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# Losses and grass silage quality in bunker silos compacted by tractor versus wheel loader



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#### ABSTRACT

The study aimed to explore whether an increase in bunker silage density obtained by turning to a heavier packing machine than a farm size tractor would reduce losses and improve grass silage quality and aerobic stability. At each of three harvests, two bunkers were packed with either a 14.5 t wheel loader (WL) or an 8.3 t tractor (T). For comparison with the bunker silages, silage was produced simultaneously in round bales with high and low chamber pressure and wrapped immediately or after delay, and in laboratory silos.

Compaction with WL increased silage dry matter (DM) density by 9 % compared with T, from 204 to 222 kg DM/m³. On average for three harvests, DM recovered as silage, or lost, was almost identical for the two packing treatments, with 870 g/kg of harvested DM recovered as feed offered to animals, 55 g/kg as wasted silage, and 75 g/kg as invisible losses due to respiration, effluent, fermentation and aerobic deterioration. However, in the harvest with lowest crop DM content, 266 g/kg, invisible DM losses with WL exceeded losses with T by 46 g/kg, of which the main portion was assumed to be caused by more effluent squeezed out by the WL. In the harvest with highest crop DM, 332 g/kg, invisible DM losses with T exceeded losses with WL by 43 g/kg, of which the main portion was assumed to be caused by poorer compaction with T, and therefore higher respiration and aerobic deterioration losses. Wasted silage DM was lower in bales than in bunkers (P=0.004). The proportion of offered silage DM from poorly compacted bales sealed after delay (867 g/kg) was similar to that of bunkers, whereas the proportion of offered silage DM from well compacted and immediately sealed bales (963 g/kg) was similar to that of laboratory silos.

Significant increases in protein bound in the neutral detergent and acid detergent fiber fractions were found in bales sealed after delay where temperatures had rised to 47 °C at wrapping. Similar levels of fiber bound protein were found in bunker silage, suggesting that they were also heated during filling. Spot samples from bunker silo shoulders were more infected by yeasts, moulds and *Clostridium tyrobutyricum* than samples from mid in bunkers and from bales. No

Abbreviations: AAT, amino acids absorbed in the intestine; ADF, acid detergent fiber; ADIP, acid detergent insoluble protein; ADL, acid detergent lignin; CP, crude protein; DM, dry matter; DOMD, digestible organic matter in dry matter; H, harvest number; IP, buffer-insoluble protein; LA, lactic acid; ME, metabolizable energy; NDF, neutral detergent fiber; NDIP, neutral detergent insoluble protein; NE, net energy; NE<sub>L</sub>, net energy lactation; NH<sub>3</sub>-N, ammonia-nitrogen; OM, organic matter; OMD, organic matter digestibility; PBV, protein balance in the rumen; SDMI, silage dry matter intake; SEM, standard error of the mean; T, tractor; TP, true protein; VFA, volatile fatty acids; VOS, digestibility of organic matter in rumen fluid *in vitro*; WL, wheel loader; WSC, water soluble carbohydrates

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differences in losses, silage composition or aerobic stability were observed between bunker silo packing with WL or T on average over three harvests.

#### 1. Introduction

The compaction of bunker silos is often found to be poor due to high initial crop layer thickness, low packing vehicle weight or insufficient packing time per tonne crop (Muck and Holmes, 2000). Because the plastic top covering of silos seldom is completely air tight, a highly compacted silage mass is necessary to maintain sufficiently anaerobic conditions. Large variation in silage density has been found within bunker silos, from 133 to 269 kg dry matter (DM)/m³ (Craig et al., 2009), and from 173 to 229 kg DM/m³ (Spiekers et al., 2009), with the highest values in the central bottom and lowest values in the top and side layers. According to Savoie and Jofriet (2003), losses in bunker silos are mainly caused by aerobic conditions during filling, storage and feed out, and to a lesser extent caused by anaerobic fermentation and release of effluent. In a replicated experiment with alfalfa, Muck et al. (2015) found higher losses and poorer silage quality in bunker silos than in bag and tower silos. Bunker silo DM losses were found to be 15.6 and 18.2 % for year 1 and 2, respectively, which was achieved with good silo management, good to excellent densities (296 and 293 kg DM/m³) and at recommended or higher feed out rates, but with evidence of beginning clostridial fermentation in year 2. Spörndly and Nylund (2017) found on average 14.1 % DM losses from farm bunker silos, including 3.4 % spoiled silage, whereas DM losses in round bales were found to be 1.1 %, with no spoiled silage. They explained the lower losses from silage stored in bales to be associated with higher DM concentrations and tighter plastic sealing compared to silage in bunker silos. In addition, the time of exposure to air between opening bales and offering silage to animals was less than that for silage fed from bunkers.

Despite the challenges with losses and risks for poorer quality outlined above, many farmers with large herd sizes prefer preservation in bunkers to round bales because of lower costs. Gjestang et al. (2004) found that for Norwegian dairy herds with more than 40 cows, total annual costs per feed unit, including silo investment and annual working expenses were lower with bunkers than bales. Cost-efficient improvements in bunker ensiling technique would thus be a welcomed achievement. The present study aimed to explore the potential in increasing density of packed grass and clover crops harvested at low to medium DM concentrations. To the authors knowledge, farm tractor and wheel loader have previously not been compared for compaction in bunker silos. The following hypothesis was tested: The increase in silage density obtained by turning from compaction with tractors to heavier wheel loaders will reduce losses and improve grass silage quality and aerobic stability.

## 2. Material and methods

# 2.1. Experimental design

The study was conducted at the Animal Production Experimental Centre at the University of Life Sciences, Ås, Norway ( $59^{\circ}40^{\circ}N$ ,  $10^{\circ}47^{\circ}E$ ; elevation 93 m.a.s.l.). In each of three grass crop harvests, primary growth (H1; June 5–6), first regrowth (H2; July 19–20), and second regrowth (H3; September 11–12) in 2016, two identical bunker silos,  $6 \text{ m} \times 27 \text{ m}$  with three 3.5 m high walls, without roof, and with maximum capacity of 300 tonnes fresh crop weight, were filled simultaneously. One silo was compacted by tractor (T) and one by wheel loader (WL). At each of three time points within a harvest, on average 2.5, 17.3 and 21.4 h after starting bunker silo filling, four round bales were produced from the same sward. Two of these were highly compacted and two were poorly compacted. Of the two bales with the same compaction, one bale was sealed immediately and one after delay until covering of the bunkers. The delays lasted between 5 and 32 h for single bales, on average 14, 29 and 16 h for H1, H2 and H3, respectively. At the same three time points within a harvest, crop harvested for the bunkers was filled into four laboratory scale silos, of which two were sealed immediately and two after delay until covering of the bunkers. The delays lasted between 5 and 30 h for single silos, on average 15, 23 and 15 h for H1, H2 and H3, respectively. In total, the study comprised 6 bunker silos, 36 round bales, and 36 laboratory scale silos.

# 2.2. Harvested crops

During each harvest, timothy-dominated grass crops from seven to nine fields were harvested. Botanical composition of each field was roughly estimated by visual inspection prior to mowing. Based on weighted averages of harvested crop dry matter (DM) from each field, botanical composition at H1, H2 and H3 was 79 %, 77 %, and 61 % timothy (*Phleum pratense*), 13 %, 19 %, and 22 % meadow fescue (*Festuca pratensis*), 4 %, 1 %, and 13 % red and white clover (*Trifolium pratense* and *Trifolium repens*), and 4 %, 3 %, and 5 % of other grass species, forbs and weeds.

# 2.3. Harvesting procedure for bunkers

#### 2.3.1. Mowing, wilting and crop transportation

Crops were mown with three mower aggregates placed in butterfly position: A Kverneland Taarup 5087 M (Kverneland Group, Klepp, Norway) without conditioner on each side of the tractor, and a Kverneland Taarup 3632 FT with conditioner in front. Following wilting to target DM concentration 250 – 300 g/kg, the three windrows were joined to one using Kverneland Taarup 9590 C Hydro rake with TerraLink Quattro Ground Contour System. Weather conditions were good during all three harvests, sunny and

with no precipitation, but with some morning dew in H3. Mean 24 h air temperatures were 16, 18 and 15 °C, for H1, H2 and H3, respectively, and respective maximum day temperatures were 20, 23 and 20 °C. Measured average wilting time in H1, H2 and H3, respectively, were 8, 11 and 30 h before raking and 5, 3 and 12 h following raking. The composite windrows were collected with a 1.8 m wide pick-up on Lely Storm 130 P precision chopper (Lely Industries, Maassluis, The Netherlands), pulled behind a JD 6175 R tractor (Deere & Company, Moline, IL, USA). The flywheel chopping system with 10 blades and 5 blowing paddles gave an expected chop length of 12 – 44 mm. GrasAAT Plus silage additive (per kg: 440 g formic acid (FA), 204 g sodium formate, 120 g propionic acid (PA), 15 g benzoic acid; Addcon Nordic, Porsgrunn, Norway), was applied at the harvester at target dosage 4 L/tonne using Cliptonpumpen (Clipton, Vejbystrand, Sweden). Two Metsjø (25 m³) and one Palmse (30 m³) trailers transported the wilted crop to the silos. All transport tractors with trailers were weighed empty (tare) trice daily, and gross weight was recorded immediately before trailers reversed towards the bunker and emptied the grass load. Every second load was filled into each of two identical bunker silos.

#### 2.3.2. Compaction procedures

One bunker silo was compacted by a John Deere 6530 Premium farm tractor (Deere & Company, Moline, IL, USA) with a 1.90 m wide Norje N106 stone fork in front (Norjes Smidesfabrik, Sölvesborg, Sweden), and weight behind, giving a total weight of 8.3 t. All four tractor wheels were equipped with Michelin Multibib radial tyres (Michelin Multibib, Clermont-Ferrand, France), in front 48 cm wide (480/65 R28) and in rear 60 cm wide (600/65 R38), with 150 kilopascal (kPa) air pressure. The other silo was compacted by a Volvo L90H wheel loader (Volvo Construction Equipment, Gothenburg, Sweden) with a 2.60 m wide Norje N985 silogrip in front, total weight 14.5 t. All four wheels were identical, with 52 cm wide Trelleborg C-800 L2 tyres, 20.5 R25 (Trelleborg AB, Trelleborg, Sweden), with 350 kPa air pressure. Simulation of downward axle pressure exerted by the two compaction machines was adapted using Terranimo (2018). Wheels of T, in front and back axles, respectively, had 0.266 and 0.375 m² contact face, exerted an average downward pressure of 77 and 55 kPa, and a maximum downward pressure of 178 and 125 kPa. Wheels of WL, in front and back axles, respectively, had 0.310 and 0.311 m² contact face, exerted an average downward pressure of 272 and 279 kPa.

The compaction machines distributed each trailer load to a thin layer to cover at least half of the total 162 m<sup>2</sup> silo area, producing a slope such that compaction machines always were able to drive over the entire crop surface. Silos were continuously compacted until next trailer load for the actual silo appeared, approximately 20 min. Harvesting for each pair of bunkers lasted two days. A thin plastic layer (0.04 mm) was placed over the crop surface overnight.

#### 2.3.3. Covering of bunkers

On the second day, a thick, black plastic (0.150 mm, Polydress Texaleen Alpha Plus; RKW Agri GmbH & Co. KG, Michelstadt, Germany) with oxygen permeability  $180 \text{ cm}^3/\text{m}^2$  in 24 h was hung over side- and end walls of the silos and fastened into the crop. When the last trailer load was filled in each silo, compaction machines worked 30 min extra. Thereafter a thin 0.04 mm plastic layer with oxygen permeability  $940 \text{ cm}^3/\text{m}^2$  in 24 h was placed over the entire crop surface and manually fastened in the slot between the crop and the thick plastic hanging on the walls. The plastic from the wall was then turned down to cover the major part of the surface, before another thick 0.150 mm black plastic of the same quality was placed over the entire surface. Total plastic consumption for bunker silos was estimated to be 0.39 g/kg crop. Old, empty tyres were manually carried into the silos, and placed side by side on the entire top surface to keep plastic down.

