Journal of Animal and Veterinary Advances 9 (19): 2485-2492, 2010
ISSN: 1680-5593

Nutrient Content and *in sacco* Digestibility of Barley Grain and Sprouted Barley

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Abstract: The studies reported in this research examined the nutrient profile of barley grain when it was sprouted hydroponically. Following sprouting, the measurement of animal response at experimental level and also in a commercial setting was performed in order to test the hypothesis that sprouting gives rise to hydroponic sprouts that give higher animal responses. In first part of the experiment, barley grain was sprouted hydroponically for a duration of 7 days. Daily sampling of the sprouts was done to assess DM concentration and also to determine the nutrient concentration on day 7 in comparison to the unsprouted grain. Results showed a 21.9% loss in DM from the original seed after sprouting for a period of 7 days. A loss of 2% GE was recorded after comparing the sprouts with the original grain. The CP, ash and all other minerals except potassium were lower in concentration on a DM basis in the barley grain than in the sprouts. This was considered to be a reflection of a loss in DM after sprouting causing a shift in concentration of these nutrients. The second phase of the experiment involved *in sacco* degradation of hydroponic barley sprouts and the unsprouted grain in the rumen of Merino sheep. There was no significant difference ($p>0.05$) in *in sacco* degradation when unsprouted grain was compared with hydroponic barley sprouts. It was concluded that the loss of 21% DM followed by a lack of difference in *in sacco* degradability disproved the presence of any advantage of sprouts over the original grain.

Key words: Barley, nutrients, *in sacco*, digestibility, sheep, reflection

INTRODUCTION

Research on hydroponically sprouted barley has shown an increase in fresh weight over the sprouting duration as well as changes in DM (Peer and Leeson 1985; Trubey *et al*., 1969). The gain in fresh weight has been mainly attributed to imbibition of water constituting up to 80-90% of the fresh weight (Sneath and McIntosh, 2003). Changes in DM occur as a result of enzymatic activities as well as in DM losses (Salunkhe *et al*., 1984; Sneath and McIntosh, 2003).

The activation of enzymes leads to hydrolysis proteins carbohydrates and lipids into their simpler components. The hydrolysis leads to increases in concentrations of amino acids, soluble sugars and fatty acids, respectively (Chavan and Kadam, 1989).

The enzymes also cause the inter-conversions of these simpler components leading to increases in quality of amino acids as well as increases in the concentrations of vitamins (Koehler *et al*., 2007; Plaza *et al*., 2003). Early research on hydroponic sprouts reported the presence of a grass juice factor that improves livestock performance (Elvehjem *et al*., 1934; Kohler *et al*., 1938). More recent research has also indicated that hydroponic sprouts are a rich source of nutrients and they contain a grass juice factor that gives an improved performance to livestock (Finney 1982; Nutrigrass, 2007).

Research on increased performance for both ruminants and monogastric animals has been reported (Finney, 1982). Workers using cattle, also reported an improvement in performance due to the feeding of hydroponic grain sprouts when compared to the original unsprouted grain (Grigorev *et al*., 1986; Tudor *et al*., 2003). Some reports, however showed there was no advantage with regards to animal performance when supplementary feeding of hydroponic grain sprouts was done (Farlin *et al*., 1971; Myers, 1974). In addition to lack of increased livestock performance, DM losses incurred as a result of sprouting (Sneath and McIntosh, 2003), tend to add to the cost of production. Due to the conflicting reports on animal performance when hydroponic sprouts are fed, further investigation needed to be done

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to provide more current information. The objectives of the current study were therefore to determine the nutrient profile of hydroponic sprouts as compared to the original grain and also to determine the degradability of the grain and sprouts *in sacco*.

MARTERIALS AND METHODS

Hydroponic unit: Hydroponic barley was sprouted in a temperature controlled room. The temperature was maintained at 25°C and continuous lighting was also provided throughout the 7 days sprouting period. Three pairs of fluorescent lamps of (Philips TLD36W/840) were used. These lamps provided an average light intensity of about 615 lux and had a wavelength of 400-700 nm. The measurement of light intensity was done at the surface of the sprouted material.

Spray watering was applied for 3 min in every 2 h for the 7 days period of sprouting. A sprinkler with a timer control device was used to achieve the set interval and running time indicated above. The excess water drained freely from the sprouting trays between waterings.

