Pasture Intake, Digestibility and Fecal Kinetics in Grazing Horses

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(ABSTRACT)

Pasture intake of grazing livestock needs to be estimated to allow determination of energy and nutrient intakes. It is commonly estimated by difference, subtracting intakes of other feeds from estimated needs for dry matter or energy. However, these estimates are often erroneous, because they do not take individual animal variation for growth, reproductive status or activity level into account.

One method that has had success in grazing ruminants has been the use of markers, or tracers, to estimate fecal output and nutrient digestibility. External markers are dosed to the animal and can be used to determine fecal output. Internal markers are an inherent part of the diet in question and can be used to determine dry matter and nutrient digestibilities. These estimates can then be used to give estimates of intake. These studies were conducted to evaluate the effectiveness of traditional marker methods in determining fecal output, digestibility, and thus intake in grazing horses.

The first trial was conducted on 8 mature mares and geldings, housed in stalls, to determine if a common external marker, Cr, could be used to determine fecal output. Horses were dosed once daily with a molasses, Cr, and hay mixture for 12 d. Feces were collected throughout the day into individual tubs so that total fecal output (TC) could be measured. Daily fecal Cr excretion values (C_t, mg/kg DM) were fit to a monoexponential equation with one rate constant (k), rising to an asymptote (C_a): $C_t = C_a - C_a \cdot e^{-kt}$. Superior fits were found when a delay (d) was incorporated into the equation, estimating the time required for Cr to enter the prefecal pool: $C_t = C_a - C_a \cdot e^{-k(t-d)}$. Estimates of fecal output (FO) were calculated using the equation: FO = Cr dose^{-d} / C_a and provided good estimates when compared to TC values.

Subsequent trials evaluated to use of internal markers and more frequent dosing of Cr to improve estimates of intake. Eight mature geldings were housed in stalls and were fed 2 hays in a replicated Latin Square design. The monoexponential equation with the delay continued to fit the data well. Thrice daily dosing of Cr improved the predictions of FO, when dosing was every 8 h. The internal marker, yttrium (Y) consistently overestimated digestibility (D). The internal markers, n-alkanes, gave a better estimate of digestibility. When the digestibility estimates were combined with the FO estimates to estimate dry matter intake (DMI, kg/d): DMI = [FO / (1-D)]*100, the combination including n-alkanes gave better estimates.

Further studies found that dosing Cr for 12 d did not improve the fit of the monoexponential equation compared to dosing for only 8 d. Marker methods that had been developed in stalls were applied to grazing horses, and results continued to be promising.

This dissertation is dedicated to my parents:

Richard L. Holland and Mary A. Holland

"Did you ever know that you're my hero, and everything I hope to be? I can fly higher than an eagle, 'cause you are the wind beneath my wings."
"The Wind Beneath My Wings" by Bette Midler

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Introduction

Accurate methods of determining voluntary intake are essential for evaluating nutrition of grazing livestock. The most common method in the field is "by difference" - subtracting amounts fed of concentrate and hay from the expected intake of an animal. Any difference is assumed to have been consumed as pasture.

A common experimental method involves using external markers to measure fecal output (Krysl et al., 1985; Galyean, 1993) and internal markers to determine digestibility (Cochran et al., 1986; Lippke et al., 1986; Cuddeford and Hughes, 1990). These results can then be combined to calculate dry matter intake. In species such as cattle and sheep, more invasive methods such as esophageal (Olson, 1991) and ruminal fistulas may be used (Judkins et al., 1990; Brandyberry et al., 1991; Luginbuhl et al., 1994a), however, these invasive methods have not been used in grazing horses.

Chromic oxide is one of the most commonly used external markers (Kleiber, 1961), because it is relatively inexpensive and easy to analyze with atomic absorption spectrophotometry. However, problems with an inconsistent passage rate when it is dosed once daily (Coleman et al., 1984; Beauchemin and Buchanan-Smith, 1989) have led to the use of continuous release capsules in ruminants (Parker et al., 1989; Luginbuhl et al., 1994b). These capsules rely on becoming lodged in the reticulo-rumen of ruminants, and therefore would not function well in nonruminants, such as horses.

Several internal markers have been tested in both ruminants and nonruminants with varying results. Acid insoluble ash gave satisfactory digestion estimates in sheep (Van Keulen and Young, 1977), however, its accuracy depended both on the type of feed consumed and analytical procedures used. Acid detergent fiber, neutral detergent fiber and acid detergent lignin have also been evaluated (Cochran et al., 1986) but they were inconsistent, when compared to total collection estimates. Other potential internal markers include rare earth elements, such as dysprosium (Turnbull and Thomas, 1987;

Moore et al., 1992) and n-alkanes (Dove and Mayes, 1991). Both have been tried in studies on ruminants and have given promising results.

These studies were conducted to test the effectiveness of chromic oxide as an external marker to predict fecal output of grazing horses. Methods were developed which involved applying a monoexponential equation to daily fecal chromium concentrations. Potential internal markers were also evaluated. These included yttrium and n-alkanes. All balance studies were first conducted in stalled horses where intake, fecal output and digestibility could be determined by weighing. Marker methods were then compared to the total collection data. Once the methods were working in stalled horses fed hay, horses were offered fresh-cut pasture in stalls. Finally, methods were tested in grazing horses using fecal collection bags.

Review of Literature

History of Digestion Studies

When digestion trials were first run, studies involved the complete weighing of offered and refused feeds (to determine intake) and total fecal output. From this, apparent digestion of dry matter could be determined with the following equation:

$$\mathbf{D, \% = [(I - F)/I]*100} \text{ or } \mathbf{D, \% = (1 - F/I) *100} \quad (1)$$

where D was dry matter digestibility, I was dry matter intake (kg), and F was dried fecal output (kg). Apparent nutrient digestibilities (ND) could be determined by analyzing feed and feces for their nutrient contents and substituting total nutrient intake (I_N) and output (F_N) into the above equation:

$$\mathbf{ND, \% = [(I_N - F_N)/I_N]*100} \text{ or } \mathbf{ND, \% = (1 - F_N / I_N)*100} \quad (2)$$

Although these trials were effective for confined animals in individual stalls or metabolism crates, these methods were difficult to apply to grazing livestock (Cordova et al., 1978). Although fecal output could be determined in grazing animals with the use of fecal collection harnesses, there was no effective way to determine intake. Another concern with these trials was the discovery that excessive handling of animals to remove and replace the harnesses was interfering with the normal grazing routine of the animals by decreasing intake, hence fecal output (Hatfield et al., 1993) as well as decreasing nutrient digestibility (Noblitt et al., 1963; Phar et al., 1971). Therefore, methods needed to be developed that would cause less interference in the daily routine of livestock.

When trials are conducted in grazing animals, indirect methods of determining fecal output, digestibility and intake require the use of substances called markers or tracers. The standard definition of an ideal fecal marker is: a substance that is indigestible, unabsorbable, and physiologically inert. It should pass through the digestive tract at a uniform rate and be easily analyzed (Maynard et al., 1979). There are two types of markers: external and internal. External markers are substances that are dosed or fed to the animal at a known rate per day. Internal markers are an inherent part of the feed being given to the animal, either naturally occurring, or mixed evenly throughout the diet.

Markers, or tracers, can be used, because the amount excreted will equal the amount consumed, as long as the marker is truly "indigestible" (Kleiber, 1961). Therefore, if I is the daily intake and F the daily fecal output, and C_I and C_F are the concentrations of the internal marker in the feed and feces, respectively, the following will be true:

$$\mathbf{I * C_I = F * C_F} \quad (3)$$

Using simple algebra, this equation can be converted to:

$$\mathbf{C_I / C_F = F / I} \quad (4)$$

which can then be substituted into the digestibility equation (1) resulting in:

$$\mathbf{D, \% = (1 - C_I/C_F)*100} \quad (5)$$

which determines dry matter digestibility without total collection of feces or weighing of feed consumed. This method has been recognized for over 120 years (Kotb and Luckey, 1972). This equation can be modified further to give estimates of digestibility of specific nutrients as well, using the concentration of a nutrient in the feed $[N_I]$ and feces $[N_F]$:

$$\mathbf{ND, \% = \{1- ([N_I]*C_I)/([N_F]*C_F)\}*100} \quad (6)$$

It is also possible to use an external marker to determine fecal output. A marker is dosed or infused daily (continuously). The marker (M) needs to be given to the animal until it reaches a plateau level in the feces, usually a minimum of 4-5 days (Pond et al., 1987). Fecal output can then be calculated:

$$\mathbf{F\ (kg/d) = M\ dose\ (g/d) / [M]\ in\ feces\ (g/kg)} \quad (7)$$

An external marker can also be given in a single, pulse dose, and fecal samples taken over the next several days so that a curve of appearance and disappearance can be calculated. The area under this curve is then used to determine F.

Estimates of F (equation 7) and D (equation 5) can then be combined to predict dry matter intake (I).

$$\mathbf{I\ (kg/d) = F\ (kg/d) / (1 - D/100)} \quad (8)$$

Use of Models in Digestion Trials

These equations have been used, with varied success, mostly because markers have not been found that consistently meet all the requirements of an "ideal" marker (as reviewed in Titgemeyer, 1997). Therefore, researchers have found that designing mathematical models, or "tracer kinetics" can be used as long as models are fit properly (Kronfeld and Ramberg, 1981) and standardized nomenclature is used (Brownell et al., 1968). Mathematical models have been applied successfully to cattle (Pond et al., 1988; Lalles et al., 1991), sheep (Grovum and Williams, 1973; Moore et al., 1992) and horses (Corino et al., 1992; Holland et al., 1998).

Most marker models for ruminants need to include two exponentials, or compartments, which relate to the complexity of the ruminant digestive system. Comparison of one- versus two-compartment models showed that the more complex model gave better predictions of fecal output and digesta flow in cattle, but that this was diet dependent (Pond et al., 1988). Cattle fed ground and pelleted straw had better predictions with the two-compartment model, where a one-compartment model gave similar results when the straw was chopped. Effects of time of dosing versus time of feeding have been noted by others, as well. Markers dosed prior to a meal will pass through at a faster rate than markers dosed after a meal (Pond et al., 1986). No single mathematical model gave adequate estimates of degradation of individual nutrients (Nocek and English, 1986). They concluded that differences were due to site and rate of degradation. Comparisons of diet retentions from algebraic and model estimates were compared by Lalles et al. (1991). Hay labeled with Yb and concentrate labeled with Ce was fed to young calves and various algebraic approximations and modeling methods were applied to the fecal marker concentrations. They concluded that both types of models gave similar estimates of total tract retention time, however, models that included a delay between compartments gave the best estimate.

The use of models has also been evaluated in sheep digestion trials. Models were compared by Grovum and Williams (1973) as a means of examining gut functions without surgical procedures. Half-times of the markers ^{51}Cr and ^{144}Ce, derived from fecal excretion curves, were related to various compartments of the digestive tract. Their results supported the use of a two-compartment model for digestion studies in sheep. Later studies by Quiroz et al. (1988) gave more credibility to the use of a two-compartment model. They found that different models were required to predict the passage of different-sized particles, requiring a different marker for each particle. Use of a single marker was sufficient, if the only interest was for fecal excretion. Moore et al. (1992) compared digesta kinetics in sheep fed either hay or a pelleted diet. Fecal output predicted by Cr was less when the pelleted ration was fed, and within 4-10% of actual fecal output. The other three markers (Yb, Er, Dy) did not give similar fecal output estimates. None of the markers gave consistent estimates of fecal output, when compared to total collection. An algebraic calculation of fecal output was compared to a one-compartment model, to determine if the more simple method was accurate (Galyean, 1993). Fecal output was calculated using the area under a curve formed from a pulse dose of Yb. The algebraic approximation method was accurate, as long as the marker could be recovered completely in the feces. The two-compartment model was usually the model of choice in most digesta passage studies. However, Thornley et al. (1995) used several models from previously published studies and found that a third compartment needed to be added to the model to describe the data. Mixing rate of the marker and diet form (i.e., hay versus hay and supplements), as well as other factors, affected how well the models fit the data.

When compared to ruminants, little research has been conducted on model development in horses. Fecal output was measured in grazing donkeys using Cr-EDTA (Lechner-Doll et al., 1992). Fecal output was 60% higher when donkeys were grazing dry pasture, than when grazing the same pasture during the growing season. However, retention time was not changed due to season. It was suggested that intake was increased when pasture digestibility was lower, and that gut fill was also higher. A two-compartment model, similar to that of Grovum and Williams (1973) was applied to horses (Bertone et al., 1989; Corino et al., 1992). This model separated the two compartments into the stomach and cecum (first compartment) and the colon (second compartment), although this separation does not make sense physiologically. In yet another study conducted on horses, the fit of a monoexponential equation to predict fecal output was compared to a two-compartment model (Holland et al., 1998). Horses were dosed with Cr daily for 12 d, with Cr measured in feces from before dosing began until the end of the trial. The data did not support the use of a second term, but the inclusion of a delay into the model, representing the time required for the marker to enter the prefecal mass, did give a good fit.