Immediately following covering, the height from top of crop surface to top of silo wall was measured with one meter intervals along the two 27 m side walls. This formed the basis for calculation of the volume of ensiled grass crop. The same measurements were done immediately before bunker silos were opened for feeding.

#### 2.4. Ensiling in round bales

At three time points during each harvest, four round bales were produced from windrows close to those simultaneously picked up for bunkers. The combined baler and wrapper, Orkel hiQ Smartbaler (Orkel, Fannrem, Norway) with 20 fixed knives, giving 52 mm theoretical chop length was used. The same silage additive, at the same target dosage as for bunkers, was applied through nozzles leading the additive directly into the bale chamber using Prodevice PDH 10 pump (Agronic, Haapavesi, Finland), regulated by Orkel steering box. Applied additive in total for four bales was manually measured at the 200 L drum attached to the tractor. Although the intention was to ensile at the same DM concentration and apply the same amount of additive for all silo types, this was unfortunately not fully obtained. Measured application rates in H1, H2, and H3, respectively, were 3.1, 3.2 and 4.1 L/t for bunkers including laboratory silos, and 3.4, 5.6 and 3.3 L/t for bales. Two bales were compacted as hard as possible by choosing maximum chamber pressure (100 %) which equals 16,000 kPa, whereas two bales were poorly compacted, using 10 % chamber pressure which equals 1600 kPa. One bale from each compaction level was immediately wrapped using inner plastic TrioBale Compressor Mantel film, 1.40 m wide, Triowrap (Trioplast, Smålandsstenar, Sweden) (H1) or net (H2 and H3), plus 8 layers of 0.75 m wide and 0.025 mm thick white Triowrap 750 plastic film. The other bale from each compaction level was applied inner plastic (H1) or net (H2 and H3), only, before all four bales were transported to the experimental site using Silagrip 2 UM-7800 (UM Underhaug, Nærbø, Norway) and weighed. Core samples for DM determination were taken from the two unwrapped bales and pooled, intended to represent all four bales harvested at the same time point. Immediately afterwards, temperature sensors were placed in each unwrapped bale some 30-40 cm from bale surface, to monitor temperature development. When bunker silos were covered, the six round bales (two bales from each of three time points) that had been stored unwrapped on average for 20 h, were reweighed and thereafter wrapped with 8 layers of plastic. For the eight layers of plastic wrapped on each bale, 2.11 g/kg crop was used. Net or inner plastic will add to the plastic consumption in bales.

#### 2.5. Ensiling in laboratory silos

At the same three time points during harvesting as for round bale production, 5 kg fresh crop harvested by the precision chopper for bunker silos, and therefore already applied silage additive, was withdrawn for ensiling in laboratory silos. Four portions, each of approximately 1 kg, were filled into plastic bags and weighed. Two bags were sealed immediately using Magic Vac Maxima vacuum system (Flaem Nuova S.p.A., Brescia, Italy) and two bags were fitted with temperature sensors and stored outdoors, on average for 18 h, until bunkers were sealed. Then all six unsealed bags were reweighed and sealed. Laboratory silo bags were stored outdoors in a non-insulated container to obtain the same storage temperature as in bunkers and bales.

#### 2.6. Fresh crop samples

Six to ten fresh crop portions were grabbed by hand from every trailer load entering the bunker silos. Within trailer load, these were mixed and used for two samples of 500 and 200 g, respectively. The 500 g portion was stored at 4  $^{\circ}$ C for 0–2 days, when it was dried at 100  $^{\circ}$ C to constant weight for DM determination. These 278 single trailer load samples included the 9 loads from which crop for laboratory silos was collected, and in addition, 9 core samples from round bales, 3 from each harvest, were dried at 100  $^{\circ}$ C to constant weight. The 200 g portions were stored at -20  $^{\circ}$ C, and later pooled to 6 or 7 samples per harvest, in total 19 samples, that were freeze dried for chemical analyses.

#### 2.7. Opening, unloading and sampling of bunkers

Following 136 days of storage, the two H1 bunker silos were opened on October 20, 2016, and grass silage offered to the university herd until February 27, 2017, in total during 130 days. The H2 bunkers were opened after 217 days of storage, and used from February 22 to June 8, 2017, during 106 days. Due to excess amount of forage at the university farm, the H3 bunkers were stored through the following summer, in total for 383 days, and fed from October 1, 2017 to February 27, 2018, during 149 days.

Silage was unloaded 3–4 days a week using Triolet TU 180 XL silo block cutter (Triolet Mullos BV, Oldenzaal, The Netherlands) mounted in front of a wheel loader. In advance, tyres on the top surface were manually removed, whereas the block cutter shore off plastic top film in the one meter depth where a silage block was removed. Silage blocks from each silo were weighed, and samples from the removed blocks, in total 1.5 – 2 kg were stored at -20 °C. Moulded or deteriorated silage that could not be fed to cattle was weighed separately, or the proportion of a weighed block that had to be wasted was visually estimated. The term "offered silage" is used for silage given to livestock as feed. Average air temperatures during unloading were -0.1 °C, 6.1 °C and 0.1 °C, and the average daily removal of silage from the face was 20, 26 and 18 cm, in H1, H2 and H3, respectively. Total precipitation from silo filling and until completed unloading was 596, 611 and 1186 mm, giving a maximum of 85, 92 and 178 tonnes of water into silage in each bunker.

The frozen silage samples were partly thawed, chopped, and pooled for various purposes. For DM determination, 22–30 pooled samples per silo (250 g-samples in duplicate), each representing 7–12 tonne fresh silage, were dried at 103 °C, weighed warm, and corrected for volatiles (see below). For other analyses, silage samples were pooled to 6 samples per bunker. Each of these 6 composite samples were shared into 3 portions that were later used for (1) analysis of fermentation quality, (2) aerobic stability test, (3) freeze dried for chemical analyses.

# 2.8. Opening and sampling of round bales and laboratory silos

At three time points during unloading of a pair of bunker silos, four round bales and four laboratory scale silos were opened. This was done in the opposite order compared to the order of ensiling, to ensure similar storage time for silage from bunkers, bales and laboratory silos.

Round bales were weighed, and bale surfaces inspected for moulded or deteriorated silage that was manually removed and weighed. Several core samples were taken and mixed to a silage sample of at least 1.8 kg from each bale.

Of the four laboratory scale silos opened at the same time, the two bags that had been immediately sealed, and the two bags subjected to delayed sealing, were parallels from which silage was pooled to a composite sample.

Samples from each bale, and each pair of laboratory silos, were shared into 4 portions that were later used for (1) analysis of fermentation quality, (2) aerobic stability test, (3) freeze dried for chemical analyses and (4) heat dried at 103 °C and corrected for volatiles for DM determination.

#### 2.9. Spot samples from bunker silo faces and round bales for microbiological analyses

At the same three time points during unloading of bunkers, when round bales and laboratory silos were opened, core samples from four spots: side, shoulder, top and mid, of the face of bunker silos and from two spots in bales: surface and mid, were taken. Spots from the bunker silo face were: (1) mid: at least 0.5 m from top surface and at least 1 m from side walls, (2) side: 0.1-0.5 m from the silo wall and at least 0.5 m from top surface; (3) top: 0.1-0.5 m from top surface and at least 1 m from silo wall; (4) shoulder: 0.1-0.5 m from the silo wall and 0.1-0.5 m from top surface. Cores from the right and left side of the silo face were

pooled for side and shoulder samples. Spots from bales: surface: the 10-15 cm outermost part; mid: 15-45 cm from surface. The corer was washed in Antibac (ethanol, isopropanol, n-propanol, water) between each sample. Samples were stored at -20  $^{\circ}$ C until analysis.

#### 2.10. Analytical procedures

#### 2.10.1. Chemical analyses

Silage samples oven dried at 103 °C to constant weight were corrected for volatiles according to NorFor (Åkerlind et al., 2011) and used to calculate the portion of crop DM that was recovered as silage or lost.

Silage samples kept undried were analysed for pH,  $NH_3$ -N, organic acids and ethanol. Methods used for undried samples from H1 and H2 were as described by Randby et al. (2010), whereas from H3, methods were modified for organic acids and ethanol: Silage samples were diluted with demineralized water and stored frozen, then thawed, filtered, diluted and subjected to a clean-up procedure with activated carbon. For lactic acid (LA) and FA the extract was analysed by high pressure liquid chromatography (HPLC) using a separation power column based on polarity, charge and particle size, at 45 °C (mobile phase,  $0.0012 \text{ M } H_2SO_4$  at 0.6 mL/min) with a UV spectrophotometric detector. For acetic acid (AA), PA, and butyric acid (BA), and ethanol, the extract was acidified and analysed by gas chromatography (GC) after adding internal standards. Carrier gas was helium with constant flow 1.5 mL/min, and temperature gradient 50-250 °C. A flame ionization (FI) detector was used, and quantification was done by comparison with internal standards.

Freeze dried fresh crop and silage samples, were equilibrated to room humidity, and milled to pass a 1.0-mm screen (Retsch SM200 cutting mill (Retsch GmbH, Haan, Germany)) prior to analyses of DM (103 °C for 4 h), ash (550 °C for 4 h), water soluble carbohydrates (WSC), crude protein (CP), true protein (TP), buffer-insoluble protein (IP), neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), neutral detergent insoluble protein (NDIP), acid detergent insoluble protein (ADIP) and digestibility of organic matter in rumen fluid *in vitro* (VOS). Water soluble carbohydrates were analysed by an enzymatic method as described by Randby et al. (2015). A Fibertec 2010 (Foss, Hillerød, Denmark) fiber analyser was used to determine NDF according to Mertens et al. (2002) using a heat stable alpha amylase and ash correction but without sodium sulphite (aNDFom). Acid detergent fiber was determined using Fibertec 8000 (Foss, Hillerød, Denmark) according to Method 973.18 (AOAC, 2000) and was corrected for ash (ADFom). Acid detergent lignin was determined by the method of Van Soest et al. (1991) using sulphuric acid and corrected for ash. The Dumas combustion method, using LECO TruMacN (Leco Corporation, St Joseph, MI, USA) was used for N analysis in freeze dried samples (CP calculated as N × 6.25), and for NDF-N and ADF-N. Buffer-insoluble protein and TP were analysed according to Licitra et al. (1996), using tungistic acid (Na<sub>2</sub>WO<sub>4</sub>) for TP. Analyses of VOS were done as described by Åkerlind et al. (2011).

# 2.10.2. Microbiological analyses of spot silage samples

For all microbiological analyses, 30 g sample and 270 mL of sterilised, quarter-strength Ringer solution (Merck) were homogenized in a Seward stomacher for 2 min. at normal intensity.

For yeast and mould analyses, 0.1 mL of each of three serial dilutions were surface spread on two malt extract agar (MEA) plates supplemented with 10 % LA. Plates were cultured aerobically for 3–4 days at 25 °C. Colonies were distinguished visually or with the help of microscope.

For analyses of *Clostridium* spores, approximately 10 mL of the homogenized initial sample dilution was exposed to heat treatment for 13 min in water bath at 80 °C, and two following dilutions were prepared from that dilution. From each dilution, 0.1 mL was surface spread on two plates with Reinforced *Clostridium* Agar (RCA; Merck) with addition of neutral red (Sigma) and cycloserine (Sigma). Plates were enumerated after 7 days of anaerobic cultivation at 37 °C. Lactate dehydrogenase (LDH) activity was tested on 3 visually similar colonies per sample as described by Jonsson (1990). Colonies positive in LDH activity were considered *Clostridium tyrobutyricum*, whereas other colonies on the same plates were considered other Clostridia species. Detection level for all microbial analyses were 50 colony forming units (cfu)/g, i.e. 1.70 log cfu/g.

