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Developing a blueprint for controlling malignant catarrhal fever (MCF) in farmed bison and buffalo in Wales

Final report

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Developing a blueprint for controlling malignant catarrhal fever (MCF) in farmed bison and buffalo in Wales

Report authors

Dr. Joseph W Angell BVSc MSc DipLSHTM PhD MRCVS
Wern Vets CYF, Department of Research and Innovation, Unit 11, Lon Parcwr Industrial Estate, Ruthin, Denbighshire, LL15 1NJ.
Telephone: 01824 703066
Email: joseph@wernvets.co.uk

Eleanor G Bentley BSc
Dept of Infection Biology and Microbiomes, Institute of Infection, Veterinary and Ecological Sciences, University of Liverpool, Liverpool Science Park IC2, 146 Brownlow Hill, Liverpool, L3 5RF.
Email: E.Bentley@liverpool.ac.uk

Professor James P Stewart
Dept of Infection Biology and Microbiomes, Institute of Infection, Veterinary and Ecological Sciences, University of Liverpool, Liverpool Science Park IC2, 146 Brownlow Hill, Liverpool, L3 5RF.
Email: jpstewart@liverpool.ac.uk

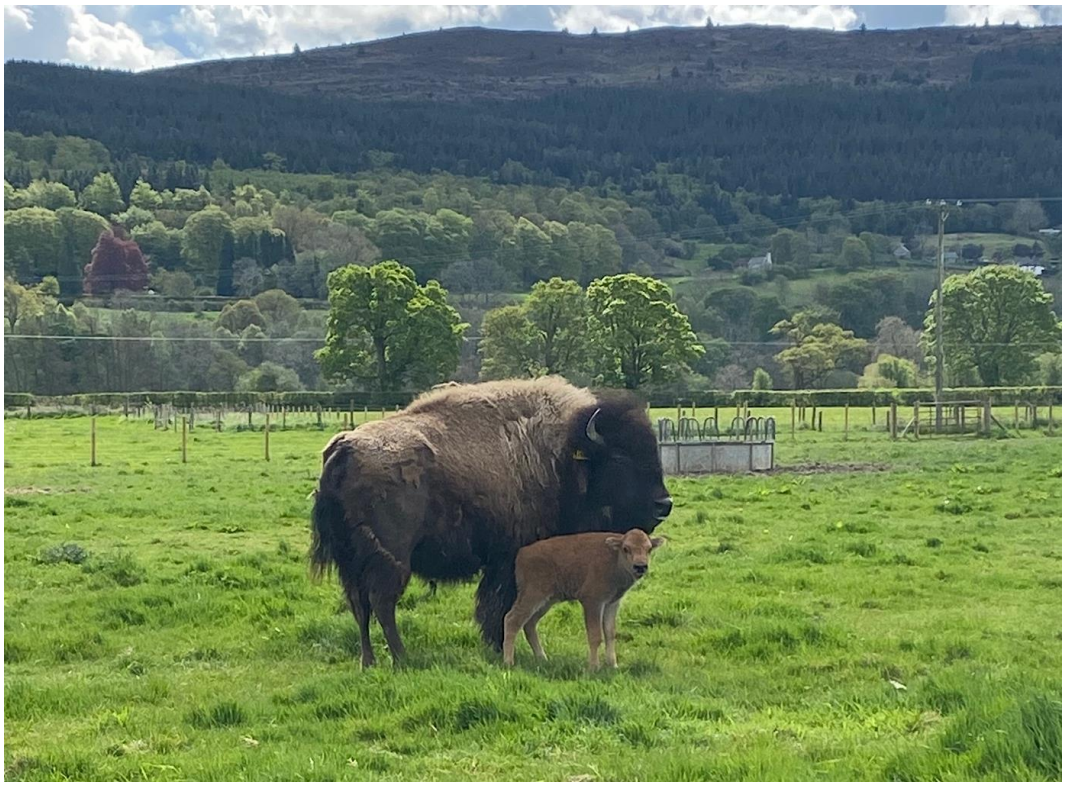

Photo: Gareth Jones

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Introduction

Bison and buffalo as diversification options in Wales

Bison farming in North America is a rapidly growing business, driven by consumer perception of bison meat as a healthier alternative to beef. In the UK and Wales, some pioneering producers have also found a ready market for bison meat. It is considered to be a healthy red meat alternative to traditional beef products, being lower in fat, cholesterol, and sodium (1-3). It also has a very similar protein content, is lower in calories and is high in iron and vitamin B-12 (Table 1).

NUTRITIONAL COMPARISONS

Per 100 Gram (3.5 oz.) Serving – Cooked Meat – Updated January 2013

SPECIES	FAT g	PROTEIN g	CALORIES kcal	CHOLESTEROL mg	IRON mg	VITAMIN B-12 mcg
BISON	**2.42**	**28.44**	**143**	**82**	**3.42**	**2.86**
Beef (Choice)	18.54	27.21	283	87	2.72	2.50
Beef (Select)	8.09	29.89	201	86	2.99	2.64
Pork	9.21	27.51	201	84	1.0	0.68
Chicken (Skinless)	7.41	28.93	190	89	1.21	0.33
Sockeye Salmon	6.69	25.40	169	84	0.50	5.67

Bison, separable lean only, cooked, roasted. USDA NDB No. 17157
Beef, composite of trimmed retail cuts, separable lean only trimmed to 0" fat, choice, cooked USDA NDB No. 13362
Beef, composite of trimmed retail cuts, separable lean only trimmed to 0" fat, select, cooked USDA NDB No. 13366
Pork, fresh, composite of trimmed retail cuts (leg, loin and shoulder), separable lean only, cooked USDA NDB No. 10093
Chicken, broilers or fryers, meat only, roasted USDA NDB No. 05013
Salmon, sockeye, cooked, dry heat USDA NDB No. 15086

Table 1: Nutritional comparisons of different meats[1].

Compared to beef in the UK, bison meat is worth considerably more, with deadweight prices (£/kg) for bison approximately twice that for cattle even at the top end of the market, and retail prices for bison meat (£/kg) frequently more than 1.5 times that for beef (Table 2). As such, bison are considered as one option for farmers looking to diversify.

[1] This table is taken from http://www.bisoncentral.com/cooking-bison/nutrition-information and is based on information from the United States Department of Agriculture Agricultural Research Service https://ndb.nal.usda.gov/ndb/search/list

Price	Beef (£/kg)[2]	Bison (£/kg)[3]	Bison (£/kg)[4]	Buffalo (£/kg)[5]	Difference (Bison-Beef)	Difference (Buffalo-Beef)
Average	£4.17[6]	£5.60			£2.25	
Retail prices						
Fillet steak	£30.06	£67.00	£54.00	£42.00	£36.45	£12.06
Sirloin steak	£18.54	£55.00	£36.50	£32.00	£38.16	£19.62
Rump steak	£14.04	£45.00	£31.25	£22.00	£31.82	£17.78
Premium mince	£5.98	£20.00	£14.80	£10.95	£13.59	£7.61

Table 2: Price comparisons between beef, bison and buffalo as deadweight and retail value in the UK.

Water buffalo are another diversification alternative. However, in contrast to bison, the primary outputs in Wales and the UK are both meat and dairy products, including whole milk, yoghurt, ice-cream, and cheese, especially mozzarella. For some of these products markets are more readily found in Asian cooking, with more widespread appeal for others.

Price	Buffalo Dairy (£/L)
Retail prices	
Raw milk (£/L)	£1.20[7]
Mozzarella (£/kg)	£24.32[8]
Ice cream (£/L)	£9.90[9]

In terms of management, buffalo can be managed similarly to dairy/beef cattle with some adaptations. Bison, however, are challenging to rear due to their temperament and sensitivity to stress. Whilst bison and water buffalo are markedly different species, with widely different farming management approaches, both have an increased susceptibility to malignant catarrhal fever (MCF), which is considered the disease-limiting factor to successful production(4). Indeed, some attempts at diversification with bison and water buffalo in the UK have failed due to MCF and finding ways to manage this disease is integral to successful diversification.

[2] Figures for November 2019 taken from AHDB Market/Supermarket reports for Great Britain
[3] Figures for Rhug Farm Estate November 2019
[4] Figures as published by Bush Farm Bison Centre http://www.bisonfarm.co.uk/Meat.html March 2022
[5] Figures for Buffalicious https://www.buffaliciousuk.com/wb-meat March2022
[6] This value is for Steers at the top of the price band, November 2019.
[7] Figures for The Milk Stop https://www.themilkstop.com March 2022
[8] Figures for Gastronomica https://gastronomica.co.uk/shop/cheese/soft-cheese/buffalo-mozzarella-buffalo-milk-125g/?gclid=CjwKCAiAg6yRBhBNEiwAeVyL0DrrR-hRCQFkWdvKkkWpa_20sdhdljqYfh548P2gtQBXKKG0FscF7RoCD-MQAvD_BwE March 2022
[9] https://www.naptonvillagestores.co.uk/product/buffalo-milk-ice-cream/ March 2022

Sheep associated malignant catarrhal fever (SA-MCF) in bison and buffalo

Malignant catarrhal fever is caused by herpesviruses of the *Macavirus* genus, namely ovine herpesvirus 2 (OvHV-2), caprine herpesvirus 2 (CpHV-2) and alcelaphine herpesvirus 1 (AlHV-1)(5). In the USA, Canada and Europe (including the UK) the sheep-associated (SA-MCF) form caused by OvHV-2 appears most commonly and has been documented in more than 30 species, however those of most interest to the UK are cattle, bison, water buffalo, deer, and pigs (5-8), and whilst sheep are considered the main reservoir host, recent work suggests a large proportion of cattle can also carry the virus subclinically (7).

Bison, buffalo and deer are considered much more susceptible than cattle with outbreaks occurring when they are kept in close proximity to sheep (9). The published reports (mostly from abroad) are striking in their severity, for example in 2003, a SA-MCF outbreak occurred on a bison feedlot in Idaho, USA. This outbreak was considered to be as a result of exposure to sheep for 19 days and resulted in a total of 825 deaths out of the 1,610 bison exposed (51.2%), with deaths peaking at a rate of 41 head per day (10). In a further report, SA-MCF cases occurred on a bison farm in western USA, with a total of 60 out of 761 bison (7.9%) dying over a 6 month period. In this case the mortality rate was related to the distance from the sheep with those at 1.6km suffering from greater losses (17.5% mortality) compared to those at 5.1km (0.43% mortality), and new cases did not stop occurring until the remaining bison had been moved to a feedlot at a distance of at least 24km from sheep (11). Other studies have shown similar devastating death rates from SA-MCF, ranging from 7.3% of bison on a ranch in Midwest USA (4) to 27.6% of bison presented to an auction market in Saskatchewan (12), and in this latter case the period of exposure to sheep was considered to be limited to a period of less than 1 day at the auction market.

In buffalo, a report from Italy demonstrated 12 cases from a farm with about 200 buffalo with 9/12 affected animals dying (13). These buffalo sometimes co-grazed with sheep from the same farm, and this was their likely source of exposure. In the same report, on another farm with 650 buffalo, six clinical cases were detected, all of which were fatal. This farm did not have its own sheep but was surrounded by other farms with sheep that could graze within 800m of the buffalo.

Clinical signs of malignant catarrhal fever

Following infection with OvHV-2, the animal develops an inflammatory response predominantly in the blood vessels and the mucosal surfaces, and the clinical signs of MCF can vary considerably depending on which blood vessels and mucosal surfaces are affected (5, 8, 14). The head and eye form is most frequently seen in cattle (5, 8, 15) with some or all of the following clinical signs: marked pyrexia (>40°C), mucoid/mucopurulent/haemorrhagic nasal discharge, ulceration of the nose/muzzle and sometimes the buccal cavity, increased lacrimation, conjunctivitis, severe keratitis/corneal oedema, hypopyon and partial/complete blindness. Diarrhoea and neurological signs may also be present. In bison and buffalo, the course of the disease may be far more rapid, sometimes simply presenting as sudden death (15). Where clinical signs are seen prior to death, depression, weight loss/wasting and diarrhoea may be more prominent, as well as haematuria, although clinical signs similar to those found in cattle may also be seen (8, 15, 16).

The following are some of the clinical signs as illustrations.

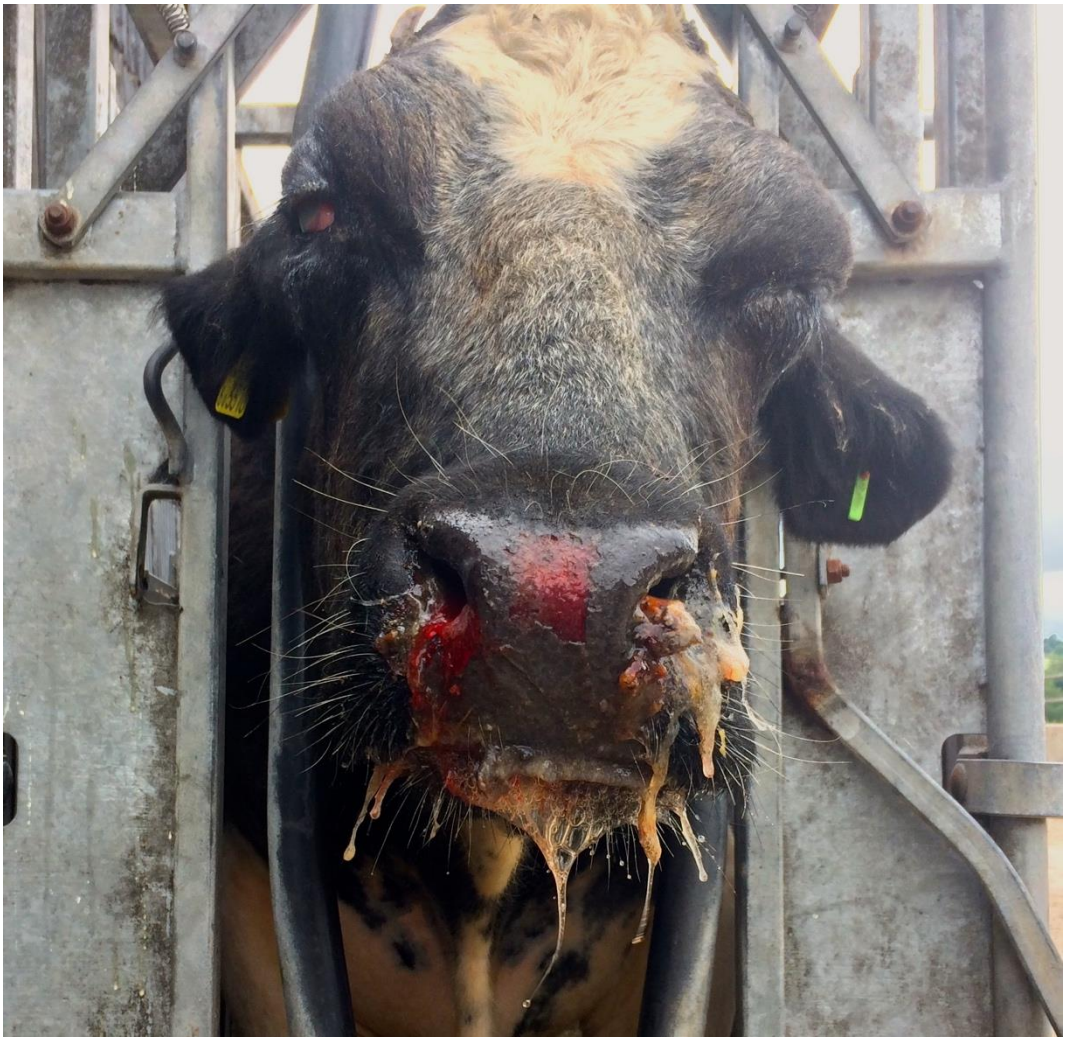

Figure 1: Mucopurulent and haemorrhagic nasal discharge. There is also some erosion to the muzzle and some periorbital swelling as a result of this bullock having collided with his surroundings due to blindness.