Use of Internal Markers for Digestibility

Lignin. Lignin has been used as an internal marker for digestibility since the late 1800's (Kotb and Lucky, 1972). It was not until the mid 1900's that researchers observed possible digestion or degradation in the digestive tract of ruminants, making it unsuitable as a marker (Fahey and Jung, 1983). Another complication with using lignin is its low concentration in more immature plants and the variability of lignin content of different plants. Although this may not pose a problems when animals are fed in stalls or pens, grazing livestock may selectively choose plants that have different lignin content than pasture samples used for evaluation.

Despite these problems, lignin is still used as an internal marker. Some researchers determined that the method of determining lignin was the culprit, so alternative analysis methods were examined (Jung et al., 1997). Acid detergent lignin and Klason lignin determinations gave similar estimates of dry mater digestibility. The Klason method did find higher levels of lignin in the forages, suggesting that the acid detergent method may cause losses of acid-soluble fractions.

Permanganate lignin was shown to underestimate digestibility by 12-18% (Shelle, 1979; Schurg, 1981). The addition of alkaline hydrogen peroxide incubation appeared to improve the recovery of lignin from plants and feces (Cochran et al., 1988). Digestibility estimates in this trial were similar to total collection estimates when sheep were fed either immature or dormant grasses. In a later digestion trial using lambs, alkaline hydrogen peroxide lignin gave variable digestibility estimates, even though lignin recovery was calculated to be near 100% (Momont et al., 1994).

Acid insoluble ash. Acid insoluble ash gave predictions of digestibility similar to weighed digestibilities in horses (Schurg, 1981) and sheep (Van Keulen and Young, 1977). However, in another study using sheep, acid insoluble ash consistently overestimated dry matter digestibility (Penning and Johnson, 1983a). Digestibilities were also overestimated when ash was used to predict digestibility in ponies, horses and white rhinoceros (Frape et al., 1982). Acid insoluble ash was used to analyze diet digestibility in pigs (McCarthy et al., 1974). Silica had to be added to the feed to increase the level of ash, and improve the digestibility predictions. Although a convenient method in housed animals, this would not be possible in grazing livestock. Sources of error when using ash were evaluated by Thonney et al. (1985). Digestibility predictions were similar to total collection when diets were high in forages containing high levels of silica. High grain diets, containing total ash contents of less than .75% were not predicted well. In a study using cattle being fed diets with varying levels of grain and hay, acid insoluble ash was found to be the best predictor of dry matter digestibility (Huhtanen et al., 1994). Other methods used in this trial (neutral detergent fiber, acid detergent fiber and various lignin fractions) did not predict digestibility accurately.

Rare Earths. Rare earth elements have been used as internal markers for digestibility predictions. Usually, they are added to a feed to increase the total amount in the diet and facilitate analysis. The two elements, Sm and La, were not suitable as markers when they were applied to soybean meal, cottonseed meal and ground corn (Crooker et al., 1982). Apparently, these tracers disassociate from the feeds and pass through the digestive tract more rapidly than the feed. Other research found minimal detachment in the rumen of the elements Yb, Er, Dy, Sa and Y, which were applied to different sized corn particles (Turnbull and Thomas, 1987). This probably indicates a difference in marking methods. Rare earth elements can be affected by time of feeding (Pond et al., 1989); therefore, fecal collections need to be taken at similar times after feeding to use these markers. It might be possible, with more accurate laboratory methods, to use rare earth elements as internal markers without adding them to the feed, as would be required in pasture situations.

Fibers. Indigestible acid detergent and neutral detergent fiber have both been used to estimate dry matter digestibility. Acid detergent fiber predicted digestibility of several forages in sheep (Penning and Johnson, 1983b), although it was mentioned that it needed to be tested with more plant species. Acid detergent fiber was also used, with success, in another experiment with sheep and steers (Nelson et al., 1990). Neutral detergent fiber has been used successfully in horses (Meacham, 1987; Moffitt, 1987).

Alkanes. Odd chain n-alkanes, components of the plant cuticular wax, are one of the newest internal markers used for digestibility prediction. Alkanes are long chain hydrocarbons, and have been found to be relatively indigestible in ruminants. When chain lengths 29, 31 and 33 were used in sheep, however, they did not give accurate estimates of digestibility (Casson et al., 1990). Concerns mentioned were incomplete recovery of alkanes in feces, and differences in alkane content of stems and leaves of plants. The use of C31 alkanes as a digestibility marker was also evaluated in dairy cattle (Ohajuruka and Palmquist, 1991). Recovery of the alkane was incomplete, leading to underestimation of dry matter digestibility in alfalfa and grass hays. N-alkanes have also been used recently to estimate digestibility in horses as well (Ordakowski, 1998). Unlike in ruminants, recoveries of all chain lengths are similar, indicating that losses of alkanes may be associated with the more complex stomach found in ruminants. It appears that recoveries of alkanes are similar for similar chain lengths. It may be possible to dose an animal with an external (or even-chain) alkane, which occur naturally in pasture in very low amounts, and then use it for estimation of fecal output. A closely related odd-chain alkane (internal marker) could then be used for digestibility estimation. This method should allow for a better estimate of intake (see below), because similar recoveries of alkanes would cancel each other (Dove and Mayes, 1991).

Studies with multiple markers. Researchers have evaluated several different internal markers in the same study in an attempt to find the best internal marker. Insoluble acid detergent fiber, insoluble neutral detergent fiber, acid insoluble ash, acid lignin, permanganate lignin, and acid detergent fiber insoluble ash were compared in a study using lambs and alfalfa hay (Undersander et al., 1987). Acid detergent fiber insoluble ash and acid insoluble ash were the best predictors of dry matter digestibility. Another study examined the four markers: insoluble acid detergent fiber, insoluble neutral detergent fiber, acid detergent lignin and acid detergent fiber with cellulose incubation (Cochran et al., 1986). In this study, cattle were fed either cubed alfalfa hay, tall fescue, tall wheatgrass hay plus soybean meal, and prairie hay. None of the markers gave estimates similar to apparent digestibilities when used with young or freshly harvested forages. Insoluble acid detergent and neutral detergent fibers gave acceptable estimates, with the alfalfa cube and tall wheatgrass plus soybean meal diets. However, permanganate insoluble ash, insoluble neutral detergent fiber, and lignin were unsuitable markers for high concentrate diets, but insoluble acid detergent fiber was a suitable marker (Tamminga et al., 1989). These same internal markers were evaluated to predict digestibility in beef steers (Sunvold and Cochran, 1991). Acid insoluble ash gave similar digestibility estimates when grass hays were fed. Lignin was found to be a suitable internal marker for both grass and legume hays.

Eleven different digestibility techniques were evaluated in a study using the diets: tall fescue hay, alfalfa hay, limit-fed tall fescue hay, limit-fed alfalfa hay, tall fescue hay plus soybean meal and alfalfa plus rolled corn, in rams (Judkins et al., 1990). Markers evaluated were: neutral detergent fiber, acid detergent fiber, acid detergent lignin, alkaline hydrogen peroxide treatment before acid detergent fiber analysis, and alkaline hydrogen peroxide treatment after acid detergent fiber extraction. All markers were compared to in vitro and in situ nylon bag methods. As would be expected, in vitro and in situ methods accurately estimated digestibility, however, none of the internal markers consistently predicted digestibility on the various diets.

Summary. It becomes apparent after reviewing the literature that there is still not one single internal marker that accurately predicts digestibility across different diets. Valid internal markers are still needed to estimate digestibility when feed consumed and fecal output cannot be weighed, as in grazing situations. One of the biggest concerns with internal markers is that most are available in only minute amounts in forages, magnifying any errors due to analysis. Many internal markers, such as silica and acid insoluble ash, are easily contaminated if soil is present on the forage or fecal samples, or if an animal consumes some soil intentionally. Plant wax alkanes are one of the best options as an internal marker.

Use of External Markers for Fecal Output

Chromium. Chromic oxide is the most commonly used external marker in nutrient digestion studies (Titgemeyer, 1997). It is most useful as a total digesta tracer, because it does not readily associate with either the fluid or particulate phase of the digesta. One of the biggest concerns with the daily use of Cr is the diurnal excretion rate. This error can be reduced with more frequent dosing and fecal sampling.

In a comparison between Yb and Cr passage rates, a difference was noted due to feeding frequency(Coleman et al., 1984). Steers were fed either two meals daily or had access to feed continuously (fed every 2 h). A pulse dose of both Cr and Yb were given intraruminally, and passage was measured in fecal samples. Dry matter intake was not affected by time of feeding, and similar passage rates of both markers were observed when animals were fed twice daily. However, when steers were continuously fed, the Cr had a slower passage rate than the Yb, which could lead to errors in fecal output estimates. The size of the particles associated with a marker can also affect passage rate (Bruining and Bosch, 1992). Cows were pulse-dosed with Cr adhered to three different sized NDF particles (<.3mm, .6-1.0 mm, and 15-25 mm) and passage of the marker was analyzed in the feces. Passage rate decreased as the size of the particle increased, indicating that Cr was probably marking passage of a certain-sized particle, and not the entire particulate phase. As noted in other studies (Ehle et al., 1984;Ramanzin et al., 1991), this indicates that Cr is probably a good marker for whole-tract kinetics, but not specific phases.

One of the earliest studies to evaluate the diurnal variation of Cr excretion was conducted in grazing cattle (Smith and Reid, 1955). In the first trial, cattle were dosed with Cr in a capsule at 700 h. Rectal grab samples were taken every 2 h on the second and sixth d of dosing. The pattern of Cr excretion was similar for all animals, with the lowest concentration being in the early afternoon and the highest excretion occurring in the evening. An interesting note was that cows had a more evenly distributed rate of excretion than steers, possibly indicating a difference in grazing behavior. When dosing was increased to twice daily, fecal samples were taken only at the time of dosing (600 and 1600 h). Marker concentration was highest in the 600 h samples, but the authors felt that the difference in Cr was a "compensatory" relationship and that recovery of Cr was still near 100%.

When diurnal variation was evaluated in horses, doses of Cr were given twice daily, at 700 and 1900 h (Haenlein et al., 1966a). Diet, either pelleted, wafered hay or loose hay, did not affect the excretion of Cr; for all horses, the lowest excretion was found in early morning (before 700 h) and the highest level was observed at about 2400 h. Another study conducted in horses found diurnal variation on Cr concentration when Cr was dosed once daily at 800 h (Cuddeford and Hughes, 1990). The diurnal variation differed in this trial than in the cattle study mentioned above; lowest Cr excretion was in late evening (around 2000-2200 h) and highest was observed just a few hours after dosing. This could be due to how the Cr was dosed; adhered to forage versus inside a capsule.

One of the ways to overcome the diurnal variation in Cr excretion is to give the dose in a continuous release bolus. The bolus is given either intraruminally or via a balling gun. Fluid within the rumen dissolves the protective wrapping so that the bolus will begin to release the Cr marker. It is assumed that the marker will be released at a constant rate, and that the bolus will remain in the reticulo-rumen. These boluses are applicable in ruminants, but are not designed for use in nonruminant herbivores, such as the horse (personal communication, Mitch Venning, Nufarm Ltd., Otahuhu, Auckland, New Zealand).

In early studies on the continuous release bolus, Cr levels reached a steady state in 5-6 d, and remained more consistent than with twice daily dosing of chromium marker (Harrison et al., 1981; Ellis et al., 1982). Later studies evaluated its use as a predictor of fecal output. The continuous release bolus gave similar estimates of fecal output when compared to total collections in stalled sheep (Laby et al., 1984). No variation in Cr release was noted regardless of age, diet or housing. These results were confirmed in other studies (Parker et al., 1989; Hatfield et al., 1991a). Other studies have not shown the same success. Diurnal variation in the release of Cr was observed by Graham et al. (1990), when the bolus was used in cattle. There were also individual animal differences. Another study evaluated the prediction of fecal output using the bolus versus total collection (Hatfield et al., 1991b). The bolus did not give accurate estimates of fecal output in stalled wethers, but was acceptable in grazing wethers. This indicates that housing and related feeding methods could affect the reliability of the continuous release bolus. One of the possible reasons for errors in fecal output estimations is the actual release of Cr versus what is predicted by the manufacturer (Brandyberry et al., 1991; Buntinx et al., 1992). Type of diet offered also affects release of the marker; the bolus works better when diets are mostly forage than when supplements or pelleted rations are used (Luginbuhl et al., 1994b). It appears that the release rate needs to be validated before the bolus is used in experimental conditions (Momont et al., 1994).

In a study comparing fecal output predictions from a pulse-dose of Cr versus a continuous infusion of Co, Cr and Co gave similar estimates (Luginbuhl et al., 1994a). Although they did not have total collections, other studies (Brandyberry et al, 1991) have shown Co to give accurate estimates of fecal output, implying that the pulse-dose of Cr gave an accurate estimate as well. Other trials have compared single doses of Cr and Yb with total collection (Moore et al., 1992). In this trial, Cr overestimated fecal output when sheep were fed hay, but underestimated output when they were fed a pelleted diet. In the same trial, Yb overestimated fecal output on both diets. In another study where Yb and Cr predictions of fecal output were compared, both underestimated fecal output when compared to total collection (93.4% versus 77.3%, of weighed output, respectively).

