, conceived in the warmest months (between June and August) of the year, and in those that delivered between July and September, the period of the year during which the highest temperatures are recorded in Italy.

On the contrary, the longest inter-calving periods were recorded between October and December, because of the OBMS technique, which retards mating till February, and in buffaloes that calved between January and March, the coldest period of the year. These subjects would conceive in spring, the mildest period of the year, characterized by temperatures between 15 and 22° C. However, conception is usually delayed until September, except for the 40% of animals, that conceive within 90 days from calving.

Therefore, we can conclude that in Italy nutrition and the warmest months, especially if a swimming-pool is present in the farm [66,67], are not able to affect the inter-calving period. The main factor, that has to be taken into account in Italy, at the latitude of 42-45, is the light stimulus. Buffaloes that deliver between January and March would conceive between March and May, which is a period, characterized by increasing day length, and delay their conception until August - September, after three months of decreasing day length. Similarly, buffaloes that deliver in the period April - September show the shortest inter-calving period, because after 58 days post partum (the useful interval in order to reach an inter-calving period of 365 days), decreasing day length begins.

When the OBMS technique is not used, in autumn also the old buffaloes (with more than 7 calving and still not pregnant after 7-9 months from last calving) spontaneously conceive. The body conditions of buffalo normally worsen after 12-15 years and hence farmers are not encouraged to eliminate the animals that are still not pregnant at the end of lactation. This is one of the causes of the prolongation of the inter-calving period.

The majority of Indian and Egyptian authors assert that the lower concentration of calving observed between January and May depends on the reduced conception rate between March and July. This phenomenon is influenced by the hot and dry climate (summer anoestrus) of this period of the year. In Italy, in farm that do not use the OBMS, a drop of calving is also observed between January and June (Figure 1), period of the year in which, unlike India, climate is either cold or mild and moderately rainy. The fertility markedly improves between July and September, period that in Italy coincides with the highest temperature (Figure 4) and THI of the year. This observation makes buffalo much different from cattle that in the hottest months of the year (July - September) show a marked decrease of fertility.

We have already highlighted that in the areas located north of the equator a drop of calving is observed between January and June.

The authors of Countries located north of the equator attribute the anoestrus of buffalo to heat stress (India, Pakistan) or to the low environmental humidity (Venezuela).

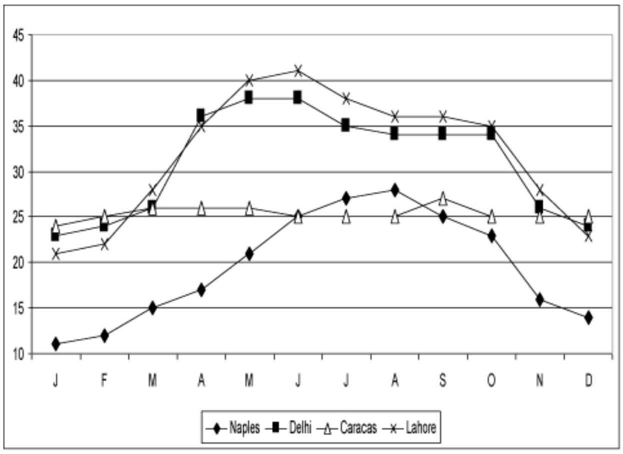

Figure 4: Monthly max temperature in Naples, Dheli, Caracas and Lahore

It is worth pointing out that, in our opinion, the decrease of calving rate between January and June-July (Figure 1) depends on the reduced reproductive activity between March and August –September. In the latter period the maximum daily temperature (Figure 4) ranges between 15C° (March) and 27C° (August) in Naples (Italy), between 25C° and 38C° in Delhi (India), between 27C° (March) and 41C° (June) in Lahore (Pakistan) and between 25C° (March) e 26C° (August) in Caracas (Venezuela).

The maximum daily temperatures that are recorded in Italy and in Venezuela make us rule out a direct action of environmental temperature on anoestrus whereas, limitedly to India and Pakistan, it is not possible to exclude that heat stress, even if it is not the main factor, contributes to aggravate the summer anoestrus. Furthermore, it is worth noting that the R.R.C. coincides (August-September) with a monthly maximum temperature of 28°C (August) and 25°C (September) in Italy, 25C° (August) and 27C° (September) in Venezuela, 34C°(September) in India and 36C° in Pakistan.