Figure 2: Crusting around the nares and mucoid nasal discharge.

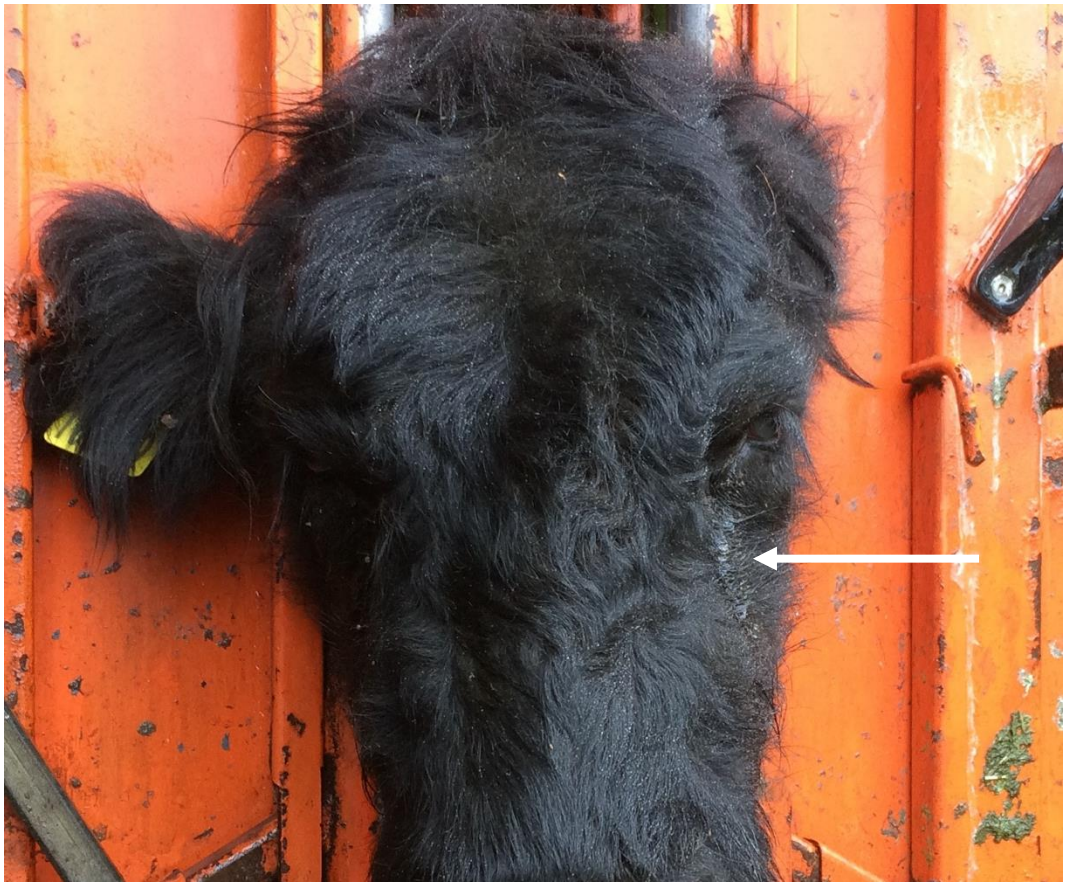

Figure 3: Increased lacrimation.

Figure 4: Ventral corneal oedema. This frequently results in impaired vision or blindness.

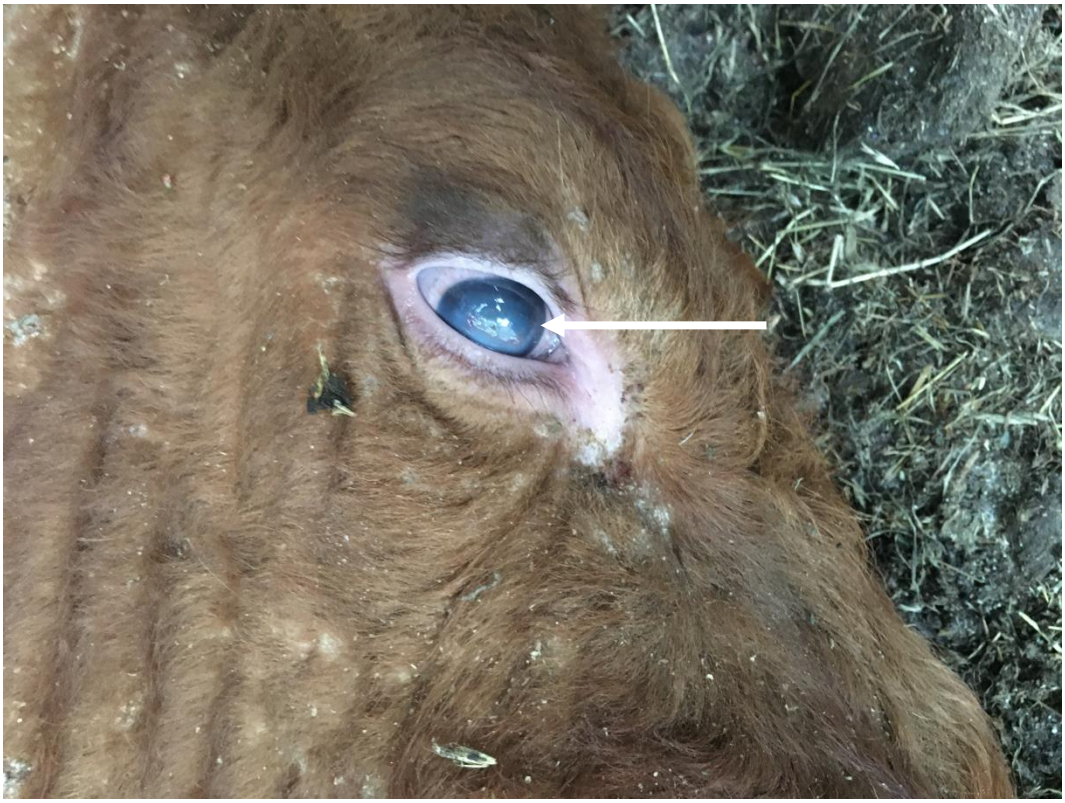

Figure 5: Centralised corneal oedema.

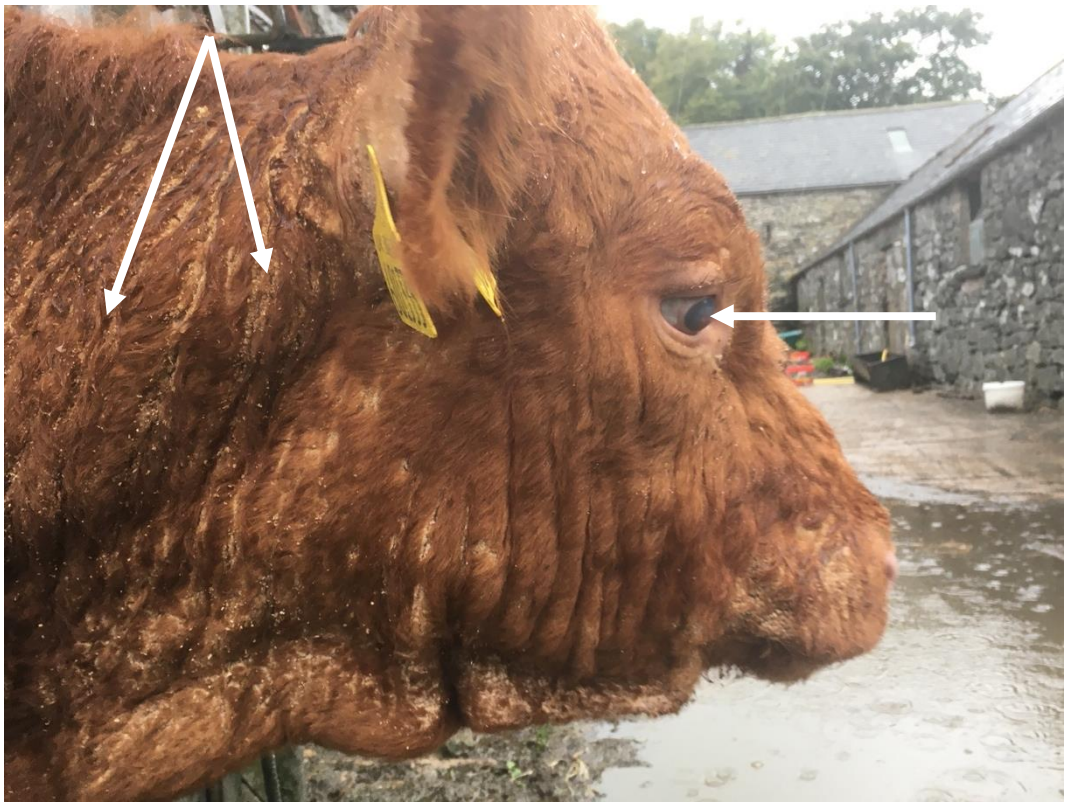

Figure 6: Abnormal skin of the neck and face (hyperkeratosis); centralised corneal oedema.

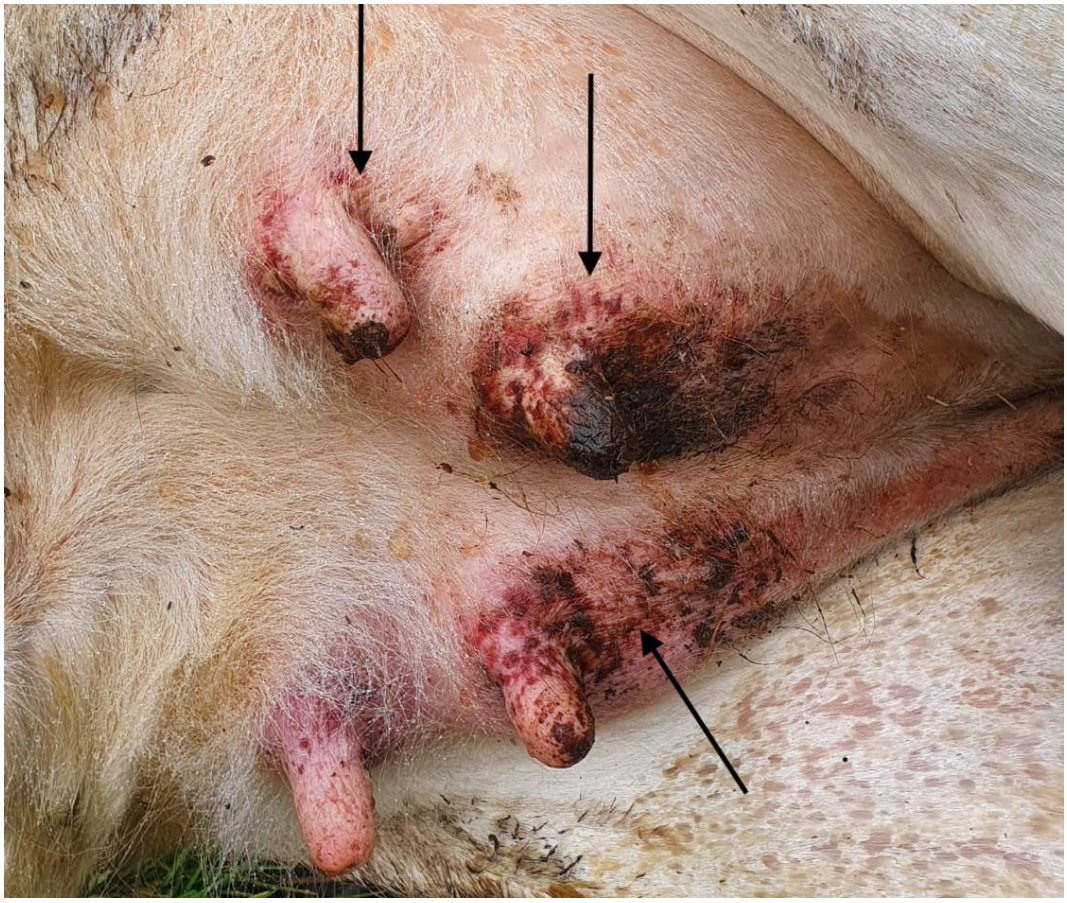

Figure 7: Ulceration and dermatitis of the teats and adjacent udder skin.

Figure 8: Pregnant females can abort.

Control of MCF on bison and buffalo farms in the UK

The control of MCF in bison and buffalo (and indeed cattle) reared in Wales is currently limited with little known about how best to approach this. Efforts are mainly focused on reducing their exposure to sheep, which is a challenge in many areas and limits these means of diversification. Other potential challenges to control include unknown interactions with other infectious diseases e.g. bovine viral diarrhoea virus (BVDv) and bovine herpes virus-1 (the causative agent of infectious bovine rhinotracheitis (IBR)). Other infectious diseases could directly affect the bison and buffalo adversely or could increase the risk of the bison and buffalo to SA-MCF through impacts on their immunity. Exposure to other infectious diseases in bison and buffalo is poorly documented in the UK although reports from other countries reveal numerous infectious diseases affecting bison and buffalo in different contexts.