Grain preparation: Seeds of Grimmett barley variety were steeped in warm water containing 0.1% (v/v) hypochlorite at 24°C for 4 h before they were transferred to perforated trays for watering. The steeped seeds were placed in plastic trays at a rate of 6.7 kg m^{-2} (100 g $tray^{-1}$). Thirty plastic trays measuring 10/15 cm each (0.15 m^2) were used. A daily random sampling of four trays was performed at approximately the same time over the 7 days period to determine DM and nutrient changes. At the time of daily sampling, the sprouts were removed from the trays and allowed to drain for at least 30 min and placed on an absorbent cloth in order to remove the surface water. The fresh sprouts were then weighed and transferred to storage at -20°C.

The daily samples taken over the 7 days period were later freeze-dried (Martin Christ, model Alpha 1-4, Osterode am Harz, Germany). Samples of Grimmett barley grain were also freeze-dried to serve as control in order to determine the DM and nutrient changes in the sprouts over the 7 days period. This was done by measuring the weights of both the hydroponic barley sprouts and dry grain.

Rumen degradation of sprouts: About Four Merino sheep, each with surgically placed rumen cannulae having an average initial body weight of 39.9±1.9 kg were used for the degradation study. The four sheep which served as four replicates were fed a basal diet of oaten and lucerne chaff (80:20, respectively) *ad libitum*. They were also given 15 g of mineral/vitamin premix and 50 g of cracked barley grain each daily. They were allowed to adjust to the experimental diet for 7 days before the rumen incubation of samples commenced.

The nylon bag study was performed as described by Orskov and McDonald (1979) and Orskov *et al.* (1980). Briefly, fresh 7 days Grimmett hydroponic barley sprouts were chopped to 1 cm lengthand then homogenized. About 10 g of the fresh sample was placed in nylon bags containing marbles. The bag and marble weight was determined before placing the samples. The nylon bags measured 70×140 mm and had a pore size of about 50 μm^2. The nylon bags containing marbles and samples were then tied with nylon fishing line before they were transferred into the rumen.

Sprout samples of cracked grain (2-3 mm) whole sprouts (1 cm length), shoot and root portions of the sprouts (1 cm length each) were incubated in nylon bags suspended in the rumens of the four sheep. These were for a comparison between cracked grain and whole sprouts and shoot versus root portions. Six nylon bags containing samples were incubated in each of the four animals and withdrawn after 6, 12, 24, 48, 72 and 96 h. After withdrawal the nylon bags were kept frozen until required then washed and placed in an oven at 60°C for 72 h for the determination of the oven-dried weight of the residue. The DM losses were then determined as a percentage loss from the original DM. The 0 h sample was determined by steeping the nylon bags containing the samples in water at 37°C for 15 min after which the bags were hand-washed under running tap water until the water was clear. For the other incubation hours (6, 12, 24, 48, 72 and 96), the bags were washed for 30 min under running tap water until the water was clear.

Nutrient analysis: Samples collected at day 7 were freeze dried, ground to pass through a 1 mm mesh screen and stored in tightly-sealed glass jars at room temperature and used for nutrient analysis. Daily sampling was done to determine dry matter loss over the 7 days period of sprouting. Three samples of dry barley seeds were also freeze-dried along with the sprouts to serve as controls. Nutrient analyses were done on dry ground samples.

Dry matter, CP, CF, ash and mineral concentrations of sprouted and non-sprouted barley were determined by the Association of Official Analytical Chemists (AOAC, 1995) methods. Gross energy was determined using a bomb calorimeter (Model C7000, Staufen, Germany).

Dry matter loss was determined by sprouting 3 trays of 100 g barley seed and freeze drying the whole contents of the trays. Three trays containing 100 g samples of dry

grain each were freeze-dried at are 51.7°C and their average DM weight served as a control. The DM loss was defined as the difference between the DM weights of the control and sprout samples after freeze-drying.

