

REPRODUCTIVE SCREENING AND ENHANCEMENT OF REPRODUCTIVE  
PERFORMANCE OF BUFFALOES (*Bubalus bubalis*) ON FEEDING  
BYPASS FAT AND AREA SPECIFIC MINERAL MIXTURE

Ashutosh Tripathi\* and Jitendra Singh Mehta

Received: 19 April 2020

Accepted: 25 December 2022

**ABSTRACT**

A total of 491 buffaloes (Murrah and non-descript) reared under unorganized system from semi-arid and arid areas of Rajasthan, India, were screened for their reproductive status. These were screened by history taking, rectal palpation, Ultrasonography and follow up. Out of these screened animals 65.59% were of normal reproductive status (Involved recently parturiated, pregnant etc. animals) while remaining included 11% cases of delayed puberty, 7.94% of delayed post-partum estrus, 5.5% of anestrus/ subestrus, 3.87% of repeat breeding, 2.85% of prolapse, 2.44% of infectious infertility and only 0.81% cases were of cystic ovarian degeneration (COD). Thirty animals were given bypass fat and area specific mineral mixture (ASMM) following parturition till 90 days postpartum, and results were compared with 20 animals from Control group. The uterus of animals given bypass fat and ASMM took significantly less time ( $24.50 \pm 0.34$  mean days) for involution than Control group ( $33 \pm 0.61$  mean days) also Experimental group took significantly less days ( $62.60 \pm 0.77$  mean days) to show first estrus postpartum than Control group ( $87.00 \pm 0.44$  mean days). First AI conception rate of Experimental

group (60%) was also better than Control group (45%).

**Keywords:** *Bubalus bubalis*, buffaloes, bypass fat, ASMM, uterine involution, first postpartum estrus, conception rate

**Abbreviations**

COD: Cystic Ovarian Degeneration

ASMM: Area Specific Mineral Mixture

AI: Artificial Insemination

NDDDB: National Dairy Development

Board

DM: Dry matter

USG: Ultrasonography

ROP: Retention of placenta

NEB: Negative energy balance

LH: Luteinizing hormone

IGF-1: Insulin like growth factor-1

PUFA: Poly Unsaturated Fatty Acids

PGFM: Prostaglandin F metabolite

**INTRODUCTION**

India reached milk production (Provisional) of around 209.96 (2020-21) million tonnes out of

which around 45% milk is produced by buffaloes (GOI, 2021). Dairy profitability entirely depends on the optimum production. For optimum production to be maintained animal need to maintain optimum reproductive efficiency i.e., a calf each year which can be possible only if animal is given optimum nutrition based on scientific recommendations along with timely therapeutic intervention in cases of reproductive disorders. Ruminants are fed mostly crop residues with low energy, protein, and minerals which limit productive and reproductive performances (Ranjan *et al.*, 2012). This can be overcome by feeding rumen bypass fat (Ca salt of fatty acid) which is partially resistant to biohydrogenation by the rumen microbes without risk of acidosis (Naik *et al.*, 2009; Block *et al.*, 2005). The optimum results were more evident at the early lactation that too 2 to 3 % of bypass fat (150 to 300 g/day) (Gargouri *et al.*, 2006). Apart from energy, macro and micro-nutrients play an important role in animal reproduction as they form components of metallo-enzymes and enzyme co-factors (Kalasariya *et al.*, 2016; Vala *et al.*, 2018) thus directly and indirectly control endocrine mechanisms. However, areas with harsh climate like semi-arid and arid regions of Rajasthan, limit this reproductive efficiency even if animal is given optimum nutrition therefore greater emphasis is to be given on the interventions needed to improve reproduction especially in buffaloes because these prefer low temperature and tropical climate. In the present project buffaloes were screened for their reproductive performance under semi-arid climate also animals were given bypass fat and ASMM to study enhancement in reproductive performance post-partum.