Ytterbium. Ytterbium has been used to estimate fecal output as a pulse-dose, a once-daily dose, and as a constantly-infused marker in sheep (Hatfield et al., 1990). The once daily dose gave more accurate estimates of fecal output than the pulse dose, when compared to total collection. However, the pulse-dose procedure gave more precise estimates, over several different housing regimens, but not in pasture situations. One concern in this study was that the bolus used to release the Yb was not releasing at a constant rate. This could have been the reason for the loss in precision. Similar problems were observed with continuous-release boluses for Cr trials.

In other studies, a pulse-dose of Yb gave accurate estimates of fecal output in stalled animals (Krysl et al., 1985; Galyean, 1993). In a follow-up trial by Krysl et al. (1988) Yb and Dy both overestimated fecal output by 15-20% when sheep were consuming prairie hay, but Yb gave a much smaller overestimate (2.5%) when sheep were consuming lucerne hay; the Dy overestimated fecal output by 50% when sheep were consuming lucerne hay. This indicates that type of diet again affects passage of dosed markers. It is also possible that time of dosing, compared to time of fecal grab sample, may influence prediction of fecal output (Prigge et al., 1981). Cattle were dosed twice daily (800 and 1600) with both Cr and Yb, and fecal samples were collected at the same times. When the two fecal samples were combined, both markers underestimated fecal output; however Cr predicted fecal output well, when just the 1600 h samples were used, indicating variation in the excretion of Cr.

A continuous release bolus has been developed for Yb and was evaluated from fecal output prediction in cattle fed either alfalfa hay or alfalfa hay plus a concentrate (Estell et al., 1990). Predictions of fecal output were 11-22% higher than weighed output (similar to previously reported range of 10-30% in grazing animals), and excretion of Yb (as evaluated in fecal samples) varied throughout the day. Part of this error was attributed to poor estimates of Yb release.

Granulated polyamide. A relatively new marker with potential for predicting fecal output is granulated polyamide (Mahler et al., 1997). These small plastic particles are administered to animals in gelatin capsules, and have been used both in a pulse-dose and a continuous dose study. Fecal samples are air-dried and then diluted to a constant volume, or ground. Granules are then recovered from the samples and weighed. One of the benefits of this marker is that no expensive lab equipment is required. Individual animals could be dosed with different colored particles so that samples could be taken from the ground, instead of taking rectal grab samples. There is some diurnal variation in the excretion of the particles, possibly due to meals or grazing time. Another concern not addressed in this article was the effect of the granules on pastures. These granules would not be biodegradable and could possibly accumulate on pastures, leading to contamination in future trials.

Summary. Although markers such as Cr and Yb, have shown potential over the years as external markers for fecal output predictions, methods need to be developed that over come some of the more common problems, like diurnal variation. One of the other concerns of dosing with external markers is the inconvenience of handling animals several times a day, and the potential stress of either using a balling gun or restraints to use a ruminal fistula one must overcome in livestock. The extra handling required may also upset the normal feeding behavior of the grazing animal. The use of a continuous release bolus could overcome this problem, in ruminants, if release rate of the marker can be refined. One advantage of horses is that they are usually trained and are used to being handled by humans. If markers can be put into a small amount appetizing of feed, the time required for dosing grazing horses would be minimal. Dosing would have to be several times a day, since continuous-release mechanisms are not applicable in horses.

Use of Markers in Combination for Intake Estimates

Alkanes. Reviews by Dove and Mayes (1991; 1996) give good summaries of how alkanes can be utilized to determine herbage intake. What follows here is a brief summary of recent research results.

One of the most promising uses of alkanes is that dosed even-chain and odd-chain alkanes inherent in forage can be used together. Studies of alkanes have shown that recoveries are improved as the chain length increases, hence a C33 alkane will have recovery closer to 100% than a C25 alkane (Mayes et al., 1986). The longer chain lengths, 29-33, are the most abundant in pasture plants. Another observation has been that markers of similar chain length have similar recoveries, and this factor indicates fecal output would not need to be estimated, because recoveries of the two markers would cancel in the calculation of intake. The paired alkanes, C32 and C33, accurately estimated herbage intake in sheep fed either fresh-cut forage or the same forage plus a barley-based concentrate (Mayes et al., 1986). The loss of alkanes, resulting in incomplete recoveries, has been attributed to absorption within the small intestine, but absorption of adjacent chain-lengths (i.e., 32 and 33) were similar (Mayes et al., 1988).

Several different even-and odd-chain length alkanes were compared in a study using mature sheep consuming fresh-cut herbage (Vulich et al., 1991). There was no difference in fecal recoveries of the chain-lengths 31, 32 or 33. Combinations of C31 and C32, and C33 and C32 gave similar estimates of dry matter intake, and these intake estimates were within 5% of the weighed intake. These pairs of alkanes also gave precise estimates of intake over several days.

Chromic oxide and various internal markers. Intake estimates may best be used for comparison between pastures/diets, not necessarily as a quantitatively correct value. Pasture intake was determined in pregnant and lactating mares by using chromic oxide and acid insoluble ash (Martin et al., 1989). Predicted digestible energy intakes were 5-20% less than expected according to the NRC (1989). The underestimation of intake could be due to the type of markers used. It is also likely that the recommendations in the NRC handbook (1989) actually underestimate maintenance requirements for horses maintained on pasture. Chromic oxide and acid insoluble ash were compared to chromic oxide and permanganate lignin for predicting digestibility of nutrients in horses (Schurg, 1981). Chromic oxide and ash predicted results within 4% of total collection digestibilities. Lignin consistently underestimated digestibility by 10-20%.

Using a chromic oxide release bolus in combination with alkaline peroxide lignin gave erroneous results on intake prediction (Momont et al., 1994). The chromic oxide bolus predicted fecal output accurately, but lignin underestimated digestibility, leading to an underestimation of dry matter intake.

Environmental impacts on digestion and intake in livestock

Diet. The effect of the addition of concentrate to an animal's intake and nutrient digestibility has been investigated in livestock. In a comparison study of horses and sheep, neither the amount of feed offered nor the addition of concentrates at several levels influenced diet digestibility, indicating limited forage:concentrate interactions (Martin-Rosset and Dulphy, 1987). In contrast, in sheep there was a decrease in total diet digestibility as amount of forage offered increased, and a decrease in forage digestibility as the level of concentrate offered increased (Martin-Rosset and Dulphy, 1987). Other studies have indicated that level of feeding can change diet digestibility. Increasing the amount of hay alone increased the apparent digestibility of fat, protein and P. However, when a portion of the diet was corn, and the level of corn was increased, dry matter, fat and protein digestibility were decreased (Ott, 1981). The addition of a supplement to the diet of grazing cattle decreased total grazing time, but increased overall diet digestibility, probably due the quality of the supplement (Krysl and Hess, 1993).

Form of the forage offered can affect intake and digestibility. Horses and sheep both consumed more hay when it was pelleted (24% and 73%) and when it was wafered (17% and 9%), than when it was fed in loose form (Haenlein et al., 1966b). In a more recent study, comparisons were made between the digestibility and intake of pelleted, cubed, chopped and long-stem alfalfa hay (Todd et al., 1995a). There was no difference in the digestibility of any forms of hay, however, the voluntary intake of the cubed hay was higher than for any other form. Alfalfa hay was offered at two levels (maintenance and 1.4x maintenance) to mature horses. Level of intake did not affect digestibility of dry matter, protein or gross energy (Todd et al., 1995b).

Season. Effects of season on forage preference were evaluated in grazing cattle and sheep (Bedell, 1968). During most of the year, sheep preferred to consume clover over grasses. Fescue was preferred during the summer when clover was dry. Cattle always showed a preference for available grasses. Ruminants consumed less fall and winter pasture than spring and summer pasture (Reed, 1978). Observed grazing differences could be due to the rate of breakdown of the forage in the reticulo-rumen; fall and winter forages are retained for a longer period. Intake of pasture was reduced during winter months in cattle, even though estimated requirements were increased (Adams et al., 1986; Johnson et al., 1998). Availability of pasture also affects intake (Woodward, 1997). Cattle and sheep were observed grazing for a shorter period of time and ruminating for longer periods when pasture was abundant. Horses grazed longer than cattle on the same pasture (Pond et al., 1995). Time spent grazing increased for both cattle and horses as available pasture decreased.

Taste preferences. Horses showed preferences for pasture mixes high in clover during spring and summer months. Perennial ryegrass and timothy also had high preference levels (Archer, 1973). In a follow-up study, red fescue and tall fescue were the most palatable during the fall. The palatability of perennial ryegrass decreased during this time of year (Archer, 1978). Sheep were observed to walk a distance for a good quality forage, instead of consuming a poor quality forage (Dumont et al., 1998). Ewes showed more reluctance to walk when the amount of available good quality forage was decreased.

Metabolic influences. Glucose administered intravenously delayed the initiation of feeding activity in ponies (Ralston and Baile, 1982a). Glucose administered via nasogastric tube also delayed to onset of feeding activity, although delay was dose-related (Ralston and Baile, 1982b). Changes in cecal volatile fatty acid content may also influence intake in horses (Ralston et al., 1983). Some factors that may affect voluntary intake in ruminants include: volatile fatty acid level, ruminal distension, and physical attributes of the feed (Forbes, 1996). Models have been proposed that may predict intake in ruminants (Allen, 1996; Illius and Jessop, 1996).

Future Studies

One of the reasons for conducting balance studies in grazing animals is to evaluate effects of different diets on the environment, especially to compare outputs of nutrients, such as N and P, that can adversely affect waterways. Another reason to evaluate pasture intake is so that new feeds can be designed that meet the optimal nutritional needs of the animals, without providing excesses or deficiencies that can lead to developmental, reproductive or performance problems.

Once models are perfected that give accurate and precise estimates of forage intake, new tracers can be applied to the concentrates, so that individual supplement intake can be measured, as well. This is especially important in feeding situations where animals are group-fed, and individual intakes cannot be determined. This should be possible with two separate markers, one for pasture and a second for the supplement, as well as simultaneous equations. Chromic oxide (Lobato et al., 1980), ytterbium (Curtis et al., 1994), tritiated water (Dove, 1984) and n-alkanes (Dove and Mayes, 1991) have been tried for this purpose in both grazing and penned sheep. Results appear promising.

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Objectives

The general objective was to develop and evaluate methods for determining pasture intake, digestibility, and fecal output using internal and external markers on two different pastures and two hay mixtures.

Achievement of this goal required the following:

1. Evaluate the use of chromium (Cr) as an external marker to estimate fecal output, and to compare Cr estimates with total fecal collection.

2. Evaluate the use of n-alkanes and yttrium as internal markers.

3. Test these marker methods in both stall and pasture situations.

4. Evaluate the use of total fecal collection harnesses in pasture situations.

5. Compare consumption and digestion of two pastures: bluegrass / white clover and tall fescue / alfalfa; and two hays: tall fescue / alfalfa and orchardgrass / alfalfa.

Chapter 1. Calculation of Fecal Kinetics in Horses Fed Hay or Hay and Concentrate[1]

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ABSTRACT

Marker methods are needed for estimating fecal output by grazing animals in studies of nutrition and environmental impact. In addition, estimates of prefecal mass and turnover time are relevant to exercise performance and certain digestive disorders. As a first step in developing marker methods for field use, a chromic oxide model of fecal kinetics was developed and tested in the context of a digestion balance experiment with stall-fed horses. The model consists of removal of feces at a constant rate from a single compartment, the prefecal mass. Four horses were fed hay, and another four were fed hay and concentrate. Balance-marker experiments were conducted for 10 d, following 7 d of adaptation. A dose of chromic oxide mixed in chopped hay and molasses was administered from a nose bag at 0700 daily for 10 d. Dry matter and Cr were measured in feeds and feces. Fecal Cr concentration (C, mg/kg DM) varied during the day, so data from total daily collections were used for model development. These fecal Cr data (C_t) at time t (days) were fitted to a single exponential, with one rate constant (k), rising to an asymptote (C_a): $C_t = C_a - C_a \cdot e^{-kt}$. Superior fits were obtained when a delay (d) was introduced between the pulse oral dose and the entry of marker into the prefecal pool: $Ct = C_a - C_a \cdot e^{-k(t-d)}$. Using pooled data, delays of 2.7 and 2.0 h gave best fits (highest estimates of R2) for pooled data from horses fed hay or hay plus concentrate, respectively. The model generated estimates of 3.4 and 3.8 kg/d of DM for fecal outputs (dose/C_a) of horses fed hay or hay and concentrate, respectively. The rate constants yielded turnover times (1/k) of 33 and 18 h, and prefecal masses [(dose/C_a)/k] of 4.6 and 2.9 kg of DM for hay or hay and concentrate groups, respectively. Using data from individual horses, mean estimates for each diet were similar to corresponding values for the pooled data. In balance experiments, feces collected were 3.7 and 4.4 kg/d, and Cr recoveries were 108 and 115% dose for the hay and hay plus concentrate diets, respectively. Marker estimates (M) were correlated with total collection estimates (T) of fecal output [M = T(.890 ± .045); r = .70, P = .041]. Adjusting for recovery improved the regression coefficient to1.009 ± .028 (r = .87, P = .002). The findings suggest that, if Cr doses are more frequent than daily and if Cr inputs other than dose can be eliminated, this method should give accurate and precise estimates of fecal output.

