My articles are organized and grouped based on the topic of the articles. The first group introduces the reader to a broader topic (feeding preferences), then gradually explores more specific topics such as the affects of systems and other influences on grazing behaviour before summarizing the "how" and "why" of buffalo grazing behaviour.

Feeding Preferences

1. Tsiobani, E. T., Yiakoulaki, M. D., & Menexes, G. (2019). Seasonal variation in water buffaloes' diet grazing in wet grasslands in Northern Greece. *Hacquetia, 18*(2), 202-212. https://doi.org/10.2478/hacq-2019-0004

Summary: This literature studied the seasonal variability effects on buffalo diet selection. This study was carried out in wet grasslands of Northern Greece from May to April of the following year. Effects of season on grazing intake have been previously studied but season in relation to diet composition has not. Herbaceous species were categorized into 3 functional groups: graminoids, legumes, and forbs. A herd of 91 water buffaloes grazed on wet grasslands daily and in the late afternoon they would return to the stable. The composition (%) of herbaceous species was calculated and recorded and a focal sampling method was applied to determine buffaloes' diet selection. Each animal was given numbered collars for identification. The first observer followed the herd constantly and observed each animal for 18 seconds, while the second observer collected the ingested vegetation for identification. There were 240 observation periods in total (2 observation periods/hour). Varying numbers of animal observations per season was a result of buffalo performing other activities such as standing, ruminating and wallowing. The results showed a significant relationship between plant functional groups and seasons. The proportion of graminoids was higher during the summer and autumn than any other season. However, forbs had the lowest proportion during that time. The proportion of legumes was highest in the summer. A significant relationship was found between buffaloes' diet and seasons. Herbaceous vegetation made up more than 90% of their diet each season. In the buffaloes' diet, a total of 66 plant species (16 graminoids, 3 legumes, 33 forbs, 7 shrubs, and 7 trees) were included. The consumption of wetland vegetation over woody shrubs could be attributed to the composition of the feed (thorns) and the nutritious qualities. Future studies could include looking at the nutritional values of vegetation and investigate which is more favourable to buffalo.

Contribution: This article gave new insight into the diet preferences of buffalo. The grazing behaviour of buffalo has been studied but there is little known about their diet preferences. This study supports previous findings that buffalo prefer grassy vegetation compared to woody shrubs. Grouping the herbaceous vegetation allowed for the researchers to study multiple species of vegetation. I chose this article as it looks at seasonal variation in buffalo diet. Diet is an aspect of buffalo grazing that is not studied often, especially with seasonal effects.

2. Barrio, J. P., Zhang, S.Y., Zhu, Z. K., Wu, F. L., Mao, X. Z., Bermúdez, F. F., & Forbes, J. M. (2000). The feeding behaviour of the water buffalo monitored by a semiautomatic feed intake recording system. *Journal of Animal and Feed Sciences, 9*(1), 55-72. https://doi.org/10.22358/jafs/68029/2000

Summary: This study investigated the feeding preferences and behaviour of indoor-stabled water buffalo using an automatic feed intake recording system. Daily number of meals, total daily intake (kg), total eating time and body weight were recorded and studied. Feed intake has been studied previously by observation but few studies have used a man-made automatic system. Each steel-sheet container was able to hold up to 20 kg of hay. Eight castrated male buffalo aged 2-3 years old were kept in individual stalls and fed grass straw with an hour of outdoor exercise provided per day for 28 days in the summer. Feed intake data continuously weighed the hay inside each feed container as the buffaloes fed. At 9 am, the initial weight of feed was measured. The total weight of hay, the start and end times of feeding for each meal and mean daily hay ingested (kg/day) for each buffalo was calculated and recorded. The weight was detected using sensors and signals were converted into digital values using a computer software. It was found that each individual preferred to have only their first meal at similar times. The meal duration was longest during the first meal (fresh hay) and consistently lower throughout the others. The first meal being the longest is typically seen in housed ruminants, the article infers that this may vary given an outdoor environment. Individuals with a higher body weight typically consumed more kg/day of hay. The intake of feed was highest in most buffalo during the first meal, then fewer and smaller meals were observed in the night. This infers that buffalo are day-time grazers rather than nighttime grazers. Number of meals varied with a maximum of 8.1 meals. Future studies explore the effects of more or less exercise on feeding/grazing behaviour.

