Abstract

The aim of this study was to determine the effect of season and parity on water buffalo calving distribution throughout the year in Venezuela. A retrospective study analyzing records of 3,192 crossbred (Murrah/Mediterranean) buffaloes including 7,790 calvings was carried out in Venezuela. Two seasons were evaluated according to the length of the photoperiod: September-February (short photoperiod; autumn-winter) and March-August (long photoperiod; spring-summer). Parity order was categorized in parity 1, parity 2, parity 3, and parity 4. Photoperiod was shorter between September and February in comparison with March-August (11.81 ± 0.08 h/day and 12.42±0.08 h/day, respectively, p < 0.05). Percentage of calving varied between the two seasons (p < 0.05), and 63.7% (95% Confidence Interval (CI): 62.5%-64.9%) of calving occurred during the season of short photoperiod in comparison with that of the season of long photoperiod (36.3%, 95% CI: 35.1%-37.5%), and this effect was observed in all parities. The proportion of calvings during the short photoperiod season was greater as parity increased. The percentage of calving during the short photoperiod season was ...
lower in primiparous water buffalo cows in comparison with that of multiparous water buffalo cows; conversely, the percentage of calving during the long photoperiod season was higher in primiparous water buffalo cows in comparison with that of multiparous water buffalo cows. Eight percent of water buffalo cows having their first calving in the long photoperiod season had the next calvings during this season. In conclusion, results confirm the seasonal reproductive behavior of water buffaloes with a concentration of calvings during the months with short photoperiod; this seasonality becomes stronger as parity increases; even though a small percentage of water buffalo cows has the ability of calving constantly during long photoperiod.

**Keywords:** Buffalo cows. Seasonality. Photoperiod. Short days. Parity.

**Resumo**

O objetivo deste estudo foi determinar o efeito da estação e paridade na distribuição do parto de búfalas durante o ano. Um estudo retrospectivo foi realizado na Venezuela, analisando os registros de 3192 búfalas mestiças (Murrah/Mediterrânea) e incluindo 7790 partos. De acordo com o fotoperíodo, duas estações foram avaliadas: setembro-fevereiro (fotoperíodo curto; outono-inverno) e março-agosto (fotoperíodo longo; primavera-verão). A ordem de parto foi categorizada em paridade 1, paridade 2, paridade 3 e paridade 4. O fotoperíodo foi menor entre setembro e fevereiro (11,81 ± 0,08 h, p < 0,05) em comparação a março-agosto (12,42 ± 0,08 h). A porcentagem de parto variou entre as duas estações (p < 0,05): 63,7% (95% intervalo de confiança [IC]: 62,5%-64.9%) dos partos aconteceram durante a estação de fotoperíodo curto em comparação com 36,3% (95% IC: 35,1%-37.5%) durante o fotoperíodo longo; esse efeito foi observado independentemente da paridade. A concentração de partos durante o fotoperíodo curto foi maior à medida que o número de partos aumentou. A porcentagem dos primeiros partos durante as estações do fotoperíodo curto e longo foi menor e maior, respectivamente, em comparação com o parto de búfalas multiparas. Oito por cento das búfalas que tiveram seu primeiro parto na estação de fotoperíodo longo tiveram seus partos seguintes durante esta mesma estação. Concluiu-se que o comportamento reprodutivo das búfalas é concentrado durante os meses com fotoperíodo curto; essa sazonalidade se torna mais forte à medida que o número de partos aumenta. Uma pequena porcentagem de búfalas tem a capacidade de parir constantemente durante a estação do fotoperíodo longo.

**Palavras-chave:** Búfalas. Sazonalidade. Fotoperíodo. Dias curtos. Paridade. Número de partos.

**Introduction**

Water buffaloes (*Bubalus bubalis*) bring important economic and social benefits to Venezuela (Menéndez-Buxadera and Verde, 2014), and in consequence, the number of water buffalo herds has been steadily increasing in recent years. A high number of water buffalo herds are reared for milk production due to the high contents of total solids present in water buffalo milk, which makes it very appealing for the cheese industry. Nevertheless, the supply of milk could be irregular because reproductive activity of water buffaloes is not uniform during the year; and in some countries this situation could represent a problem in the market of products derived from water buffalo milk (Carcangiu et al., 2011; Zetouni et al., 2014).