RESULTS AND DISCUSSION

Fresh weight and DM changes in sprouts: Grains of barley gained weight over the 7 days sprouting period as a result of water imbibition. Fresh sprouts weighed about 1.75 times their original pre-steeped weight after 1 day, 2.0 times after 2 days, 2.3 times after 3 days, 2.7 times after 4 days, 3.3 times after 5 days, 3.6 times after 6 days and 3.7 times after 7 days (Fig. 1). The DM concentration of the hydroponic barley sprouts was 44.6% after 1 day, 39.7% after 2 days, 34.6% after 3 days, 29.1% after 4 days, 22.9% after 5 days, 20.9% after 6days and 19.7% after 7 days of sprouting (Fig. 2) while that of the freeze-dried barley grain (control) was 98.7%. The 19.7% concentration of DM in the 7 days sprouts clearly indicated the presence of water (about 80%). There was a gradual decline in DM content of the sprouts from day 1-7 of sprouting, peaking at an average DM loss of 21.9% on day 7 (Fig. 3). The hydroponic barley sprouts were 13-16 arecm in height at the end of the sprouting period of 7 days and had a substantial root mass at the base (Fig. 4).

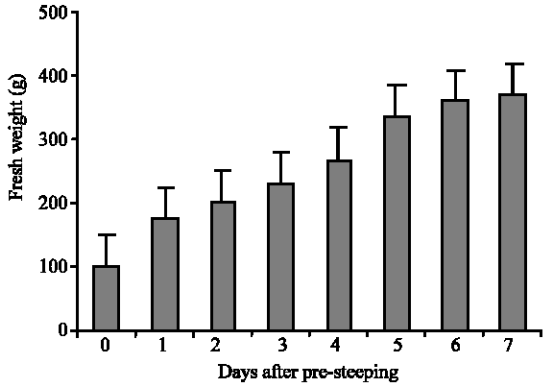

Fig. 1: Daily increase in fresh weight of grimmett barley during spouting

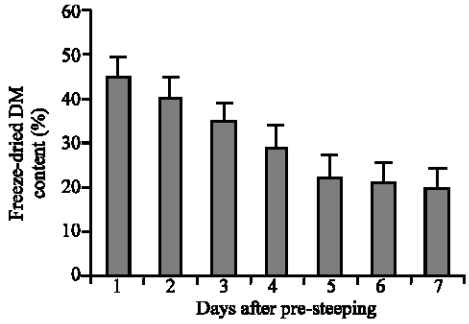

Fig. 2: Dry matter content of fresh grimmett barley after pre-steeping

Nutrient profile of Grimmett hydroponic barley sprouts and grain: The DM content, gross energy, crude

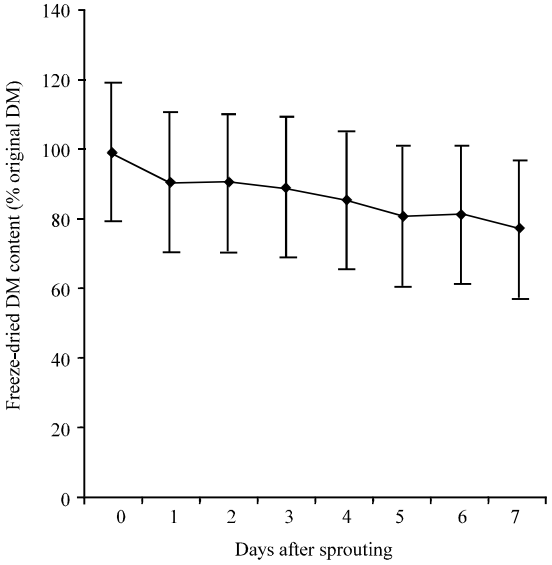

Fig. 3: Dry matter lodd in grimmett barley over 7 day sprouting period

(a)

(b)

Fig. 4: Hydroponic barley sprouts at day 7 (a) and harvested to be fed to animals (b) both showing a massive root mass

Table 1: Nutrient profile of grimmett barley grain and sprouts

Grimmett barley	DM	GE*	CP	Ash	Ca	K	Mg	P	S	Al	Cu	Fe	Mn	Mo	Na	Zn
	(%DM)									(ug g^{-1} DM)						
Grain	90.5	15.3	12.6	2.0	0.03	0.49	0.10	0.22	0.14	1.4	5.1	39.6	15.3	1.0	148.1	16.7
Sprouts	90.2	15.0	15.4	4.3	0.06	0.26	0.13	0.26	0.17	5.5	13.4	52.0	12.2	1.7	830.9	23.0

*Gross energy (MJ kg^{-1}) on a DM basis

protein and mineral profiles of the sprouted Grimmett barley as well as the non-sprouted grain are shown in Table 1. The original barley grain had higher DM and gross energy values than the sprouted barley (90.5 vs. 90.2% and 15.25 vs. 15.04 MJ kg^{-1}), respectively. The crude protein, ash and all the minerals except potassium were lower in concentration on a DM basis in the barley grain than in the sprouts.