Contribution: This article gives insight into feed intake recording systems and how they can be better adjusted to observe feeding behaviour. Previously, the force exerted by water buffalo when pushing the container to eat was excessive for the sensors, thus this was revised. The article supports the findings that meals excluding the first are not well-synchronized in time of feed or amount of intake. I included this article because it gives quantitative insight into feeding preferences of buffalo that can be used to increase welfare and weight gain of the animal.

Effects of systems on grazing behaviour

3. Almeida, J. C. F., Joset, W. C. L., Noronha, R. P. P., Barbosa, A.V. C., Lourenço Júnior, J. B., & Silva, J. A. R. da. (2019). Behavior of buffalo heifers reared in shaded and unshaded pastures during the dry season on Marajó Island, Pará, Brazil. *Acta Scientiarum Animal Sciences*, *41*(1), 1807-8672. https://doi.org/10.4025/actascianimsci.v41i1.43088

Summary: This article explored the differences in grazing, rumination and idling of marsh and river buffalo heifers reared in silvopastoral systems with shade (WS) and without shade systems (WOS) in November, the driest month. Silvopastoral systems manually integrate trees and shrubs as means for shade. The behaviour of buffalo in relation to their mean temperature and humidity index (THI) in different environments has not been studied in depth. Twenty buffalo heifers with a mean age of 24 months were numbered 1-20 with non-toxic paint and randomly split into two groups; WOS paddocks (enclosed field) and WS silvopastoral system paddocks. Behaviour was examined through visual observation and sampling method over 72 consecutive hours, in 60-minute forms divided into 12 periods of 5 minutes; morning (6:00-9:55 a.m.), intermediate (10:00 a.m.-1:55 p.m.), afternoon (2:00-5:55 p.m.), evening (6:00-9:55 p.m.), night (10:00 p.m.-1:55 a.m.), and early morning (2:00-5:55 am). Grazing was time spent on foraging, chewing and swallowing while rumination was observed time of jawbone movement without grazing. Idling was time spent standing or lying without any activity. THI was calculated and wind speed (WSP) was measured using a digital anemometer. THI was higher in the WOS group in all shifts except the night and early morning, demonstrating that unshaded environments caused thermal stress in buffalo. The WSP was highest during the evening shift, intermediate and afternoon shifts, respectively. Overall, the WS group grazed more, with a larger difference in the afternoon and evening with high THI. Overall, the WOS group ruminated less but idled more to reduce internal heat production. These infer improved welfare and productivity of buffalo reared in shaded systems. Questions arise on how these results differ in the wet season.

Contribution: This article provides more insight into the effect of shade in relation to THI and WSP and how they affect buffalo behaviour. In previous studies, the THI and WSP have not been calculated and used to explain the results of the study. It allows for readers to synthesize the effect of shade on thermal stress of buffalo and the resulting effects on their behaviour. I chose this article in order to investigate the best conditions to promote welfare, increased grazing and ruminating behaviour in buffalo. The study expands our understanding of the grazing behaviour of buffalo in varying conditions.

4. Galloso-Hernández, M. A., Soca-Pérez, M., Dublin, D., Alvarez-Díaz, C. A., Iglesias-Gómez, J., Díaz-Gaona, C., & Rodríguez-Estévez, V. (2021). Thermoregulatory and feeding behavior under different management and heat stress conditions in heifer water buffalo (*Bubalus bubalis*) in the tropics. *Animals*, *11*(4), 1162. https://doi.org/10.3390/ani11041162

Summary: This study explored the behaviour of heifer buffalo under moderate and intense heat stress conditions in two different systems; silvopastoral system (SPS) with (*Leucahena leucocephala)* trees and conventional system (CVS) without trees. SPS manually integrates trees and shrubs while CVS is not modified. The objective of the study was to investigate the grazing, browsing, rumination, water intake, sheltering behaviour, and wallowing of buffalo under two heat stress conditions and systems. Four experimental conditions were applied; CVS in intense and moderate thermal stress, SPS in moderate and intense thermal stress. The experiment was