There are currently no vaccines commercially available for MCF. Two experimental vaccines have been developed. The first is against wildebeest-associated MCF and has been used successfully in cattle in Africa to limit infection and mortality (17). The second is against OvHV-2 associated MCF – the disease seen in the UK. This second vaccine is an exciting development and was used in this project.

Aims and Objectives of the project

Aim

The primary question addressed in this project was: 'is it possible to develop a blueprint for the control of MCF in farmed bison and buffalo in Wales?'.

Null hypothesis: It is not possible to control MCF in bison/buffalo herds in Wales

Objectives

In order to answer this question and to explore whether this null hypothesis can be disproved, various sub-questions were identified, and they form the basis of the objectives and approach used:

1. Have the farmed bison and buffalo already been exposed to OvHV-2 in the past and has MCF disease been identified in the herds?

2. Do other species, known to potentially carry OvHV-2, and kept in close proximity to the farmed bison and buffalo carry OvHV-2 and therefore pose a risk of infection?

3. Have the farmed bison and buffalo been exposed to other infectious diseases that could potentially increase their risk of developing MCF following exposure to OvHV-2?

4. Can use of a vaccine against OvHV-2 be successfully incorporated into a disease control programme?

Objective 1 – investigation of exposure to OvHV-2 in the bison/buffalo
Objective 2 – determine whether other species in close contact with the bison/buffalo carry OvHV-2
Objective 3 – investigate exposure to other infectious diseases that may potentially increase the risk of bison/buffalo developing SA-MCF
Objective 4 – make use of a vaccine against OvHV-2 on one farm with a known severe SA-MCF disease problem, and as far as is possible, assess whether it is practically useful in this context.
Objective 5 – management and control of SA-MCF on farms in the operational group
Objective 6 –formulate a strategy for control of SA-MCF on bison/buffalo farms in Wales.
Objective 7 – local, national, and international knowledge exchange

The main outcome used to determine success or failure will be evidence of MCF disease in bison/buffalo on the study farms during the study period.

As the project developed, we also sought to opportunistically investigate SA-MCF disease outbreaks on other farms with cattle, linked to the project through proximity. This flexible responsive approach enabled greater farmer engagement and a more refined understanding of the specific disease dynamics in Wales.

Approach, methodologies, and results

Objective 1 – investigation of exposure to OvHV-2 in the bison/buffalo

Approach

Heparinised blood samples were taken from the bison/buffalo from each farm for analysis to determine their exposure to OvHV-2. The sampling was coordinated to occur within a single 12 month period and to coincide with another management event e.g. TB testing to minimise any additional stress on these animals.

In addition, herd records were analysed to determine the historic disease incidents and patterns in mortality.

Rhug Estate

In March 2020, the herd of bison (n=8) were blood sampled and the presence of virus detectable within the blood was determined by qPCR. At this stage **no virus was detectable by qPCR in the blood samples from the bison**. Blood samples were also analysed by antibody ELISA specific to the OvHV-2 gB protein, and **all bison were positive at this stage**. Concurrently, a review of the clinical records revealed several confirmed and suspected cases of MCF over the previous years with a steady and continued decline in herd size down to the remaining 8 animals.

As such, whilst no virus was detected to be circulating at the time of sampling, all the bison had been exposed to OvHV-2 at some point as evidenced by the positive antibody test. Additionally, the evidence from the clinical records demonstrated historic exposure at the farm level due to the presence of clinical cases.

In May 2020, a calf died and was examined post-mortem. The cause of death was not due to SA-MCF but as a result of cardiac fibrosis as a result of suspected selenium deficiency (18) (Figure 9). However, a small amount of OvHV-2 was also detected in the pre-scapular lymph node post-mortem and indicated a possible latent infection.

Buffalo Dairy

In June 2020, 19 buffalo were blood sampled and the presence of virus detectable within the blood was determined by qPCR. At this stage, **a small amount of virus (<0.01 viral copies per µgDNA) was detectable by qPCR in the blood samples from four of the buffalo.** Blood samples were also analysed by antibody ELISA specific to the OvHV-2 gB protein, and **all buffalo were negative at this stage**. A review of the farm and clinical records did not reveal any confirmed cases of MCF, but there was one case suspected some years previously by the farm veterinary surgeon, although this was never confirmed.

Cors Dyfi

The buffalo were due to return to Cors Dyfi in spring 2020 after over wintering on another farm and were due to enter the project at this point. However, as a consequence of the restrictions imposed due to Covid-19 and the logistical difficulties surrounding their care and transport, the buffalo were permanently rehomed at another location and left the project. Cors Dyfi were keen to remain in

the project as an interested party but were unlikely to have any other direct part until Objective 7 – knowledge exchange.

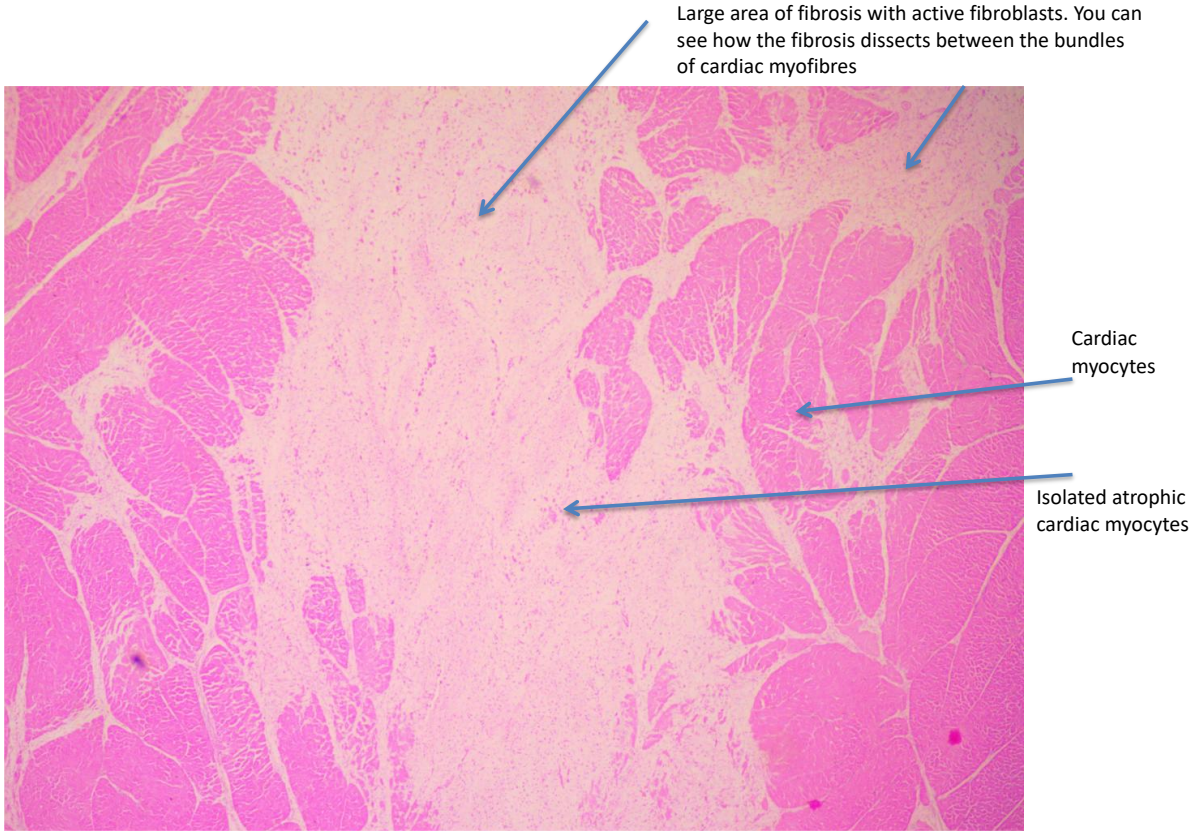

Figure 9: Haematoxylin and eosin stained cardiac tissue from a bison calf that died with cardiac fibrosis, possibly as a result of chronic selenium deficiency (photo courtesy of Hayley Crosby-Durrani).

Objective 2 – determine whether other species in close contact with the bison/buffalo carry OvHV-2

Approach

Blood was taken from a representative sample of other livestock on the farm e.g. sheep and cattle to determine the presence of OvHV-2 and exposure to this virus in these other species. In each case a random sample of adult animals was tested to maximise the chances of detecting a pathogen if present. In addition, tissue samples were obtained opportunistically from culled male deer on one farm and analysed for the presence of OvHV-2 by qPCR. We aimed to sample 10 animals from each risk group (for example 10 cattle and 10 sheep). The different species available on each farm varied between farms and it was not possible to sample animals grazing nearby but not owned by the members of the operational group.

Rhug Estate

Concurrent with the sampling of the bison, 10 cattle and 10 sheep were blood sampled and the presence of virus detectable within the blood was determined by qPCR. Sixteen male deer were opportunistically sampled between October and December 2021 with qPCR carried out on one mediastinal or sub-mandibular lymph node from each animal. **All samples were negative for OvHV-2 by qPCR.**

Buffalo Dairy

Concurrent with the sampling of the buffalo, 10 sheep were blood sampled and the presence of virus detectable within the blood was determined by qPCR. **One sheep had a small amount of virus (<0.01 virus copies per µg DNA) detectable in the blood sample.**

Objective 3 – investigate exposure to other infectious diseases that may potentially increase the risk of bison/buffalo developing SA-MCF

Approach

Concurrent with the sampling of the bison and buffalo for OvHV-2 additional blood samples were obtained for commercial viral and bacterial antigen and antibody tests carried out by laboratories of the Animal and Plant Health Agency (APHA), together with trace element tests carried out by NUVetNA laboratories. The tests were selected to include a comprehensive screen of potential pathogens that could cause disease in the bison/buffalo, and/or impact upon their immune status. Indeed, animals infected with any other disease are already undergoing a stress challenge and therefore become more likely to succumb to any other infection. Exposure to the following diseases was investigated:

Bovine Viral Diarrhoea virus (BVDv)

This viral disease is caused by viruses of the *Pestivirus* genus and infection of susceptible animals can result in failure of conception, abortion, malformed foetuses, still births, and the birth of persistently infected (PI) carrier offspring. Infection can also weaken the immune system leaving animals more susceptible to other infections. Bison are known to be susceptible to BVDv and can become PI (19).

Leptospirosis

This bacterial disease is caused by serovars of the bacterium *Leptospira interrogans*. It is unclear how likely this disease will impact bison and buffalo, however, exposure to *L. interrogans* serovars has been reported and there is no reason not to suspect it could impact bison and buffalo adversely (20). In other species it causes abortion in pregnant females, as well as nephritis and haemolytic anaemia (21).

Johne's Disease

Johne's disease is caused by *Mycobacterium avium* subspecies *paratuberculosis*. It causes a chronic lymphogranulomatous enteritis particularly affecting the ileum and jejunum, which can result in chronic diarrhoea, wasting and death (22, 23). Due to these chronic changes, infected animals may become immunocompromised and become more susceptible to other infectious diseases.

Neosporosis

This parasitic disease is caused by the protozoan parasite *Neospora caninum* with incursion occurring through contact with infected dog faeces. In cattle, once infected, animals are infected for life. However, during pregnancy infected females can either abort or transmit the parasite through to their offspring resulting in the birth of persistently infected animals. These offspring if female, can also go on to abort or further transmit the parasite on to their offspring. Viable *N. caninum* parasites have been isolated from buffalo (24) and antibodies have previously been detected in bison (25). Again, infected animals may be more prone to other infections during the period that the parasite is active.

Infectious bovine rhinotracheitis (IBR)

This viral disease is caused by bovine herpes virus-1 and is found worldwide. The symptoms can appear similar to the head and eye form of SA-MCF with a mucopurulent nasal discharge, pyrexia,

abortion in pregnant females, and pneumonia (26). It is unclear how big a problem this could be in bison as reports of infection and disease are scarce, however, in cattle the disease is widespread and can cause large outbreaks in naïve herds as well as contributing to multipathogen respiratory disease complex in calves (27). Infected animals could be more prone to other infectious diseases, particularly those affecting the respiratory tract, and symptomatic animals could present similarly to those with SA-MCF.

Mycoplasma bovis

In the USA *Mycoplasma bovis* has become a major concern in bison production with several large outbreaks reported in recent years (28-30). Symptoms vary, but can include lethargy, joint infections, lameness, respiratory disease/pneumonia, weight loss, mastitis, and abortions. In cattle other symptoms have also been reported (31). Infections can vary in severity, and damage from infection can increase the risk of other infections developing secondarily as a result of immune compromise and damage to affected tissues. In cattle, infections can become chronic with persistent shedding for many months (31).

Trace elements

Optimal nutrition underpins healthy livestock and whilst fresh and conserved forage should readily supply the energy and protein needs of bison and buffalo, the available forage can over or under supply various nutrients. For different species and breeds this can result in toxicity or deficiencies. These rarely result in acute mortality however chronic under or over supply can impair the health and performance of the animals. In Wales, recent work highlighted the likelihood of deficiencies in various trace elements in grazed pasture with a resultant reduction in optimal performance (32). Several minerals and trace elements are involved in the immune system and deficiencies in these can result in impaired immunity and an increased susceptibility to infection (32).

Rhug Estate

In March 2020 concurrent with the sampling of the bison for OvHV-2, the whole herd of bison (n=8) were tested for the following pathogens (

Table 3). Antigen and antibody tests were carried out on an individual basis. Parasitic tests were carried out on pooled faecal samples. Trace element tests were only possible on 5/8 bison due to the volume of blood available; insufficient blood was obtained from three animals. However, following on from other work the mean results from five samples is likely to give a reasonable estimate for this population of eight bison (32).