Key Words: Feces, Turnover, Feces Composition, Chromium

Introduction

The fecal output of grazing animals is relevant to environmental contamination, feed intake, digestibility, water balance, power:weight ratio and exercise performance, and certain digestive disorders (Martin et al., 1989; Oldham and Tamminga, 1995; Kronfeld, 1996). Fecal kinetics can be represented as a simple model, the removal of feces at a constant rate from a thoroughly mixing prefecal mass or compartment, and quantified by markers or tracers. Following the administration of a known dose of a marker or tracer into a system, a series of samples is taken, the temporal response of the marker concentration is observed, and the data are subjected to mathematical analysis (Brownell et al., 1968). The flow rate of the marked material can be calculated from the magnitude of the response (noncompartmental analysis). In addition, rate constant(s), hence turnover time(s) and exchanging mass(es) or compartment size(s), can be calculated from the shape of the response (compartmental analysis). Noncompartmental and compartmental analysis have been applied to marker studies in ruminants and horses (Blaxter et al., 1956; Grovum and Williams, 1973; Orton et al., 1985; Pond et al., 1988; Bertone et al., 1989). Our objective was to develop a simple, convenient and mathematically correct method for estimating fecal kinetics in grazing animals, and the first step involved the testing a marker model in the context of a digestion balance experiment done on stall-fed horses.

Materials and Methods

Eight Thoroughbred horses, aged 2 to 10 yr, were housed in 14-m^2 box stalls with sealed cinder block walls and asphalt floors and were hand-walked daily for 15 min to provide limited exercise. Four horses were fed a mixture of orchardgrass and alfalfa hay

(**HAY**), at a rate of 2% of BW per day. The other four were fed the same hay plus 3.0 kg/d of pelleted concentrate (**H&C**, PurePride 200, Purina Mills, St. Louis, MO that contained processed grain by-products, grain products, roughage products, molasses products, calcium carbonate and salt). Feeds were offered at 0700 and 1600. Orts were collected and weighed each morning. Horses were adapted to the diets for 7 d, after which combined balance and marker experiments were conducted for 10 d.

Feed samples were taken every second day for 10 d. They were weighed, dried and combined, and duplicate samples were submitted to the Virginia Tech Forage Testing Laboratory, which follows standard procedures (Robertson and VanSoest, 1977; AOAC, 1990), for analysis of CP, ether extract (**EE**), ash, ADF and NDF (Table 1). Assays of NDF in the concentrate were not repeatable, so concentrate NDF was calculated from ADF using an equation based on data for barley, corn, oats, wheat and soybean meal (NRC, 1989): NDF = 6.81 + 1.15 x ADF (SE = 2.11; $r^2 = .81$, $P = .014$). Then the NDF for H&C was calculated as a weighted average. Nonstructural carbohydrate (**NSC**) was calculated by difference: NSC = 100 – %CP – %EE – %NDF – %ash. The marker, 27.4 g of Cr, in the form of 40 g of Cr_2O_3 (Chromic Oxide Sesqui; Fisher Scientific, Fair Lawn, NJ) was thoroughly mixed into 310 g of the hay (chopped to 2 cm) and molasses mixture. This mixture was offered in a nose bag and completely consumed immediately before the morning feed was offered.

Feces were collected every 2 h, and each day's output was weighed and mixed thoroughly. A representative daily sample (10% of wet weight) was dried in a forced-air oven. On the last day, diurnal fecal samples were collected at 0600, 1200, 1800 and 2400 from two horses in each group. These daily and diurnal fecal samples were used for Cr analysis (see below). Dried fecal samples were pooled for the 10 d and duplicate samples were submitted for proximate analysis by the Virginia Tech Forage Testing Laboratory (Robertson and VanSoest, 1977; AOAC, 1990).

For Cr analysis, fecal samples were collected as above. Also, samples of the hay, concentrate, and Cr_2O_3 mixture were collected every 2nd d and were weighed and dried.

All samples were ground to pass a .5-mm screen in a Cyclone Mill (UDY Corporation, Fort Collins, CO) and analyzed for Cr by atomic absorption spectrophotometry using an air-acetylene flame after digestion with nitric and perchloric acids (Sandel, 1959). Two dilutions of each Cr sample were analyzed in duplicate, and five dilutions of a standard solution of potassium dichromate were used for calibration.

Digestible DM (**D**, %) was calculated from the balance data for daily intake (**I**, kg of DM) and daily fecal output (**FO**, kg of DM):

$$D = 100(I - FO)/I$$

Digestibilities of CP, EE, ash, NSC, NDF and ADF were calculated similarly from the balance data.

Fecal Cr concentrations (**C_t**, mg/kg DM) at time *t* (days) were analyzed separately for each horse (labeled A through H) and pooled for those fed HAY (A through D) or H&C (E through H). The pooled data were used for the initial model development, in which the C_t data were fitted to a monoexponential rising to an asymptote (C_a):

$$C_t = C_a - C_a \cdot e^{-kt}$$

using a graphics and curve-fitting program (SlideWrite Plus 3.0 for Windows, Advanced Graphics Software, Inc., Carlsbad, CA). Estimates of C_a were used to calculate daily fecal output (**FO**, kg/d):

$$FO = (\text{Cr dose})/C_a$$

The rate constant (k, d^{-1}) was used to calculate its reciprocal, the turnover time (**TT**, d), and hence to calculate the compartment size or prefecal mass (**PFM**, kg):

$$PFM = FO \times TT = FO/k$$

This equation defines the PFM in terms of tracer kinetics (rather than as any mass defined by anatomical location, although the PFM probably resides partly or entirely in the large bowel).

Further development of the model concerned the delay between oral administration of marker and its entry into the PFM (Figure 1), following procedures for tracer kinetics (Brownell et al., 1968; Kronfeld and Ramberg, 1981). Then the model was applied to the separate data for each horse. Data and model parameters were summarized as means and standard errors. Effects of diets were compared by non-paired *t*-tests.

Results

Balance Experiments. Mean intake of DM was 18% greater and mean fecal output was 20% greater for horses in the H&C group than for those in the HAY group (Table 2). When expressed per unit of BW, however, no differences between HAY and H&C groups were found for DM intake or fecal output. Apparent digestibility of DM was unchanged by diet, but it was higher for NSC and lower for NDF in horses fed concentrate (Table 3). Fecal DM expressed as a percentage of wet weight was 18.5 ± .59% in the HAY group and 21.9 ± .82% in the H&C group ($P = .003$); corresponding fecal wet weights were 19.9 ± 1.14 and 20.2 ± 1.09 kg/d.

The Cr provided was .153 ± .094 g/d in HAY, .116 ± .058 g/d in H&C, and .557 ± .003 g/d in the marker carrier in addition to the 27.4-g dose. Thus, the mean daily dose of Cr was 28.11 g for HAY and 28.07 g for H&C, and these estimates of Cr dose were used instead of 27.4 g in the kinetic analysis. Fecal Cr concentrations on the last day in four horses were 10.35 ± 2.72, 2.65 ± 1.21,1.07 ± .34 and 8.34 ± 1.49 mg/kg of DM at 0600, 1200, 1800 and 2400, respectively.

Model Development. In the marker experiments, mean C_t for the HAY group and for the H&C group on each day in the 10-d balance experiment were fitted to monoexponential curves rising to asymptotic values (Figure 2). Agreement between observed and predicted values was good; the R^2 were .93 and .87, and the F-values were 140 and 68 ($P < .001$), for HAY and H&C, respectively.

The delay (Figure 1) was represented by a separate compartment, and the C_t data were fitted to an equation with two exponential terms. Adjusted R^2 decreased from .93 and .87 to .90 and .84 for HAY and H&C, respectively; corresponding F-values declined from 140 and 69 to 32 and 19. The standard errors were larger than the means of parameters in the second term (data not shown). Hence, these data did not support a two-compartment model (Brownell et al., 1968), such as those proposed for sheep and horses (Blaxter et al., 1956; Grovum and Williams, 1973; Bertone et al., 1989).

The delay (*d*, d) was then represented by a decrease in time (*t*) in the monoexponential equation:

$$C_t = C_a - C_a \cdot e^{-k(t - d)}$$

This equation could not be solved by the curve-fitting program, so a series of values of *d* from .05 to .2 d (1.2 to 4.8 h) were included in the equation, and the best fit was found to two significant figures in eight iterations. More precision was obtained by fitting a quadratic equation to R^2 and *d*:

$$R^2 = a + bd + cd^2$$

and calculating the *d* corresponding to the peak value of R^2 (the best fit):

$$d = -b/2c$$

Delays were .1122 d (2.002 h) and .0834 d (2.693 h) for the HAY and H&C diets, respectively. Best fits to this model (Figure 1) were obtained with R^2 values of .95 and .90, and F-values of 158 and 77 ($P < .00001$), for the HAY and H&C diets, respectively. The model parameters and dependent estimates of FO, TT and PFM are listed in Table 4.

Individual Data. The C_t data for individual horses were fitted to the exponential model with or without the delay. In each case, inclusion of the delay conferred a slight advantage in regard to R^2 and F (Table 5), although the differences between mean estimates of C_a and k with or without the delay were less than one SE (data not shown). Similarly, approximations of C_a calculated as the mean C_t for the last five d of marker administration were 96.4 ± 1.8% of the true C_a and were within one SE of the mean C_a. Estimates of FO, TT and PFM using the delay model (Figure 1) are summarized in Table 6. Fecal output was unchanged by diet, but TT and PFM were approximately halved by the H&C diet.

Comparing the marker data to corresponding collection data, fecal outputs using the external marker were correlated with corresponding measures by collection (Figure 3a). The fecal recoveries of Cr based on C_a and total collections of feces were 108 ± 5% and 115 ± 5% for HAY and H&C diets, respectively. Adjusted estimates of FO calculated from Cr recovery, instead of dose (Blaxter et al., 1956; Titgemeyer, 1997), were closely related to total collections (Figure 3b).

Discussion

The results indicate that a one-compartment model can be used to represent fecal kinetics in horses. In addition, the findings suggest ways of improving data generation, especially for application of this model to grazing animals.

Balance Experiment. Apparent digestibility of DM was not improved by replacing hay with concentrate, which reduced ADF content by 19%. Because digestibility is usually increased by a reduction in dietary fiber, this result was surprising. Increased digestibility of NSC, which occurs mainly in the small intestine, was counteracted by decreased digestibility of fiber, which occurs mainly in the large intestine and was probably impaired by the decreased time allowed for fermentation of H&C (VanSoest, 1977). Feces were approximately 2% drier for the H&C diet, probably because the feces contained less fiber, which binds water.

The 79% coefficient of variation in fecal Cr concentration during the day confirmed previous findings in the horse (Haenlein et al., 1966). This variation emphasized the need for thorough mixing of the 24-h output in the present experiment and the need for more frequent dosing in field studies using grab samples.

Marker Experiment. The one-compartment model applies calculations more widely applied to tracer- and indicator-dilution than to marker experiments (Kronfeld and Ramberg, 1981; Merchen, 1988). It yielded good fits to the data. Previous studies of two-compartment systems in sheep, cattle, and horses have combined marker methods for estimating transit time and mean retention time with kinetic estimates of rate constants and, in some models, an age-dependent (time-dependent) rate function that substitutes for one rate constant (Blaxter et al., 1956; Grovum and Williams, 1973; Pond et al., 1988; Bertone et al., 1989). In most cases, SE of rate constants (or functions) have not been published, and in others large SE relative to k values have not led to model rejection. In our model development, a two-compartment model was rejected, mainly because estimated SE were greater than corresponding parameter estimates for the second compartment. Relatively large SE may devolve from insufficient or imprecise data. Alternatively, failure to determine a second mixing compartment may reflect the simpler digestive tract of horses. The rumen-reticulum, which serves as an initial mixing chamber, and which complicates kinetic analysis in ruminants, is absent in horses. The delay in the equine one-compartment model represents movement of the oral dose of marker to the PFM with no appreciable mixing. The consistent improvement in fits of the data supports the need for introducing the delay into the model.

In a typical tracer dilution experiment on a system in a steady state, the transport or flow rate is measured as a function of the magnitude of the temporal response, which is the integral of the tracer concentration in the sampled compartment as it changes with time (Brownell et al., 1968; Kronfeld and Ramberg, 1981). This magnitude can be calculated as the area subtended by the time-concentration curve following a pulse dose of tracer, or as the asymptotic concentration during a continuous infusion, the latter being the convolution integral of the former (Meier and Zierler, 1954). The area under the curve can be calculated from an exponential equation, approximated algebraically (Galyean, 1993), measured with a planimeter, or determined gravimetrically. The asymptotic concentration during a continuous administration can be calculated precisely from an equation, as here, or be approximated as an average plateau value (Merchen, 1988; Cuddeford and Hughes, 1990). In compartmental analysis (Brownell et al., 1968), additional information on the structure of the system is obtained from the shape of the temporal response by analyzing the whole curve and determining the rate constant(s), which can be used to calculate the turnover time and mixing mass or compartment size, as in the present model (Figure 1, Table 4).