#### 2.10.3. Aerobic stability test

Silage samples of 700 g were placed in perforated plastic bags in perforated polystyrene boxes at 20  $^{\circ}$ C. The temperature of the silages and the ambient temperature were logged every 4 h for 30 days. Silages were regarded stable until silage temperature reached 2  $^{\circ}$ C above ambient temperature. Because room temperature sensing was done by a sensor placed 1.5 m above the floor, room temperature at the floor, where silage samples were monitored, were only 17.5  $^{\circ}$ C for samples from H1. Room sensor was lowered to floor position for H2 and H3, and ambient temperature 20  $^{\circ}$ C was obtained.

#### 2.11. Calculations

Silage *in vivo* organic matter (OM) digestibility (OMD) was calculated according to Lindgren (1983), using the equation OMD % =  $-2.0 + 0.90 \times \text{VOS}$ , where VOS is the digestibility of organic matter in rumen fluid *in vitro*. Silage concentrations of metabolizable energy (ME) and net energy lactation (NE<sub>L</sub>) per kg DM were calculated according to Van Es (1978) using the equations MJ ME =  $15.1 \times \text{DOMD}$ , and MJ NE<sub>L</sub> =  $0.6 \times (1 + 0.004 \times ((Q \times 100)-57)) \times \text{ME} \times 0.9752$ , where DOMD = digestible OM in DM, Q = ME/gross energy (GE), and GE =  $18.4 \times \text{MJ/kg}$  DM. Silage concentrations of metabolizable protein expressed as amino acids absorbed in the intestine (AAT), and protein balance in the rumen (PBV), were calculated according to Madsen et al. (1995) using analysed CP values and the ME values based on feed analyses. Digestible carbohydrates in silages were calculated according to Spörndly (2003),

where carbohydrates, g/kg DM =  $922.0 \times (1.46 \times \text{CP})$ , and carbohydrate digestibility =  $(31.4 + (3.89 \times \text{ME}))/100$ . Constant factors of 0.80 for rumen protein degradation and 0.82 for intestinal digestibility of undegradable amino acids in silages were used (Spörndly, 2003). Silage DM intake (SDMI) index was calculated according to Huhtanen et al. (2007). Analytical results of the feed protein fractions TP, IP, NDIP and ADIP are presented as A, B1, B2, B3 and C, with decreasing solubility and rate of enzyme degradability according to Van Soest (1994): Protein fraction A = Non protein nitrogen (NPN, calculated as CP-TP), B1 = True buffer soluble protein (BSP, calculated as TP-IP), B2 = Neutral detergent soluble protein, calculated as IP-NDIP, B3 = Neutral detergent insoluble protein (calculated as NDIP-ADIP (insoluble in neutral detergent but soluble in acid detergent)), C = Acid detergent insoluble protein (ADIP).

#### 2.12. Statistical analyses

All data was analysed using SAS (release 9.4, 2002–2012; SAS Institute inc., Cary, NC, USA). Fresh crop and silage weights, densities, losses, and weighted averages of bunker silage chemical composition were analysed using the PROC MIXED procedure by the model:  $Y_{ik} = \mu + S_i + H_k + e_{ik}$ , where  $\mu$  = general mean,  $S_i$  = the effect of bunker silo compaction i,  $H_k$  = effect of harvest k, and  $e_{ik}$  is the random residual error. All results were presented as least square (LS) means. Due to only 6 observations, the silo compaction  $\times$  harvest interaction could not be estimated (Tables 2–4).

Round bale weights at ensiling and opening, and densities, were analysed using the PROC MIXED procedure by the model:  $Y_{ijk} = \mu + S_i + D_j + H_k + S_i \times H_k + D_j \times H_k + S_i \times D_j + e_{ijk}$ , where  $\mu = \text{general mean}$ ,  $S_i = \text{effect of compaction } i$ ,  $D_j = \text{effect of sealing time } j$ ,  $H_k = \text{effect of harvest } k$ ,  $S_i \times H_k$ ,  $D_j \times H_k$ ,  $S_i \times D_j$  are interactions, and  $e_{ijk}$  is the random residual error. The RANDOM statement was included for the nine time points, three at each harvest, when round bales and laboratory silos were produced (Table 5).

Losses, silage chemical composition and aerobic stability from bunkers, bales and laboratory silos were analysed using the PROC MIXED procedure by the model:  $Y_{ik} = \mu + S_i + H_k + S_i \times H_k + e_{ik}$ , where  $\mu =$  general mean,  $S_i =$  silo treatment i, i = 1–8,  $H_k =$  effect of harvest k,  $S_i \times H_k =$  the effect of interaction, and  $e_{ik}$  is the random residual error. The RANDOM statement was included for the nine time points when round bales and laboratory silos were sampled. Of the six analysed samples from each bunker silo, two samples were assigned each of the three time points within harvest. Treatment means were separated using the PDIFF statement, and contrasts were estimated using the ESTIMATE statement. Silo treatment i: 1. Bunker compacted by tractor, 2. Bunker compacted by wheel loader, 3. Poorly compacted and immediately sealed bales, 4. Poorly compacted bales with delayed sealing, 5. Highly compacted and immediately sealed bales, 6. Highly compacted bales with delayed sealing. 7. Laboratory silos immediately sealed, 8. Laboratory silos with delayed sealing. The same model was also used to evaluate bales alone, i = 3–6 (Table 6–8).

The frequency of spot samples with detected microbial growth from the four points in bunker silo faces and from surface and mid in bales was analysed with Chi-square test using the PROC FREQ procedure. Additionally, the number of detected colonies per g sample in bunker silo faces, and surface and mid in round bales, was analysed using the PROC MIXED procedure by the model  $Y_{ikl} = \mu + S_i + H_k + P_l + S_i \times P_l + e_{ikl}$ , where  $\mu =$  general mean,  $S_i =$  the effect compaction i,  $H_k =$  effect of harvest k,  $P_l =$  point in bunker silo face or bale l,  $S_i \times P_l =$  the effect of interaction, and  $e_{ikl}$  is the random residual error. Points in bunker silo face or bale l: 1. Mid, 2. Side, 3. Top, 4. Shoulder, 5. Bale surface, 6. Bale mid (Table 9).

The frequency of spot samples with detected microbial growth from the three harvests, and from bunkers and round bales, was analysed with the Chi-square test using the PROC FREQ procedure. Additionally, the number of detected colonies per g sample in the three harvests and the six silo treatments was analysed using the PROC MIXED procedure by the model  $Y_{ik} = \mu + S_i + H_k + e_{ik}$ , where  $\mu$  = general mean,  $S_i$  = the effect silo treatment i, i = 1-6,  $H_k$  = effect of harvest k, and  $e_{ik}$  is the random residual error. Silo treatment i was as described for analysis of chemical composition. The same model was also used to evaluate bales alone, i = 3-6. The silo treatment  $\times$  harvest interaction was insignificant for all variables and therefore excluded. Values from bunkers were means over 4 points in the silo face, and values from bales were means over 2 points: surface and mid (Table 10).

Microbial counts were  $\log_{10}$  transformed. Negative results (below detection limit) were defined as half of the detection limit, i.e. 25 cfu/g = 1.40 log cfu/g (Vissers et al., 2007a; Gismervik et al., 2015). Results were considered statistically significant at P < 0.05, and P-values between 0.05 and 0.1 were considered to indicate trends.

Table 1

Chemical composition of fresh grass crops ensiled in bunkers during three harvests, as weighted averages of dry matter yields from all fields within harvest.

Harvest	N	DM, g/kg	g/kg	g DM	Pı	otein frac	ctions <sup>1</sup> , g	N/kg tot	al N		g/kg	DM			g/kg		Per kg	DM	
		g/ Kg	OM	СР	A	B1	B2	В3	С	WSC	NDF	ADF	ADL	OMD	DOMD	MJ ME	$\mathrm{MJ}\;\mathrm{NE}_\mathrm{L}$	g AAT	g PBV
1	6	284	931	121	306	30.3	406	217	40.8	87.6	627	358	36.4	0.674	627	9.7	5.58	68.9	2.2
2	7	266	921	149	244	47.6	409	263	36.0	97.8	564	318	28.3	0.740	682	10.7	6.28	74.1	20.4
3	6	332	917	142	285	57.1	326	276	55.4	95.1	597	332	32.4	0.711	652	10.2	5.91	71.4	18.8

<sup>&</sup>lt;sup>1</sup> Protein fractions: A = Non protein nitrogen (NPN, calculated as CP-TP), B1 = True buffer soluble protein (BSP, calculated as TP-IP), B2 = Neutral detergent soluble protein, calculated as IP-NDIP, B3 = Neutral detergent insoluble protein (calculated as NDIP-ADIP (insoluble in neutral detergent but soluble in acid detergent)), C = Acid detergent insoluble protein (ADIP).

#### 3. Results

#### 3.1. Bunker silo fresh crop composition, crop and silage weights, densities and losses

Weighted compositional averages of analyses of crop harvested for bunker silos indicated that the H1 crop was rather mature, with low OMD, DOMD, content of energy (ME and  $NE_L$ ) and protein (CP, AAT and PBV), and high content of NDF and ADF (Table 1). The H3 crop was intermediate, whereas H2 was of the highest nutritional value.

The highest fresh crop volume and weight was ensiled in each bunker in H2, with lower and similar amounts in H1 and H3 (Table 2). Because DM concentration was lowest in H2 and highest in H3, the amount of crop DM was only slightly higher in H2 than in H3, with less in H1. Density in kg/m³ was highest in H2, but on DM basis, density was highest in H3. Compaction by WL produced a higher fresh crop density than compaction by T directly following silo covering (P = 0.008). The same tendency was apparent on crop DM basis (P = 0.06). During the ensiling period, silage volumes shrank slightly more for silos compacted by T than WL, and less in H1 than in H2 and H3, which contributed to smaller, and not significant differences between T and WL in silage density in kg and kg DM at feeding, although the numeric differences were similar as for fresh crop.

In H1 and H3, the wet weight of silage removed from the bunker silos exceeded the amount of ensiled fresh crop (Table 2), giving negative values for invisible losses (Table 3), defined as the calculated difference between the amount of crop filled in the silos and the total amount of silage (offered plus wasted) removed. In this study, silage effluent is a portion of invisible losses, whereas absorbed rainwater decreases invisible losses on wet weight basis.

On wet weight basis, the proportion of silage that could be offered to cattle tended to be higher when crop was compacted by T than WL (P = 0.06). This was due to a lower amount (more negative) of invisible losses for T than for WL (P = 0.07). On DM basis, no differences in the proportion of offered silage, wasted silage or invisible losses were detected between silos compacted by T or WL.

#### 3.2. Bunker silage chemical composition

In all harvests, silage removed from silos was wetter than the fresh crop ensiled (Table 1 and 4). The difference in DM concentration was most pronounced for the most heavily wilted crop (H3), where it reached 49 g/kg. Numerically, OM concentrations of silage were slightly higher than in the parent crop, and CP concentrations were higher or similar. Fresh crop contained on average, per kg N, 277 g of fraction A, 679 g N of fraction B, and 44 g of fraction C. On average for silages, proportions per kg N had changed to 542 g of A, 412 g of B and 46 g of C fractions. There were no significant differences in silage composition and digestibility according to bunker silo compaction.