Reproductive activity of buffaloes tends to be seasonal, with a higher activity occurring in the season when photoperiod is short (Baruselli et al., 2001; Phogat et al., 2016; Zicarelli, 2017; Gasparrini, 2019). Photoperiod duration varies according to the month and latitude. In the Northern Hemisphere, it begins to diminish during autumn (September-November), and the shortest photoperiod occurs in winter (December-March); while in the Southern Hemisphere, short photoperiod occurs from March to September (Autumn-Winter in Southern Hemisphere) (Petersen et al., 2017).

Reproductive seasonality is a behavior that guarantees the survival of calves (Zicarelli, 2010; Gasparrini, 2019) and is orchestrated by the pineal gland and melatonin (Parmeggiani et al., 1994). However, an influence of the variation of the forage supply throughout the year cannot be ruled out in tropical areas (Zicarelli, 2010). In the case of water buffaloes and other species, reduction of the
photoperiod increases melatonin production and secretion, which in turn promotes the synthesis and secretion of GnRH, the releases of gonadotropins, and ovarian activity (Bittman and Karsch, 1984; Parmeggiani et al., 1994; Ramadan, 2017).

In order to address the effect of seasonal reproductive behavior on the supply of milk, water buffaloes are incorporated into a de-seasonalization practice in some countries, the out-of-season breeding strategy (OSBS) (Zicarelli, 2010). This practice is based, in many cases, on the use of hormonal protocols to induce the estrus and the ovulation so that calving occurs during the period of low reproductive activity (long photoperiod) (Bhat et al., 2014; Carvalho et al., 2016). However, this can be an expensive alternative, especially in developing countries, in which many of the hormones must be imported. Therefore, identifying additional factors that regulate the seasonal behavior of buffaloes is of interest in order to design strategies that allow a more uniform distribution of births throughout the year or a higher concentration of these at the time of greatest reproductive activity, according to the demands of the market or the needs of farmers. The objective of this study was to determine the effect of season and parity on water buffalo calving distribution throughout the year in Venezuela.

Material and methods

This was a retrospective study analyzing reproductive records of 3,192 crossbred (Murrah/Mediterranean) water buffalo cows from three herds including 7,790 calvings occurring in a period of ten years. The farms were located in the same agroecological area in Catatumbo and Jesus María Semprún counties, Zulia State, Venezuela. Animals grazed in pastures consisting of Brachiaria humidicola and Brachiaria arrecta in one and two-day paddock rotations. Mechanical milking was performed twice a day after a short sucking by the water buffalo’s own calf to stimulate milk letdown. During the milking, the calf remained bound to the water buffalo forelimb, and after milking the calves suckled their mothers to remove residual milk from the udder. A water buffalo bull for each 25 cows was present continuously during the year. The farms maintained computerized and paper records of each water buffalo cow, information about month of calving and calving order (until parity 4) was abstracted for this study. Information about duration of photoperiod/darkness for El Guabayo city (Catatumbo county) and Casigu el Cubo city (Jesus Maria Semprun county) was obtained from Time and Date AS©. Since both cities are nearby and with a similar latitude, the average of the values of photoperiod/darkness from both cities was used.

Statistical analysis

From the date of calving the corresponding month was obtained and a categorical variable called season of calving was created according to the photoperiod duration, with the following values: September-February (short photoperiod), corresponding to autumn-winter; and March-August (long photoperiod), corresponding to spring-summer. The length of photoperiod and of darkness were regressed on season using linear mixed models (PROC MIXED in SAS) with random intercepts (months). Parity was categorized in parity 1, parity 2, parity 3, and parity 4. The distribution of calving per photoperiodical season was assessed using an unconditional logistic regression, estimated using a GEE approach, independent working correlation, and robust standard errors.