The main assumption in the model is that chromic oxide particles are mixing thoroughly in a prefecal mass of material, which consists of water with potentially dissolved or suspended DM, from which the feces are removed and represent samples. The PFM probably resides mainly in the large bowel, but it was defined above in terms of the method of its determination (marker dilution) and does not necessarily coincide exactly with the contents of the cecum and large colon, which range from 3 to 7 kg of DM in horses (Alexander, 1972; Bertone et al., 1989). Note that this model assumes that the chromic oxide is not mixing in the delay component, which lies between the mouth and the PFM. In contrast, thorough mixing of the marker throughout the contents of the small intestine would be necessary for the estimation of digestibility and would probably call for an even distribution of the marker throughout the feed, for example, by mordanting Cr to fiber (Uden et al., 1980).

The flow of digesta has been differentiated into a two-phase system, particulate and fluid (Faichney, 1975). This partition is an approximation because particles of different sizes and fluid at different sites (e.g., close to or far from the gut wall) probably flow and mix at different rates. Chromic oxide forms small insoluble particles that may not flow like any other particulate species but are recovered as DM in the feces, so Cr has been regarded as a tracer or marker of prefecal and fecal DM in this model. The term *marker* may be preferred because Cr is not strictly a tracer, as usually defined (Brownell et al., 1968), of the nonhomogeneous DM in this system. Typical methods of tracer kinetics have been used to build this model, however, rather than simpler marker calculations commonly used in animal nutrition (Merchen, 1988). Chromic oxide was selected in part because it was the least expensive of likely external markers, and this method is being developed for eventual use on large numbers of grazing animals, so cost, convenience, and safety were considered.

Chromic oxide was used previously to estimate mean retention time, 38 h, in horses fed alfalfa hay (Vander Noot et al., 1967). Estimates of mean retention time of particles from hay using other markers have ranged from 23 to 36 h (reviewed by Orton et al., 1985). Similar estimates for mixtures of hay and concentrates have ranged from 23 to 33 h. Estimation of mean retention time has usually been based on marker appearance in feces (following Blaxter et al., 1956), and this estimate corresponds approximately to the delay plus turnover time in the present model, which would total 20 h for H&C and 35 h for HAY. The reduced time available for fermentation probably contributed to the decreased in apparent digestibilities of NDF and ADF in horses fed H&C, compared to those fed HAY.

The differences in PFM between the HAY and H&C diets, 1.76 kg DM for the pooled data (Table 4) and 3.01 kg DM in the individual horses, are approximately one-fifth to one-tenth of corresponding wet weights, 10 to 30 kg, of material residing presumably mainly in the large bowel. The impact of such a weight difference, 2 to 6% of BW, on the competitive performance of a sporting or recreational horse would be substantial, especially because such a horse would likely be consuming two to three times its maintenance requirements, and the dead weight of bowel contents would be commensurately higher (Kronfeld, 1996).

The estimated recovery of daily Cr output was approximately 3.3 g or 12% higher than the known daily Cr intake. A possible extra source of Cr would be coprophagy, which would tend to increase C_t but also to decrease FO, so that the calculated recovery of Cr in the feces may not be affected. The water was assayed for 26 minerals by the university's Facilities Department, and the Cr concentration was reported as < .01 (ug/ml. The excess Cr recovery would have required a Cr concentration of nearly 120 (ug/ml in 30 L of water intake, so the water does not seem to be its source.

The model estimates of FO correlated with collection estimates, and the coefficient of .89 reflects the higher C_a than would be expected from the recovery. When the dose is adjusted for the recovery, then the correlation is improved and the mean marker estimate equals the mean collection estimate of FO. This result suggests that if extraneous sources of Cr can be eliminated the present model should yield accurate and precise estimates of FO. The diurnal variation of C_t suggests that more frequent dosage of Cr would be necessary for application of the marker method in the field, where only a series of grab samples of feces would be available.

Implications

A simple model of fecal kinetics using chromic oxide as a marker was developed and tested in stall-fed horses. The results suggest ways to improve this approach for use in grazing animals. The method also allows estimation of a prefecal mass and its turnover time, which could be relevant to exercise performance.

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Table 1. Contents[a] of CP, ether extract, ash, NDF, ADF and Cr in DM of hay, concentrate, and a hay:concentrate mixture

Nutrient,	Hay	Concentrate[b]	H&C[c]
	------------------% DM[d]-----------------		
CP	13.8 ± .5	14.8 ± .1	14.1
Ether extract	1.4 ± .1	1.9 ± .2	1.5
Ash	9.2 ± .5	6.0 ± .1	8.2
ADF	32.0 ± .7	11.4 ± .3	25.9
NDF	60.1 ± .4	19.9 ± 2.1	46.2
NSC[e]	15.5	57.4	28.1
Cr	.018 ± .011	.0014	.013

[a] Analysis of CP, ether extract, and ash (AOAC, 1990); ADF and NDF (Roberston and Van Soest, 1977); chromium (Sandel, 1959).

[b] The concentrate was Pure Pride 200 (Purina Mills, St Louis, MO).

[c] H&C = hay and concentrate, a weighted average of hay:concentrate, 7:3.

[d] Mean ± SE, n = 4.

[e] NSC = nonstructural carbohydrate estimated by difference: NSC = 100 - (%CP + %Ash + %ether extract + %NDF).

Table 2. Daily DM intake and fecal output of four horses fed hay alone (HAY) and four fed a hay:concentrate mixture (H&C, 7:3)

Variable	HAY	H&C[a]	SEM	P
Weight, kg	474	529	21	.11
DM intake, kg/d	8.52	10.09	.31	.016
DM intake, g/kg of BW daily	18.0	19.1	.41	.13
Fecal DM, kg/d	3.68	4.42	.23	.062
Fecal DM, g/kg of BW daily	7.80	8.34	.35	.32

Table 3. Digestibilities (%) of DM, CP, ether extract, ADF, NDF and nonstructural carbohydrate (NSC) in four horses fed hay (HAY) and four fed a hay:concentrate mixture (H&C, 7:3)

Nutrient	HAY	H&C[a]	SEM	P
DM	56.8	56.2	4.7	.91
CP	69.3	63.0	2.1	.072
Ether extract	15	22	7.6	.54
Ash	42.7	36.8	4.0	.33
ADF	46.6	40.1	2.2	.084
NDF	53.2	42.2	3.1	.045
NSC	69.6	82.2	1.2	.001

Table 4. Estimates of fit of pooled fecal Cr concentration data (Figure 2) to a one-compartment model (Figure 1), model parameters, and calculated variables in horses fed hay (HAY) or a hay:concentrate mixture (H&C, 7:3)

Estimate	HAY	H&C
R^2	.946	.896
F	77	158
P- value	.0000	.0000
Cr_a, mg/kg[a]	8.32	7.42
SE	.3050	.3257
t-statistic	27.3	22.8
P-value	.0000	.0000
-k, d^{-1}[b]	.733	1.323
SE	.131	.403
t-statistic	5.58	3.28
P-value	.0003	.009
TT, h[c]	32.7	18.1
Delay, h[d]	2.69	2.00
FO, kg/d[e]	3.38	3.80
PFM, kg[f]	4.61	2.87

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[a] Cr_a is the asymptotic value of fecal Cr concentration.

[b] k is the rate constant.

[c] TT is turnover time of the prefecal mass, TT = -1/k

[c] Delay is the time between the administration of Cr and its entry into the prefecal mass

[d] FO is fecal output, FO = Dose of Cr/Cr_a.

[e] PFM is prefecal mass or mixing compartment, PFM = FO x TT.

Table 5. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment (Figure 1) in trials on eight horses

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.725	21.1	.0008	.750	23.9	.0005
B	.795	31.1	.0002	.807	33.5	.0001
C	.665	17.8	.0014	.702	21.2	.0007
D	.843	43.0	.0000	.860	49.2	. 0000
E	.588	12.9	.0043	.626	15.1	.0026
F	.738	25.3	.0004	.775	30.1	.0002
G	.755	27.8	.0003	.790	34.0	.0001
H	.627	15.1	.0025	.673	18.5	.0012
A to D[b]	.940	140.2	.0000	.946	157.7	.0000
E to H	.884	68.5	.0000	.896	77.5	.0000

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[a] Delay was 2.69 h for horses fed hay (A to D) and 2.00 h for horses fed hay and concentrate (E to H).

[b] Pooled data as in Figure 2.

Table 6. Fecal kinetics in four horses fed hay (HAY) and four fed a hay:concentrate mixtures (H&C, 7:3) estimated with a one-compartment model with a delay[a] between time of administration and entry of marker into the compartment

Variable	HAY	H&C	SEM	*P*-value
Fecal output, kg DM/d	3.39	3.82	.27	.36
Fecal output g/kg of BW daily	7.20	7.26	.54	.93
Turnover time, h	43.3	18.3	6.7	.042
Prefecal mass, kg DM	5.96	2.95	.81	.040
Prefecal mass, g/kg of BW daily	12.67	5.60	1.71	.027

[a]The delays were .1122 and .0834 d for HAY and H&C, respectively.

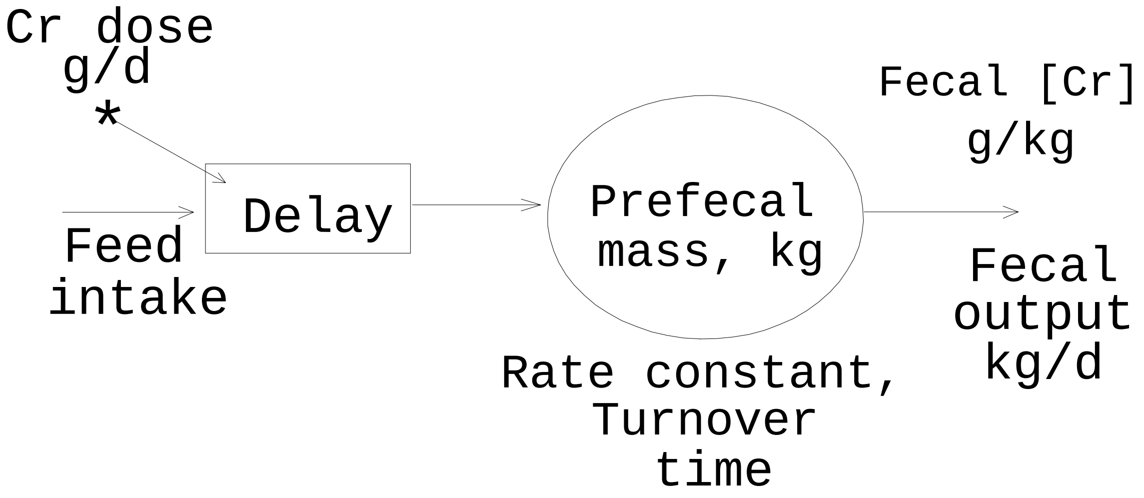

Figure 1. One compartment model of the prefecal mass (PFM, kg) or mixing compartment, and fecal output (FO, kg/d), with a delay (*d*, d) between oral administration of Cr dose (*) and the entry of this Cr into the PFM. The fecal Cr concentrations (mg/kg DM) at time *t* (d), C_t, rise to an asymptotic value, C_a, and can be used to determine a single rate constant, k (d^{-1}): $C_t = C_a - C_a \cdot e^{-k(t - d)}$.

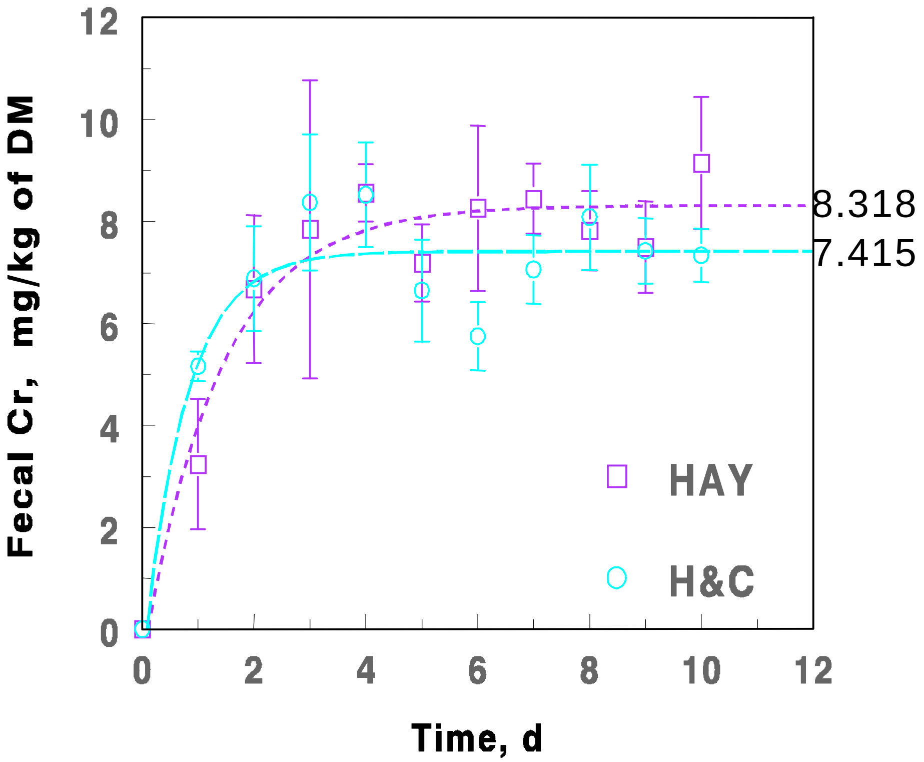

Figure 2. Mean daily fecal concentrations of Cr, C_t, for horses fed hay (HAY, open squares, SE flags) or hay-and-concentrate (H&C, closed circles) are plotted against time, and the data are fit to a one-compartment model (Figure 1).