#### 3.3. Round bale weights and densities of fresh crop and silage. Temperatures and occurrence of mould in bales and laboratory silos

The crop preserved in round bales from H1, H2 and H3 was wilted to 257, 294 and 359 g DM/kg, respectively (Table 5). H1 bales contained less fresh crop in kg and kg DM, in total and per m³, compared with bales from H2 and H3. Although H2 bales were heaviest on fresh weight basis, H3 bales contained most DM. Similar differences between the three harvests were apparent for bale silage weights in kg and kg DM as for fresh crop.

Table 2

Effect of bunker silo compaction by tractor (T) or wheel loader (WL) in three harvests on fresh grass crop and silage weights, dry matter concentrations and densities.

					Fresh	n crop					Sil	age		
Harvest	Compaction	N	Volume m <sup>3</sup>	Kg	DM g/kg	Kg DM	Kg per m <sup>3</sup>	Kg DM per m <sup>3</sup>	Volume m <sup>3</sup>	Kg	DM g/kg	Kg DM	Kg per m <sup>3</sup>	Kg DM per m <sup>3</sup>
1	T	1	314	205600	288	59262	655	189	303	214720	267	57436	710	190
1	WL	1	308	218630	280	61113	711	199	298	221940	269	59681	745	200
2	T	1	388	293690	261	76521	757	197	346	289220	243	70420	837	204
2	WL	1	355	291990	272	79511	824	224	329	276300	252	69557	841	212
3	T	1	330	221100	338	74688	669	226	292	236460	278	65854	811	226
3	WL	1	305	227240	327	74395	746	244	265	240260	286	68815	907	260
1		2	311	212115	284	60188	683	194	300	218330	268	58559	727	195
2		2	371	292840	266	78016	790	211	337	282760	248	69988	839	208
3		2	317	224170	333	74542	708	235	278	238360	282	67335	859	243
SEM			6.93	3685	5.97	833	5.25	4.25	5.51	5339	1.89	1016	23.4	7.23
P			0.04	0.007	0.03	0.008	0.009	0.04	0.03	0.03	0.01	0.03	0.098	0.08
	T	3	344	240130	292	70157	694	204	313	246800	262	64570	786	206
	WL	3	322	245953	291	71673	760	222	297	246167	268	66018	831	224
	SEM		5.66	3009	4.87	681	4.29	3.47	4.50	4400	1.55	830	19.1	5.91
	P		0.12	0.30	0.74	0.26	0.008	0.06	0.12	0.93	0.10	0.34	0.24	0.17

**Table 3**Effect of bunker silo compaction by tractor (T) or wheel loader (WL) in three harvests on crop recovered as offered silage and wasted silage, and sum of invisible losses through respiration, effluent, fermentation and aerobic deterioration.

				g/kg crop			g/kg crop DM	
Harvest	Compaction	N	Offered silage	Wasted silage	Invisible losses	Offered silage	Wasted silage	Invisible losses
1	Т	1	961	83.1	-44.4	892	77.1	30.8
1	WL	1	952	63.0	-15.1	916	60.7	23.4
2	T	1	919	65.4	15.2	859	61.1	79.7
2	WL	1	894	52.1	53.7	827	48.1	125.2
3	T	1	1026	43.2	-69.5	846	35.6	118.3
3	WL	1	1007	50.8	-57.3	881	44.5	75.0
1		2	957	73.1	-29.8	904	68.9	27.1
2		2	907	58.8	34.5	843	54.6	102.5
3		2	1016	47.0	-63.4	863	40.1	96.7
SEM			4.07	7.22	6.67	18.0	6.87	22.3
P			0.006	0.23	0.02	0.25	0.18	0.22
	Т	3	969	63.9	-32.9	866	57.9	76.3
	WL	3	951	55.3	-6.2	874	51.1	74.5
	SEM		3.32	5.89	5.45	14.7	5.61	18.2
	P		0.06	0.41	0.07	0.72	0.48	0.95

Average initial temperatures in the six round bales fitted with temperature sensors at each harvest were 22.4, 25.3 and 24.6 °C for H1, H2 and H3, respectively. Immediately before wrapping, average temperatures were 31.2, 47.0 and 40.5 °C, for H1, H2 and H3, giving respective temperature increases of 8.8, 21.7 and 15.9 °C, and temperature increases per h of aerobic storage of 0.6, 0.8 and 1.2 °C. Temperatures increased consistently more in poorly compacted than in highly compacted bales, and reached on average 41.5 and 37.6 °C at wrapping, respectively, which was 18.2 and 12.7 °C above initial temperature (P = 0.06, not presented in Table).

During bale opening, some effluent was observed, but not weighed, in 4 bales from H1 and 4 bales from H2, where initial crop contained 210 and 245 g DM/kg, respectively. Moulded silage was wasted from 3, 6 and 10 of the 12 bales in H1, H2 and H3, respectively, which constituted respectively 0.1, 5.8 and 20.8 kg per bale.

Highly compacted bales contained in the range 32–38% more weight than poorly compacted bales measured in fresh crop and silage bale weights in kg and in kg DM, and in total and per  $m^3$  (P < 0.001). Compared with immediate sealing, bales subjected to delayed sealing had similar fresh crop weights, but lower silage weights in kg and kg per  $m^3$  (P = 0.04), and tended to have lower silage DM weights, in total and per  $m^3$  (P = 0.07).

Crop withdrawn from trailer loads for ensiling in laboratory silos contained on average 255, 291 and 405 g DM/kg in H1, H2 and H3, respectively (not presented in Table). Average initial temperatures in the six laboratory silos fitted with temperature sensors at each harvest were 23.7, 25.6 and 26.4  $^{\circ}$ C for H1, H2 and H3, respectively. Immediately before silo sealing, average temperatures were 25.7, 32.1 and 23.4  $^{\circ}$ C, for H1, H2 and H3, giving respective temperature increases of 1.9, 6.5 and -2.9  $^{\circ}$ C, and temperature increases per h of aerobic storage of 0.2, 0.3 and -0.3  $^{\circ}$ C. Temperatures in two laboratory silos prepared at 15:00 h during the first day in H1, with initial temperatures of 23.4 and 24.6  $^{\circ}$ C, were recorded to have 35.2 and 37.1  $^{\circ}$ C the following day at 13.00 h, however temperatures decreased by nearly 10  $^{\circ}$ C until silos were sealed 8.5 h later, at 21.30 h.

# 3.4. Harvested crop recovered as offered silage or wasted silage, and invisible losses in bunkers, round bales and laboratory silos

Laboratory silos and immediately sealed round bales had the highest proportions of offered silage on wet weight basis, on average 987 g/kg, that were significantly higher than in bales sealed after delay (Table 6). On DM basis, the proportion of offered silage from bunkers, on average 870 g/kg, was similar to that of poorly compacted bales sealed after delay, 867 g/kg, but contrasts (not presented in Table) revealed significantly lower proportion of offered silage from bunkers than from immediately sealed bales, 956 g/kg (P = 0.0496), and from laboratory silos, 963 g/kg (P = 0.03). Contrasts also showed that wasted silage from bunkers, on average 54.5 g/kg on DM basis, was significantly higher than the average from all bales (P < 0.001), and also from the poorly compacted bales sealed after delay (P = 0.02). No significant differences were found in invisible DM losses, but invisible DM losses tended to be higher from poorly compacted bales sealed after delay, 108 g/kg, than from highly compacted and immediately sealed bales, 25 g/kg (P = 0.07).

#### 3.5. Fermentation quality, chemical composition and aerobic stability of silage from bunkers, round bales and laboratory silos

Dry matter content, concentrations of BA in DM, and pH, NH<sub>3</sub>-N in N, and SDMI index were higher in silages from H3 than from H1 and H2 (Table 7). Concentrations of WSC were lower and of ethanol higher in H1 than in H2 and H3. The quantitatively most important fermentation acids, LA and AA, did not differ between harvests, however TA were higher in H1 than in H3. Formic acid was found in highest amount in H2 and lowest amount in H3, whereas propionic acid was found in higher amount in H3 and H2 than in

Effect of bunker silo compaction by tractor (T) or wheel loader (WL) in three harvests on chemical composition of silage as weighted averages of dry matter removed from each bunker.

	g AAT g PBV				69.5 32.6		9.0 19.5	67.6 11.7						68.7 21.7		
Per kg DM	MJ NE <sub>L</sub> g						5.92	5.60						5.86		
	MJ ME	6.7	9.7	10.6	10.4	10.2	10.2	9.7	10.5	10.2	0.058	0.02	10.2	10.1	0.047	0.42
g/kg	DOMD	629	627	229	999	650	653	628	671	652	3.82	0.03	652	648	3.12	0.49
	OMD	0.679	0.678	0.740	0.725	0.717	0.716	0.679	0.733	0.717	0.004	0.02	0.712	902.0	0.003	0.35
	ADL	41.5	39.1	35.9	36.5	41.5	41.6	40.3	36.2	41.6	0.80	0.08	39.6	39.1	99.0	09.0
g/kg DM	ADF	363	366	324	332	343	342	365	328	343	2.25	0.01	343	347	1.84	0.33
g/kg	NDF	605	289	531	543	529	551	262	537	540	9.82	0.08	222	561	8.04	0.65
	WSC	23.0	21.3	18.1	11.4	30.8	26.9	22.2	14.8	28.9	1.25	0.03	24.0	19.9	1.02	0.11
	С	46.6	45.2	42.2	43.5	57.6	55.0	45.9	42.9	56.3	1.00	0.02	48.8	47.9	0.82	0.52
Protein fractions $^2$ , g N/kg total N	B3	102	115	151	156	91	120	109	154	106	6.11	0.049	115	130	4.99	0.16
actions², g N	B2	255	247	249	252	314	316	251	251	315	3.04	0.007	273	272	2.48	0.80
Protein fr	B1	30.7	29.5	31.7	24.5	22.2	11.8	30.0	28.1	17.0	2.25	0.09	28.2	21.8	1.84	0.13
	Α	299	564	526	523	515	498	292	525	202	4.38	0.02	236	528	3.57	0.28
DM	CP	127	130	152	153	142	139	129	153	141	1.53	0.02	140	141	1.25	0.87
g/kg DM	ОМ	927	924	915	918	206	912	926	916	910	2.19	0.07	916	918	1.79	09.0
DM	g/kg	270	271	241	248	279	287	271	245	283	1.89	0.009	263	569	1.55	0.13
	$N^1$	1	1	1	1	1	1	7	7	7			က	က		
Compact.		H	WL	H	WL	H	ML						T	WL	SEM	Ь
Harvest		1	1	7	7	က	3	1	2	က	SEM	Ь				

1 Chemical composition based on weighted averages of 6 analysed silage samples from each bunker. Statistics based on the weighted yearly average from each bunker, i.e. 2 silos from each harvest, and 3 silos compacted by tractor and 3 silos compacted by wheel loader.

<sup>2</sup> Protein fractions: A = Non protein nitrogen (NPN, calculated as CP-TP), B1 = True buffer soluble protein (BSP, calculated as TP-IP), B2 = Neutral detergent soluble protein, calculated as IP-NDIP, B3 = Neutral detergent insoluble protein (calculated as NDIP-ADIP (insoluble in neutral detergent but soluble in acid detergent)), C = Acid detergent insoluble protein (ADIP). H1. Silage aerobic stability was considerably lower in H2 than in H1 and H3. Silages from H1 had higher proportion of protein fraction A than H2 and H3 silages (Table 8). Harvest 2 silage had highest and H1 silage lowest B3 fraction, whereas H3 silage had higher C fraction than H1 and H2 silages. Higher ADF concentration was found in H1 and H3 silage than in H2 silage, and ADL concentration was higher in H3 silage than in H1 and H2 silage. Harvest 2 silage contained highest and H1 silage lowest energy and protein values, shown by significant differences in DOMD, ME, NE and AAT.