## MATERIALS AND METHODS

### Location of the study

Animals from semi-arid area (in and around Bikaner) of Rajasthan, India were screened for various reproductive disorders by history taking, rectal palpation, ultrasonography and follow up at regular intervals. Total sum of 491 animals were screened.

### Animals and Treatment group

The rumen-protected bypass fat (EnerFAT) is the Ca salt of a fatty acid consisting of a mixture of long-chain saturated and unsaturated fatty acid based on palm oil fatty acid distillate, a commercial product constituting 84% of fat (on DM basis). Thirty Post parturient buffaloes were taken in group as experimental and 20 animals as Control group and subjected to feeding by pass fat (source of energy) and ASMM from day of parturition to 90 days post-partum. The bypass fat and ASMM were given at rate of 20 g/ kg milk and 100 g/ day to buffaloes, respectively. The animals were screened at every seven days (starting 14 days postpartum) for assessing uterine involution. The effect of feeding of bypass fat and ASMM on uterine involution, day of first estrus post-partum and first estrus conception rate were studied.

### Statistical analysis

All statistical procedures were carried out as per Snedecor and Cochran (1994).

## RESULTS AND DISCUSSIONS

Reproductive screening revealed that buffaloes are less prone towards cystic ovarian degeneration (Agarwal *et al.*, 2005) as mentioned

in Table 1 and Figure 1 however up to 29% cases of COD (out of total ovarian affections) have been reported but that too from the culled animals also more cases were in right ovary (Srinivasan *et al.*, 2017), in the present research all the animals were of age less than 10 years however in previous reports these were found to be of old age groups (Srinivasan *et al.*, 2017).

These animals showed highest cases of delayed puberty, delayed post-partum estrus and anestrus/ subestrus which may be due to nutritional deficiency together with poor unorganized management and climatic stress of semi-arid and arid region (Warriach *et al.*, 2015).

Infectious infertility cases were having history of dystocia/ retention of placenta/ prolapse during parturition. Out of total 12 cases 8 were having history of dystocia while 03 were with the history of retention of placenta while 01 case was having history of prolapse during parturition. Previously also it has been reported to be potent cause of infertility problems in buffaloes concurrent to parturition and postpartum contamination of pathogenic bacteria commonly (Azawi, 2010).

The incidence of Prolapse case was around 2.85% which is slightly lower than reported previously with 3 to 16% incidence (Nanda and Sharma, 1982; Bhatti *et al.*, 2006; Mishra *et al.*, 1997; Rabbani *et al.*, 2010; Akhtar *et al.*, 2012; Al-Saeed *et al.*, 2017; Kumar and Singh, 2009; Khtri *et al.*, 2013; Thakur *et al.*, 2017). All these animals were multiparous and of age group more than 10 years, none of the case involved primiparous animals. Out of total 14 cases more than 50% cases (08/14) were related to difficult or assisted birth while (04/14) could not shed their placenta in time however 02 cases were not having any history of above-mentioned reasons also none of these two animals prolapsed before, above mentioned

history involve the predisposing factors reported previously also (Ingawale *et al.*, 2020).

Feeding bypass fat significantly increased rate of involution of uterus compared to Control group as mentioned in Table 2. As has been reported before that these fats contain poly unsaturated fatty acids (PUFA) which enhance levels of PGFM in serum thus stimulating secretion of PGF2 $\alpha$  which favors uterine involution (Mattos *et al.*, 2004). However, some authors could not find significant effects of bypass fat over uterine involution in cattle (Nirwan *et al.*, 2019)