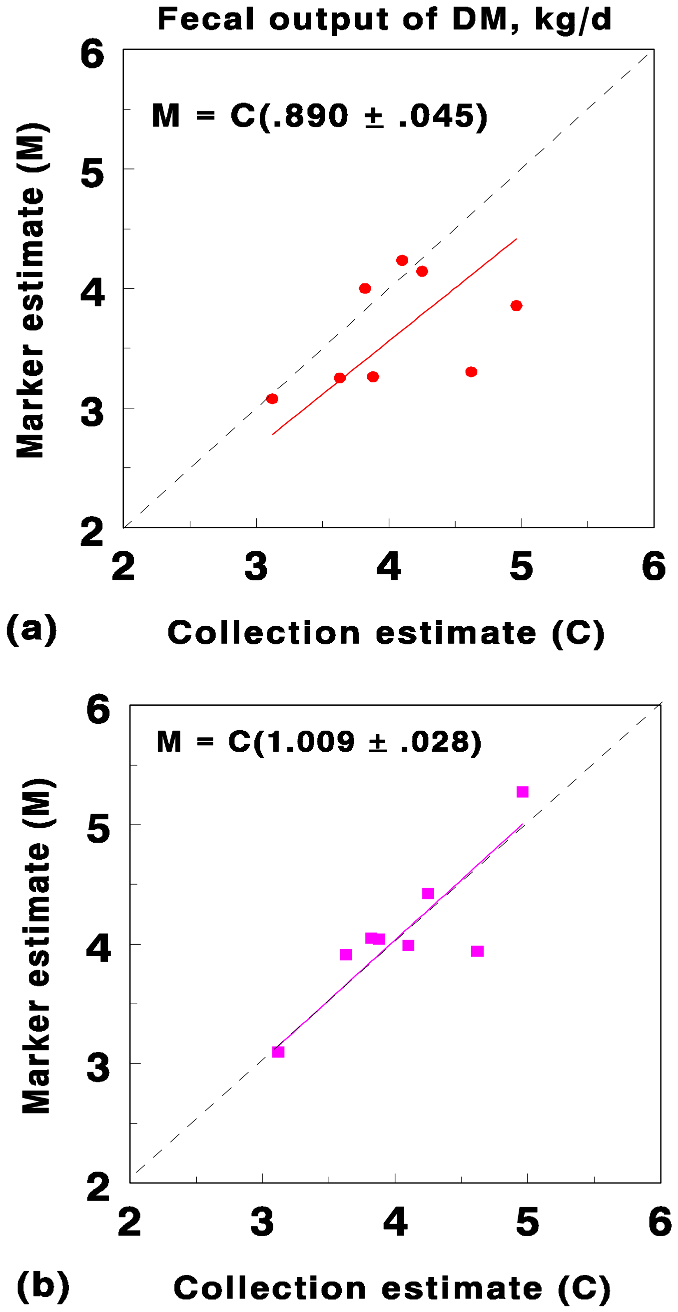

Figure 3. Fecal outputs predicted by marker dilution were correlated with collection values, both unadjusted (a) and adjusted (b) for Cr recoveries in feces. The correlation coefficient was improved by adjustment from .70 (*P* = .041) to .87 (*P* = .002).

Chapter 2. Diurnal Variation of Chromic Oxide Excretion in Horses Dosed with Chromic Oxide Either Once or Thrice Daily

ABSTRACT

Chromic oxide is one of the most commonly used external markers for fecal output (FO) predictions in digestion and intake studies. However, Cr can give erroneous results due to variation in Cr excretion over a 24 h period. This study was conducted to determine if the diurnal variation could be overcome if daily dosing was increased from once to thrice daily. Four trials were conducted to evaluate marker methods for predicting fecal output by applying a monoexponential equation with one rate constant (k), rising to an asymptote (C_a) to daily fecal Cr (C_t, g/kg DM). Chromium was dosed either once or 3 times daily. Fecal grab samples were taken either every 6 h, every 8 h, or at 700, 1300 and 1900 h during the last day of Cr dosing. These individual samples were analyzed for Cr. When Cr was dosed once daily there was variation throughout the day. When Cr was dosed every 8 h, there was no difference between individual fecal samples. However, when Cr was dosed every 6 h from 700 h to 1900 h, there was some variation in excretion of Cr. This indicates that dosing 3 times daily is sufficient to overcome diurnal variation, as long as the dosing is spaced evenly throughout the day. If Cr is to be used in grazing studies, a fourth dose of Cr may need to be administered.

Key Words: Fecal Output, Chromium, Dosing, Diurnal Variation

Introduction

One of the problems associated with using chromium (Cr) as a marker in digestion and intake studies is that excretion of Cr in the feces may have diurnal variation in excretion in the feces. This change in fecal Cr concentration affects how well fecal output is predicted.

Although once daily dosing would be convenient, horses dosed before the morning feed had fecal Cr concentrations that varied from about 50 mg/kg fecal DM to almost 600 mg/kg DM throughout a 24 h period (Cuddeford and Hughes, 1990). It was concluded that Cr would not make an acceptable marker for this reason. However, in an earlier study where Cr was dosed twice daily, the diurnal variation was less, and Cr was found to be an acceptable marker (Haenlein et al., 1966). One method of marker administration that has been used in ruminants is the use of a continuous release bolus (Brandyberry et al., 1991; Hatfield et al., 1991; Buntinx et al., 1992). This bolus is administered via a balling gun and is assumed to remain in the reticulo-rumen, continuously releasing chromium, for 20 d. It was found that twice-daily dosing of Cr gave similar output estimates to both total collection and the bolus estimates (Hatfield et al., 1991). In other studies, the bolus was found to overestimate FO. One of the problems associated with the bolus was inconsistent release of Cr throughout the day. Although the slow-release bolus would be convenient, it is not suitable for use in horses (personal communication, Mitch Venning, Nufarm Ltd.). Therefore, the only solution to using Cr in horses would be to find a dosing regimen that could overcome diurnal variation.

The objective of these trials was to compare diurnal variation in fecal Cr excretion in horses dosed once or thrice daily with a chromic oxide mixture. Time of day of dosing was also evaluated. A third objective was to compare the accuracy of a mixture of fecal grab samples to a more systematic mix of daily fecal excretion.

Materials and Methods

In each trial where diurnal variation was evaluated, mature Thoroughbred horses were used. In the first trial, 4 horses were used. In all subsequent trials, 8 geldings were used.

Trial 1. During the first trial, the marker, 27.4 g of Cr, in the form of 40 g of Cr_2O_3 (Chromic Oxide Sesqui; Fisher Scientific, Fair Lawn, NJ) was thoroughly mixed into 310 g of the hay (chopped to 2 cm) and molasses mixture. This mixture was offered in a nose bag and completely consumed immediately before the morning feed was offered at 700 h). In this trial, horses were offered one of two diets, four horses were fed a mixture of orchardgrass and alfalfa hay (**HAY**), at a rate of 2% of BW per day. The other four were fed the same hay plus 3.0 kg/d of pelleted concentrate (**H&C**, PurePride 200, Purina Mills, St. Louis, MO that contained processed grain by-products, grain products, roughage products, molasses products, calcium carbonate and salt). Feeds were offered at 0700 and 1600 h. On the last day, diurnal fecal samples were collected at 0600, 1200, 1800 and 2400 h from two horses in each group. These diurnal fecal samples were used for Cr analysis (see below).

Trial 2. Geldings were housed in individual box stalls, 14 m^2, for the duration of each of 2 trials and fed one of 2 hays during each trial in a replicated 2 x 2 Latin Square design. The two hays used were a tall fescue (*Festuca arundinacea*) / alfalfa *(Medicago sativa)* blend (TF/A), and an orchardgrass (*Dactylis glomerata*) / alfalfa *(Medicago sativa)* blend (OG/A). Both hays were harvested at the Middleburg Agricultural Research and Extension Center the previous summer.

Horses were fed hays at 2% of their BW per day initially, and this was adjusted to maintain constant body weight as determined by weekly weighing. The daily ration was divided into three meals per day at 700, 1500 and 2300 h. Horses were fed their respective hay for 7 d prior to the beginning of each trial to allow for diet accommodation.

Each trial was conducted for 17 d. During the first 3 d, baseline fecal samples were collected. On d 4, horses began to be dosed three times daily, at 700, 1500, and 2300 h, with a “greenola bar” containing 6.68 ± .32 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for eight d, from d 3 until d 11. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected three times per day, when bars were fed. On the last day of dosing, a subsample was taken from each fecal grab sample so analysis of the diurnal excretion of Cr could be determined. Samples of feces were ground through a .5 mm screen Thomas-Wiley Laboratory Mill (Model 4, Thomas Scientific, Swedesboro, NJ). Analysis of Cr in "greenola bars" and fecal samples was as described below.

Trial 3. Geldings were paired by weight and divided into two groups of four. Each group was placed into one of two 8-acre pastures for the first trial and then switched to the other pasture for the second trial in a replicated 2 x 2 Latin Square design. One pasture was predominately bluegrass (*Poa pratensis*) and white clover (*Trifolium repens*) (BG/WC), while the second was predominately tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*) (TF/A). Horses were placed on their respective pastures 2 wk prior to the beginning of data collection. Two horses from each group were removed from the pastures before data collection and housed in individual box stalls, 14 m^2, for the duration of each trial.

Each trial was conducted for 15 d. During the first 3 d, baseline fecal samples were collected. On d 4, horses began to be dosed three times daily, at 700, 1300, and 1900 h, with a “greenola bar” containing 6.40 ± .48 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for eight d until d 11. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected three times a day, when bars were fed. . On the last day of dosing, a subsample was taken from each fecal grab sample so analysis of the diurnal excretion of Cr could be determined. Analysis of Cr in "greenola bars" and fecal samples was as described below.

Trial 4. Geldings were paired by weight and divided into two groups of four. Each group was placed into one of two 8-acre pastures for the first trial and then switched to the other pasture for the second trial in a replicated 2 x 2 Latin Square design. One pasture was predominately bluegrass (*Poa pratensis*) and white clover (*Trifolium repens*)

(BG/WC) while the second was predominately tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*) (TF/A). Horses were placed on their respective pastures 2-wk prior to the beginning of data collection.

Each trial was conducted for 15 d. During the first 3 d, baseline fecal samples were collected. On d 4, horses began to be dosed three times daily, at 700, 1300, and 1900 h, with a "greenola bar" containing 5.98 ± .45 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for eight days, from d 3 until d 11. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected three times a day, when bars were fed. On the last day of dosing, a subsample was taken from each fecal grab sample so analysis of the diurnal excretion of Cr could be determined.

Cr Analysis. For Cr analysis, fecal samples were collected as above. During Trial 1, samples of the Cr_2O_3 mixture were collected every 2nd day and were weighed and dried, and ground to pass a .5 mm screen in a Cyclone Mill (UDY Corporation, Fort Collins, CO). During Trials 2, 3, and 4, two "greenola bars" were set aside from each batch (approximately 12 per trial) for analysis of Cr. Samples of feces from Trials 2, 3, and 4 were ground through a .5 mm screen Thomas-Wiley Laboratory Mill (Model 4, Thomas Scientific, Swedesboro, NJ). "Greenola bars" were finely ground in a coffee grinder (Electric Miracle Seed&Coffee Mill, Model MC200, Whatman Scientific, Hillsboro, OR).

One concern during Trials 2, 3, and 4 was that the daily mixture of the three grab samples was not representative of daily Cr excretion. Therefore, it was decided to make a weighed mix from the individual grab samples collected for diurnal variation and compare that to the larger mixes made for each day of the trial. After individual samples were dried and ground, .5g of each sample from one day was weighed and mixed thoroughly. This mixed sample (Cm) was compared to the mixed grab samples for the day (Cd).

Analysis of Cr in doses (either mixture or bars) and feces was done by atomic absorption spectrophotometry using an air-acetylene flame after digestion with nitric and perchloric acids (Sandel, 1959). Two dilutions of each Cr sample were analyzed in duplicate, and five dilutions of a standard solution of potassium dichromate were used for calibration.

Differences between the diurnal samples was compared by the GLM procedure of SAS with horse, diet, time and diet*time in the model. Differences between daily excretion of Cr predicted from grab samples (Cd) and from a mix of the diurnal samples (Cm) was compared using the GLM procedure of SAS with horse, diet, method and diet*method in the model.