Rumen degradation of sprouts: The results of DM loss from whole sprouts versus cracked barley grain are shown in Fig. 5. Dry matter loss from shoot versus the root portions are shown in Fig. 6. In Fig. 5, the initial degradation of the whole sprout was significantly higher than the cracked grain after 6 h of incubation in the rumen, but thereafter from 12-96 h, there were no significant differences ($p>0.05$) between the whole sprouts and cracked grain. There were however, remarkable differences in degradation of the root and shoot portions. This was shown by the higher rate ($p<0.05$) of degradation for the shoot portions when compared to the roots (Fig. 6)

Fresh weight and DM changes in sprouts: The weight gain of barley with pre-steeping and subsequent sprouting is consistent with earlier reports. Reports by Trubey *et al.* (1969) indicated an increase in fresh weight of oats sprouted for 3 and 6 days. The fresh weight of the 6 days sprouts was higher than the 3 days sprouts due to the continuing uptake of water. The root development with time was also greater giving the older sprouts more absorptive capacity. Ueno *et al.* (1966) also found an increase in fresh weight of barley seedlings in a 6 day growing period.

Commercial sprouts growers have reported fresh weight increases of between 6 and 10 fold (i.e., 1 kg of seed yielding about 6-10 kg of fresh sprouts), (Sneath and McIntosh, 2003) while trial yields from experiments indicated a 5-8 fold increase in fresh weight with sprouting of grain. Pre-steeping of the barley seed prior to germination and sprouting enables the seed to imbibe water and swell.

This swelling of the seed later allows for the splitting of the seed coat as well as part of the endosperm, thereby allowing even more water to be absorbed by the seed as irrigation continues. Depending on whether nutrient solution is used rather than water, there may also be absorption of minerals by the roots which would increase the ash content of the sprout and could add even more to the final weight. The role of photosynthesis in increasing the weight of sprouts grown under lights

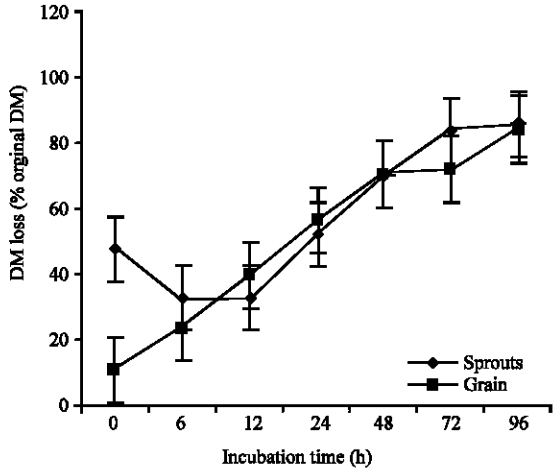

Fig. 5: Nylon bag degradation of barley sprouts and cracked barley grain in the rumen of sheep fed oaten chaff

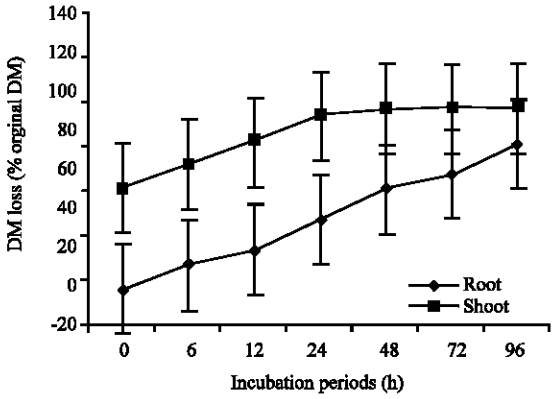

Fig. 6: Nylon bag degradation of fresh root and shoot portions of grimmett barley sprouts in the rumen of sheep fed oaten chaff

has been reported, although such increases may not be of great significance in the young growing plant. There was an increase in loss of DM content in a sprouting trial where no light was provided but the rate of loss of DM slowed down after day 4 in lighted treatments in the trial when leaves began photosynthesizing (O'Sullivan, 1982). This was in agreement with reports that lighting of sprouts has some effect only from day 3 of sprouting (Morgan *et al.*, 1992). The weight increase of sprouts in the current trial was 3.7 times the original grain indicating

a lower value compared to reports of 5-8 times increase indicated by commercial operators. The difference may be due to seed quality and grain variety used, nutrient solution used during sprouting, lighting, irrigation frequencies, seed treatment, water quality and pH, seeding densityand temperature or the degree of drainage of free water prior to weighing.