Bacterial and Viral Pathogens	**n/n (%) positive**		**Interpretation**
BVD antigen (PCR)	0/8	(0)	No evidence of any PI animals
BVD antibody	1/8	(12.5)	One animal previously exposed to BVD
L.hardjo MAT antibody	0/8	(0)	No evidence of previous exposure
Johne's antibody	0/8	(0)	No evidence of previous exposure
N. caninum antibody	0/8	(0)	No evidence of previous exposure
IBR antibody	1/8	(12.5)	One animal previously exposed to IBR
Mycoplasma bovis antibody	7/8	(87.5)	Seven animals previously exposed to *M. bovis*
Parasitic pathogens (n=8 samples)		**Result**	**Interpretation**
Pooled *D. viviparous*		Neg	Lung worm not detected
Pooled *F. hepatica* FEC		5 epg	Low adult *F. hepatica* burden
Pooled Rumen fluke FEC		9 epg	Low burden
Pooled nematode FEC		50 epg	Low burden
Pooled coccidial oocyst FEC		350 opg	Low/normal burden
Trace elements (n=5 samples)	**Mean**	**sd**	**Reference range**	**Interpretation**
Plasma selenium µmol/l	0.27	0.11	0.5-1	Marginally deficient
Plasma copper µmol/l	4.26	2.74	9.4-19	Deficient
Plasma zinc µmol/l	13.56	2.23	12.3-18.5	Adequate
Plasma cobalt nmol/l	25.81	17.49	>5	Adequate

Table 3: Pathogen screening test results for the bison at Rhug Estate, March 2020. Normal plasma trace element ranges for bison are unknown and so those for cattle were used as supplied by NUVetNA laboratories. EPG: eggs per gram; OPG: oocysts per gram.

One year later in March 2021, due to the small size of the herd, an opportunity presented itself for the purchase of eight more female bison in an attempt to increase the herd size. These were sampled and tested in the same way as the original bison, except that antibodies to *Mycoplasma bovis* were not tested for on this occasion (
Table 4).

Bacterial and Viral Pathogens	n/n (%) positive		Interpretation
BVD antigen (PCR)	0/8	0	No evidence of any PI animals
BVD antibody	0/8	0	No evidence of previous exposure
L.hardjo MAT antibody	0/8	0	No evidence of previous exposure
Johne's antibody	0/8	0	No evidence of previous exposure
N. caninum antibody	2/8	25.0	Two animals previously exposed to *Neospora*
IBR antibody	0/8	0	No evidence of previous exposure
Trace elements (n=8 samples)	**Mean**	**sd**	**Reference range**	**Interpretation**
Plasma selenium µmol/l	0.97	0.11	0.5-1	Optimal/high
Glutathione peroxidase	276.25	47.68	80-150	High
Plasma copper µmol/l	13.85	2.65	9.4-19	Optimal
Caeruloplasmin	22.64	6.46	15-35	Optimal
Caeruloplasmin:plasma cu	1.54	0.36	~2	Possible thiomolybdate problem
Superoxide dismutase	1681	158	2000-2500	Low
Plasma zinc µmol/l	13.56	2.23	12.3-18.5	Optimal
Plasma cobalt nmol/l	25.81	17.49	>5	Optimal
Plasma inorganic iodine pool (n=8 samples) µg/l	116.0		>100	Normal

Table 4: Pathogen screening test results for the purchased bison on entry to Rhug Estate, March 2021. Normal trace element ranges for bison are unknown and so those for cattle were used as supplied by NUVetNA laboratories.

Buffalo Dairy

In June 2020, concurrent with the sampling of the buffalo for OvHV-2, the buffalo were tested for the following pathogens (

Table 5). Antigen and antibody tests were carried out on an individual basis. Parasitic tests were carried out on pooled faecal samples. Trace element tests were carried out on an individual basis apart for plasma inorganic iodine which was carried out on three random pools of samples.

Bacterial and Viral Pathogens		**n/n (%) positi**		**Interpretation**
BVD antigen (PCR)		0/19	0	No evidence of any PI animals
BVD antibody		0/19	0	No evidence of previous exposure
L. hardjo MAT antibody		0/19	0	No evidence of previous exposure
Johne's antibody		0/19	0	No evidence of previous exposure
N. caninum antibody		0/19	0	No evidence of previous exposure
IBR antibody		1/19	5.3	One animal previously exposed to IBR
Mycoplasma bovis antibody		5/19	26.3	5 animals previously exposed to *M.*
	Suspect	8/19	42.1	*bovis*

Parasitic pathogens (n=10 samples)	**Eggs/oocysts per**	**Interpretation**
Pooled *D. viviparous*	Negative	Lung worm not detected
Pooled *F. hepatica* FEC	0 epg	*F. hepatica* not detected
Pooled Rumen fluke FEC	0 epg	Rumen fluke not detected
Pooled nematode FEC	0 epg	Nematodes not detected
Pooled coccidial oocyst FEC	0 opg	Coccidial oocysts not detected

Trace elements (n=19 samples)	**Mean**	**sd**	**Reference Range**	**Interpretation**
Plasma selenium µmol/l	0.44	0.23	0.5-1	Marginally low
Plasma copper µmol/l	12.91	2.09	9.4-19	Optimal
Plasma zinc µmol/l	12.16	1.68	12.3-18.5	Sub-optimal
Plasma cobalt nmol/l	4.86	2.25	>5	Marginally low
Plasma inorganic iodine pool 1 (n=6 samples) µg/L	112.3		>100	Optimal
Plasma inorganic iodine pool 2 (n=6 samples) µg/L	68.2		>100	Low
Plasma inorganic iodine pool 3 (n=7 samples) µg/L	60.8		>100	Low

Table 5: Pathogen screening test results for the buffalo at Buffalo Dairy, June 2020. Normal trace element ranges for buffalo are unknown and so those for cattle were used as supplied by NUVetNA laboratories. EPG: eggs per gram; OPG: oocysts per gram.

Objective 4 – make use of a vaccine against OvHV-2 on one farm with a known severe MCF disease problem, and as far as is possible, assess whether it is practically useful in this context.

The primary research for the SA-MCF vaccine was carried out prior to start of this project (33), and, it was only used at Rhug Estate for the following reasons: 1. Rhug Estate had a historically high disease incidence and whilst the other farms in this project were likely to be exposed to OvHV-2, they as yet do not have a severe problem with MCF. 2. The production of the vaccine is currently expensive, technically difficult, and time consuming, and therefore the funds available limit the use within this project to one farm. 3. The use of this vaccine is only possible under the veterinary cascade (34); in this project this vaccine was used under the veterinary cascade on an individual case basis as it was deemed necessary by the prescribing veterinary sturgeon. Expert advice on the use of this vaccine in this context was sought from DEFRA, the Health and Safety Executive (HSE) and the Veterinary Medicines Directorate (VMD). Where no licensed veterinary medicine exists one may be formulated for use in a specific disease context if prescribed by a veterinary surgeon under the VMD prescribing cascade (34). In doing so, the prescribing veterinary surgeon takes responsibility for its use, maintains the appropriate records and observes the relevant health and safety legislation. In this context, the high disease incidence and the lack of any licensed medicine justify the use of this vaccine in this context.

Due to the nature of the vaccine being classed as a novel genetically modified organism, a risk assessment for work with genetically modified micro-organisms was carried out and approved by the GM Safety Committee of the University of Liverpool and the Health and Safety Executive of the UK. Modified Vaccinia Ankara is classified as a category 1 hazard (lowest hazard group) under the Advisory Committee on Dangerous Pathogens framework due to multiple attenuating mutations over six separate genomic loci and a wide safety profile during vaccine campaigns where humans have been deliberately inoculated. Any risk to human health was therefore considered extremely low/negligible (35). However, due to the use of this recombinant organism, appropriate containment procedures were implemented within the 'Contained Use' framework.

Vaccination and methodology

Vaccination occurred in April 2020. At inoculation each animal was restrained using a squeeze type crush and inspected for any signs of ill health or disease by a veterinary surgeon (JA). Animals were body condition scored as for cattle (36). As all eight bison appeared healthy and in good body condition with no external signs of disease, each one was then injected with 2ml of the reconstituted vaccine solution, corresponding to 5×10^8 plaque forming units of virus, given by deep intramuscular injection in the middle third of the neck on the left-hand side. Tuberculin testing had been carried out 72 hours previously on the right-hand side of the neck. Injection was carried out by an experienced veterinary surgeon (JA) using a closed unit sterimatic automatic injector gun (MSD Animal Health) fitted with a 1.5 inch 16 gauge luer-lock needle (Figure 10). The needle was disinfected between each animal by way of the sterimatic bung containing 2.5% glutaraldehyde solution. To facilitate accurate administration a $10cm^2$ patch of hair was clipped from the injection site prior to administration (Figure 11). Immediately after inoculation (Figure 12) the injection site was inspected for any signs of leakage from the injection site and then immediately disinfected using surgical spirit sprayed onto the site (Figure 13). At no time was any leakage observed to have occurred. Following injection each animal was then immediately released into a secure holding pen. It was expected that viral clearance within each animal would occur within 48 hours (35), however

to ensure food safety and public confidence, and following advice from DEFRA and the Veterinary Medicines Directorate, a 28 day meat withhold was imposed.

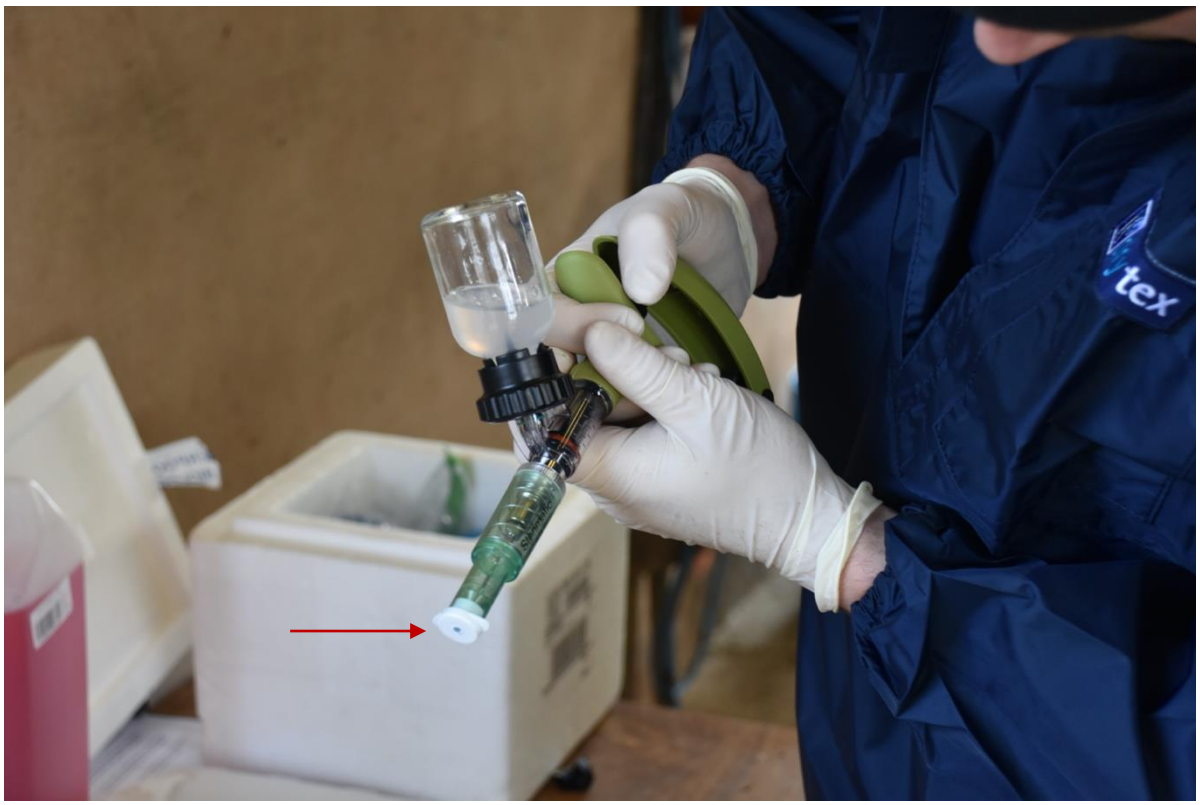

Figure 10: Vaccine and closed unit sterimatic injector system. The arrow indicates the sterimatic bung used to disinfect the needle automatically at each injection.

Figure 11: Hair was clipped from the injection site to facilitate injection and containment of the vaccine.

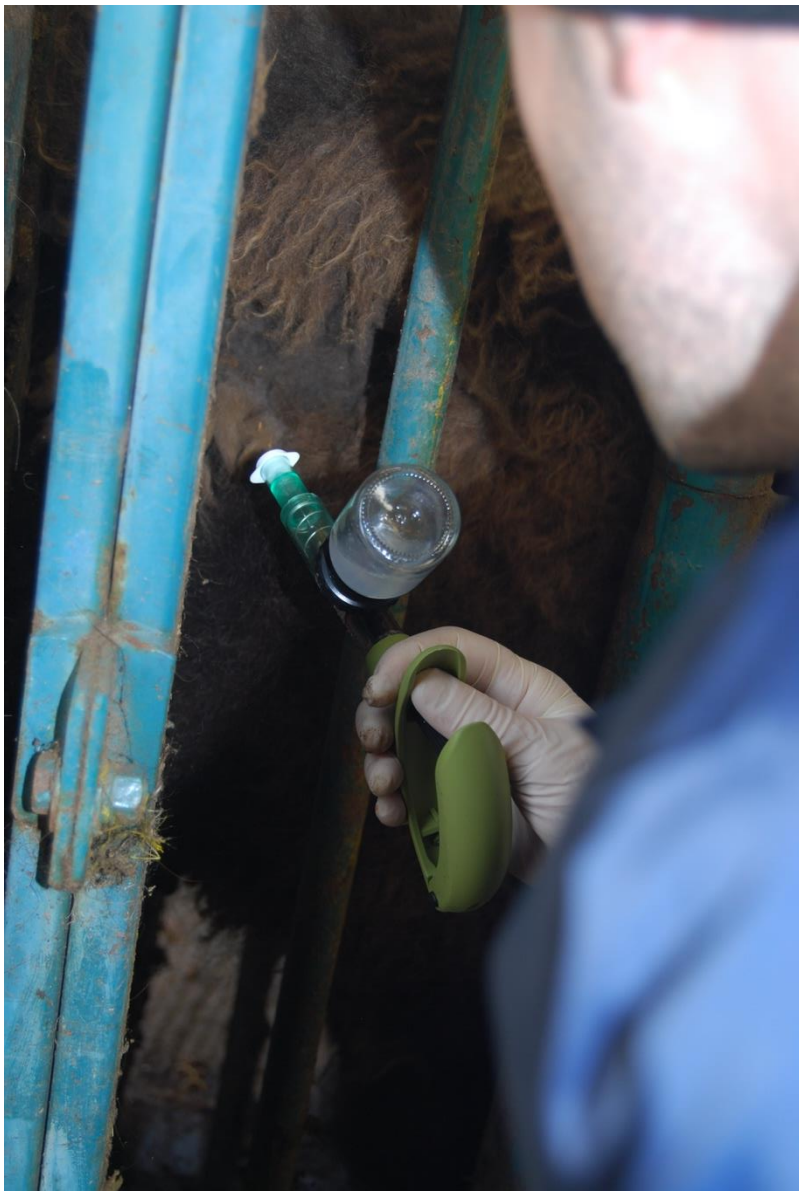

Figure 12: Injection of the vaccine in the clipped site in the neck of the bison.