The effect of parity on the distribution of calvings per photoperiodical season was evaluated using the same logistic regression just mentioned, adding parity as a predictor. Similarly, the distribution of calving per month was assessed using an unconditional multinomial logistic regression with robust standard errors calculated at the cluster level (water buffalo cow). The effect of parity on the distribution of calvings per month was evaluated using the same multinomial logistic regression just mentioned, adding parity as a predictor. Analyses were performed in SAS 9.4 and Stata 15.1. Results are reported as means ± standard error of the mean (SEM) or as proportions and 95% Confidence Intervals (CI). Differences were considered statistically significant when $p$ values were ≤ 0.05.
Results

The duration of photoperiod and darkness varied significantly between seasons. Photoperiod was shorter and darkness longer between September and February (Figure 1, Table 1) as it is expected from a country in the Northern Hemisphere. The percentage of calving varied among months (p < 0.05) (Figure 2); and a higher proportion of calving (p < 0.05) occurred during the season of short photoperiod (63.7%, 95% CI: 62.5-64.9) in comparison with that of the long photoperiod season (36.3%, 95% CI: 35.1-37.5).

The season affected the percentage of calving regardless of parity. Percentages of calving were higher during September-February in the four parity categories (Table 2, Figure 3). It was observed that the proportion of calving during short photoperiod was greater as the number of calving increased. Moreover, it was observed that the percentage of calving during long photoperiod season was higher in primiparous water buffalo cows than that of multiparous water buffalo cows (Table 2), especially during April and May (Figure 3), when the proportion of calvings was 9.1% in primiparous water buffalo cows in comparison with 4.4%, 4.3%, and 7.4% in water buffalo cows with 2, 3, and 4 calvings, respectively. Additionally, 8% of water buffalo cows having their first calving in the long photoperiod season, had the next three calvings in the same season.

Figure 1 - Monthly duration of photoperiod and darkness. Combined data from El Guayabo (8.62443° - 72.3389°) and Casigua el Cubo (8.75722° - 72.535°) at Zulia State, Venezuela.

Table 1 - Duration of photoperiod and darkness (hours) according to the season. Combined data from El Guayabo (8.62443° - 72.3389°) and Casigua el Cubo (8.75722° - 72.535°) at Zulia State, Venezuela

| Season of calving     | Photoperiod (h) (means ± se) | Darkness (h) (means ± se) |
|-----------------------|------------------------------|---------------------------|
| Short photoperiod     |                              |                           |
| September - February  | 11.81 ± 0.08°                | 12.19 ± 0.08°             |
| Long photoperiod      |                              |                           |
| March - August        | 12.42 ± 0.08°                | 11.58 ± 0.08°             |

Note: Values in the same column with different superscript letters are significantly different (p < 0.05).
Effect of season and parity on water buffalo calving distribution throughout the year in Venezuela

**Figure 2** - Monthly distribution of the percentage of calvings in crossbred Murrah/Mediterranean buffaloes at Zulia State in Venezuela.

**Figure 3** - Monthly distribution of percentage of calving according to parity in crossbred Murrah/Mediterranean buffaloes at Zulia State in Venezuela.
Discussion

In this study we determined the distribution of calvings of water buffaloes throughout the year and evaluated the effect of seasons with different photoperiod/darkness duration on the distribution of calvings of water buffaloes. In addition, the influence of parity on the percentage of calvings in each photoperiodical season was evaluated.