This study showed a gradual decrease in DM concentration of the original grain following pre-steeping and sprouting from day 1-7. Chavan and Kadam (1989) reported a decrease from the original dry weight of seed during soaking and subsequent sprouting processes due to leaching of materials and oxidation of substrates from the seed. The leaking of the solutes from the seeds is fastest at the start of water uptake and comes to a halt after about one day (Simon, 1984). Solutes that leak include proteins, amino acids, sugarsorganic acids and inorganic ions. Seed soaking apart from causing the leaching of nutrients, also initiates a series of events that lead to oxidation of the substrates stored in the grain causing a loss in DM. When water is imbibed into the embryo of the seed, it dissolves the plant hormone Giberrelic Acid (GA) which is transported with water to the aleurone layer. In the aleurone layer it activates DNA which translates the amino acids present into the enzyme, amylase. The amylase is then released into the endosperm where it catalyses the hydrolysis of starch into its component glucose units. The glucose is then used for metabolic activities within the young growing plant, its oxidation generating ATP and releasing carbon dioxide. The loss of DM through respiration in the young plant when compared to the gains of photosynthetic activities brings about a net loss in DM when sprouting is completed. In a 7 day sprout, photosynthesis commences around day 5 when the chloroplasts are activated and this does not provide enough time for any significant DM accumulation. The lighting provided at the level of the sprouting tray in the current study was 615 lux. This compares to about 100,000 lux for normal sunlight on a clear summer day. In a lighted experiment, DM losses continued to increase from a value of 5.2% after 3 day to 12.3% after 6 day, probably reflecting the losses due to respiration and the negligible amount of photosynthesis by young seedlings (Trubey *et al*., 1969).

Nutrient profile of barley grain and sprouts: The DM content in the day 7 sprouts showed a loss which reflected a lower value than the original barley grain. There was also a lower energy value for the sprouts on a DM basis when compared to the original grain. The soluble carbohydrates used to provide the energy to maintain the metabolic activities of the young plant are oxidized during plant respiration. The continuation of such a process depletes the food reserves of the endosperm in the seed without any adequate replenishment from photosynthesis by the young plant. A short growing cycle as is the case with sprouting provides little chance for DM accumulation. From all literature consulted so far, there has been no report of gain in DM above the original grain DM input. For all the reports summarised by Sneath and McIntosh (2003), a DM loss ranging from 7- 47% was reported for short cycle sprouting. The energy content of the sprouts when compared to the original grain on a DM basis followed a similar pattern (lower energy in sprouts) as was the case for total DM content. Most of the losses in DM in the sprouts were as a result of respiration, an energy requiring process which shows why there was lower energy on a DM basis in the sprouts.

Crude protein, ash and all minerals except potassium were lower in concentration on a DM basis in barley grain when compared to the sprouts. This was likely due to a change in weight of the carbohydrates used in providing energy for the young seedling through respiration as there was a 21% loss in DM after sprouting. The change in the case of CP% was likely due to the loss in carbohydrate as DM, by the sprouts due to the fact that there was no nitrogen source added externally to the water used for irrigation during sprouting. This CP% increase was therefore not a likely true increase (Chavan and Kadam, 1989; Cuddeford 1989; Lorenz 1980; Peer and Leeson, 1985; Morgan *et al*., 1992; Flynn and O'Kiely, 1986).

Rumen degradation of hydroponic barley sprouts: The results shown in Fig. 5 indicate an initial significant difference in degradation between the sprouts and cracked barley grain (0-6 h incubation times). It is likely that the sprouts being hypotonic to the rumen environment might have had some uptake of DM, most likely minerals in solution making them heavier than when they were placed in the rumen thereby showing an apparent decrease in digestibility in this 0-6 h period. This trend abated after 6 h assumedly due to the sprouts DM being removed by enzymatic digestion. Presence of enzymes (proteases) in plant cells in lytic vacuoles has been reported (Feller, 1986; Matile, 1997). These enzymes have been reported to commence the initial degradation of proteins in the rumen in the first few hours of ingestion of forage (Kingston-Smith *et al*., 2005).