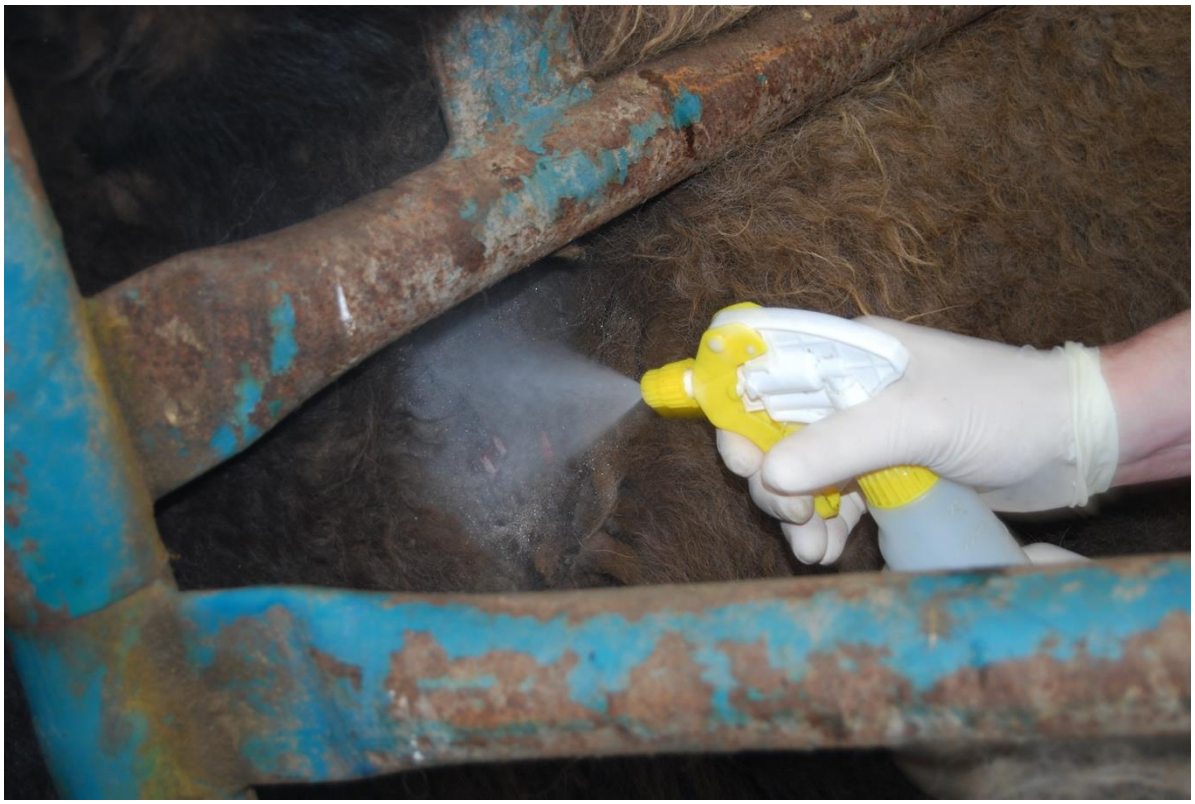

Figure 13: Disinfection of the vaccination site with surgical spirit immediately after vaccination.

Post vaccination

All eight bison were isolated and contained in the secure holding pen for seven days immediately adjacent to the crush and handling pens. This was to contain any faeces, urine or other secretions shed immediately post vaccination. Strict biosecurity measures were imposed including the prevention of entry by all personnel and sealing off this part of the farm for the duration. The bison were supplied with enough hay and water to exceed the time spent in confined isolation. To ensure the biosecurity measures were complied with, a daily, randomly timed inspection was carried out by the veterinary surgeon (JA). This also enabled distant visual examination of the bison to monitor for any signs of ill health.

Immediately post vaccination of the herd, the race, crush, and vaccine administration area were disinfected with a solution of 2% w/v Virkon (LANXESS). In addition, the vaccine administration equipment including the vaccine vial, gun, needles, and all waste were contained and submerged in 2% w/v Virkon (LANXESS) for 24 hours before being disposed of through a recognised clinical waste handling company.

After the seven days of isolation and confinement had elapsed, the bison were released back to their usual pasture environment. The surface of the isolation pen was then sampled whereby the person sampling walked in a zig-zag fashion throughout the pen wearing boot socks, which were subsequently removed before a plain cotton swab was drawn firmly five times across the contaminated surface from each sock. In a similar fashion, the animal handling system was also sampled. These swabs were then frozen at -20°C prior to analysis.

Second vaccination

Following the purchase of the new bison in 2021, the seven of the eight original bison (one had died as a result of chronic cardiac injury and stress (Table 6)), one new calf and the eight new bison were all vaccinated or revaccinated in June 2021 in an identical way to the first. The original bison had now received two doses of vaccine and the new ones and the calf one dose. In order to minimise stress, the original bison were also due a booster multivalent clostridial vaccine (Bravoxxin 10; MSD Animal Health) which was also administered concurrently, but on the opposite side of the neck to the MCF vaccine.

Laboratory tests

Vaccine quality

Prior to use, a quality assessment of the vaccine was carried out by conducting a metagenomic sequence analysis using a MinION nanopore DNA sequencer (Oxford Nanopore Technologies) to confirm the absence of other infectious agents as well as testing for any bacterial growth on agar plates.

Virus and antibody tests

Post thawing, the swabs used to sample the post vaccination housing environment and handling facilities were analysed for the presence of MVA by PCR.

Blood samples were taken immediately prior to both vaccination events from the coccygeal or jugular veins for other clinical reasons (e.g. trace element or other pathogen testing). In addition, some of the drawn blood was processed for OvHV-2 detection by qPCR and antibody testing using ELISAs specific for IgG to the MVA construct and also the gB protein. Post vaccination, blood samples were also obtained at other clinical/treatment events, at 42 days and 400 days post vaccination, to determine whether antibodies to the vaccine could be detected.

For the OvHV-2 and antibody tests, the blood was processed within hours of sampling. Plasma and buffy coat were extracted and then frozen separately at -20°C. Samples were then thawed immediately prior to analysis.

For the qPCR analyses, DNA was extracted from buffy coat using a DNeasy Blood & Tissue kit according to manufacturer's instructions. 100ng of each sample was run in qPCR using TaqMan™ Universal PCR Master Mix, with primers at a final concentration of 0.5uM. Cycling conditions were 50°C for 2 minutes, 95°C for 13 minutes then 39 cycles of 95°C for 15 seconds then 60°C for 1 minute.

For antibody quantification, high protein binding plates were incubated overnight with either heat inactivated WT-MVA virus or gB protein. Plates were blocked with 2% milk powder 0.05% Tween PBS then incubated with samples diluted in blocking buffer at 37°C for 1 hour then washed. Plates were incubated with Goat anti-Bovine IgG HRP (ThermoFisher) diluted in blocking buffer at 37°C for 1 hour before being washed a final time then detected with TMB substrate which was stopped with sulfuric acid. Absorbance was read at 450nm.

Results

Clinical observations

At each visit for the immediate seven day period post-vaccination, the bison were observed to be either standing and eating, or lying and cudding. The bison continued to behave normally throughout their confinement and at no time did any bison exhibit any signs of ill health such as any nasal discharge, reduced appetite, increased or decreased respiratory rates, lying apart from the herd, or appearing distressed in any way.

No deaths confirmed as SA-MCF were observed during the study period April 2020 to December 2022 (Table 6). Ovine herpes virus-2 was detected in 3/6 bison that died during the study period, however pathology did not indicate MCF and virus was detected at a low level. It is possible that latently infected animals may express some virus due to challenges to the immune system when undergoing other health challenges.

During the study period six bison died and were available for post mortem examination. For each animal, tissue samples of either one or a combination of rete mirabile, lymph node, or spleen were analysed for OvHV-2 by PCR/qPCR. As part of the diagnostic process various ancillary tests were carried out as indicated through the diagnostic process. The overall diagnostic result together with abnormal results pertinent to the health of the rest of the herd are summarised in
Table 6. At various times OvHV-2 was detected by PCR but histopathology consistent with SA-MCF was not detected. It is possible that some lesions were obliterated due to the rapid autolysis of the carcasses however in most cases another cause of death was considered more likely. The OvHV-2 could have been a latent infection with recrudescence as a result of stress perimortem.

Visually, throughout the study period, the bison appeared in an improved condition compared to that prior to the study. On average, mean body condition scores increased from 2.5/5 to 3.3/5. It is difficult to know how much effect the vaccination had on their overall condition as other interventions e.g. anthelmintic treatment and trace element supplementation will also have contributed.

No adverse events through giving both the SA-MCF vaccine and the multivalent clostridial vaccine concurrently were observed.

Date of death	Signalment	Cause of death	Evidence of SA-MCF	Other abnormal findings pertinent to the health of the rest of the herd	Comments pertinent to the health of the rest of the herd
17/05/2020	Bison calf, male, 8 months old	Chronic cardiac injury and fibrosis; stress of handling 2 days earlier	OvHV-2 detected by PCR.	Marginal liver tissue selenium deficiency; plasma selenium deficient.	Suspected chronic selenium deficiency. OvHV-2 detected but pathology not consistent with MCF. Possible latent infection.
10/10/2021	Bison bull, 6 years old	Terminal septicaemia	None	• Fibrinosuppurative meningitis • Suppurative bronchopneumonia	*Histophilus somni* should be considered.
19/10/2021	Bison heifer, 3 years 3 months old	Unknown – possible bacterial brain infection and chronic parasitism	OvHV-2 detected by PCR.	• Occasional cardiac sarcocysts. • Lice eggs detected. • Faecal worm egg count 250 trichostrongyle epg	OvHV-2 detected but pathology not consistent with MCF. Possible latent infection.
03/11/2021	Bison cow, 6 years old	Shooting; euthanasia due to chronic weight loss – parasitic gastroenteritis	OvHV-2 detected by PCR.	• Lungworms identified • Total faecal worm egg count 350 epg • Heavy/fatal burden of *Ostertagia ostertagii* 18,560 parasites • Heavy burden of *Trichostrongylus axei* 4,640 parasites • Marginal copper deficiency • Marginal selenium deficiency	OvHV-2 detected but pathology not consistent with MCF. Possible latent infection.
20/01/2022	Bison calf, female, 6 months old	Diarrhoea and severe chronic weight loss – parasitic gastroenteritis	None	• Total faecal egg count 650 epg	
16/03/2022	Bison cow, 3 years old	Unknown – suspected stress related post TB test	None	• Multiple cardiac sarcocysts	

Table 6: Summary of post-mortem examination findings.

A significant cause of death in 3/6 bison was as a result of parasitism with abomasal and intestinal nematodes, resulting in diarrhoea and wasting. In addition, the first death was likely to be due to a chronic selenium deficiency. Finally, in several cases stress was considered to at least have contributed to death of the bison if not conclusively a specific cause. Other incidental but important findings included other parasites, including lung worms and *Sarcocystis* spp., together with marginal copper and selenium deficiencies, despite supplementation with slow-release trace element boluses.

Laboratory results

Vaccine quality

The results of the vaccine quality assessment using the MinION sequencer indicated a high level of purity (97.4%) and there was no bacterial growth on agar plates.

Environmental swab samples

No MVA DNA was detected by PCR on any of the swabs used to sample the post vaccination isolation housing and the handling facilities.

qPCR detection of OvHV-2

No OvHV-2 was detected in any of the blood samples pre- (n=8) or post- (n=8) vaccination at any time point. OvHV-2 was detected by PCR in some of the post-mortem tissues in bison that died from other causes (Table 6).

Antibody results

Blood IgG for the MVA vaccine construct, as detected by ELISA, demonstrated a significant increase in antibodies 42 days post vaccination (Figure 14). Blood IgG specific to the gB protein as detected by ELISA was not significantly different in the samples obtained pre vaccination or those obtained 42, or 400 days post vaccination (Figure 15).

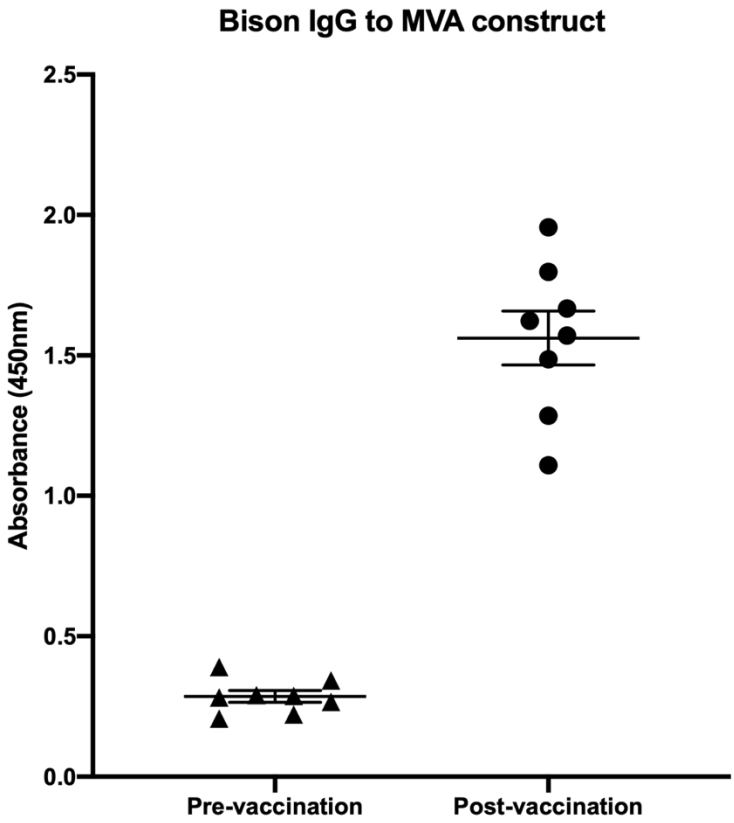

Figure 14: Immunoglobulin G for the MVA vaccine construct ELISA data for the bison, pre and 42 days post vaccination with the SA-MCF vaccine.