The monthly rainfall recorded in Italy and Venezuela between September and October (R. R. A.) is not different from that observed between March and May (Italy) and between June and August (Venezuela) when the lower conception rate is observed (Figure 5).

On the contrary, in India and in Pakistan the rain season takes place between July and September. Therefore, the stop of reproductive activity in Italy and Venezuela cannot be attributed to the rainfall; on the other hand, in India and Pakistan the reproductive activity is good also in October

– November (calving of August and September), months in which the temperature is lower and the rainfall is already minimal.

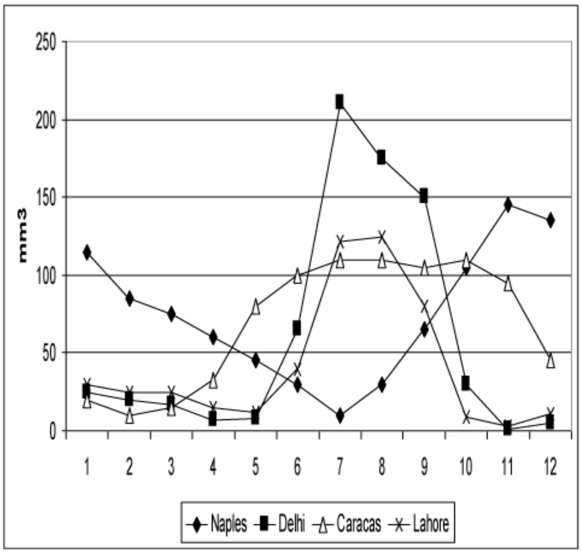

Figure 5: Monthly rainfall (mm)

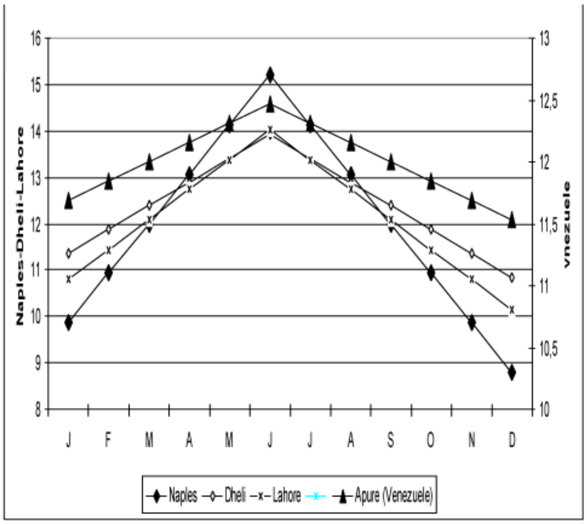

Figure 6: Trend of daily light hours in Naples, Dheli, Appure (Venezuele), Lahore

However, the trend of daily light hours, although with different daily values, is shared by all the areas of breeding situated north of the equator (Figure 6). Interestingly, a 4-year retrospective analysis of data (B Gasparrini, unpublished observations) obtained in an in vitro embryo production laboratory showed that a significant decrease of blastocyst rate is observed between April-June compared to October-December, whereas intermediate values were recorded between July and September and between January and March. It results that the drop of oocyte developmental competence coincides with the spring months that, at our latitudes are characterized by mild environmental temperatures; this pattern confirms that the light stimulus plays the most critical role in determining seasonality.

South of the equator Baruselli et al., demonstrated that calving gathering accentuates proportionally at increasing distance from the equator (Figure 2). It is not possible to show any relationship between daily maximum temperature, rainfall and the calving calendar (Figure 2) whereas an evident relationship exists between the latter and the daily light hours at different latitudes (Figure 7).

Throughout this paper, the seasonality of the females has been considered. However, the phenomenon may affect also the males and the effects are clear when the animals are maintained in free mounting condition. Recently, we observed in the same farm a higher pregnancy rate between January and April in buffaloes inseminated by AI, rather than in animals that were bred together with the bull, probably because the AI avoided the negative effect of the bull.

Furthermore, we have recently verified [68] that in April only 23%, 31% and 29% of the bulls showed values higher than the average value respectively of testosterone, diidro testosterone and androstenedione.