Feeding bypass fat significantly reduced mean days required to show first estrus post-partum (Table 2) by preventing negative energy balance (NEB) (poor nutrition and milk flushing) and thus shorten duration of post-partum acyclicity. Animals with low Body Condition Score are more prone to acyclicity (Baruselli *et al.*, 2001) so feeding bypass fat in the diet shows prompt response in cyclicity and fertility (Lopes *et al.*, 2009; Tyagi *et al.*, 2010; Aardema *et al.*, 2014). Conception rates too increased in groups fed bypass fat and ASMM (Table 3) as negative energy balance mediates its effects through glucose, insulin, insulin-like growth factor-1 (IGF-1) and luteinizing hormone (LH) (Beam and Butler, 1999) also it mobilize non-esterified fatty acid and triglyceride to the liver where their accumulation severely deteriorates fertility (Bertics *et al.*, 1992; Crowe *et al.*, 2014). However, a recent report differs from the above finding where feeding bypass fat to dairy cattle could not significantly reduce the time to show first estrus (Nirwan *et al.*, 2019).

Feeding ASMM along with bypass fat showed promising effects on the reproductive performance (early involution of uterus, less days required to first estrus and improved conception rates) (Table 2 and 3) as minerals are the

Table 1. reproductive status of buffaloes screened.

S. No.	Reproductive disorder	No. of animals (Total= 491)	Remarks
1	Animals with normal reproduction	322 (65.59%)	Involved recently parturited, pregnant etc. animals
2	Repeat breeder	19 (3.87%)	-
3	COD	04 (0.81%)	03/ 4 Right ovary
			01/4 Left ovary
4	Infectious infertility	12 (2.44%)	08/12 Dystocia
			03/12 ROP
			01/12 Prolapse
5	Prolapse	14 (2.85%)	08/14 Dystocia/ assisted birth
			04/14 Retention of placenta
			02/14 no previous history of prolapse
6	Anoestrus/ subestrus	27 (5.50%)	Unorganized rearing, poor nutrition
7	Delayed puberty	54 (11%)	
8	Delayed post-partum estrus	39 (7.94%)	

Table 2. Studies on the effect of feeding bypass fat and ASMM on involution and first estrus post-partum in buffaloes.

Parameter	Buffalo (Murrah and Non-descript)	
	Experimental group	Control group
Involution (Mean days post-partum)	24.50±0.34 <sup>a</sup>	33±0.61 <sup>b</sup>
First estrus (Mean days post-partum)	62.60±0.77 <sup>a</sup>	87.00±0.44 <sup>b</sup>

Different superscripts within row differ significantly ( $P \leq 0.01$ ).

Table 3. Studies on the effect of feeding bypass fat and ASMM on first AI conception rate in post parturient buffaloes.

Parameter	Buffalo (Murrah and Non-descript)	
	Experimental group	Control group
First A.I. conception rate	18/30 (60%)	9/20 (45%)