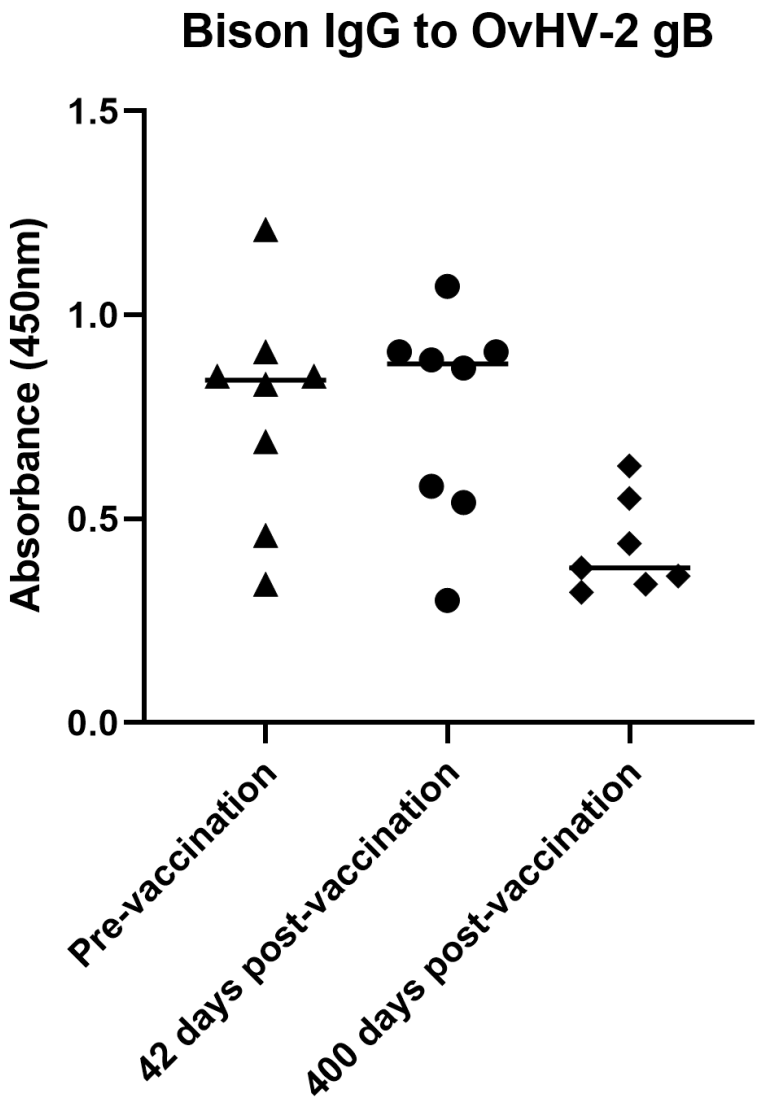

Figure 15: Immunoglobulin G ELISA data for the bison, pre vaccination and 42 and 400 days post vaccination.

Discussion

In this study, the herd of bison were successfully inoculated with a novel SA-MCF vaccine on two separate occasions. No clinical adverse events were noted. Containment of the vaccine was achieved on both occasions within the animals, with no detection of vaccinal genetic material outside the animals in any of the environmental swabs. This is important under the GMO (Contained Use) Regulations 2014.

No vaccine associated adverse events were detected and the bison appeared to be clinically well as a result of the vaccine. A death occurred in a poorly grown calf 44 days after vaccination, but two days after handling for trace element treatment and blood sampling. Following an investigation, this death was not considered to be as a result of the vaccination but as a result of stress through handling and concurrent chronic cardiac fibrosis as a result of a chronic selenium deficiency.

In addition to the calf, five other deaths occurred during the post vaccination study period. However, none of these were as a result of SA-MCF although some virus was detected in 3/6 cases. Latent, sub-clinical infection has been demonstrated in cattle (7, 37) and given the other pathologies noted it is considered likely that the detection of the virus in these instances was as a result of latent virus infection. These deaths and resultant detection of OvHV-2 illustrate the sensitivity of bison to stress and disease, and the importance of optimising the overall health of the animal. Any stressor (handling, nutritional, environmental, infectious etc.) could result in impairments to immunity with an increased likelihood of recrudescence of latent viruses, including OvHV-2.

It is not possible to say whether the vaccine prevented infection or disease in the bison, or whether one or two inoculations afforded a different clinical outcome. To determine this a randomised controlled challenge study design would be required which was outwith the scope of this study. However, given the setup of the farm, the bison were at times throughout the study period in close proximity to sheep, and at specific times in close proximity to sheep under stress e.g. lambing time. Two lambing periods were experienced during the study period and at that time the bison were located within 100-200m of the lambing shed for a period of 5 weeks. At no time were the bison more than 500m away from sheep. Given this close contact and the demonstrable safety of the vaccine further investigation should be carried out to establish efficacy.

Laboratory analyses indicated a significant increase in circulating immunoglobulins specific to the MVA vaccine construct 42 days post vaccination (Figure 14). This is indicative of a successful immunological response in the bison. Antibodies specifically to the gB protein did not show any significant increase in concentration post vaccination compared to pre-vaccination concentrations (Figure 15). Whilst this is less encouraging, it is still unclear as to what antibody concentration is required for protection or a reduction in pathology. Antibodies, produced by B lymphocytes, can be important in fighting virus infections. Herpesviruses become latent and so effector T-cells are equally important for an effective vaccine. Vectored vaccines based on MVA are highly effective at generating potent T-cell responses and importantly the generation of memory T-cells (38). Recent work has demonstrated a specific role for infected T-lymphocytes in the inflammatory processes seen in MCF, alongside monocytes and locally proliferating macrophages (39). Immunological memory generated by a vaccine resulting in memory T-cells may be advantageous in the rapid deployment of specific effector T-cells post viral challenge resulting in the rapid destruction of infected cells and a consequent reduction in pathology. Further work is now needed to understand the specific immune response to this MVA vector vaccine and whether this hypothesis holds true.

Objective 5 – management and control of MCF on farm

The disease profile for each herd was analysed from all the available data and a control strategy developed tailored to each farm as a result. These were issued in the form of written reports and discussed verbally with each farmer. The reports were also made available to each farmer's own veterinary surgeon. These were reviewed regularly and revised as necessary. The final Health Plans with relevant disease control measures are included in Appendix 1 and Appendix 2.

After deployment, the farms were monitored for a further 12 months to assess the success of the control package implemented on each farm. The farmers were contacted at regular intervals by telephone. For Buffalo Dairy no significant health events occurred during the monitoring period, and production was also reported to be good.

For Rhug Estate, overall, the general health of the bison appeared to be improved, as evidenced by the visual improvements and the measurable improvements in body condition. Six deaths did occur although SA-MCF was not a cause in any of them. The information gathered from the investigations carried out for each death resulted in updates and improvements to the health plan, with improvements made to monitoring and management of the health of the herd. For example, more regular faecal egg counts are now carried out to monitor the parasite burden of the herd as well as routine trace element supplementation. These management approaches are now established and will continue after the cessation of the project.

Stress remains a significant and difficult factor to manage. Farmed animals inevitably need handling for management events e.g. parasite control, trace element supplementation and statutory testing e.g. for bovine tuberculosis. Combining these necessary events where possible in order to minimise further handling is essential to minimise the number of stress events experienced by the bison. In addition, trying to arrange these events to occur separately to concurrent sheep stress events e.g. lambing, would be prudent in order to limit the risk to the bison as much as possible.

Objective 6 – formulate a strategy for control of MCF on bison/buffalo farms in Wales

It is possible to farm both bison and buffalo successfully in Wales with respect to SA-MCF control. However, both species are not without their challenges and an understanding of the biology of the virus and its epidemiology can assist in successful management.

In this project Buffalo Dairy exist in general isolation from sheep and although four animals were found to be latently infected with virus detectable by qPCR, this isolation from sheep is likely to have contributed to the zero incidence of SA-MCF in this project.

The bison at Rhug Estate were always situated in generally close proximity to sheep. Deaths occurred, but after the deployment of the management and control plan none were attributable to SA-MCF. Whilst the vaccine may well have contributed to this success, the data is not strong enough to make firm conclusions as to specific efficacy.

For farmers seeking to establish or develop existing herds the following is recommended:

1. Establish the herds as far away as possible from sheep, with where possible, at least a 3km distance from the nearest sheep. Whilst this may still not be far enough to totally eliminate windborne aerosol transmission, further distances allow for a greater dispersal of any infected aerosol thereby reducing the risk.
2. Optimise the nutrition of the animals ensuring an appropriate forage based diet with supplementation with energy, protein and minerals based on a sound evidence based approach. Well nourished animals are likely to be more resistant to infectious diseases. Use appropriate downstream monitoring and make adjustments as necessary. There is some evidence bison could be particularly vulnerable to low copper diets (40) so testing and intervening where necessary could be beneficial.
3. Manage parasitic diseases carefully – adult animals may not always have low parasite burdens so regular pooled faecal egg counting e.g. on a monthly basis, is recommended to detect any increase in parasitism early. Treat affected groups promptly applying the COWS (41) recommendations as for other bovines.
4. Manage other infectious diseases intensively through appropriate testing and control strategies, with regular monitoring and review of their effectiveness. For example, vaccination against common infectious diseases e.g. BVD, IBR and clostridial diseases can readily be carried out.
5. The SA-MCF vaccine used in this study was experimental and is unavailable for use on other farms at this stage. However, the evidence gathered in this project is encouraging and we would hope that to enable more farmers to explore bison and buffalo farming in Wales as a diversification option that appropriate funding, possibly from Welsh Government, could be forthcoming to allow rapid development of this vaccine.

Objective 7 – knowledge exchange

The farmers in this project were communicated with regularly to ensure understanding of the data for their operation and the implications for management. Written reports were supplied throughout documenting the data and advice as necessary. Written health plans were compiled and supplied to each farmer to provide a basis for the health management of their herds (Appendix 1; Appendix 2). These were also made available to their own veterinary surgeon.

Data and conclusions from this project were also presented at the World Buiatrics Conference 2022 in Madrid, under the Infectious Diseases theme https://www.wbc-madrid2022.com/index.php/en/. This was carried out in the form of two talks, and a poster from an allied project was also presented. This international conference attended by >3,000 delegates from across the world served as a platform to disseminate the experiences and findings from this project to a wide audience. Discussions were also held with delegates with regard to the experiences reported here and comparisons made with the experiences of other delegates in their local areas.

A meeting is planned in summer 2023 in association with the Moredun at Rhug Estate where results of this project will be further disseminated. Applications are also in the process of being made to present elements of this work at meetings of the British Cattle Veterinary Association and the Sheep Veterinary Society – specialist divisions within the British veterinary profession.

Opportunistic outbreak investigations of SA-MCF cases on cattle farms in proximity to the project

During the project, three outbreaks of SA-MCF in cattle were detected on farms not originally included in the project, but situated close to one or other of the project farms. We took the opportunity to obtain samples from the cattle and sheep on these farms where possible in order to gain a deeper understanding into the disease. The inclusion of these farms has been really useful in being able to study the disease dynamics in real time within a herd. The scarcity of bison and buffalo herds within Wales, and in the UK in general, necessitate that cattle herds may offer information pertinent to the project farms. The proximity of these farms has also served to improve our understanding of the wider local risk in general.

The data from these outbreak investigations has been prepared as case report series for publication in a scientific journal, in that it may be peer reviewed and serve to reach a wide audience that may benefit from the data and insights gained from these investigations.

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Appendix 1 – Final health plan for Rhug Estate

Health, welfare and biosecurity plan for BISON at Rhug Estate

Date of report: 11th February 2021

Veterinary Surgeon: Joseph Angell BVSc MSc DipLSHTM PhD MRCVS

This document represents an attempt by the above named veterinary surgeon to evaluate the specific situation and risks on this unit on the basis of the past history, relevant examinations, treatments and laboratory procedures performed on the unit. Where possible cost-effective control strategies are proposed. It does not represent an exhaustive list of the diseases and disease risks present, it must be read in conjunction with the health plan and no liability is accepted for any omissions or future disease cases that may arise. It must be emphasized that the disease situation on any farm is constantly changing and therefore continuous veterinary input is required to keep track of it. This document should not be copied or re-produced without the written permission of Wern Veterinary Surgeons.

Farm Details

Name	Gareth Jones	Phone Number	01490 413000
Address	Rhug Estate	Fax Number	
	Corwen	Mobile phone no	07887 911771
	Denbighshire	Email address	gareth.jones@rhug.co.uk
	LL21 0EH		
		Holding Number	56/318/5001/04
Species	Bison	Number	8-16

Summary of routine actions showing the timing of routine tests, vaccinations and treatments required

At TB testing

At day 1 of TB test:

1. Take blood samples for trace element testing/monitoring from 8 weaned/adult animals.
2. Take blood samples for Johnes testing in all adults **over two years of age.**
3. Take blood samples from **5 youngstock** to test for BVD antibodies.
4. Take pooled faecal samples for fluke egg identification and gastrointestinal nematode counts.

At day 2 of TB test

1. Treat for PGE, fluke and external parasites as necessary based on test results.
2. Administer COSECURE bolus (adjust dose based on body mass).
3. Administer Macavax MCF vaccine (with appropriate containment carried out).
4. Administer lungworm vaccine if necessary.
5. Administer clostridial vaccine to youngstock (<1 year of age).

Autumn

1. Administer live IBR vaccine to all animals over 2 weeks of age. To bison between 2 weeks and 3 months of age it should be given intranasally. To bison over 3 months of age it should be given intramuscularly.
2. Administer clostridial vaccine to youngstock (<1 year of age).

Biosecurity

The current herd has been tested and shown to currently be free from BVD, leptospirosis, Johnes disease and neosporosis. Antibodies to infectious bovine rhinotracheitis (IBR) were detected in one animal, and antibodies to *Mycoplasma bovis* were detected in 7/8 animals.

Contact between bison and other animals on the farm

The bison graze separately from all other animals on the farm and contact with other animals should be avoided where possible to minimize the risk of introducing an infection. Sheep, cattle and goats can both transmit infections to bison via nose-to-nose contact and in some cases by aerosols which can be carried by wind. **Sheep in particular should be managed as far away as possible from the bison (ideally greater than 3 miles!) to reduce the risk of the transmission of OvHv-2.** This risk is increased during periods of stress for the sheep e.g. lambing, weaning, shearing, dipping etc.