Apart from bales that were poorly compacted and sealed after delay, silage from bales and laboratory silos had higher DM concentrations than bunkers (Table 7). Consistently, bales and laboratory silos had higher pH and WSC concentrations than bunkers, whereas treatment differences were not found for NH<sub>3</sub>-N. Contrasts (not presented in Table) showed that LA and TA concentrations were highest in bunkers, intermediate in laboratory silos and lowest in bales (P < 0.005) and that AA and BA concentrations were higher in bunkers than in laboratory silos and bales (P < 0.02). However, laboratory silos with delayed sealing did not differ significantly from bunkers in LA, AA and TA concentrations, and bales that were poorly compacted and sealed after delay, did not differ significantly from bunkers in AA concentration. Butyric acid was not detected in laboratory silos, but in 7 of 36 samples from round bales, and in 23 of 36 samples from bunkers. All 12 bunker silo samples from H3 contained BA, and all analyses showed low levels ( $\leq$  1.51 g BA/kg DM). Ethanol concentrations were lower in bunkers than in bales and laboratory silos (P < 0.001). Contrasts showed that SDMI indexes were in general higher in bales than in bunkers (P < 0.001), whereas laboratory silos were equal to bales if immediately sealed, and not different from bunkers if sealed after delay. Aerobic stability did not differ between silage treatments, however, when round bales were considered separately, delayed sealing tended to decrease aerobic stability (P = 0.006).

On N basis, laboratory silos with delayed sealing had less of protein fraction A than all other treatments (P < 0.001; Table 8), and contrasts (not presented in Table) showed that bales had higher A-fraction than bunkers and laboratory silos (P = 0.001). In bales, a profound effect was found of sealing time for all protein fractions. The most soluble fractions, A and B1, were found in highest proportions in immediately sealed bales (P < 0.01), whereas the less soluble fractions, B2, B3 and C, were found in highest proportions in bales sealed after delay (P < 0.001). Similar effects were found of sealing time in laboratory silos, but differences were significant only for fractions A and B3 (P < 0.001). For fractions A, B2, B3 and C, bunker silos differed significantly from immediately sealed bales, but did not differ from bales that were sealed after delay. For fraction B1 the same was true for bunker T. No effect of compaction was found in protein fractions, neither in bunkers nor in bales, apart from a difference between T and WL silage for fraction B1 (P = 0.047).

Immediately sealed bales and laboratory silos had lower NDF concentrations than the corresponding silages with delayed sealing (P < 0.001). Bunker silos did not differ from bales and laboratory silos with delayed sealing, but, with one exception (bunker T vs. immediately sealed poorly compacted bales), differed significantly from those sealed immediately.

Although variations in ADF concentrations were small and differences in general insignificant, contrasts showed that delayed sealing time in laboratory silos significantly increased ADF concentrations from 334 to 352 g/kg DM (P = 0.01), and that also bunker silo ADF concentration, 345 g/kg DM, was significantly higher than in immediately sealed laboratory silos (P = 0.04). No differences were found among silage treatments in ADL concentrations, nor in measures of digestibility nor in calculated energy and protein values.

# 3.6. Microbiological composition of spot silage samples from bunker silo faces and round bales

Yeast, mould and spores of *C. tyrobutyricum* were more frequently detected in bunker shoulder samples compared with mid, side, and top samples of bunkers, and surface and mid samples in bales (P = 0.001; Table 9). Also, a higher number of colonies of yeast, mould and spores of *C. tyrobutyricum* were found per g shoulder sample (P < 0.001), compared with the other mentioned samples. *C. tyrobutyricum* spores were less frequently detected in samples from mid in bunkers, and had a lower number of detected colonies per g sample, than side and top samples in bunkers (P < 0.001). However, mid in bunkers had a higher frequency of samples with detected *C. tyrobutyricum* colonies (8 of 18 samples) compared with mid samples from bales (5 of 36 samples; P = 0.01). Bunker shoulder samples had a higher number of detected colonies of other *Clostridia* species than mid and side samples in bunkers and samples from bales (P = 0.02). Bunker shoulder samples had higher pH than samples from all other points in bunkers and bales, and mid samples from bunkers had lower pH than samples from surface and mid in bales (P < 0.001). No differences in microbiological composition or pH were found between surface and mid samples in round bales.

Spot sample pH was higher in H3 than in H1 and H2 (P < 0.001), but microbiological composition did not differ significantly between harvests (Table 10). Yeasts tended to be detected in higher frequencies in samples from T bunkers than from WL bunkers or round bales (P = 0.06). Mould tended to be detected in higher frequencies in samples from bunkers than from bales (P = 0.06), and the number of mould colonies detected per g sample was significantly higher in T bunkers than in immediately sealed or highly compacted bales (P = 0.045). Clostridium tyrobutyricum spores were detected in higher frequencies in samples from bunkers than bales (P < 0.001), and also the number of detected C. tyrobutyricum colonies was higher in bunkers than in bales (P < 0.001).

Spores of *C. tyrobutyricum* were detected in 2 of 35 immediately sealed round bales versus in 12 of 36 bales sealed after delay (Table 10). This effect of delayed sealing was highly significant (P = 0.004) whereas no effect on spore incidence was found of bale compaction. Further, a significant interaction indicated that bales that were both poorly compacted and sealed after delay contained a higher number of *C. tyrobutyricum* colonies per g sample than other round bales (P = 0.01). Spores of other *Clostridia* spp. were detected in higher frequencies (P = 0.04), and in a higher number of colonies (P = 0.03), in bales sealed after delay compared with immediately sealed bales.

#### 4. Discussion

#### 4.1. Bunker silo fresh crop and silage weights, densities and losses

To obtain flexibility in ensiling and feeding, a farm usually has at least two silos. In Norway, silos of the same size as the experimental bunkers are used in farms with average herd size, 28 dairy cows, or less. To avoid heating in bunkers during feed out, round bales are often used in late spring, summer, and early autumn. The experimental silos were, however, substantially smaller than most bunkers on commercial farms other places in the world. Heating during silo filling, and feed out at high temperatures, may be even more challenging than in the present study. However, larger silos have relatively less surface area, that may reduce challenges with air ingress after covering. The 9% higher density of crop and crop DM immediately after silo covering by compaction with WL than T (Table 2) was most likely an effect of the higher packing vehicle weight, in line with Muck and Holmes (2000). Compared with the parent crop (Table 1 and 2), DM concentrations in silage (Table 4) based on samples withdrawn at every silage removal and corrected for volatiles, were consistently lower than fresh crop DM, whereas OM concentrations in DM were similar for crop and silage. This suggests that only a minor proportion of the DM differences between crop and silage could be attributed to plant respiration. The magnitude of the DM differences in H1 and H3 where fresh crop DM was highest, and the negative invisible losses on wet weight basis (Table 3), suggest that absorption of rainwater into silages was the main cause. The largest crop-to-silage differences in DM appeared in H3, where crop was driest.

In H1, only little effluent was visually observed in the collection tank following ensiling, but in H2, effluent drainage from silos was larger, and initiated immediately following ensiling, before rainwater entered silos. Unfortunately, effluent amounts could not be measured, and would anyway have included rainwater. A tiny flow of seepage from the silo face to the drainage system in front of silos was observed for all silos during the entire unloading period.

Fresh H1 crop was not only drier, but also more mature and therefore stiffer than the H2 crop, which reduces the pressure on the liquid fraction and diminishes effluent production (Reynolds and Williams, 1995). The higher value of invisible losses from WL silo than from T silo in H2 might, at least partly, be due to a higher amount of silage effluent squeezed out by the heavier packing vehicle. From a grass crop containing 224 g DM/kg, Randby (1997) recovered 5.6 % of crop DM in effluent mainly due to self compaction in tower silos, and as a three year average, 11 % of crop DM was recovered in effluent with an average crop DM of 193 g/kg. McEniry et al. (2007) found that increasing weight to compact herbage increased effluent production, in line with Reynolds and Williams (1995) who found that effluent amount increased by a factor of five between the smallest and largest values of top pressure, and by a factor of two between the highest and lowest values of "Young's modulus", that describes mechanical properties of the ensiled crop such as chop length, forage type and maturity. Grasses containing up to 400 g DM/kg may produce effluent if pressure is high enough (Kirsch et al., 1955, quoted by McDonald et al., 1991). When additional effluent is released due to increased pressure, also effluent DM concentration, and thereby DM losses, is expected to increase.

The long storage time for H3 silos, 12.6 months, including a summer, might have increased respiration loss during storage, which depends on silo permeability, forage density or porosity, the moisture content, and O<sub>2</sub> diffusion coefficient. Respiration loss may amount to 0.5–1.5 % per month, being almost negligible (< 0.5 % per month) if the coverage is perfect (Savoie and Jofriet, 2003). The coverage is, however, seldom perfect, and Ashbell and Weinberg (1992, quoted by Savoie and Jofriet, 2003) indicated that the

Table 5

Effect of harvest and ensiling practices (compaction and sealing time) on fresh crop and silage weights, dry matter concentrations and densities of round bales.

						Fresh cro	pp				Silage		
Harvest	Compaction	Sealing	N	Kg	DM g/kg	Kg DM	Kg per m³	Kg DM per m <sup>3</sup>	Kg	DM g/kg	Kg DM	Kg per m <sup>3</sup>	Kg DM per m <sup>3</sup>
1			12	724	257	185	447	115	713	240	171	441	106
2			12	812	294	239	501	148	800	269	214	494	132
3			$12^{1}$	742	359	266	458	164	733	366	263	453	162
SEM				25.2		19.2	15.6	11.9	23.9	20.9	12.2	14.8	7.5
P				0.06		0.02	0.06	0.02	0.048	0.001	< 0.001	0.048	< 0.001
	Poor	Immed.	9	653	304	199	403	123	649	296	190	401	117
	Poor	Delay	9	651	304	197	402	122	637	283	174	394	107
	High	Immed.	$9^1$	873	304	265	539	163	866	293	253	535	157
	High	Delay	9	861	304	261	532	161	843	295	246	521	152
	SEM			15.9		11.4	9.8	7.0	15.2	14.5	8.7	9.4	5.4
	P	Compactio	n	< 0.001		< 0.001	< 0.001	< 0.001	< 0.001	0.62	< 0.001	< 0.001	< 0.001
	P	Sealing		0.35		0.36	0.36	0.36	0.04	0.58	0.07	0.04	0.07
	P	Comp. × Se	aling	0.52		0.76	0.53	0.75	0.46	0.45	0.49	0.46	0.50

 $<sup>^{1}</sup>$  One missing value of silage bale weight. For silage weight in kg, kg DM, kg per  $m^{3}$  and kg DM per  $m^{3}$ , SEM should be multiplied by 1.005 (harvest 3) or 1.006 (high compaction and immediate sealing).