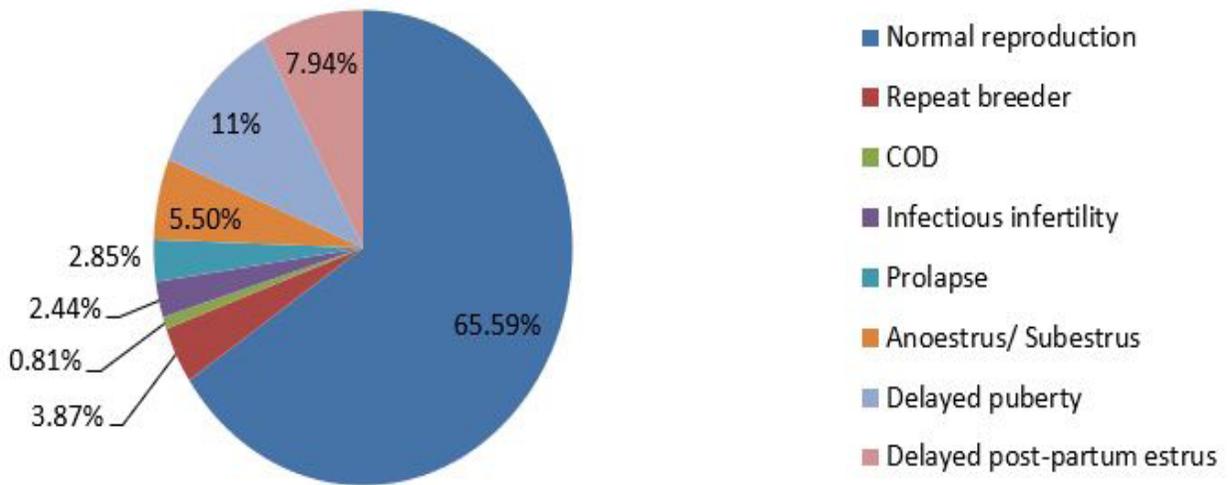


Figure 1. Reproductive status of screened buffaloes.

components of hormones and thus directly regulate the endocrine activity. Being indispensable part of carbohydrate, protein and nucleic acid metabolism these command production of reproductive and other hormones and hence affecting postpartum fertility (Kumar *et al.*, 2011).

### ACKNOWLEDGEMENT

Authors acknowledge, Indian Council of Agricultural Research and Rajasthan University of Veterinary and Animal Sciences, Bikaner for providing all the financial and administrative support for conducting the present research.

### REFERENCES

- Aardema, H., B.M. Gadella, C.H.A. Van de Lest, J.F.H.M. Brouwers, T.A.E. Stout, B.A.J. Roelen and P.L.A.M. Vos. 2014. Free fatty acid levels in fluid of dominant follicles at the preferred insemination time in dairy cows are not affected by early postpartum fatty acid stress. *J. Dairy Sci.*, **98**(4): 2322-2336. DOI: 10.3168/jds.2014-7970
- Agarwal, S.K., S.K. Singh and R. Rajkumar. 2005. Reproductive disorders and their management in cattle and buffalo: A review. *Indian J. Anim. Sci.*, **75**(7): 858-873.
- Akhtar, M.S., L.A. Lodhi, M.M. Ayaz, Z.I. Qureshi and G. Muhammad. 2012. Prevalence of puerperal period reproductive disorders in Nili-Ravi buffaloes of different parity in district Bahawalpur, Pakistan. *Pak. J. Vet. Anim. Sci.*, **2**: 79-82. Available on: <http://www.jvas.com.pk/doc/2012/V-2-2/4.pdf>
- Alexander, G., R.Z. Prabhakara and J. Rama Prasad. 2002. Effect of supplementing sheep with sunflower acid oil or its calcium soap on nutrient utilization. *Asian Austral. J. Anim.*, **15**(9): 1288-1293. DOI: 10.5713/ajas.2002.1288
- Al-Saed, A.H.N., S.A. Abd and S.G. Al-Maliki. 2017. A comparison between prolapse and mineral deficiency in Iraqi local cattle and buffalo cows. *Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences*, **2**(5): 32-40. DOI: 10.26479/2017.0205.03
- Azawi, O.I. 2010. Uterine infection in buffalo cows: A review. *Buffalo Bull.*, **29**(3): 154-171. Available on: <https://www.cabdirect.org/cabdirect/FullTextPDF/2011/20113095762.pdf>
- Baruselli, P.S., V.H. Barnabe, R.C. Barnabe, J.A. Visintin and R. Porto. 2001. Effect of body condition score at calving on postpartum reproductive performance in buffalo. *Buffalo Journal*, **17**: 53-65.
- Beam, S.W. and W.R. Butler. 1999. Effects of energy balance on follicular development and first ovulation in postpartum dairy cows. *J. Reprod. Fertil.*, **54**: 411-24. DOI: 10.1530/biosciprocs.4.032
- Bertics, S.J., R.R. Grummer, C. Cadorniga-Valino and E.E. Stoddard. 1992. Effect of prepartum dry matter intake on liver triglyceride concentration and early lactation. *J. Dairy Sci.*, **75**(7): 1914-1922. DOI: 10.3168/jds.S0022-0302(92)77951-X
- Bhatti, M.S., I. Ahmad, N. Ahmad., L.A. Lodhi and M. Ahmad. 2006. Epidemiological survey of genital prolapse in buffaloes kept under different systems and serum micro mineral contents. *Pak. Vet. J.*, **26**(4): 197-200. Available on: <http://www.pvj.com.pk/>