done by recording 72 observations in 10 minute intervals of the above activities of 9 heifer buffalo (12-18 months of age), during a 12 hour day cycle (6:00 to 18:00 h) in the months of May and November, for 3 days per experimental condition. The number of animals and time spent doing each activity were recorded and the relative variables were grouped. Active feeding behaviour consisted of grazing and browsing while feeding behaviour was the sum of active feeding behaviour, rumination and water consumption. Thermoregulatory behaviour was the sum of shading behaviour and wallowing. Browsing was affected in both moderate and intense heat stress conditions with more time (h) spent in SPS than CVS. Grazing was not affected in moderate heat stress but was affected in intense heat stress with more time spent in SPS. Feeding behaviour was affected overall with more time spent in SPS during intense heat stress conditions and least time spent in SPS during moderate heat stress. Thermoregulating behaviour was affected with more time spent in SPS during moderate and intense heat stress. These findings are crucial to understand how to increase the well-being of buffalo and promote weight gain. Questions are raised on the mechanisms involved in how heat stress affects buffalo.

Contribution: This article adds insight into the effects of trees on buffalo grazing and thermoregulating behaviour in heat stress. It is not known whether trees reduce wallowing time, help thermoregulating behaviour while also increase grazing time. This study expands on previous findings that better daily weight gain in buffalos were found in silvopastoral systems when compared to conventional systems by exploring why this occurs. I chose this article to better understand how different environmental factors (such as heat stress) may affect the grazing behaviour of buffalo. This is important as heat conditions change depending on the location of the buffalo.

5. De la Cruz-Cruz, L., Guerrero-Legarreta, I., Ramirez-Necoechea, R., Roldan-Santiago, P., Mora-Medina, P., Hernandez-Gonzalez, R., & Mota-Rojas, D. (2014). The behaviour and productivity of water buffalo in different breeding systems: A review. *Veterinarni Medicina, (59)*4, 181-193. https://doi.org/10.17221/7479-VETMED

Summary: This review focuses on the feeding behaviour and productivity of water buffalo under various production systems in relation to meat and milk production. Although the relationship between each system and feeding behaviour has been studied, comparing systems in relation to production yield has not. The article focused on three production systems; Silvopastoral systems, outdoor systems and intensive systems. Silvopastoral systems manually integrate trees and shrubs, outdoor systems are free-range and intensive systems utilize confinement while providing feed supplements. The objective of this article was to review the effects of these systems on feeding behaviour and weight gain in relation to milk and meat production in buffalo. The review cumulated findings from sources with common parameters (meta-analysis); 1) Captive buffaloes are put in at least one of the systems 2) Animals are provided a range of vegetation 3) Observations of feeding and/or weight gain were taken and

averaged. Silvopastoral systems showed a relationship between increase in foraging activities to wallowing, welfare and homeostasis. They attenuate thermal stress through trees and shrubs, promoting daily weight gain (g/animal) and milk production. The free-range aspect of outdoor systems allowed for increased walking and grazing. A daily weight gain and increase in milk production was measured to be less than that of silvopastoral. Milk and meat production for these two pasture based systems decreased during the dry season. Intensive systems showed consistent and the most daily weight gain, while also reducing slaughtering age by providing buffaloes with balance feed supplements. However, the welfare of buffalo decreased and an increase in stress was suggested by aggravated buffalo behaviour. Overall, the article synthesizes that intensive systems tend to be most advantageous solely for increased meat and milk production. Questions are raised around the effect of genetic phenotype on a buffalo's ability to gain weight.

Contribution: This review article adds insight to previous knowledge on the effects of three systems on feeding and weight gain of water buffalo, in relation to meat and milk production by comparing them to conclude which system is most advantageous. It supports previous study findings while giving a new perspective through a comparative lens. I included this review because it explores how different aspects affect what is known about grazing behaviour in buffalo; The effect of trees and shrubs in silvopastoral systems, the effect of free-range in outdoor systems and the effect of confinement and supplemental feed in intensive systems.

Environmental and climatic influences on grazing behaviour

6. Napolitano, F., Grasso, F., Saltalamacchia, F., Martiniello, P., Bilancione, A., Pacelli, C., & Rosa, D. (2016). Grazing behaviour of buffalo heifers. *Italian Journal of Animal Science, 6*(2), 19-22. https://doi.org/10.4081/ijas.2007.s2.1256