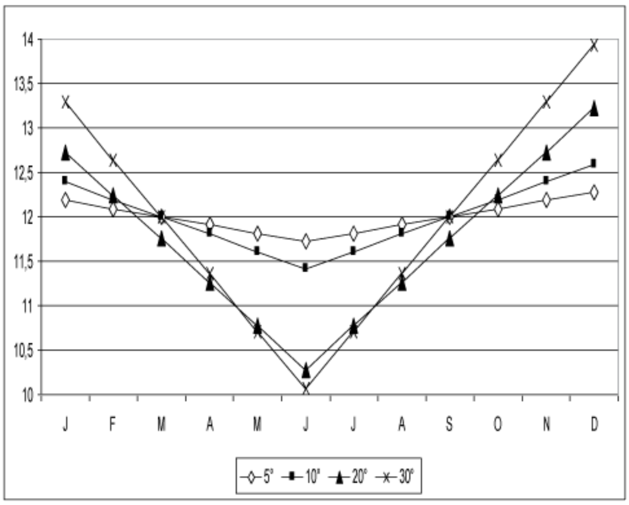

Figure 7: Trend of daily light hours in Brazil in function of latitude (°)

Strategies to enhance reproductive performance

The delayed puberty and the consequent older age at first calving depend on both inappropriate weaning techniques and inadequate feeding during growth. With a rationing characterized by energy density of 0.8 MUF (MUF = Milk unit forages = 1700 kcal NEl) and 12.5-13% protein content on dry matter, together with forage/concentrate ratio of 50-60%, age at first calving is 28-32 months.

Embryonic mortality in buffalo species is primarily due to a reduced secretion of progesterone by corpus luteum. The importance of progesterone concentration during the first weeks of pregnancy for reducing embryonic mortality has been demonstrated in both cattle [69] and buffalo [70-72]. In cattle, several treatments have been utilized usually within day 5 after conception to enhance progesterone secretion by the existing corpus luteum or to induce ovulation and formation of an accessory corpus luteum (Mann 2002). However, while treatments on day 5 post-insemination do not have any effect in reducing embryonic mortality in buffalo delayed treatment of buffaloes with GnRH agonist, hCG or progesterone on Day 25 after AI reduces embryonic mortality [73,74].

Although the light/dark ratio is the main factor affecting reproductive efficiency, another important factor is the satisfaction of the buffalo phisiological need of water for bathing.

The swimming-pool presence reduced the not pregnant buffaloes/corpora lutea found at rectal examination ratio (NP/CL) in buffaloes that calved between April and August, because it acted clearly as a tool against heat stress. NP/CL ratio, as expression of anomalous oestrous cycle or embryonic mortality, may be proposed as a specific tool for evaluating buffalo welfare.

The treatments for anoestrus are based on the use of progesterone devices combined with PMSG. However, the results are influenced by the year/farm effect and therefore are variable. The above treatments in natural mating conditions do not have an immediate impact, but especially in primiparous, they have a beneficial effect on the R.R.C.. Unsatisfactory responses are perhaps more useful because they lead to assess the environmental causes that determine the failures.

For instance, we have demonstrated that increased space and better welfare conditions improve fertility in Italy. In 21 farms in which the OBMS is performed we observed that 38.1%, 52.4% and 4.8% of the farms increased fertility rate respectively on June, July-August and September (L Zicarelli, unpublished observations). Out of the 8 farms that resumed fertility in June, 6 (75%) had at their disposal either covered sheds, that shorten the day length on average by 2 h during the year, or wide open spaces in which buffalo cows can move for at least 6 h/day or swimming pools. Out of the 11 farms that resumed fertility in July only 2 (18,2%) had swimming pools available (6/8 vs. 2/11; P < 0,05).

In a farm in which a variable number of heads have been bred for 10 years in the same space, an increase of reproductive activity in June was recorded when the buffaloes bred were on average 352 and in July when the number of buffaloes were on average 451. Between January and June 77% and 68% calving was observed respectively for 352 and 451 buffaloes (P < 0,01).

These finding suggest that the effects of season can in part be attenuated by improving the welfare status of the animals.

Conclusions

The seasonality in buffalo species is influenced by the light/dark ratio throughout the year. In some Countries the seasonality is influenced by nutritional factors. Furthermore, the improvement of the welfare of the subjects (swimming-pool, space, brightness of the stalls) can increase the percentage of calving between March - June. Delayed treatment of buffaloes with GnRH agonist, hCG or progesterone on Day 25 after AI can reduce the embryonic mortality in the months in which daylight hours increase. A significant improvement in reproductive efficiency can be achieved by increasing the culling rate from 10% -14% to 25% -30%, and hence eliminating older subjects and those with reproductive problems.

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