Table 6

Effect of harvest, silo type (bunkers, round bales or laboratory silos), and ensiling practices (compaction and sealing time) on harvested crop recovered as offered silage or wasted silage, and "invisible" losses through respiration, effluent, fermentation, and aerobic deterioration.

						g/kg crop			g/kg crop D	M
Harvest	Silo type	Compaction	Sealing <sup>1</sup>	N	Offered silage	Wasted Silage <sup>2</sup>	Invisible losses	Offered silage	Wasted Silage <sup>2</sup>	Invisible losses
1				20	978ª	24.5	3.2 <sup>b</sup>	934	23.1	48.4
2				20	963 <sup>b</sup>	25.0	18.3°	889	23.0	93.4
3				19 <sup>3</sup>	981 <sup>a</sup>	35.3	$-7.7^{a}$	945	34.1	29.7
SEM					4.35	6.3	1.26	22.2	6.6	22.3
P					0.01	0.42	< 0.001	0.19	0.42	0.13
	Bunkers	Т		3	969 <sup>abc</sup>	63.9 <sup>a</sup>	-32.9 <sup>a</sup>	866	57.9°	76.3
	Bunkers	WL		3	951 <sup>ab</sup>	55.3ab	$-6.2^{b}$	874	51.1 <sup>bc</sup>	74.5
	Bales	Poor	Imm	9	988°	5.8 <sup>c</sup>	6.7°	950	6.7 <sup>a</sup>	43.4
	Bales	Poor	Del	9	953 <sup>a</sup>	26.9 <sup>b</sup>	20.4 <sup>e</sup>	867	25.3ab	107.6
	Bales	High	Imm	8 <sup>3</sup>	985°	11.4 <sup>bc</sup>	4.0°	963	12.5 <sup>a</sup>	25.0
	Bales	High	Del	9	973 <sup>ab</sup>	6.1°	$20.5^{\rm e}$	937	6.8 <sup>a</sup>	56.7
	Labsilos	-	Imm	9	988 <sup>c</sup>		11.6 <sup>d</sup>	964		36.0
	Labsilos		Del	9	987°		$12.9^{d}$	962		38.1
	SEM	Bunkers			10.0	12.0	2.9	51.3	12.4	51.4
	SEM	Bales and Labs	ilos		5.8	6.9	1.68	29.6	7.2	29.7
	P	Treatment <sup>4</sup>			< 0.001	< 0.001	< 0.001	0.17	0.004	0.64
	P	Treatment × F	Iarvest		< 0.001	0.42	< 0.001	0.87	0.45	0.85

Means with different letters differ at P < 0.05.

top 30 cm layer of silage had between 12 and 78 % DM loss during an eight months period depending on the tightness of the cover, similar to findings by Bolsen (1997, quoted by Savoie and Jofriet, 2003). For the present H3 silos filled to 3 m height, this would amount to 3–20% DM loss on a total silo basis for 12 months. Silage density is considerably lower in the top than the bottom layer (Spiekers et al., 2009) which increases the vulnerability of the top layer.

In February 2018, when unloading was nearly completed in H3 silos and ambient temperatures were down to minus 10 °C, heating was observed by the shoulders of back walls in both silos, where wheels of packing vehicles had poor access. Silage was wasted from both silos, and, also earlier during unloading some silage was wasted due to heating. Total amount of wasted silage, and differences between the two silos, were small. Still, the higher value of invisible losses from H3 silage compacted by T than WL might be caused by higher aerobic storage and unloading losses due to the 12 % poorer crop compaction of T than WL on wet weight basis. Losses during unloading depend on the density of the silage, the ambient temperature, the aerobic stability of the silage, and feed-out rate, which determines the duration that silage is exposed to air (Muck et al., 2003). During formation of surface waste material, DM is lost in gaseous form, and the amount of wasted silage is therefore usually underestimated (McDonald et al., 1991). In the present study, such an underestimate would give an equal overestimate of invisible losses, and not influence the amount of offered silage.

Bunker silo DM densities were equal or higher than minimum recommendations for grass silage by Savoie and Jofriet (2003), Wilkinson (2005), and Spiekers et al. (2009). Fermentation DM losses could not be determined separately, but were likely in the range 10 − 30 g/kg at the actual DM concentrations (Savoie and Jofriet, 2003). Daily feed out rate was in line with the recommended ≥ 20 cm (Savoie and Jofriet, 2003) for H1 and H2, and slightly below for H3. The average DM losses in the present study, 130 g/kg, of which 55 g/kg was wasted silage, was the same as estimated for bunker silos by Savoie and Jofriet (2003), similar to 141 g/kg, of which 34 g/kg was wasted silage by Spörndly and Nylund (2017), and similar or less than 156 and 182 g/kg, of which 65 and 28 g/kg was wasted silage, found by Muck et al. (2015) in a two-year study with alfalfa. Köhler et al. (2019) found on average 90 g/kg total DM losses from grass crops and 70 g/kg from maize.

Although no differences in the proportion of offered silage, wasted silage or invisible losses on DM basis were detected between silos compacted by T or WL on average over three harvests, results from individual harvests suggest that the heaviest compaction vehicle was not the best choice for the leafy low DM H2 crop. When used intensively on the chopped crop in thin layers in H2, WL may have squeezed out considerably more DM through effluent than suggested by Savoie and Jofriet (2003), based on data from Bastiman and Altman (1985) from past times when WL possibly were not used for silo packing. A proportion of the 45.5 g/kg crop DM higher invisible losses observed in the WL than in the T silo in H2 was apparently effluent DM. On the other hand, WL may have

<sup>&</sup>lt;sup>1</sup> Imm=immediate sealing Del=delayed sealing.

<sup>&</sup>lt;sup>2</sup> No silage was wasted from the laboratory scale silos, because the sealing was perfect and prohibited aerobic deterioration. Therefore, laboratory silos were not included in statistical evaluation of wasted silage, so mean values from each harvest are from 14, 14 and 13 observations (harvests 1, 2, and 3), of which 2 are from bunkers.

<sup>&</sup>lt;sup>3</sup> One missing roundbale. SEM must be multiplied by 1.02 (harvest 3) and 1.08 (bales with high compaction and immediate sealing).

<sup>&</sup>lt;sup>4</sup> Treatments: Eight treatments are the combination of silo types, compaction, and sealing: 1. Bunkers packed by tractor (T), 2. Bunkers packed by wheel loader (WL), 3. Poorly compacted and immediately sealed bales, 4. Poorly compacted bales sealed after delay, 5. Highly compacted and immediately sealed bales, 6. Highly compacted bales sealed after delay, 7. Immediately sealed laboratory silos, 8. Laboratory silos sealed after delay.

Effect of harvest, silo type (bunkers, round bales or laboratory silos), and ensiling practices (compaction and sealing time) on fermentation quality, intake potential and aerobic stability of silages.

Harvest	Silo	Compaction Sealing <sup>1</sup>	Sealing <sup>1</sup>		g/kg		g/kg N				8/k	g/kgDM					Aerobic
	rype			z	DM	Hq	NH3-N	WSC	LA	FA	AA	PA	BA	TA	Ethanol	SDMI	мар., п
1				30	251ª	4.21 a	87 a	23 a	53	10.4 b	10.6	1.0 a	0.1 a	75 b	8.6 b	94 a	409 b
2				30	264 a	4.12 a	80 a	52 b	49	12.1 °	0.6	1.4 b	0.2 a	$71^{ab}$	4.1 a	в 26	154 a
3				30	353 b	4.64 b	$120^{\ b}$	48 b	42	5.7 a	10.4	1.5 b	0.3 b	e 09	5.2 a	$103^{\ b}$	305 b
SEM					13.1	0.063	7.4	4.6	3.7	0.45	0.93	0.07	0.05	4.4	0.64	1.15	37.7
Ь					< 0.001	< 0.001	< 0.001	< 0.001	0.10	< 0.001	0.41	< 0.001	0.02	0.04	< 0.001	< 0.001	< 0.001
	Bunkers	L		18	262 а	4.08 a	81	24 a	<sub>5</sub> 69	6.7 a	11.8 b	1.2 a	0.5 °	3 68	2.6 a	95 a	272
	Bunkers	WL		18	267 а	4.03 a	66	19 a	<sub>2</sub> 29	6.4 a	$13.0^{\  m b}$	1.1 a	0.6 <sup>cd</sup>	.89°	2.5 a	95 a	307
	Bales	Poor	Imm	6	$296^{\text{ bc}}$	4.42 °	101	53 с	34 a	$11.6^{\  m b}$	8.1 a	$1.5^{\rm b}$	< 0.01 <sup>a</sup>	55 a	7.6 b	$100^{\circ}$	307
	Bales	Poor	Del	6	283 ab	4.59 <sup>d</sup>	106	43 bc	34 a	10.4 b	10.7  ab	$1.2^{ab}$	0.3 bc	56 a	7.7 b	99 pc	264
	Bales	High	Imm	6	293 bc	4.42 °	103	54 c	27 a	14.5 °	8.0 a	$1.9^{\circ}$	$0.03^{\mathrm{ab}}$	51 a	6.4 b	$101^{\circ}$	317
	Bales	High	Del	6	295 bc	4.45 °	85	49 bc	35 a	11.7 b	8.4 a	$1.5^{b}$	$0.2^{ab}$	57 a	6.6 b	99 pc	254
	Labsilos		Imm	6	310 °	4.33 bc	66	48 bc	55 b	7.4 a	8.9 a	$1.2^{\rm a}$	< 0.01 <sup>a</sup>	72 b	7.6 b	96 pc	294
	Labsilos		Del	6	300 c	4.28 b	93	36 <sub>P</sub>	$62^{\rm bc}$	6.6 a	$11.2^{ab}$	$1.0^{\rm a}$	< 0.01 <sup>a</sup>	$80^{ m pc}$	6.7 b	97 ab	299
	SEM	Bunkers			9.37	0.046	7.3	3.6	3.3	0.56	06.0	0.08	0.07	3.8	0.54	0.94	33.5
	SEM	Bales and Labsilos	silos		11.3	0.057	10.0	4.5	4.3	0.79	1.22	0.12	0.09	5.0	0.70	1.21	44.2
	Ь	Treatment <sup>3</sup>			< 0.001	< 0.001	0.32	< 0.001	< 0.001	< 0.001	0.002	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.91
	Ь	Treatment × Harvest	Harvest		< 0.001	0.04	0.01	< 0.001	< 0.001	< 0.001	0.01	0.03	0.008	< 0.001	0.001	0.01	0.01

Means with different letters differ at  $P\,<\,0.05$ .

<sup>&</sup>lt;sup>1</sup> Imm = immediate sealing Del = delayed sealing.

<sup>&</sup>lt;sup>2</sup> Aerobic stability, hours.

<sup>3</sup> Treatments: Eight treatments are the combination of silo types, compaction, and sealing: 1. Bunkers packed by tractor (T), 2. Bunkers packed by wheel loader (WL), 3. Poorly compacted and immediately sealed bales, 4. Poorly compacted bales sealed after delay, 5. Highly compacted and immediately sealed bales, 6. Highly compacted bales sealed after delay, 7. Immediately sealed laboratory silos, 8. Laboratory silos sealed after delay.