Summary: This article investigated the effects of season on river buffalo grazing behaviour. Buffalo were fenced in an area where approximately 10% of the canopy vegetation was woodland and the rest was occupied by grassy habitat. Previous studies covered the grazing behaviour of other herbivorous animals but little is known about the grazing time of buffalo. For a year starting in November, sixteen buffalo heifers around the age of 8-9 months from the same herd were free to graze throughout the experiment. From December to March, buffalo were kept in a barn and supplemented dehydrated beet-pulps and hay. From April to October, continuous focal sampling was used on a buffalo chosen at random and observations were taken between 5:30-15:00 hours with the start and stop time tape-recorded. The behaviours observed were posture (standing or lying), grazing, location (in the sun or shade), walking, resting, rumination and other. When grazing, the type of vegetation eaten was recorded and for each observation session, the proportion of time spent on each behaviour was calculated. Six observation sessions were taken for each season; Spring (April-mid June), summer (mid-June to mid-September) and autumn (mid-September to October). The results show that the ingestion of woody or shrub

vegetation was rarely observed and the proportion of time spent grazing was not affected by season. Buffalo were found to lay and ruminate more in the summer and autumn than in the spring where there was an increase in walking. Animals were found more in the shade laying down in the summer months. These results infer that river buffalo are grazers rather than browsers because of the type of vegetation ingested (little to no shrub). Questions are raised from this study on the grazing time of buffalo in the evening (observation sessions not included in this study).

Contribution: This study provides more insight into the proportion of time spent on grazing by buffalo. Not much is known about the grazing behaviour of buffalo, especially when looking at the types of vegetation that they ingest. It contradicts previous studies that show that grazing activities are higher during the dry season. In this experiment, season had no effect on grazing time. I included this article because not many other studies look at grazing time and other behaviours with focal animal sampling and calculated proportions. It helps us categorize buffalo as grazers rather than browsers.

7. Tatipikalawan, J. M., Nurtini, S., Sulastri, E., & Widi, T. S. M. (2019). Utilization of lutur in the traditional grazing system of buffalo production in Moa Island – Maluku, Indonesia. *IOP Conference Series: Earth and Environmental Science, 387*(1), 12070. https://doi.org/10.1088/1755-1315/387/1/012070?

Summary: The article explored the use of *lutur* fences in traditional buffalo grazing systems. The fences used were made of a one-meter high limestone called *lutur* and enclosed the space used for farming practices and buffalo grazing pastures. Previous studies have investigated size of pasture in relation to buffalo grazing but little studies are done on the effects of communal pastures and farming systems. The study was carried out on Moa Island on 6 villages by 261 selected buffalo farmers with at least 3 years of experience each. The size of *lutur* pastures ranged from 0.7 to 1 hectares, holding up to 250-600 buffalo each. In addition to having their own pastures, farmers with smaller herds of buffalo shared land called communal *lutur*. These were practiced with 3 main characteristics; 30-140 days per year of 5-7 hours daily grazing time, schedules for grazing were discussed among participating farmers, groups of 2-3 per village with 20-25 farmers each and 1-2 times annual grazing frequency. The data analyzed was taken descriptively from each selected participant through semi-structured questionnaires, focus group discussions and direct observations. It was found that *lutur* pastures made buffalo keeping management easier, especially during the rainy season when farmers needed to care for sick and pregnant buffaloes. Communal *lutur* allowed for the use of land and labour to be more efficient by overcoming declining grazing land and reduced patterns of livestock mobility because of increased human populations. However due to the closed off space, an increase of inbreeding was found amongst each herd. This created a problem for farmers as albino buffaloes and hanging horned buffaloes decreased in body size. This study allowed farmers to explore

proximate influences on buffalo grazing behaviour while also investigating varying methods of farming to increase production and welfare of buffalo.

Contribution: This article took on a different perspective than previous studies by looking at effects of fencing and farming systems on buffalo grazing behaviour. The study explores human influences on buffalo in relation to production and welfare. It supports previous studies that find reduced mobility leads to inbreeding in animals. I chose this article as it takes on a new perspective of grazing behaviour and the effects of human influences rather than other articles that look at environmental factors.