Results and Discussion

Trial 1. Fecal Cr concentrations on the last day in four horses were quite variable (Table 2). The 600 h sample was not different from the 2400 h sample ($P = .523$) but was different from both the 1200 and 1800 h samples ($P < .018$). The 1200 and 1800 h samples were not different ($P = .372$), but 1800 h was different from 2400 h ($P = .012$). The coefficient of variation between these samples was 79%, indicating large variation between sampling times. These results confirm that once daily dosing of a Cr mix is not adequate to overcome diurnal variation in output, as found by Cuddeford and Hughes (1990).

Trial 2. Changes in Cr levels were not different between TF/A and OG/A ($P = .271$), so data were combined. There were no differences between the diurnal samples ($P > .085$), indicating that dosing Cr every 8 h overcame the diurnal variation (Table 2). The coefficient of variation was only 7% between these samples, indicating that the diurnal variation observed in the first trial was overcome. When the grab sample mix

(Cd) and diurnal mix (Cm) were compared, no differences were found between diets (P = .652), so again, data for both diets were combined. Cd and Cm did not differ (P = .069), but both were different from a sample collected from the TC sample for the same day (P = .0001) (Table 3). In one study where once daily versus twice daily dosing was compared, no difference was found between the methods (Smith and Reid, 1955). However, dosing times were not spread evenly throughout the day (600 and 1600 h), and may have affected the results.

Trial 3. Changes in diurnal samples were different between BG/WC and TF/A in this trial (P = .051) so data were analyzed separately for each diet. When horses were on the BG/WC pasture, differences in diurnal samples were not different (P > .149), with a coefficient of variation of 11%. When horses were consuming TF/A, there was a difference between the 700 and 2300 h samples (P = .009), and an increased coefficient of variation of 25% (Table 2). There was also a diet difference when the Cd samples were compared with Cm (P = .046), so again, data was analyzed separately for each diet (Table 3). There was no difference in Cr level in either the BG/WC pasture (P = .484), and the TF/A pasture (P = .585). Differences in diurnal excretion of Cr due to diet has also been observed in other species either due to the time an animal spends grazing (Owens and Hanson, 1992) and even the type of forage being consumed (Parker et al., 1989).

Trial 4. Diurnal differences were not different between diet (P = .113), but there was a difference between sampling times (Table 2). Samples taken at 700 h were not different from samples taken at 1300 h (P = .953). However, 700 h samples were different from 1900 h samples, and 1300 h samples were different from 1900 h samples (P = .002). The coefficient of variation between these samples was 25%; an improvement over the once daily dosing, but not as consistent as dosing every 8 h. When Cd was compared to Cm, no difference was found (P > .254) when horses were consuming either diet. These samples were also similar to the analysis of Cr in TC samples from the same day (P > .274).

There has been some speculation that a steady flow of a marker may never be achieved, due to the variation in time spent grazing, as well as the variation in the time of day when grazing occurs (Owens and Hanson, 1992). Since the animals themselves are not continuously grazing for 24 h, the marker cannot be expected to be excreted continuously. It is also possible that incomplete mixing may occur within the large bowel when grazing occurs in phases. This would affect the excretion of Cr. Along these lines, more frequent dosing should help to overcome these differences.

Implications

Results from these trials indicate that Cr needs to be dosed at least 3 times a day, to improve results in marker studies. Separating the doses evenly throughout the day gives the best result. The pasture trials indicate that dosing Cr at 700, 1300 and 1900 h cannot overcome the diurnal variation in Cr excretion, and that a fourth or possibly fifth dosing, later in the day may be necessary, even if fecal samples are not taken at that time.

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Table 1. Recipe for "greenola bars"[a] for fecal kinetic studies.

Ingredients	
Oat-based sweet feed	2.16 kg
Cr_2O_3	800 g
Liquid molasses	1 L
Beer	380 mL

Instructions

Mix Cr_2O_3, molasses and beer thoroughly, making sure Cr_2O_3 is well-blended. Add sweet feed slowly, making sure all particles are covered. Divide mixture between 4 well-oiled 12"x15" pans. Put into oven set at low temperature. When mixture begins to set, cut into 20 bars per tray. Put back into oven until completely dry. Remove from pans and place each bar into an individual bag for ease of handling.

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[a] Adapted from Practical Horseman. 1996. 24(4):122. Horse Cookie recipe

Table 2. Comparison of diurnal variation in fecal Cr excretion (means±SE), mg/kg DM.

Trial 1. Once daily dosing

Time	Cr
600	10.35 ± 2.72^{a}
1200	2.65 ± 1.21^{b}
1800	$1.07 \pm .34^{b,c}$
2400	8.34 ± 1.49^{d}

^{a,b} P < .018

^{c,d} P < .012

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Trial 2. Thrice daily dosing

Time	Cr
700	$4.27 \pm .279^{a}$
1500	$3.88 \pm .333^{b}$
2300	$3.76 \pm .250^{c}$

^{a,b} P = .180

^{b,c} P = .689

^{a,c} P = .085

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Trial 3. Thrice daily dosing

BG/WC

Time	Cr
700	$4.64 \pm .724^{a}$
1300	$5.35 \pm .736^{b}$
1900	$4.31 \pm .606^{c}$

^{a,b} P = .321

^{b,c} P = .149

^{a,c} P = .642

TF/A

Time	Cr
700	$4.99 \pm .294^{a}$
1300	$3.83 \pm .574^{b}$
1900	$3.02 \pm .482^{c}$

^{a,b} P = .110

^{b,c} P = .257

^{a,c} P = .009

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Trial 4. Thrice daily dosing

Time	Cr
700	$6.18 \pm .547^{a}$
1300	$6.13 \pm .505^{b}$
1900	$3.79 \pm .513^{c}$

^{a,b} P = .953

^{b,c} P = .002

^{a,c} P = .002

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Table 3. Evaluation of differences between a composite of daily grab samples [Cd] and an even mix of diurnal samples [Cm] (means SE) [a]. Comparison of fecal collection methods.

Trial 2

Method	Cr	CV
[Cd]	3.40 ± .20[b]	24
[Cm]	3.88 ± .269[c]	28
[TC sample]	4.77 ± .404[d]	34

[b,c] P = .069

[c,d] P = .001

[b,d] P = .0001

==

Trial 3

BG/WC

Method	Cr	CV
[Cd]	5.21 ± .719[b]	39
[Cm]	4.80 ± .634[c]	37

[b,c] P = .484

TF/A

Method	Cr	CV
[Cd]	4.31 ± .259[b]	17
[Cm]	4.00 ± .292[c]	21

[b,c] P = .585

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Trial 4

Method	Cr	CV
[Cd]	5.38 ± .357[b]	27
[Cm]	5.46 ± .318[c]	23
[TC sample]	5.02 ± .366[d]	29

[b,c] P = .820

[c,d] P = .254

[b,d] P = .358

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[a] [Cd] is the fecal Cr concentration found in a combination of 3 grab samples.

[Cm] is the fecal Cr concentration found in a mix made from .5 g of each individual grab sample from the last day of dosing.

[TC sample] is the Cr concentration found in a subsample of the TC for the last day of dosing.

Chapter 3. Fecal Output, Digestibility, and Intake of Pastures Predicted by Marker Methods in Grazing Horses. Comparison of 12 D versus 8 D Dosing.

ABSTRACT

Marker methods, using Cr, were developed to estimate fecal output in mature horses. Initially, these methods were used in stalled horses consuming either hay or hay and concentrate. Horses were dosed for 12 d and a monoexponential equation with one rate constant (k), rising to an asymptote (C_a), was applied to the daily fecal Cr data. This study was done to determine if 8 d dosing was sufficient to give accurate and precise estimates of fecal output (FO): FO = Cr dose (d^{-1}) / C_a. In this study, 8 horses were divided into two groups, and were placed on either bluegrass/white clover or tall fescue/alfalfa pastures in a replicated 2x2 Latin Square design. Horses were fed Cr in "greenola bars" which contained 5.91 ± .32 g Cr/bar. Bars were fed at 700, 1300 and 1900 h for 12 d. Fecal samples were collected 3 x daily. Pasture and fecal samples were analyzed for Cr, yttrium (Y), n-alkanes, and nutrients. Daily fecal data (Ct, g/kg DM) were fit to a monoexponential equation that included a delay (d) which represented the time required for Cr to enter the prefecal mass. The equation was used to calculate C_a with the data for the 12 d of dosing or, for only the first 8 d of dosing. The use of the yttrium (Y) and n-alkanes were evaluated as internal markers to determine dry matter digestibility (D). Estimates of FO and D were then used to predict dry matter intake (DMI): DMI = FO / (1-D). Differences were found between digestibility estimates of Y and n-alkanes. Fecal kinetics were similar when either 12 d or 8 d dosing was used. This indicates that horses can be dosed for only 8 d, and still give reliable estimates of fecal output.

Key Words: Fecal Output, Intake, Digestibility, Fecal Kinetics

Introduction

Methods of administering external markers have varied across studies. Often, markers, such as Cr and Yb, have been given in a single dose and then the rate of disappearance of marker from the feces has been plotted (Galyean, 1993; Susmel et al., 1996). The area under the plotted curve is then used to estimate fecal output. Another method is to administer a marker continuously, either through fed markers (Pond et al., 1989; Holland et al., 1998) or through a time-release bolus (Brandyberry et al., 1991; Hatfield et al., 1991; Buntinx et al., 1992). Once the fecal marker concentration reaches a plateau, the level can be used to predict fecal output (FO) with the equation: FO = marker dose^{-d} / Cr concentration in feces.

One of the concerns with continuous infusion of a marker, is determining how long the marker needs to be dosed before a consistent fecal concentration is reached. It has been estimated that a minimum of 4 d is required for Cr to reach this state. Fecal samples collected after this time have been used to estimate fecal output (Haenlein et al., 1966; Hatfield et al., 1991). Often, however, Cr will overestimate fecal output when used in this manner. Using the daily fecal concentrations of Cr from the first day of dosing to predict an asymptote value of fecal concentration has been proposed to predict fecal outputs more accurately than an averaged plateau value (Holland, et al., 1998).

Internal markers have been used to predict dry matter and nutrient digestibilities in herbivores (Cochran et al., 1987; Owens and Hanson, 1992). Some markers, such as lignin, have been deemed unsuitable due to apparent digestion or degradation in the digestive tract (Fahey and Jung, 1983). Others, such as acid insoluble ash, have given conflicting results, sometimes being an acceptable marker (Schurg, 1981; Cuddeford and Hughes, 1990) while other times being unacceptable (Miller et al., 1986). Other markers that have shown promise are n-alkanes (Ohajuruka and Palmquist, 1991; Dove and Mayes, 1996).

The primary objective of this study was to evaluate how dosing Cr for 8 d, versus dosing for 12 d affected the prediction of a fecal asymptote, hence fecal output. Other objectives were to evaluate the use of yttrium and n-alkanes as internal markers for digestibility predictions, and to combine fecal output and digestibility predictions to predict intake in grazing horses.

Materials and Methods

Eight mature Thoroughbred geldings, average weight 560 ± 26 kg, were used in these studies, which were conducted during the months of August and September, 1996. Geldings were paired by weight and divided into two groups of four. Each group was placed into one of two 8-acre pastures for the first trial and then switched to the other pasture for the second trial in a replicated 2 x 2 Latin Square design. One pasture (BG/WC) was predominately bluegrass (*Poa pratensis*) and white clover (*Trifolium repens*); the second (TF/A) was predominately tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*). Horses were placed on their respective pastures 2-wk prior to the beginning of data collection.

Each trial was conducted for 14 d, with a 14 d break between periods. On the first day, a baseline fecal samples was collected. On d 2, horses began to be dosed three times daily, at 700, 1300, and 1900 h, with a “greenola bar” containing 5.91 ± .32 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for twelve d until d 14. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected from all horses three times daily, about the time bars were fed. Fecal samples from the same day were combined, weighed, dried in a forced-air oven to a constant weight, and then reweighed for DM determination. Total collection (TC) of feces was attempted on all horses during the first trial. One gelding would not accept the fecal collection harness (Wyoming Tent Co., Cheyenne, WY), and a second quickly developed a skin rash on his back under the bag. After 5 d, all fecal collection harnesses were removed because of problems with sores under harness straps and rashes. On the days when collections were successful, bags were emptied 3 times daily in separate tubs for each horse. Feces from each 24 h period, including individual grab samples, were weighed, mixed in a small cement mixer (Red Lion Utility Mixer, Model RLX3-Type C, Monarch Industries, Bloomington, MN) and subsampled. Mixed grab samples and TC subsamples were weighed, dried to constant weight in a forced air over, and weighed again for DM determination.

Pasture samples were collected using methods adapted from Blaser et al. (1986). Samples were collected during each trial using hand-held electric clippers with a 10.2 cm wide edge. Samples were clipped across the pasture in a random fashion until 1-2 kg, wet weight, was collected. These samples were weighed, dried in a forced-air oven, and weighed again for DM determination. These samples were used for nutrient analysis. Between trials, pasture samples were collected for botanical composition. A stick, 1.83 m, was thrown randomly into each acre. The pasture to the left of the stick was clipped using hand-held electric clippers. Total samples for each pasture were hand separated into edible plants and weeds (Table 2). Samples were weighed wet, to give a total weight and then all samples were dried so that percentages of wet and dry pasture could be determined (Table 2).