Results of the present study coincide with previous reports (Zicarelli, 1994; Montiel-Urdaneta et al., 1997; Pérez Rojo et al., 2016; Cerón-Muñoz et al., 2017; Gunwant et al., 2018a) showing that water buffalo cows tend to exhibit a seasonal reproductive activity, with calvings concentrated during months with decreasing photoperiod (Figures 1, 2 and 3). In Venezuela, it was reported that 82.77% of calvings occurred between September and December (Montiel-Urdaneta et al., 1997) and 70.98% occurred between September and February (Montiel-Urdaneta, 2006), while Colmenares et al. (2007) reported that 83.3% of calvings occurred between August and November. Seasonality of calvings is a consequence of the concentration of estrus and conception during months with short photoperiod, in which there are high plasma levels of melatonin (Parmeggiani et al., 1994; Kassim et al., 2008) that promote the secretion of gonadotropins; thus, both heifers and postpartum water buffalo cows show ovarian activity and become pregnant. After a gestation of 314.04±8.30 days (Montiel-Urdaneta, 2006), calving occurs during the short photoperiod season. In Colombia, Sánchez et al. (2017) observed 61.9% of calvings between September and February, and this concentration of calving was associated with lower temperature and higher rainfall. In Cuba, 76% of calvings of Buffalipso water buffaloes, occurred between June and September (Almaguer et al., 2013), while in India (Bundelkhand region), 69.6% of calvings of Murrah water buffaloes occurred between September and February (Kushwaha et al., 2011). The effect of photoperiod on reproductive activity of water buffaloes is confirmed by the fact that calvings are concentrated between February and May in the Southern Hemisphere, when the photoperiod is also short (Zicarelli 2010, 2017).

In the present study, 63.7% and 36.3% of calvings occurred during the short and long photoperiod seasons, respectively, and these percentages changed slightly, but significantly, according to parity. Seasonal behavior increases with parity from 60.4% of calvings in the short photoperiod season in first-calving water buffalo cows to 67.2% of calvings in the short photoperiod season in fourth-calving water buffalo cows (p < 0.05). The percentage of heifers having their first calving during long photoperiod season was higher than the percentage of multiparous buffaloes calving in this season, and this was similar to the results of Cerón-Muñoz et al. (2017). Heifers are less sensitive to photoperiod than water buffalo cows who have previously had a calf, and have less seasonal behavior, probably because they have higher melatonin levels during

| Parity    | Short photoperiod September-February (%) | 95% CI* (%) | Long photoperiod March-August (%) | 95% CI* (%) |
|-----------|-----------------------------------------|-------------|----------------------------------|-------------|
| First     | 60.6a (n = 1916)                         | 58.9 - 62.3 | 39.4ab (n = 1244)                | 37.7 - 41.1 |
| Second    | 64ab (n = 1340)                          | 62.0 - 66.1 | 36ab (n = 753)                   | 33.9 - 38.0 |
| Third     | 67.2ab (n = 1009)                        | 64.8 - 69.6 | 32.8ab (n = 493)                 | 30.4 - 35.2 |
| Fourth    | 67.2ab (n = 696)                         | 64.4 - 70.1 | 32.8ab (n = 339)                 | 29.9 - 35.6 |

Note: Values in the same column with different lowercase superscripts are significantly different (p < 0.05). Values in the same row with different uppercase superscripts are significantly different (p < 0.05). *95% Confidence interval.
the hours of light, being able to come in estrus and become pregnant during long photoperiod season (Zicarelli, 1994; Borghese et al., 1995; Cerón-Muñoz et al., 2017). In addition, it has been observed that heifers maintained in an extended photoperiod during winter had a higher growth rate and an early onset of puberty, without affecting the levels of melatonin compared to controls (Roy et al., 2016). Farmers practicing the OSBS can take advantage of this, and with an adequate nutritional management, they can prepare heifers to calve during the long photoperiod season to produce more milk during this season. Nevertheless, this strategy implies that primiparous water buffaloes must have an especial management to prevent anestrus and infertility and thus guarantee a short conception interval. Primiparity and calving during long photoperiod season affect negatively the reproductive performance (Hassan et al., 2017; El-Tarabany, 2018; Nava-Trujillo et al., 2018) and this affects negatively the herd’s profitability (Cicek et al., 2017; Hassan et al., 2017).

Additionally, in the present study it was observed that 8% of water buffaloes having the first calving in the long photoperiod season, had the next three calvings in the same season. A predisposition to calving during the long photoperiod season could exist and identifying the subpopulation of heifers with non-seasonal behavior could be an advantage and will facilitate the implementation of the OSBS. Carcangiu et al. (2011), Luridiana et al. (2012) and Gunwant et al. (2018b) observed that water buffaloes with the genotype TT in the gene of melatonin receptor 1A were able to mate and calve during the long photoperiod season, with this behavior being repetitive in the following years (Carcangiu et al., 2011). Although buffalo milk production is more profitable than cow's milk production (Menghi et al., 2007) and farmers seek non-seasonal calvings in some countries (Zicarelli, 2010; Gasparini, 2019), season of calving could affect the profitability of buffalo herds. Hassan et al. (2017) under Egyptian conditions showed that water buffaloes calving in winter produced more milk, had a higher reproductive efficiency, and a better economic efficiency than water buffaloes calving in the other seasons. Therefore, the implications of maintaining or not the calving seasonality will depend on the context of each region or country and the goals of each farmer.