Added animals

When purchasing animals from sources with an unknown disease status:

1. **Check TB status of source herd before purchase and any appropriate testing has been carried out.**
2. **Isolate animals for one month in a separate field or pen. Adopt strict biosecurity measures for all personnel, feed and equipment.**
3. **Arrange for blood sampling of all purchased animals for MCF associated viruses, Johnes antibodies, BVD antigen, IBR antibodies, *Neospora* antibodies and *Leptospira* antibodies. In addition, sampling for trace elements may also be beneficial in order to consider whether the new animals require similar or different supplementation compared with that for the existing herd.**
4. **In addition, treatment for liver fluke and worms and external parasites should be considered, unless they have recently been treated prior to purchase.**
5. **Vaccination using the novel recombinant Macavax MCF vaccine should be carried out as soon as possible, with appropriate containment accounted for (see separate containment document which includes the protocols necessary for this).**

Infectious diseases

Malignant catarrhal fever

Malignant catarrhal fever (MCF) has caused many problems in this herd over the years. In the UK it is caused by herpesviruses of the *Macavirus* genus, namely ovine herpesvirus 2 (OvHV-2) and caprine herpesvirus 2 (CpHV-2), and whilst sheep are considered the main reservoir host, a large proportion of cattle can also carry the virus subclinically. Bison have been found to be very susceptible to infection and minimizing contact between bison and species potentially carrying and shedding these herpes viruses is essential in aiding control. Sheep in particular may shed large quantities of virus when under stress (e.g. lambing etc.) and the virus may infect bison 3 miles away via airborne aerosol carriage. Aerosols containing virus disperse in the air and spread out, and therefore **maintaining as great a distance as possible from the sheep** will reduce the risk of infection even if it is not possible to maintain optimal distances and eliminate all risk.

Vaccination annually in the Spring (concurrent with TB testing) with the Macavax MCF vaccine may help prevent disease, although its efficacy is not fully established. Disease may still occur if the immunity of the bison is adversely affected in some way or if there is sufficient virus to 'overwhelm' any protective immunological response. Until it bears a UK license this live recombinant vaccine must be used under Class 1 GMO Contained Use (GMO(CU)) regulations. The containment use risk assessment and contained use forms are attached.

Neospora caninum

This is a parasite shed by dogs, particularly ones that ingest fresh bovine tissues, with placentas and calves being a particular risk, hence farm dogs that have access to any carcasses/afterbirths and hounds fed on possibly occasionally undercooked offal represent a high risk as contaminators of silage, mixed rations in the barriers or simply of fields grazed, in this case by heifers particularly. Thus, prompt carcass disposal and sealed (dog proof) storage are essential.

Once animals are infected by ingesting the parasite it becomes dormant in muscle tissue until immune system changes induced by pregnancy cause its activation, multiplication and migration via the maternal blood to settle in the foetus which may either be killed and aborted immediately or after a period of mummification, or itself become latently infected until, in the case of females, they themselves become pregnant. Thus, vertical spread from mother to daughter is very effective. Spread is also possible from an infected dam to other calves via pooled colostrum. Direct horizontal transmission between animals, other than colostrum, is not possible.

The following actions should be considered:

1. **Imported cows/heifers could be Neospora carriers so blood test on entry (see added animals above).**
2. **Blood test any aborting bison in case of previously undetected infection.**
3. **Ensure that the risk of dogs contaminating pasture and feed are minimized by:**
    i. **Minimize the exposure of your own dogs to cleansings and afterbirths.**
    ii. **Prohibit visitors from allowing dogs to roam the yards/fields.**
    iii. **Consider the risks from 'foreign dogs' crossing your fields (the parasite can survive, at least low grade fermentation, hence silage can be contaminated in the field).**

Bovine viral diarrhoea (BVD) virus

Currently there is no evidence of BVD within the bison herd. As such, provided the bison can be kept completely separate from cattle, sheep and goats, the risk of infection can be managed. Contact with other species, particularly purchased cattle will greatly increase the risk of infection possibly resulting in abortion and immunosuppression.

The following actions should be considered:

1. **Imported bison could be BVD carriers so blood test on entry (see added animals above).**
2. **Blood test any aborting bison in case of previously undetected infection.**
3. **Have tissues from any aborted foetuses tested in case of previously undetected infection.**
4. **Consider vaccinating the herd for BVD if there are concerns about effectively managing disease incursion risks.**

Infectious bovine rhinotracheitis (IBR)

There is evidence that the herd has been exposed to IBR infection in the past, with 1/8 animals demonstrating antibodies to this virus in March 2020. However, as yet we have not determined that any clinical disease has been associated with IBR, and clinical diseases has not been reported to my knowledge in the USA or Canada either. There are risks of exposure to this virus in the bison herd via wind-borne aerosols and recrudescence of latent infection within the herd, however it is unclear whether this will cause any clinical or subclinical effects. **Vaccination of all bison with a live vaccine in Autumn could be carried out as a precaution and may help reduce any risk of clinical disease or sub-clinical immunosuppressive effects.**

Leptospirosis

There is currently no evidence of exposure to leptospirosis within the herd. However, infection can spread from in contact cattle or sheep via shared grazing, water courses or contaminated feed.

The following actions should be considered:

1. **Imported bison could be carriers of leptospires so blood test on entry (see added animals above).**
2. **Blood test any aborting bison in case of previously undetected infection.**
3. **Have tissues from any aborted foetuses tested in case of previously undetected infection.**
4. **Consider vaccinating the herd for leptospirosis if there are concerns about effectively managing disease incursion risks.**

Johnes disease

There is currently no evidence of infection with Johnes disease (*Mycobacterium avium* subspecies *paratuberculosis* (MAP)) within the herd. Infection could be introduced via an infected animal, wildlife, infected watercourses, contaminated feed, contaminated equipment/housing etc.

The following actions should be considered:

1. **Imported bison could be carriers of the causative organism and this is the probably the most likely route of entry so blood test all animals on entry (see added animals above).**

2. **Blood and faecal test any bison showing symptoms of chronic weight loss, diarrhoea, ventral oedema or failure to thrive, in case of previously undetected infection.**
3. **Consider annual screening all bison over 2 years of age for Johnes antibodies in order to monitor for any undetected disease.**

Mycoplasma bovis

Recent testing revealed 7/8 animals with exposure to *Mycoplasma bovis*. In cattle, this disease can cause respiratory problems in calves, mastitis in lactating cows and joint infections. We would expect similar issues in bison. Bison infected with MCF may develop secondary infection with *Mycoplasma bovis* as well, making the disease worse. Currently, we do not know how much of a specific problem it is causing or whether it is exacerbating disease.

The following actions should be considered:

1. **Test future cases of respiratory disease/mortality to determine the extent to which *Mycoplasma bovis* is contributing to the clinical signs.**
2. **If it becomes apparent that *Mycoplasma bovis* is contributing to the clinical picture, there is a new vaccine Myco-B we can try which may help reduce the clinical signs although I am unsure as to its efficacy in bison and reports from the USA are unclear. There are also autogenous bacterin vaccines produced and it could be possible to source one of these or develop one should it be necessary.**

Parasitic diseases

Liver fluke

Liver fluke infection can cause weight loss, anaemia, reduced milk yield and poor fertility and in growing animals can be fatal. **Faecal testing the herd at day 1 of TB testing would be recommended to determine if adult liver fluke infection is present.** Treatment can then be carried out at day 2 of TB testing if necessary. Due to the timing of TB testing in spring, treatment with an adulticide e.g. Trodax would be recommended if liver fluke infection is detected.

Parasitic gastroenteritis complex

So far this has caused very few issues, however, remain vigilant for signs of disease among growing animals. **Faecal testing the herd at day 1 of TB testing would be recommended to determine if a severe infection is present.** Treatment can then be carried out at day 2 of TB testing if necessary.

Lungworm

Lungworm issues have been identified historically, however more recently it has not been a problem. Lungworm can be carried by earthworms and projected into the air by airborne fungal spores so biosecurity cannot be relied upon to keep pastures free. Problems may have occurred previously due to impaired immunity. If necessary, a lungworm vaccine could be given to young stock at day 2 of TB testing.

Skin parasites

Monitor the herd for signs of scratching and contact the vets to discuss if this is noted.

Other potential disease issues

There are many other diseases that could cause issues. However, so far they have not caused any problems or been detected. That said, they are worth being aware of and remaining vigilant for and appropriate actions can be carried out if and when necessary. A list of some possible issues with some advice is provided on the next page.

Other infectious or infective diseases to be considered		
Disease	Possible clinical effects	Control programme or Advice
Salmonella dublin	Scouring adult cows, particularly after stress periods such as calving. Abortions in late gestation. Vague illness in calves.	Have any scouring adults examined by vet +/or faecal culture. Send in faeces sample from any calves with bloody scour.
Salmonella typhimurium	Scouring adult cows, particularly after stress periods such as calving.	
Tuberculosis		The herd is currently clear of TB infection. Biosecurity procedures should prevent cattle to bison transmission. Long term the local disease situation will dictate the risk from wildlife with badger visitation of supplementary feeding areas possible, and avoidable only by badger fencing.
Respiratory Syncitial Virus (RSV)	These will manifest themselves as calf pneumonia – usually between 6 weeks and 12 months of age.	Test if pneumonia becomes a problem.
Parainfluenza 3 Virus (PI 3)		
Manhaemia spp.		
Haemophilus Somnus		
Rotavirus	These will manifest themselves as calf diarrhea – usually between 0 and 3 weeks of age.	Unlikely to be an issue with managing these animals outside, however if necessary send in faeces sample from 2nd scouring calf in a batch.
Coronavirus		
E. Coli K99		
Cryptosporidium parvum		

Coccidiosis	Common problem from faecal contamination of feed and water troughs and from calves eating straw bedding. Manifests itself as scour, illthrift and in severe cases bloody diarrhea with severe straining in calves over 3 weeks old.	Minimise faecal ingestion. Veterinary examination and testing of scouring calves over 3 weeks old.
Clostridial diseases	Sudden deaths, usually in bison at grass, although younger calves can be infected.	**Report all sudden deaths to vets.** **May be necessary to vaccinate calves if an issue, although two doses 4-6 weeks apart are required.**
Ringworm		Avoid borrowing handling equipment etc. Housing can harbor ringworm spores for several years if previously contaminated by cattle. A vaccine is available if necessary.

Nutrition

Currently the bison are grazed on good quality grass pastures, with supplementary silage offered in the winter. This approach should generally meet the nutritional needs of the herd without additional feeding. However, some trace element deficiencies have been identified and have resulted in clinical disease and death.

Grass samples taken from two fields used by the bison have been used to inform this advice, as has the latest blood analyses.

Selenium

One of the forage samples shows the available selenium to be below optimal. In addition, for the blood samples obtained, the plasma selenium sits on average in the marginally low range although two individuals are within the deficient range. Also, the postmortem of a calf with evidence of poor growth revealed some heart muscle damage that was likely as a result of chronic selenium deficiency. Therefore, it is likely that the bison may benefit from some selenium supplementation and this could be supplied in a slow release bolus, which is likely to be most easily administered at day 2 of TB testing. **Some supplementation may be beneficial.**

Copper

The available copper in both forage samples was low. Also, the concentration of copper antagonists was high. The average plasma copper concentration measured very low for the blood samples tested, with three animals more severely affected. Therefore, it is likely that the bison may benefit from some copper supplementation and this could be supplied in a slow release bolus, which is likely to be most easily administered at day 2 of TB testing. **Some supplementation may be beneficial.**

Zinc

The available zinc concentration in both forage samples was low. However, on examination of the blood samples analysed, one animal had a plasma zinc concentration just below the normal range although the average of the group sits well within the normal range. **No supplementation is required but keep this under review.**

Cobalt

The cobalt concentration fro one of the forage samples was within the normal range but was low for the other sample. Based on the blood samples analysed cobalt was on average high. This is unlikely to cause a problem but no further supplementation is needed. **No supplementation is required.**

Iodine

The concentration of iodine in both forage samples was low, however there is currently no animal data available on this. I would expect there to be at least a marginal deficiency of iodine and some blood investigation would be beneficial to be able to determine the correct level of supplementation.

Note

Trace element management **testing** is just one part of assessing whether trace elements are needed. The interpretation is supplied based on the current knowledge of the farm history particularly with regards to production and health, and this of course may change and vary over time. There is always the risk of over supplying animals with trace elements so where values are only marginally outwith the normal or expected range any real improvements may not be seen and there is a risk of toxicity as well.

Actions to be considered:

1. **Plan to take blood samples from 8 individuals at day 1 of TB testing to monitor the trace element situation. Include tests for copper, selenium, cobalt, zinc and iodine.**
2. **Plan to have liver samples stored/analysed for trace elements from fallen stock or slaughter animals. This can give a longer-term indication of trace-element nutritional assimilation allowing more fine tuning to take place.**
3. **Plan to assess the trace element composition of the available forage on a yearly basis.**
4. **Give trace element boluses containing Copper and Selenium at day 2 of TB testing. Adjust the dose based on body mass.**

Appendix 2 – Final health Plan for Buffalo Dairy

Health, welfare and biosecurity plan for BUFFALO at Buffalo Dairy

Date of report: 11th February 2021

Veterinary Surgeon: Joseph Angell BVSc MSc DipLSHTM PhD MRCVS

This document represents an attempt by the above named veterinary surgeon to evaluate the specific situation and risks on this unit on the basis of the past history, relevant examinations, treatments and laboratory procedures performed on the unit. Where possible cost-effective control strategies are proposed. It does not represent an exhaustive list of the diseases and disease risks present, it must be read in conjunction with the health plan and no liability is accepted for any omissions or future disease cases that may arise. It must be emphasized that the disease situation on any farm is constantly changing and therefore continuous veterinary input is required to keep track of it. This document should not be copied or re-produced without the written permission of Wern Veterinary Surgeons.

Farm Details

Name	Duncan and Julie Aitkenhead	Phone Number	
Address	Buffalo Dairy	Fax Number	
	Ty Mawr	Mobile phone no	07967 976272
	Llanon	Email address	buffalodairy@gmail.com
	SY23 5LZ		
		Holding Number	
Species	Buffalo	Number	~50

Biosecurity

A proportion of the current herd has been tested and so far, there has been no detection *Neospora caninum* antibodies, Johnes antibodies, *Leptospira hardjo* antibodies, BVD antibodies or BVD antigen and as such there is a low probability that these diseases are circulating in this herd.