Newly ingested feeds are colonised by microbes in the rumen but the degradation is not immediate. Bacteria

are known to colonise the feed at about 8 h, protozoa after 2 h and fungal spores after 15 min (Grenet, 1997). The fungal spores however, took 3 h or more to produce hyphae to degrade the plant cell wallsand make them fragile for bacterial and protozoal degradation. It is also pertinent to note that freezing and thawing of bags could also have led to loss of material from the plant cells in the current study.

Both sprouts and cracked barley grain contain fibrous materials in the form of a seed coat. This may further explain why there was no significant ($p>0.05$) difference in *in sacco* digestibility of both grain and sprouts for the rest of the incubation hours except at 72 h.

The large difference ($p<0.05$) in digestibility between shoot and root components in the current study was as expected because of the compositions of the two fractions. Week old plant shoots are easily degraded in the rumen. These young shoots are usually not fibrous in nature because the deposition of fibre commences later during the jointing stage (Nutrigrass, 2007). The root portion in this study comprised of an interwoven succulent and tender root mass with the seed husk which housed the endosperm at seed formation and maturity. The husk of barley seed is usually high in fibre that is made up of cell wall polysaccharides such as cellulose and hemi-cellulose that are usually more resistant to degradation than the young succulent shoots.

Studies on plant maturity and leaf to stem ratio as they affect degradability and animal performance indicated a higher organic matter intake organic matter digestibility and faster particulate passage rates for immature hay fractions than mature fractions in steers. The differences in the parameters mentioned were as a result of lower passage rates for the mature hay. The particle size of the stem portions in the study were greater ($p<0.05$) than leaves (Lamb, 1996). The highest quality part of forage is the leaf tissue (Griffin and Jung, 1983; Nelson and Moser, 1994) and ruminant animals prefer diets that are leafy and non-stemmy where the leaves are characterized by low to intermediate tensile strength; the tensile strength is usually associated with cellulosic material and lignification which occurs more rapidly in stems than in leaves with advance in maturity (Morrison, 1980). A forage composition that allows rapid degradation to 1mm size encourages faster passage through the reticulo-omasal orifice (Minson, 1990) this indicates a lower retention time which enhances voluntary intake (Minson 1990). The shoot portion in the current study was the most likely to be rapidly degraded to a 1 mm size, making it more likely to have a faster passage rate out of the rumen.

Surface area for microbial attachment and subsequent degradation is another aspect of digestibility that favours the leaf portion above the stem. Laredo and Minson (1975b) and Poppi *et al.* (1980, 1981) observed that sheep consumed more tropical grass leaves than stems. They were of the opinion that the higher intakes for leaf than for stem were related to higher surface area of leaf material that allowed for greater bacterial degradation and a shorter retention time within the reticulo-rumen. In related studies (Ulyatt *et al.*, 1986) observed that when forage availability was not restrictedintake was inversely related to the proportion of indigestible fibre in the feed and the length of time the feed was retained in the rumen. Other studies showed the relationship between particle size and passage rate was an inverse one (Ehle, 1984; Laredo and Minson, 1975a).

CONCLUSION

Results of the present study indicate that the loss in DM was 21.9% over the sprouting period of 7 days. Sprouting brought about a loss of total DM as result of respiration of the germinating seed and loss of solutes. The loss in total DM led to corresponding increases in the crude protein and mineral concentrations per unit of DM. There was however a decrease in energy value of the sprouts compared to the original barley grainwhich was expected because of the energy requiring metabolic activities taking place during sprouting. This energy was provided as carbohydrate from the seed endosperm and was respired, producing carbon dioxide as a loss to the system measured as DM. This DM loss represents a loss in digestible energy that would otherwise have been available to the animal for productive purposes. From Table 1, it can be calculated that a 2% loss of ME was recorded after sprouting.

A 21% loss in DM during sprouting without a significant improvement in DM digestibility *in sacco* represents a considerable reduction in total digestible energy. These results indicate that the nutritive value of the original grain was higher than that of the resulting sprouts.

The *in sacco* degradation results comparing the non-sprouted barley grain and the hydroponic barley sprouts did not indicate the presence of a grass juice factor enhancing the degradation of the sprouts above the barley grain.

ACKNOWLEDGEMENTS

The researchers would like to acknowledge the technical assistance of Graham Chaffey, Jenny Wittig, Graham Taylor and David Creed during the conduct of the

experiment. The research was made possible by the University of New England Scholarships (International Postgraduate Research Scholarship and University of New England Research Assistantship).

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