8. Tsiobani, E. T., Yiakoulaki, M. D., Hasanagas, N. D., Menexes, G., & Papanikolaou, K. (2016). Water Buffaloes grazing behaviour at the Lake Kerkini National Park, Northern Greece. *Hacquetia, 15*(2), 133–142. https://doi.org/10.1515/hacq-2016-0015

Summary: This article investigated the monthly variation of water buffaloes' grazing behaviour. Each month varies in climate, daylight hours and quality of vegetation, all affecting the grazing behaviour of buffalo. In previous literature, low intensity grazing systems have been studied in relation to season but none have explored the monthly change in behaviour. This study aimed to explore 3 main objectives; 1. Grazing routes of buffalo and distance travelled in order to meet nutritional needs 2. Time devoted to grazing-related activities per month 3. Type of land grazed on each month. Grazing-related activities were categorized as feeding, moving, wallowing, standing, ruminating, drinking, and lying. This experiment took place from November to October the following year. Research was conducted on grasslands consisting mainly of herbaceous species with some woody species present, and agricultural land containing crops such as alfalfa, barley, wheat, rye and oats. Focal sampling was used to analyze the behaviour of 6 female buffalo aged four-years old, which were identified and marked numerically. Buffalo were analyzed for two consecutive days per month in one-hour observation protocol forms, divided into six observation periods of 10 minutes per animal. Grazing-related activities were recorded every 15-seconds. In terms of distance travelled, the longest distance was in June (10.7 km/day) and the shortest in September (2.9 km/day). This infers that buffalo must travel further in summer months to satisfy their nutritional needs. Monthly variations of time spent on each grazing-related activity was found and more time was spent feeding in May than any other month. Buffaloes grazed on grasslands from May to July, while in August to April they grazed on agricultural land. These results deduce that grassland quality declines during the colder months. Buffaloes Questions are raised regarding the effect of age, sex and size on grazing behaviour.

Contribution: This article gives new insight into how different months affect the grazing behaviour of water buffalo. Since this subspecies of buffalo is considered endangered, not many studies have been done on their grazing behaviour especially in relation to months. This study

does support previous findings regarding the movement of buffalo (distance travelled) while providing new findings on social interaction. I have included this article as it focuses on an endangered species that is not well researched and takes into account the effects of varying months on a broad category of grazing-related behaviour.

Explaining Feeding behaviour

9. Vega, R. S. A., Del Barrio, A.N., Sangel, P.P., Katsube, O., Canaria, J. C., Herrera, J. V., Lapitan, R. M., Orden, E. A., Fujihara, T., & Kanai, Y. (2010). Eating and rumination behaviour in Brahman grade cattle and crossbred water buffalo fed on high roughage diet. *Animal Science Journal, 81*(5), 574-579.
https://doi-org.ezproxy.lib.ucalgary.ca/10.1111/j.1740-0929.2010.00784.x

Summary: This experiment compared feeding, nutrient digestibility and muscle mastication of tropical grade buffalo and crossbred water buffalo. Muscle mastication is used to determine how well food is chewed and absorbed. Previous studies have not looked at muscle diameter of buffalo in relation to rumination efficiency. Buffaloes' behaviours were analyzed on video camera recording to determine frequency and duration of activity; feed intake, rumination, chews, lying and standing position. Eight heads of both water buffalo and tropical grade buffalo were individually penned and fed Napier grass as much as necessary. Each animal was used to determine nutrient digestibility, while three of each buffalo subspecies' behaviours were observed for 24-hours on the 107th, 109th, and 111th days of the digestion trial. On day 112, these 6 buffalo were slaughtered to analyze the muscle mastication by determining the diameter of muscle involved in rumination. Observations were divided into three phases; Phase 1 (07.30-14.30); Phase 2 (14.31-21.30) and Phase 3 (21.31-07.29). The data showed a correlation between eating and ruminating times, concluding that the best time to sample these together would be together from 14.30-07.30 hours. The higher feed intake and slower/lesser chews of buffalo resulted in comparable nutrient digestibility of both species. This could be explained by a larger or more well-developed diameter of muscle. In crossbred buffalo, the duration of standing was shorter but the frequency of drinking and defecation was higher. It was concluded that slower chewing and a larger diameter of muscle resulted in more efficient rumination. In addition, less frequent shifts from lying to standing position and a longer duration of lying likely indicated better energy efficiency. Questions are raised on the difference of average muscle diameter and body mass between species and how this affects the results.

Contribution: This article gives new insight into the relationship of feeding behaviour, nutrient digestibility and muscle mastication in different species of buffalo. Previous studies have looked at feeding behaviour and nutrient digestibility individually but none have looked at the correlation of them to determine rumination and intake efficiency. I chose this article as it looked at both the behaviour aspect of feeding as well as the digestibility and muscle mastication. These

in combination give the reader more background information as to why the observed behaviour is the way it is.