Effect of harvest, silo type (bunkers, round bales or laboratory silos), and ensiling practices (compaction and sealing time) on chemical composition, protein fractions, digestibility, and calculated energy and protein values of silages. Table 8

Harves	Harvest Silo type Compact. Sealing	Compact.	Sealing <sup>1</sup>		g/kg DM	M		Protein frac	Protein fractions <sup>2</sup> , g N/kg total N	kg total N			g/kg DM			g/kg		Pr kg DM	DM	
				z	ОМ	8	A	B1	B2	B3	O	NDF	ADF	ADL	OMD	DOMD	MJ ME	MJ $NE_L$	g AAT	g PBV
1				30	919	144	588 b	19.9	266	84 a	42.2 a	571	354 b	38 a	0.685 a	630 a	9.8 a	5.63 a	67.7 a	26.4
2				30	917	155	520 a	27.2	272	142 c	39.1 <sup>a</sup>	536	326 a	36 a	$0.722^{b}$	662°	$10.3^{\circ}$	6.04 °	69.4 <sup>c</sup>	34.3
က				30	916	137	524 a	26.2	279	117 b	53.7 b	549	351 b	46 b	$0.705^{\rm b}$	646 b	$10.1^{\  m b}$	5.83 b	68.6 b	18.1
SEM					3.0	9.9	17.9	3.02	8.0	8.1	3.06	11.5	2.0	1.6	0.0068	4.9	0.09	0.063	0.25	6.45
Ь					0.77	0.16	0.02	0.19	0.51	< 0.001	0.003	0.10	< 0.001	< 0.001	0.002	< 0.001	< 0.001	< 0.001	< 0.001	0.21
	Bunker	L		18	916	140	535 pc	28.1 b	273 b	115 b	48.9 <sup>d</sup>	555 bc	344	40	0.711	652	10.2	5.91	68.8	21.0
	Bunker	MI		18	918	141	529 b	21.7 a	272 <sup>b</sup>	$130^{\  m bc}$	47.7 cd	$_{ m po}$ $099$	346	39	0.707	648	10.1	5.86	68.7	21.9
	Bales	Poor	Imm	6	921	145	<sub>p</sub> 909	$30.6^{\rm \ p}$	246 a	77 a	41.1 <sup>a</sup>	543 ab	339	39	0.701	646	10.0	5.82	68.5	26.8
	Bales	Poor	Del.	6	916	145	514 b	$20.2^{a}$	$281^{\rm b}$	136 °	$49.1^{d}$	273 d	347	40	0.704	645	10.0	5.83	68.5	26.6
	Bales	High	Imm	6	917	150	<sub>p</sub> 209	$29.2^{b}$	255 a	e8 a	41.0 <sup>a</sup>	536 a	343	38	0.702	643	10.0	5.80	68.4	31.1
	Bales	High	Del.	6	919	146	525 b	$22.1^{ab}$	277 <sup>b</sup>	133 <sub>bc</sub>	43.8 abc	265 cd	345	43	0.698	642	10.0	5.78	68.3	28.6
	Labsilos		Imm	6	917	146	561 °	$19.2^{a}$	287 b	<sub>e</sub> 06	42.2  ab	531 a	334	44	0.706	647	10.1	5.84	9.89	27.2
	Labsilos		Del.	6	918	145	477 a	24.3 ab	286 <sup>b</sup>	167 <sup>d</sup>	46.1 bcd	554 bc	352	38	0.702	644	10.0	5.81	68.5	26.8
SEM	Bunkers				2.1	4.4	13.0	2.64	6.2	7.0	2.07	2.6	4.1	1.9	0.0047	3.5	90.0	0.045	0.18	4.29
SEM	Bales and Labsilos	Labsilos			2.5	2.0	15.8	3.46	7.8	9.5	2.42	8.7	5.3	2.6	0.0054	4.3	0.08	0.054	0.22	4.92
Ь	Treatment <sup>3</sup>	£3			0.55	0.31	< 0.001	0.047	< 0.001	< 0.001	< 0.001	< 0.001	0.26	09.0	0.19	0.33	0.30	0.29	0.30	0.22
Ь	Treatmen	Freatment $\times$ Harvest	بب		< 0.001	0.03	0.28	< 0.001	< 0.001	0.002	0.76	< 0.001	0.17	0.11	0.01	0.14	0.10	0.097	0.08	90.0

Means with different letters differ at P < 0.05.

<sup>1</sup> Imm = immediate sealing Del = delayed sealing.

<sup>2</sup> Protein fractions: A = Non protein nitrogen (NPN, calculated as CP-TP), B1 = True buffer soluble protein (BSP, calculated as TP-IP), B2 = Neutral detergent soluble protein, calculated as IP-NDIP, B3 = Neutral detergent insoluble protein (calculated as NDIP-ADIP (insoluble in neutral detergent but soluble in acid detergent)), C = Acid detergent insoluble protein (ADIP).

<sup>3</sup> Treatments: Eight treatments are the combination of silo types, compaction, and sealing: 1. Bunkers packed by tractor (T), 2. Bunkers packed by wheel loader (WL), 3. Poorly compacted and immediately sealed bales, 6. Highly compacted bales sealed after delay, 7. Immediately sealed laboratory silos, 8. Laboratory silos sealed after delay. contributed to reduced losses of similar size caused by lower pore volume and thereby reduced respiration losses during storage and unloading from the high DM crop in H3. The design of the present experiment did not allow statistical tests to support or reject such a causal relationship. Still, it is in line with increased resistance to compaction of matrixes with increasing mechanical strength (Richard et al., 2004), which in this case illustrates that the dry H3 crop required more intense compaction than the leafy and wet H2 crop, in order to reduce porosity.

#### 4.2. Round bale and laboratory silo fresh crop and silage weights, densities and losses as compared with bunkers

Wet weight densities, 403 and 535 kg/m<sup>3</sup> for poorly and highly compacted round bales, respectively (Table 5), were far below 705 kg/m<sup>3</sup> as recommended for bunkers (Holmes and Bolson, 2009). Silage DM density of poorly and highly compacted round bales was 52 % and 72 % of average DM density in bunker silos, respectively, in line with Bernardes et al. (2018), who pointed out that bales usually have a lower density than forage packed in bunkers. Recommended target DM density for baled silage was suggested by Jennins (2011, quoted by Coblenz and Akins, 2018) to be 162 kg DM/m<sup>3</sup>, as obtained with highly compacted bales in the present study. Five of the 36 round bales were calculated to have negative DM losses which was probably due to unrepresentative sampling of the parent crop as mentioned by Gross and Averdunk (1968, quoted by McDonald et al., 1991) to be the main error in measurements of losses, as also experienced by others (Köhler et al., 2019).

The higher quantity of surface mould from the drier H3 bales compared with H1 and H2 bales was possibly due to higher air-filled pore volumes due to higher silage DM concentrations (McEniry et al., 2007) and longer storage time, partly in summer temperatures. Increased temperatures increase air diffusion through polyethylene (PE) membranes (Skjervheim, 1992) facilitating fungal growth and aerobic respiration. McEniry et al. (2007) observed increased wet weight losses when laboratory silos were subjected to air infiltration, similar to the higher invisible losses observed in bales subjected to delayed sealing in the present study (Table 6). However, delayed sealing in laboratory silos involved only a minor increase in invisible losses.

The higher proportion of wasted silage DM and lower proportion of silage DM suitable for feeding to animals from bunkers compared with highly compacted round bales (Table 6) revealed challenges with storage procedures in bunkers. Whereas combined balers and wrappers complete the silage making process and initiate anaerobiosis 2 – 3 min after the crop is picked up, bunker silo production is critically dependent on perfect manual work during the final covering stage. Also, at that stage, the crop in bunkers has already been stored more or less aerobically for hours. Although a sidewall PE layer was lapped over the forage to give an extra protection against air leakage to the silo shoulders in this study, the biggest amounts of rotten and moulded silage that were wasted were found just at that point. Ruppel et al. (1995) considered losses and quality changes to be more extreme in bunker silos than in tower silos because of greater surface for oxygen penetration, less perfect covering, and greater dependence on management practices during filling and feedout. Removal of plastic top films prior to unloading of bunkers opens for air ingress to the silage surface in advance of unloading. This may increase silage deterioration especially in silo shoulders that usually have lower density than the central silage surface. This problem was minimized in the present study because plastic was removed with the block cutter, however, removal of tyres in advance may have loosened the plastic cover and facilitated air ingress.

# 4.3. Fermentation quality, chemical composition and aerobic stability of silage from bunkers, round bales and laboratory silos

At all harvests, crop was successfully wilted for 13-42 h without precipitation. The crop had fairly high WSC concentration, low or

Table 9

Effect of four sampling points: mid, side, top and shoulder from the face of bunkers, and of two sampling points: surface and mid in round bales, on microbiological quality and pH in grass silage.

Silo	Sampling	Tot	Y	east	M	ould	C. tyro	butyricum	Other	Clostridia spp.	
type	point	N	$N^1$	Log cfu/g <sup>2</sup>	N <sup>1</sup>	Log cfu/g <sup>2</sup>	N <sup>1</sup>	Log cfu/g <sup>2</sup>	$N^1$	Log cfu/g <sup>2</sup>	pН
Bunkers <sup>3</sup>	Mid	18	0	1.57 <sup>a</sup>	1	0.96 <sup>a</sup>	8	1.73 <sup>a</sup>	1	1.43 <sup>a</sup>	4.10 <sup>a</sup>
	Side	18	1	1.63 <sup>a</sup>	0	0.88 a	13	2.53 b	3	1.66 <sup>a</sup>	4.21 ab
	Top	18	1	1.62 a	3	1.17 a	11	2.76 b	3	1.70 ab	4.29 ab
	Shoulder	18	7	3.11 b	11	3.38 b	17	5.01 <sup>c</sup>	5	2.06 b	5.82 <sup>c</sup>
Bales <sup>4</sup>	Surface	35	0	1.57 <sup>a</sup>	1	0.97 a	9	1.59 a	1	1.45 <sup>a</sup>	4.44 <sup>b</sup>
	Mid	36	2	1.74 <sup>a</sup>	2	1.00 a	5	1.55 <sup>a</sup>	3	1.54 <sup>a</sup>	4.47 <sup>b</sup>
$X^2$ or SEM			29.6	0.28-0.31	46.8	0.40-0.43	44.5	0.14-0.20	9.4	0.10-0.14	0.09-0.13
P			< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.09	0.02	< 0.001

Means with different letters differ at  $P\ < 0.05$ .

Number of samples above detection limit.

<sup>&</sup>lt;sup>2</sup> Detection limit = 1.70 log cfu/g (50 cfu/g) for yeast, mould, *C. tyrobutyricum* and other *Clostridia* species. Concentrations in samples below detection limit are set to half of detection limit i.e. 1.40 log cfu/g (25 cfu/g).

Means of compaction by tractor and wheel loader.

<sup>&</sup>lt;sup>4</sup> Means of poorly and highly compacted and of immediately and delayed sealed bales.

medium protein concentration (Table 1), and an effective acid based silage additive was applied. According Weissbach and Honig (1996), well fermented silages should be expected. However, several samples from H3 silages contained NH<sub>3</sub>-N above 100 g/kg N (Table 7) that is suggested as an upper level for well fermented grass silages (Eurofins, 2019). Concentrations of fermentation acids and ethanol were within recommended ranges. Fermentation was clearly more restricted in bales than in bunkers, with higher pH and WSC concentrations and lower concentrations of fermentation acids, with laboratory silos being intermediate. These differences in the extent of fermentation could be ascribed to at least three reasons: (1) Bunker silages and laboratory silages were more finely chopped than round bale silage, and therefore a greater portion of sugars were available for bacterial fermentation (Muck et al., 2003), (2) Laboratory silos and round bales achieved a higher DM concentration than bunkers, which restricted fermentation to a greater extent (Coblenz and Akins, 2018), and (3) Bales were applied a higher rate of silage additive than bunkers and laboratory silos, that also restricted silage fermentation to a greater extent (Jaakkola et al., 2006). Differences in analysed silage FA concentrations were mainly due to the different additive application rates. The higher calculated SDMI potential of round bale silage than of bunker silage could be ascribed to the lower extent of fermentation. Higher silage density per se did not influence the extent of fermentation, neither in bunkers (WL vs T) nor in bales (high vs poor compaction), in line with McEniry et al. (2007).

Although increased silage density diminishes ingress of air into the silage mass during unloading, no effect of the higher density obtained by WL compared with T was found on aerobic stability over three harvests, neither within any harvest. In H2, where average stability was considerably lower than in H1 and H3, silage in the two bunkers had the numerically highest stability, significantly higher than poorly compacted round bales sealed after delay, and laboratory silages both with and without delay (P = 0.003), as could be expected from density differences and air ingress. High yeast and mould counts may decrease aerobic stability (Wilkinson and Davies, 2012), however, counts were similar for all harvests and for all sampling points, apart from shoulders (Tables 9 and 10). Samples for aerobic stability were withdrawn from silage removed by block cutter for feeding, where deteriorated silage from shoulders was not included. The low ambient air temperature, 17.5°C, during measurement of aerobic stability in H1, versus 20°C in H2 and H3, may have increased measured stability in H1. Low WSC in H1 silage and relatively high BA concentrations in H3 silage might have contributed to high aerobic stability in those harvests. However, the low average ambient temperatures during feed out, -0.1 and 0.1°C at H1 and H3, respectively, versus 6.1°C in H2, may be the main reason (Wilkinson and Davies, 2012).