pdf-files/26\_4/page%20197-200.pdf

- Block, E., W. Chalupa, E. Evans, T. Jenkins, P. Moate, D. Palmquist and C. Sniffen. 2005. Calcium salts are highly digestible. *Feedstuffs*, **77**(30): 20-25.
- Crowe, M.A., M.G. Diskin and E.J. Williams. 2014. Parturition to resumption of ovarian cyclicity: Comparative aspects of beef and dairy cows. *Animal*, **8**(Suppl. 1): 40-53. DOI: 10.1017/S1751731114000251
- Gargouri, A., G. Caja, R. Casals and I. Mezghani. 2006. Lactational evaluation of effects of calcium soap of fatty acids on dairy ewes. *Small Ruminant Res.*, **66**(1-3): 1-10. DOI: 10.1016/j.smallrumres.2006.03.004
- GOI. 2021. *Annual Report. 2021-22*. Department of Animal Husbandry and Dairying, Ministry of Fisheries, Animal Husbandry and Dairying, Government of India, India.
- Ingawale, M.V., M. Siddiqui, M.G. Thorat, S.P. Waghmare and C.H. Pawshe. 2020. Management of post partum complete eversion of uterus in graded Murrah buffalo. *Buffalo Bull.*, **39**(1): 125-28. Available on: [https://kukrdb.lib.ku.ac.th/journal/BuffaloBulletin/search\\_detail/result/399040](https://kukrdb.lib.ku.ac.th/journal/BuffaloBulletin/search_detail/result/399040)
- Kalasariya, R.M., A.J. Dhimi, K.K. Hadiya, K.S. Mungad, V.P. Ramani and S.C. Parmar. 2016. Impact of peripartum nutritional supplementation on plasma minerals profile and postpartum fertility in buffaloes. *International Journal of Environmental Science and Technology*, **5**(6): 3749-3759.
- Khatri, P., D. Das, I. Kaka, M.U. Samo and B. Bhutto. 2013. Influence of environmental temperature on post partum reproductive potential of Khundi buffaloes. *Journal of Veterinary Advances*, **3**(4): 139-145.
- Kumar, R. and R. Singh. 2009. Incidence of utero-vaginal prolapse among the buffaloes under field conditions of western Uttar Pradesh. *Indian J. Anim. Sci.*, **79**(8): 847-849.
- Kumar, S., A.K. Pandey, W.A.A. Razzaque and D.K. Dwivedi. 2011. Importance of micro minerals in reproductive performance in livestock. *Veterinary World*, **4**(5): 230-233. Available on: <http://www.veterinaryworld.org/Vol.4/May%20-%202011/Importance%20of%20micro%20minerals%20in%20reproductive.pdf>
- Lopes, C.N., A.B. Scarpa, B.I. Cappelozza, R.F. Cooke and J.L.M. Vasconcelos. 2009. Effects of rumen-protected polyunsaturated fatty acid supplementation on reproductive performance of *Bos indicus* beef cows. *J. Anim. Sci.*, **87**(12): 3935-3943. DOI: 10.2527/jas.2009-2201
- Mattos, R., C.R. Staples, A. Arteché, M.C. Wiltbank, F.J. Diaz, T.C. Jenkins and W.W. Thatcher. 2004. The effects of feeding fish oil on uterine secretion of PGF<sub>2</sub>α, milk composition, and metabolic status of periparturient Holstein cows. *J. Dairy Sci.*, **87**(4): 921-932. DOI: 10.3168/jds.S0022-0302(04)73236-1
- Mishra, U.K., R.G. Agrawal and R.K. Pandit. 1997. Clinical study on prolapse of genitalia in Murrah buffaloes. *Indian Journal of Animal Reproduction*, **18**(2): 124-126.
- Naik, P.K., S. Saijppaul, A.S. Sirohi and M. Raquib. 2009. Lactation response of cross bred dairy cows fed on indigenously prepared rumen protected fat - A field trial. *Indian J. Anim. Sci.*, **79**(10): 1045-1049.
- Nanda, A.S. and R.D. Sharma. 1982. Incidence and etiology of prepartum prolapse of vagina in buffaloes. *Indian J. Dairy Sci.*, **35**: 168-171.