**Conclusion**

Water buffalo cows exhibit a seasonal reproductive activity with a concentration of calving during months of short photoperiod, September-February in Venezuela; this seasonal behavior is influenced by parity and becomes stronger as parity increases; heifers seem to be less sensitive to photoperiod, which leads to a higher proportion of first calvings during the months with long photoperiod (March to August, > 12 hours). A small percentage of water buffalo cows has the ability of calving constantly during long photoperiod season.

**References**

Almaguer PY, Font PH, Quirino CR, Montes I, Rosell R, Barzaga R, et al. Estacionalidad de los partos en hembras bubalinas, tipo Buffalipso, de la empresa agropecuaria Bayamo. Actas Iberoam Conserv Anim. 2013;3:78-83

Baruselli PS, Bernardes O, Braga DP, Araujo DC, Tonhati H. Calving distribution throughout the year in buffalo raised all over Brazil. VI World Buffalo Congress; 2001 May 21-23; Maracaibo, Venezuela; 2001. p. 234-9.

Bhat GR, Dhaliwal GS, Ghuman SPS. Low breeding season and estrus synchronization in buffalo (Bubalus bubalis). Ind J Vet Sci. 2014;2(1):28-33.

Bittman EL, Karsch FJ. Nightly duration of pineal melatonin secretion determines the reproductive response to inhibitory day length in the ewe. Biol Reprod. 1984;30(3):585-93.

Borghese A, Barile VL, Terzano GM, Pilla AM, Parmeggiani A. Melatonin trend during season in heifers and buffalo cows. Bubalus Bubalis. 1995;1:61-4.

Carcangiu V, Mura MC, Pazzola M, Vacca GM, Paludo M, Marchi B, et al. Characterization of the Mediterranean Italian buffaloes melatonin receptor 1A (MTNR1A) gene and its association with reproductive seasonality. Theriogenology 2011;76(3):419-26.
Cerón-Muñoz MF, Agudelo-Gómez DA, Ramírez-Arias JP. Estacionalidad de partos de búfalas en Colombia. Livestock Res Rural Dev. 2017;29(2):Article#38.

Gīcek H, Tandogan M, Uyarlar C. Financial losses due to fertility problems in Anatolian dairy buffalo. Indian J Anim Res. 2017;51(6):1144-8.

Colmenares O, Bello R, Herrera P, Birbe B, Martínez N. Non-genetic factors affecting calving interval and weaning weight in a buffalo herd located in well drained savannas, Guárico state, Venezuela. Ital J Anim Sci. 2007;6(Suppl 2):1354-6.

El-Tarabany MS. Survival analysis and seasonal patterns of pregnancy outcomes in Egyptian buffaloes. Livest Sci. 2018;213:61-6.

Gasparrini B. Effect of reproductive season on embryo development in the buffalo. Reprod Fertil Dev. 2018;31:68-81.

Gunwant P, Pandey AK, Kumar A, Singh I, Kumar S, Phogat JB, et al. Polymorphism of melatonin receptor (MTNR1A) gene and its association with seasonal reproduction in water buffalo (Bubalus bubalis). Anim Reprod Sci. 2018b;199:51-9.

Gunwant P, Pandey AK, Singh I, Phogat JB, Kumar S, Kumar S. Seasonal Variation of Calving in Murrah Buffaloes at Organized Dairy Farm. Int J Pure App Biosci. 2018a;6(1):1283-7.

Hassan FAM, Ali MA, El-Tarabany MS. Economic impacts of calving season and parity on reproduction and production traits of buffaloes in the sub-tropics. Environ Sci Pollut Res Int. 2017;24(11):10258-66.