The strong increase in NPN (protein fraction A) in silage on the expense of true available protein (fraction B) in the crop is a normal change that is caused by hydrolysis of protein to amino acids (Van Soest, 1994). Heat denatured B3 protein has high digestibility, but digests at slower rates than B1 and B2, whereas fraction C is formed through the Maillard heat reaction, which is detectable at 30 °C but more important above 50 °C, and renders the protein indigestible with properties like lignin (Van Soest, 1994). The highest measured temperatures in H1, H2 and H3 bales, respectively, were 49.1, 49.2 and 58.4 °C, all in bales with poor compaction, and where sealing was delayed with at least 28 h, with respective maximum temperatures in laboratory silos of 37.1, 36.4 and 28.1 °C. Carpintero and Suarez (1992) heat treated alfalfa at 40, 53 and 78 °C prior to ensiling, and observed increased proportion of TP with increasing temperature.

**Table 10**Effect of harvest, silo type (bunkers or round bales), and ensiling practices (compaction and sealing time) on microbiological quality and pH in grass silage. Means of four sampling points from the face of bunkers: mid, side, top, shoulder, and two sampling points in bales: mid and surface.

Harvest	Silo type	Compaction	Sealing	Tot		Yeast		Mould	C. tyre	obutyricum	Oth	er <i>Clostridia</i> spp.	
				N	$N^1$	Log cfu/g <sup>2</sup>	N <sup>1</sup>	Log cfu/g <sup>2</sup>	N <sup>1</sup>	Log cfu/g <sup>2</sup>	N <sup>1</sup>	Log cfu/g <sup>2</sup>	pН
1				48	4	1.63	2	1.28	24	2.24	4	1.51	4.31 <sup>a</sup>
2				48	5	1.72	9	1.65	21	2.01	6	1.59	4.34 <sup>a</sup>
3				47	2	1.52	7	1.18	18	1.91	6	1.61	4.86 <sup>b</sup>
X <sup>2</sup> or SE	M				1.31	0.15-0.25	5.0	0.23-0.64	1.32	0.179-0.182	0.59	0.091-0.092	0.110-0.111
P					0.52	0.64	0.08	0.12	0.52	0.40	0.74	0.69	< 0.001
	Bunkers	T		36	7	2.07	8	1.87 <sup>b</sup>	25	$3.18^{b}$	7	1.77	4.68
	Bunkers	WL		36	2	1.61	7	1.69 ab	24	2.83 <sup>b</sup>	5	1.66	4.53
	Bales	Poor	Imm	18	0	1.43	1	1.10 a	1	1.42 a	0	1.40	4.43
	Bales	Poor	Del	18	1	1.68	2	1.43 ab	8	1.98 a	3	1.71	4.50
	Bales	High	Imm	17	1	1.52	0	1.07 <sup>a</sup>	1	1.43 a	0	1.40	4.42
	Bales	High	Del	18	0	1.43	0	1.07 <sup>a</sup>	4	1.47 <sup>a</sup>	1	1.47	4.46
SEM	Bunkers	_				0.20		0.43		0.20		0.103	0.125
SEM	Bales					0.26-0.27		0.47		0.29		0.146-0.150	0.176-0.181
$X^2$					10.4		10.5		41.2		8.3		
P	Treatment	.3			0.06	0.13	0.06	0.045	< 0.001	< 0.001	0.14	0.17	0.81

Means with different letters differ at P < 0.05.

<sup>&</sup>lt;sup>1</sup> Number of samples above detection limit.

<sup>&</sup>lt;sup>2</sup> Detection limit = 1.70 log cfu/g (50 cfu/g) for yeast, mould, *C. tyrobutyricum* and other *Clostridia* species. Concentrations in samples below detection limit are set to half of detection limit i.e. 1.40 log cfu/g (25 cfu/g).

<sup>&</sup>lt;sup>3</sup> Treatments: Six treatments are the combination of silo types, compaction, and sealing: 1. Bunkers packed by tractor (T), 2. Bunkers packed by wheel loader (WL), 3. Poorly compacted and immediately sealed bales, 4. Poorly compacted bales sealed after delay, 5. Highly compacted and immediately sealed bales, 6. Highly compacted bales sealed after delay.

The profound decrease in protein solubility in bales subjected to delayed sealing was probably caused by temperature increases during initial aerobic storage. The similarity of protein fractions in bales subjected to delayed sealing and in bunkers suggests that grass crops ensiled in bunkers were also heated. Spörndly and Nylund (2017) measured at least 30 °C in farm bunker silos immediately after filling, and in some cases 40 °C. Van Soest (1994) stated that larger masses are more prone to heating because there is less surface for radiant losses. Also, Forristal and O'Kiely (2005) mentioned that bales have a higher surface to volume ratio than bunker silos and, as a result, core temperatures in bales decline more rapidly than in bunker silos. However, in the 10 round bales stored aerobically during a night, no decrease in core temperature was observed the following morning, which was the case for one-kg laboratory silos, and therefore the heat effect on protein may have been somewhat lower and more inconsistent in laboratory silos than in bales.

The consistent increase in NDF concentrations in bales and laboratory silos subjected to delayed sealing, and the similarity of NDF levels in bunkers with bale silages sealed after delay, are apparently a measure of heat denatured protein bound to the NDF fraction. Loss of soluble carbohydrates due to prolonged plant respiration in silages subjected to delayed sealing might also have caused the observed increase in NDF concentration. Loss of carbohydrates would, however, concurrently have increased silage CP concentration, which was not observed. The smaller differences found in ADF concentrations with delayed sealing is consistent with smaller increases in the protein C fraction with delayed sealing and suggests that only a small portion of protein was subjected to Maillard reactions and rendered as indigestible protein in ADF. The effect in ruminant nutrition of decreased rate of protein degradation due to moderate spontaneous heating depends on the animal, the total feed ration and type of production, but might increase microbial output and N efficiency, and therefore be beneficial (Chaudhry and Webster, 1993). However, the heat is produced by crop respiration that surely consumes carbohydrates and adds losses of DM and energy. The magnitude of these losses is unknown, and results regarding changes in energy density and DM recovery in spontaneously heated bales due to delayed sealing are conflicting (Coblenz and Akins, 2018). The VOS analysis did not reveal any effect of delayed sealing on OMD, which concurs with the observation that only a minor N proportion, about 5 g/kg of N was subjected to heat damage, and about 62 g/kg N was moderately protein denatured. This was calculated as differences between immediate and delayed sealed bales, for protein fractions C and B3, respectively.

# 4.4. Microbiological composition of spot silage samples from bunker silo faces and round bales

Bunker silo shoulders were often visually moulded, which indicated air ingress during storage. Less frequently, mould was observed on top of silage. Vissers et al. (2007a) found that silage in the surface layer of clamps had low penetration resistance, i.e. low density, and higher levels of butyric acid bacteria, yeasts and mould than samples from the mid of silos, in line with results from shoulder samples in the present study (Table 9). They proposed that vulnerable points in large silos are initially exposed to aerobic growth of moulds and yeasts, that later facilitates growth of *C. tyrobutyricum* in anaerobic niches. Exactly the same seemed to happen in the present study, where both yeast, mould and *C. tyrobutyricum* spores were found in significantly higher concentrations in shoulder samples compared with all other sampling points in bunkers and bales. Vissers et al. (2007b) proposed that farmers should aim for a concentration of butyric acid bacteria in silages of less than 3 log cfu/g, and prevent concentration from exceeding 5 log cfu/g, to obtain the goal of less than 3 log cfu/L in farm tank milk. In the present study, only bunker shoulder samples contained > 5 log cfu/g of *C. tyrobutyricum* spores.

Greater air exposure due to weaker compaction during silo filling, may explain the tendency of higher frequencies of yeasts in samples from T than WL silage (Table 10), in line with higher yeast levels found in silages with aeration by McEniry et al. (2007) and Knicky et al. (2016). Also, prolonged silo filling increases temperatures that further stimulates yeast growth (Wilkinson and Davis, 2012). The higher frequencies and concentrations of moulds detected in samples from bunkers than bales concur with the higher proportion of wasted silage from bunkers than bales. However, it contrasts with the much higher silage density in bunkers than in bales that in theory should reduce porosity and protect against air and fungal growth.

The eight plastic layers on bales, in total 0.20 mm thick, seemed to protect better against air ingress than the three plastic layers, in total 0.34 mm thick, covering the shoulders of bunkers. However, moulded, wasted round bale silage amounted to 0.2, 7.2 and 30.0 g/kg DM in H1, H2 and H3, respectively (P = 0.01; not presented in Table), which suggests that 8 plastic layers were sufficient for the low DM round bales, 257 g DM/kg, in H1, but not for high DM bales, 359 g DM/kg, in H3 (Table 5) when stored through a warm summer. Also, in bunkers the amount of wasted silage was numerically higher in H3 than in H1 and H2.

Several authors have documented large differences in DM density within a bunker silo (Craig et al., 2009; Spiekers et al., 2009). The effect of insufficient plastic cover on fungal growth depends on silage density in proximity to the plastic seal, not to entire silage density. Wheels of packing vehicles working in bunkers do neither exert pressure to the area closest to the silo side walls, nor close to the back wall. Holmes (2006) stated the need for an increased number of packing tractor passes near bunker silo walls to increase density in that area of the silo, and the need for extra packing effort on top layers to increase density in those layers most exposed to oxygen. Spörndly and Nylund (2017) found that temperatures during storage were often higher along the walls than in mid of silos, which suggested that heat was produced by yeasts and other aerobic microorganisms due to air ingress. The high number of *C. tyrobutyricum* colonies found in bales that were both poorly compacted and sealed after delay, compared with other bales, may illustrate conditions in bunker shoulders: Delayed or poor plastic covering may to a greater extent facilitate clostridial growth if silage density close to the plastic is low rather than high. In their prospects for future silage production towards 2050, Wilkinson and Muck (2019) propose robotic packing of bunkers. Small, heavy robots carrying wheels on all four edges, driving completely against all walls with no damage, working continuously during silo filling, and for hours after completed filling, might solve the main problems in today's bunker silage production.

The hypothesis on reduced losses, higher grass silage quality, and improved aerobic stability in a denser bunker silage produced by a heavier compaction vehicle, was rejected.

#### 5. Conclusions

No differences in losses or silage quality were observed between bunker silo packing with WL or T on average over three harvests. These results are, however, only applicable to the crops studied. Invisible losses from the wet and least fibrous crop (H2) were higher after packing with WL than T, which suggests higher effluent losses. Invisible losses from the driest and most fibrous crop (H3) were higher after packing with T than WL, which suggests higher respiration and aerobic deterioration losses. Although statistics cannot confirm these results obtained with only two silos within a harvest, these observations suggest that packing pressure should be tuned to crop wetness and morphology and that a minimum wet weigh crop density of 705 kg/m³ should be targeted. When crop for round bale silage got heated prior to plastic sealing, the amount of fiber bound protein in silage increased. High filling rate, to avoid heating, is therefore recommended for bunkers. Bunker silage density did not influence silage quality in those parts of the silo where air ingress was avoided. However, density is critical in surface layers, especially in shoulders where average spore concentration of Clostridium tyrobutyricum was > 5 log cfu/g. This was higher than in all other sampled spots from bunkers and bales. The importance of high-quality work in the finishing phase of bunker silo production cannot be over emphasized. Packing vehicles must produce a dense and smooth surface, with extra focus on the area close to walls. Manual work with plastic coverage must focus on the same areas for maximal protection against air ingress. Less manual work is required in round bale production, which therefore, in some cases, may be preferable.

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# CRediT authorship contribution statement

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#### **Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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