- Nirwan, S.S., J.S. Mehta, A. Kumar, P. Kumar, A. Kumar and V. Singh. 2019. Effects of bypass fat on postpartum reproductive performance in dairy cattle. *Indian J. Dairy Sci.*, **72**(2): 194-200. DOI: 10.33785/IJDS.2019.v72i02.011
- Rabbani, R.A., I. Ahmad and L.A. Lodhi. 2010. Prevalence of various reproductive disorders and economic losses caused by genital prolapse in buffaloes. *Pak. Vet. J.*, **30**(1): 44-48.
- Ranjan, A., B. Sahoo, V.K. Singh, S. Srivastava, S.P. Singh and A.K. Pattanaik. 2012. Effect of bypass fat supplementation on productive performance and blood biochemical profile in lactating Murrah (*Bubalus bubalis*) buffaloes. *Trop. Anim. Health Pro.*, **44**(7): 1615-1621. DOI: 10.1007/s11250-012-0115-3
- Snedecor, G.W. and W.G. Cochran. 1994. *Statistical Methods*, 8<sup>th</sup> ed. Iowa State University Press, Iowa, USA.
- Srinivasan, T., A.W. Lakkawar, K.C. Varshney, S.M. Raju and C. Thandavamurthy. 2017. Pathology of cystic ovarian degeneration in buffaloes. *International Journal of Livestock Research*, **7**(4): 180-187. DOI: 10.5455/ijlr.20170312054143
- Thakur, D., J.R. Kumar, S. Pradeep and A. Yadav. 2017. Feeding patterns, nutritional status of available feeds during advanced pregnancy and incidence of reproductive and metabolic disorders in buffaloes of Indore district of Madhya Pradesh. *Indian J. Anim. Nutr.*, **34**: 50-55. DOI: 10.5958/2231-6744.2017.00008.1
- Tyagi, N., S.S. Thakur and S.K. Shelke. 2010. Effect of bypass fat supplementation on productive and reproductive performance in crossbred cows. *Trop. Anim. Health Pro.*, **42**(8): 1749-1755. DOI: 10.1007/s11250-010-9631-1
- Vala, K.B., A.J. Dhama, F.S. Kavani, S.C. Parmar and M.M. Pathan. 2018. Effect of peripartum nutritional supplementation on plasma profiles of macro minerals and postpartum fertility in Jaffarabadi buffaloes. *Indian Journal of Veterinary Sciences and Biotechnology*, **14**(1): 22-27. DOI: 10.21887/ijvsbt.v14i1.12992
- Warriach, H.M., D.M. McGill, R.D. Bush, Wynn and K.R. Chohan. 2015. A review of recent developments in buffalo reproduction - A review. *Asian Australas. J. Anim. Sci.*, **28**(3): 451-455. DOI: 10.5713/ajas.14.025910.5958/2231-6744.2017.00008.1
- Warriach, H.M., D.M. McGill, R.D. Bush, Wynn and K.R. Chohan. 2015. A review of recent developments in buffalo reproduction - A review. *Asian Australas. J. Anim. Sci.*, **28**(3): 451-455. DOI: 10.5713/ajas.14.0259