Kassim NSI, Afify AA, Hassan HZ. Effect of photoperiod length on some reproductive traits and hormonal profiles in buffalo heifers. American-Eurasian J Agric Environ Sci. 2008;3(4):646-55.

Kushwaha BP, Singh S, Maity SB. Seasonality of calving in Bhadawari and Murrah buffaloes in Bundelkhand, India. Buffalo Bulletin. 2011;30(4):256-61.

Luridiana S, Mura MC, Pazzola M, Paludo M, Cosso G, Dettori ML, et al. Association between melatonin receptor 1A (MTNR1A) gene polymorphism and the reproductive performance of Mediterranean Italian buffaloes. Reprod Fertil Dev. 2012;24(7):983-7.

Menéndez-Buxadera A, Verde O. Componentes de (co) varianza de la producción de leche de un rebaño bufalino venezolano estimados con modelos de lactancia completa o del día de control. Zootecnia Trop. 2014;32(1):63-75.

Menghi A, Corradini E, De Roest K. Profitability of buffalo’s milk in the province of Latina (Italy) in 2004 and 2005. Ital J Anim Sci. 2007;6(Suppl 2):1390-3.

Montiel-Urdaneta NS. Algunos aspectos reproductivos e inseminación artificial en búfalas. Proceedings X Seminario de Pastos y Forrajes; 2006 Apr 20-22; Maracaibo, Venezuela. 2006. p. 174-86.

Montiel-Urdaneta N, Rojas N, Angulo F, Hernández A, Zuleta J, Cahuao N, et al. Efecto de algunos factores ambientales sobre la estacionalidad en los partos en búfalas. Arch Latinoam Prod Anim. 1997;5(Supl. 1):423-5.

Nava-Trujillo H, Escalona-Muñoz J, Carrillo-Fernández F, Parra-Olivero A. Effect of parity on productive performance and calving interval in water buffaloes. J Buffalo Sci. 2018;7(1):13-6.

Parmegiani A, Di Palo R, Zicarelli L, Campanille G, Esposito L, Seren E, et al. Melatonin and reproductive seasonality in the buffalo cow. Agric Ric. 1994;153:41-8.

Pérez Rojo AF, Bernal Calderon A, Herrera Rios AC. Seasonality and influence on birth of females buffaloes. Ces Med Vet Zootec.. 2016;11(3):193.

Phogat JB, Pandey AK, Singh I. Seasonality of buffalo reproduction. Int J Plant Anim Environ Sci. 2016;6(2):46-54.
Ramadan TA. Role of Melatonin in Reproductive Seasonality in Buffaloes. In: Payan-Carreira R (E.). Theriogenology. London, UK: IntechOpen; 2017. p.545-54.

Roy AK, Singh M, Kumar P, Kumar BSB. Effect of extended photoperiod during winter on growth and onset of puberty in Murrah buffalo heifers. Vet World. 2016;9(2):216-21.

Sánchez JA, Romero MH, Vela YJS. Estacionalidad reproductiva de la hembra bufalina (Bubalus bubalis). Rev Investig Vet Perú. 2017;28(3):606-18

Carvalho NA, Soares JG, Baruselli PS. Strategies to overcome seasonal anestrus in water buffalo. Theriogenology 2016;86(1):200-6.

Zetouni L, Camargo GM, Fonseca PDS, Cardoso DF, Gil FM, Hurtado-Lugo NA, et al. Polymorphisms in the MTRN1A gene and their effects on the productive and reproductive traits in buffaloes. Trop Anim Health Prod. 2014;46(2):337-40.

Zicarelli L. Management under different environmental conditions. Buffalo J. 1994;2:17-38.

Zicarelli L. Enhancing reproductive performance in domestic dairy water buffalo (Bubalus bubalis). Soc Reprod Fertil Suppl. 2010;67:443-55.

Zicarelli L. Influence of Seasonality on Buffalo Production. In: Precisse GA, editor. The Buffalo (Bubalus bubalis)-Production and Research. Sharjah, UAE: Bentham Science Publishers; 2017. p.196-224.