Antibodies to infectious bovine rhinotracheitis (IBR) were detected in one animal, and antibodies to *Mycoplasma bovis* were detected in 5 animals with suspect results in a further 8 animals (n tested = 19).

Contact between buffalo and other animals on the farm

The buffalo graze separately from all other animals on the farm with the sheep flock managed some miles away. Contact with other animals should be avoided where possible to minimize the risk of introducing an infection. Sheep, cattle and goats can both transmit infections to buffalo via nose-to-nose contact and in some cases by aerosols which can be carried by wind. **Sheep in particular should be managed as far away as possible from the buffalo (ideally greater than 3 miles!) to reduce the risk of the transmission of OvHv-2.** This risk is increased during periods of stress for the sheep e.g. lambing, weaning, shearing, dipping etc.

Added animals

When purchasing animals from sources with an unknown disease status:

6. **Check TB status of source herd before purchase and any appropriate testing has been carried out.**
7. **Isolate animals for one month in a separate field or pen. Adopt strict biosecurity measures for all personnel, feed and equipment.**
8. **Arrange for blood sampling of all purchased animals for MCF associated viruses, Johnes antibodies, BVD antigen, IBR antibodies, *Neospora* antibodies and *Leptospira* antibodies. In addition, sampling for trace elements may also be beneficial in order to consider whether the new animals require similar or different supplementation compared with that for the existing herd.**
9. **In addition, treatment for liver fluke and worms and external parasites should be considered (bearing in mind off-license treatment and appropriate milk/meat withdrawl periods), unless they have recently been treated prior to purchase.**

Infectious diseases

Malignant catarrhal fever

Malignant catarrhal fever (MCF) has so far not been detected in this herd. In the UK it is caused by herpesviruses of the *Macavirus* genus, namely ovine herpesvirus 2 (OvHV-2) and caprine herpesvirus 2 (CpHV-2), and whilst sheep are considered the main reservoir host, a large proportion of cattle can also carry the virus subclinically. Buffalo have been found to be very susceptible to infection and minimizing contact between buffalo and species potentially carrying and shedding these herpes viruses is essential in aiding control. Sheep in particular may shed large quantities of virus when under stress (e.g. lambing etc.) and the virus may infect buffalo some miles away via airborne aerosol carriage. Aerosols containing virus disperse in the air and spread out, and therefore **maintaining as great a distance as possible from the sheep** will reduce the risk of infection even if it is not possible to maintain optimal distances and eliminate all risk.

Through the EIP project a novel vaccine against MCF has been used in bison and this could be caried out in the buffalo if the disease risk situation changes. Currently the likelihood of disease incursion is considered to be low provided the biosecurity measures and current grazing practices remain the same, although it cannot fully be eliminated. In addition, the efficacy of this vaccine has not yet been fully established as it is very much still in the development stage and it has yet to be deployed in buffalo. If vaccination were to be considered, it would need to be carried out under Class 1 GMO Contained Use (GMO(CU)) regulations with approval from the Health and Safety Executive. In brief, this would require containment of the buffalo after vaccination. It would mean housing for one week and disposal of any milk during this period, together with other measures. I would be happy to help arrange this for you and detail the measures you would need to take if you wanted to investigate this possibility further.

After vaccination disease could still occur if the immunity of the buffalo were adversely affected in some way or if there were sufficient virus to 'overwhelm' any protective immunological response. Until it bears a UK license this live recombinant vaccine must be used under Class 1 GMO Contained Use (GMO(CU)) regulations. The containment use risk assessment and contained use forms are available if necessary.

Neospora caninum

This is a parasite shed by dogs, particularly ones that ingest fresh bovine tissues, with placentas and calves being a particular risk, hence farm dogs that have access to any carcasses/afterbirths and hounds fed on possibly occasionally undercooked offal represent a high risk as contaminators of silage, mixed rations in the barriers or simply of fields grazed, in this case by heifers particularly. Thus, prompt carcass disposal and sealed (dog proof) storage are essential.

Once animals are infected by ingesting the parasite it becomes dormant in muscle tissue until immune system changes induced by pregnancy cause its activation, multiplication and migration via the maternal blood to settle in the foetus which may either be killed and aborted immediately or after a period of mummification, or itself become latently infected until, in the case of females, they themselves become pregnant. Thus, vertical spread from mother to daughter is very effective. Spread is also possible from an infected dam to other calves via pooled colostrum. Direct horizontal transmission between animals, other than colostrum, is not possible.

The following actions should be considered:

4. **Imported cows/heifers could be Neospora carriers so blood test on entry (see added animals above).**
5. **Blood test any aborting buffalo in case of previously undetected infection.**
6. **Ensure that the risk of dogs contaminating pasture and feed are minimized by:**
    i. **Minimize the exposure of your own dogs to cleansings and afterbirths.**
    ii. **Prohibit visitors from allowing dogs to roam the yards/fields.**
    iii. **Consider the risks from 'foreign dogs' crossing your fields (the parasite can survive, at least low grade fermentation, hence silage can be contaminated in the field).**

Bovine viral diarrhoea (BVD) virus

Currently there is no evidence of BVD within the herd. As such, provided the buffalo can be kept completely separate from cattle, sheep and goats, the risk of infection can be managed. Contact with other species, particularly purchased cattle will greatly increase the risk of infection possibly resulting in abortion and immunosuppression.

The following actions should be considered:

5. **Imported buffalo could be BVD carriers so blood test on entry (see added animals above).**
6. **Blood test any aborting buffalo in case of previously undetected infection.**
7. **Have tissues from any aborted foetuses tested in case of previously undetected infection.**
8. **Consider vaccinating the herd for BVD if there are concerns about effectively managing disease incursion risks.**

Infectious bovine rhinotracheitis (IBR)

There is evidence that the herd has been exposed to IBR infection in the past, with 1/19 animals demonstrating antibodies to this virus in June 2020. However, there have been no recorded disease incidents indicating IBR infection. Given the risk of infection via wind-borne aerosols and recrudescence of latent infection within the herd, **vaccination of all buffalo with a live vaccine in Autumn could be considered.**

Leptospirosis

There is currently no evidence of exposure to leptospirosis within the herd. However, infection can spread from in contact cattle or sheep via shared grazing, water courses or contaminated feed.

The following actions should be considered:

5. **Imported buffalo could be carriers of leptospires so blood test on entry (see added animals above).**
6. **Blood test any aborting buffalo in case of previously undetected infection.**
7. **Have tissues from any aborted foetuses tested in case of previously undetected infection.**
8. **Consider vaccinating the herd for leptospirosis if there are concerns about effectively managing disease incursion risks.**

Johnes disease

There is currently no evidence of infection with Johnes disease (*Mycobacterium avium* subspecies *paratuberculosis* (MAP)) within the herd. Infection could be introduced via an infected animal, wildlife, infected watercourses, contaminated feed, contaminated equipment/housing etc.

The following actions should be considered:

4. **Imported buffalo could be carriers of the causative organism and this is the probably the most likely route of entry so blood test all animals on entry (see added animals above).**
5. **Blood and faecal test any buffalo showing symptoms of chronic weight loss, diarrhoea, ventral oedema or failure to thrive, in case of previously undetected infection.**
6. **Consider annual screening all buffalo over 2 years of age for Johnes antibodies in order to monitor for any undetected disease.**

Mycoplasma bovis

Recent testing revealed antibodies to *Mycoplasma bovis* were detected in 5 animals with suspect results in a further 8 animals (n tested = 19). However, there have been no recorded disease incidents. In cattle, this disease can cause respiratory problems in calves, mastitis in lactating cows and joint infections. We would expect similar issues in buffalo. Buffalo infected with MCF may develop secondary infection with *Mycoplasma bovis* as well, making the disease worse. Currently, we do not know how much of a specific problem it is causing or whether it is exacerbating disease.

The following actions should be considered:

3. **Test future cases of respiratory disease/mortality to determine the extent to which *Mycoplasma bovis* is contributing to the clinical signs.**
4. **If it becomes apparent that *Mycoplasma bovis* is contributing to the clinical picture, there is a new vaccine Myco-B we can try which may help reduce the clinical signs although I am unsure as to its efficacy in buffalo.**

Parasitic diseases

Liver fluke

Liver fluke infection can cause weight loss, anaemia, reduced milk yield and poor fertility and in growing animals can be fatal. **Faecal testing individual milking animals at drying off would be recommended to determine if liver fluke infection is present. In addition, pooled faecal testing of youngstock in the autumn and/or at housing is recommended.** Treatment can then be carried out on a case-by-case basis as necessary. Due to the seasonal nature of the fluke life cycle the exact treatments recommended will vary depending on the time of year, the previous grazing history and the type of animal e.g. youngstock or adult milking cow.

Parasitic gastroenteritis complex

So far this has caused very few issues, however, remain vigilant for signs of disease among growing animals. **Faecal testing the youngstock during the summer and autumn would be recommended to determine if a severe infection is present.** Treatment can then be carried out on a case-by-case basis as necesarry.

Lungworm

Lungworm has not caused any issues historically. Lungworm can be carried by earthworms and projected into the air by airborne fungal spores so biosecurity cannot be relied upon to keep pastures free. If necessary, a lungworm vaccine could be given to young stock prior to first grazing.

Skin parasites

Monitor the herd for signs of scratching and contact the vets to discuss if this is noted.

Other potential disease issues

There are many other diseases that could cause issues. However, so far they have not caused any problems or been detected. That said, they are worth being aware of and remaining vigilant for and appropriate actions can be carried out if and when necessary. A list of some possible issues with some advice is provided on the next page.

Other infectious or infective diseases to be considered		
Disease	Possible clinical effects	Control programme or Advice
Salmonella dublin	Scouring adult cows, particularly after stress periods such as calving. Abortions in late gestation. Vague illness in calves.	Have any scouring adults examined by vet +/or faecal culture. Send in faeces sample from any calves with bloody scour.
Salmonella typhimurium	Scouring adult cows, particularly after stress periods such as calving.	
Tuberculosis		The herd is currently clear of TB infection. Biosecurity procedures should prevent cattle to bison transmission. Long term the local disease situation will dictate the risk from wildlife with badger visitation of supplementary feeding areas possible, and avoidable only by badger fencing.
Respiratory Syncitial Virus (RSV)	These will manifest themselves as calf pneumonia – usually between 6 weeks and 12 months of age.	Test if pneumonia becomes a problem.
Parainfluenza 3 Virus (PI 3)		
Manhaemia spp.		
Haemophilus Somnus		
Rotavirus	These will manifest themselves as calf diarrhea – usually between 0 and 3 weeks of age.	Unlikely to be an issue with managing these animals outside, however if necessary send in faeces sample from 2nd scouring calf in a batch.
Coronavirus		
E. Coli K99		
Cryptosporidium parvum		

Coccidiosis	Common problem from faecal contamination of feed and water troughs and from calves eating straw bedding. Manifests itself as scour, illthrift and in severe cases bloody diarrhea with severe straining in calves over 3 weeks old.	Minimise faecal ingestion. Veterinary examination and testing of scouring calves over 3 weeks old.
Clostridial diseases	Sudden deaths, usually in bison at grass, although younger calves can be infected.	**Report all sudden deaths to vets.** **May be necessary to vaccinate calves if an issue, although two doses 4-6 weeks apart are required.**
Ringworm		Avoid borrowing handling equipment etc. Housing can harbor ringworm spores for several years if previously contaminated by cattle. A vaccine is available if necessary.

Nutrition

Currently the buffalo are grazed on good quality grass pastures, with supplementary silage offered in the winter and concentrate feed supplied to the milking animals. This approach should generally meet the nutritional needs of the herd without additional feeding. However, some trace element deficiencies have been identified although so far have not been noted to be involved in any observed disease processes.

It would be worth obtaining grass and silage samples periodically to ascertain the trace element status of the farm and to gradually build a picture of the farm over time in order to determine what the available forage can supply to the grazing herd. Sometimes testing can be carried out subsidised by some of the mineral companies.

Selenium

Based on the recent blood values, the plasma selenium sits on average in the sub-optimal range although two individuals were within the optimal range. It is possible that the buffalo may benefit from some selenium supplementation, however if they are being fed concentrate feed i.e. the milking animals, then it is likely they will be obtaining sufficient selenium that way and any more could result in over supplementation with a risk of toxicity. For those animals just on forage with no concentrate feeding, some selenium supplementation is likely to be beneficial and this could be supplied in a slow-release bolus or via a drench given every 3 months. If you were to do this, monitoring their response would be advised to detect whether an oversupply occurs. **Some supplementation may be beneficial.**

Copper

Seven individuals had plasma copper concentrations just below the normal range and the average was well within the normal range. As such, there was no evidence that the buffalo need any additional copper and supplementation could result in toxicity. **No supplementation is required.**

Zinc

Nine individuals had plasma zinc concentrations just below the normal range although the average sits within the normal range. A such, the buffalo may benefit from additional zinc but the benefits seen may be minimal. **Some supplementation may be beneficial.**

Cobalt

Cobalt is on average marginally low. For those animals grazing outside this is likely to cause very little problem as we would expect the cobalt concentration of the forage to increase over the winter period as a result of the expected increase in rainfall. In addition, for any cattle being fed concentrate feed then they are also likely to be receiving enough cobalt via this route. **No supplementation is required.**

Iodine

Two of the pooled results are just below the normal range (<100) and one is within the normal range (>100). Again, it is unlikely that supplementation is likely to be beneficial unless problems are observed. **No supplementation is required.**

Note

Trace element management **testing** is just one part of assessing whether trace elements are needed. The interpretation is supplied based on the current knowledge of the farm history particularly with regards to production and health, and this of course may change and vary over time. There is always the risk of over supplying animals with trace elements so where values are only marginally outwith the normal or expected range any real improvements may not be seen and there is a risk of toxicity as well.

Actions to be considered:

5. **Plan to take blood samples from 8 individuals annually to monitor the trace element situation. Include tests for copper, selenium, cobalt, zinc and iodine.**
6. **Plan to have liver samples stored/analysed for trace elements from fallen stock or slaughter animals. This can give a longer-term indication of trace-element nutritional assimilation allowing more fine tuning to take place.**
7. **Plan to assess the trace element composition of the available forage on a yearly basis.**