Samples of pastures and feces were ground through a .5 mm screen Thomas-Wiley Laboratory Mill (Model 4, Thomas Scientific, Swedesboro, NJ). Composite samples were made from the pasture samples and from the feces from the last 4 d of dosing. These composites were submitted for analysis of DM, CP, EE, NSC, NDF, ADF, Ca, P, K, Mg, Na, Fe, Zn, Cu, Mn, Mo, S and ash by standard methods (Robertson and Van Soest, 1977; AOAC, 1990) at the Northeast DHIA Laboratory (Ithaca, New York). Pasture analysis can be found in Table 3.

The twelve "greenola bars" were finely ground in a coffee grinder (Electric Miracle Seed&Coffee Mill, Model MC200, Whatman Scientific, Hillsboro, OR). Analysis of Cr in "greenola bars", pasture composites, fecal composites, daily fecal samples and TC subsamples used atomic absorption spectrophotometry with an air - acetylene flame after samples were digested with nitric and perchloric acids (Sandel, 1959). Two dilutions of each Cr sample were analyzed in duplicate, and five dilutions of standard solution of potassium dichromate were used for calibration. Yttrium (Y) was analyzed in pasture and fecal composite samples by inductively coupled plasma spectroscopy after samples were digested with nitric and perchloric acid. Alkane content was analyzed following preparation methods developed by Ordakowski (1998) and using gas chromatography (Model 5890A, Hewlett Packard, Wilmington, DE).

Dry matter digestibility (D, %) was determined by using a ratio of the internal markers (M), either Y or alkanes:

$$D = (1- C_I / C_F)*100$$

where C_I is the [M] in the feed samples and C_F is the [M] in the fecal samples.

Nutrient digestibilities (ND) were calculated using the internal markers Y and n-alkanes:

$$ND=[1- (C_I / C_{F)}\ (N_I /\ \%N_F)]*100$$

where N_I (%DM) is the concentration of a nutrient in the feed samples and N_F (%DM) is the concentration of a nutrient in the fecal samples.

Pooled daily fecal Cr concentrations (C_t, mg/kg DM) for all 8 horses on one diet were fit to a simple, monoexponential model rising to an asymptote (C_a) using a graphics and curve-fitting program (SlideWrite Plus 4.0 for Windows, Advanced Graphics Software, Carlsbad, CA) as previously described (Holland et al., 1998):

$$C_t = C_a - C_a \cdot e^{-kt}$$

where Ct was fecal Cr concentration (mg/kg DM) at time t (days). The asymptotic value of fecal Cr determined fecal output (FO):

$$FO = (\text{Cr dose}/C_a).$$

The rate constant (k, d^{-1}) determined its reciprocal, the turnover time (TT, d). The compartment size, or mass, of the prefecal pool (PFM, kg) was calculated:

$$PFM = FO \times TT = FO/k.$$

Fecal outputs predicted by C_a were compared to total collection values. FO and D predicted from markers were used to calculate dry matter intake (DMI, kg/d):

$$DMI = FO/(100 - D)$$

and these predicted intakes were compared.

The data were summarized as means and SEMs, and analyzed by the GLM procedure of SAS (SAS Institute, Inc., Cary, NC) with horse, diet, method and all interactions in the model. Differences in digestibilities predicted by Y and alkanes were compared. Differences in predicted fecal outputs from C_t and C_a were compared to TC. Intake predictions from markers were compared. Means were compared using Fisher's protected LSD.

Results and Discussion

Digestibilities. Estimates of D using Y or alkanes are presented in Table 4. Digestibilities of BG/WC and TF/A pastures were not significantly different overall ($P > .465$). Nutrient digestibilities predicted by C27 were not different from digestibilities predicted by either C31 or Y (Table 4). C27 was different from C29 ($P < .046$). Except for dry matter digestibility, there was no difference in nutrient digestibilities between C29 and either C31 ($P > .093$) or Y ($P > .112$). Digestibility of pasture is most often related to plant maturity (Burns et al., 1997), usually determined by fiber content of the pasture (Darlington and Hershberger, 1968). Nutrient content of these pastures differed slightly (Table 3) in CP, ADF and NDF content. The higher CP content and lower fiber(s) content of the TF/A pasture gives reason for the higher digestibility predictions for this pasture.

For BG/WC, all methods predicted dry matter digestibility similarly ($P > .066$). Y and C27 had differing nutrient digestibility predictions for all other nutrients ($P < .042$). Digestibility predictions between Y and C31 were different for NDF ($P = .016$) and ash ($P=.025$). The rest of the comparisons between methods were not different ($P > .068$). When TF/A was the pasture being grazed, C27 and C29 predicted DM, CP, ADF, AND NDF digestibilities differently ($P < .015$). C29 and Y also had differing nutrient digestibilities for DM, CP, ADF, NDF, EE, AND NSC ($P < .046$). The rest of the digestibility comparisons between methods were not different ($P > .070$).

Dry matter digestibilities for forages usually range between 55 and 70% when consumed by horses, so predictions from the C27 and C31 alkanes are probably the best estimates in this study. The variability in the yttrium predictions from one pasture to the other indicates this may not be a good marker of digestibility in grazing animals, even though it did appear satisfactory when used in stalled animals (Journal Papers 4 and 5). Other rare earth elements have been tested as internal markers in horses (Todd et al., 1995). Dysprosium underestimated digestibilities; authors determined that recovery of the marker was incomplete.

Alkanes have been used successfully in ruminants as markers for digestibility. This is due to the relatively high recovery of the alkanes in the feces (Dove and Mayes, 1991). It has been found that the percent recovered increases with increasing chain length, therefore, only the higher length alkanes were used in this trial. These alkanes are also the most abundant in forages and therefore there is less chance for error in analysis. Digestibilities found here using alkanes were similar to those predicted in other studies on horses (Ordakowski, 1998).

Fecal Output. Fecal output estimates are summarized in Table 5. C_a (asymptotic prediction of fecal Cr concentration) and C_d (plateau level of fecal Cr concentration, last 4 d of dosing) did not yield similar predictions, when compared to TC ($P = .0001$). However, C_a and C_d were not different from each other ($P = .432$). When the two periods were combined, there was no difference in FO predictions from C_a or C_d ($P = .452$). There was also no diet effect on the ability of these two methods to predict FO ($P > .560$). Horses consuming BG/WC had higher total FO than those grazing TF/A ($P = .031$). Since the TF/A pasture had higher apparent nutrient digestibilities, more of the forage was utilized, leading to lower fecal output.

Intake. Intakes (Table 6) predicted by C_a and C29 were not different from C_a and C31, or C_a and Y ($P > .346$). Intake prediction of C_a and C27 in combination was different from C_a in combination with C29, C31, and Y ($P < .02$). Prediction results were similar when placed on a by-diet reference. C_a and C27 predictions were different from

C29 and Y combinations ($P < .01$), but not C31 ($P > .06$)when horses were consuming BG/WC. When horses were grazing TF/A, C_a and C27 were different from C29 and 31 ($P < .05$), but not Y ($P > .346$).

Average estimates of dry matter intake are between 1.4 and 1.8% of an animal's BW (NRC, 1989). Therefore, intake estimates using FO predictions and Y and the C29 and C31 alkanes probably are close to actual intakes. Intakes for grazing animals would be higher than for stalled animals, due to time spent foraging, so estimates as high as 3% of BW are within an expected range. This is supported by work conducted with grazing cattle, where intakes ranged from 1 to 3 % of the animals' BW (Cordova et al., 1978). Intake estimates in pregnant and lactating mares ranged from 1.5 to 2.5 % of BW (Martin et al., 1989). In this study, C27 was not an effective marker, although it did predict digestibility similar to TC in the hay trial (Chapter 4).

Fecal Kinetics. When the monoexponential equation was applied to Cr data without a delay, it gave good fits to both the BG/WC data ($r^2 = .891$) and the TF/A data ($r^2 = .826$). Comparisons between 12 d dosing and 8 d dosing indicated that the addition of 4 extra days of dosing did not improve the fit for either BG/WC or TF/A pastures (r^2 from .891 to .898 for BG/WC and .826 to .881 for TF/A) although there was a slight decrease in the corresponding F-statistic. Data for individual horses is shown in Table 7. Since the 8 d dosing data fit well, it was used in the remainder of the comparison. Sutton et al. (1977) evaluated how length of collection period affected nutrient digestibilities in mature geldings. Increasing the collection period from 3 to 7 d did not affect results. It appears that once marker excretion reaches a plateau level, that additional fecal collection will not improve results.

The fit of the monoexponential equation improved when a delay of 1.80 h and 2.38 h were applied to BG/WC and TF/A, respectively (Table 8). This delay (*d*, d) was added to the model as a decrease in time (t) in the original equation:

$$C_t = C_a - C_a \cdot e^{-k(t-d)}$$

Estimates of *d* were added to the equation manually until the best fit (using improvements in r^2) was found. This model represents a delay of the marker entering the prefecal pool, as described in tracer kinetic studies (Kronfeld and Ramberg, 1981). Daily fecal concentrations of Cr (C_t), were plotted against time to determine C_a (Figure 1). Model parameters and estimates of TT, FO and PFM for the equation with delay are given in Table 8. Delays for these pastures (1.80 h and 2.38 h for BG/WC and TF/A, respectively) were much shorter than reported previously (Holland et al., 1998). This is probably related to the high moisture content of pasture, when compared to hay or hay and concentrate diets. These pastures were between 65 and 80% moisture (Table 3) and this alone would increase passage rate, therefore decrease TT.

When the two equations, with and without the delay, were applied to individual horse data, all but horse A fed TF/A improved with the delay (Table 9 a and b). Delays are inserted into models for ruminant passage studies as well, not only to estimate marker transit times, but also in an attempt to estimate time required for mixing to occur in the various compartments of the complex stomach (Grovum and Williams, 1973; Pond et al., 1988).

Estimates of fecal kinetics are given in Table 10. Fecal output and PFM were smaller for horses consuming TF/A (Table 10) and could be related to the higher quality pasture and higher digestibility. Another factor that may influence fecal kinetics is the selectivity of the grazing animal (Cherney et al., 1990; Hitchcock et al., 1990). Pasture samples are collected randomly, but probably do not mimic the plants and plant parts consumed by the horses. In ruminants, esophageal fistulas have been used to give estimates of forage intake, but these methods are not applied to horses

Implications

It will be possible to predict fecal output and nutrient digestibilities using an 8 d dosing schedule, instead of the 12 d schedule. Yttrium did not appear to be an effective marker in grazing horses, even though it gave good digestibility estimates in stalled horses. Alkanes may provide more reliable digestibility estimates in grazing animals, although no one alkane is consistent across all plant species. Delays of external markers entering into the prefecal mass is influenced by diet. Predicted DMI was higher than reported estimates, indicating that reported estimates do not adequately express nutrient requirements of grazing horses.

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Table 1. Recipe for "greenola bars"[a] for fecal kinetic studies.

Ingredients

Oat-based sweet feed	2.16 kg
Cr_2O_3	800 g
Liquid molasses	1 L
Beer	380 mL

Instructions

Mix Cr_2O_3, molasses and beer thoroughly, making sure Cr_2O_3 is well-blended. Add sweet feed slowly, making sure all particles are covered. Divide mixture between 4 well-oiled 12"x15" pans. Put into oven set at low temperature. When mixture begins to set, cut into 20 bars per tray. Put back into oven until completely dry. Remove from pans and place each bar into an individual bag for ease of handling.

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[a] Adapted from Practical Horseman. 1996. 24(4):122 Horse Cookie recipe.

Table 2. Botanical composition (BC) of bluegrass/white clover (BG/WC) and tall fescue/alfalfa (TF/A) pasture.

BG/WC[a]

Plant	% of wet pasture	% of dry pasture
Bluegrass (*Poa pratensis*)	45.34	59.01
White Clover (*Trifolium repens*)	5.42	3.42
Tall Fescue (*Festuca arundinacea*)	36.66	26.08
Orchardgrass (*Dactylis glomerata*)	2.39	2.48
Yellow Millet (*Setaria glauca*)	3.15	2.17
Plantain (*Plantago major*)	2.93	1.86
Misc. Weeds	2.27	3.73

[a] Percent DM of total BC sample 34.92

TF/A[a]

Plant	% of wet pasture	% of dry pasture
Tall Fescue (*Festuca arundinacea*)	62.93	48.79
Alfalfa (*Medicago sativa*)	2.74	3.46
White Clover (*Trifolium repens*)	4.77	3.81
Bluegrass (*Poa pratensis*)	.95	2.08
Orchardgrass (*Dactylis glomerata*)	2.38	2.08
Dandelion (*Taraxacum officinale*)	.24	.35
Plantain (*Plantago major*)	12.28	9.00
Misc. Weed	13.71	30.45

[a] Percent DM of total BC sample 34.45

Table 3. Nutrient and internal marker content[a] of pastures, means ±SE.

Nutrient	bluegrass / white clover	tall fescue / alfalfa
DM, %	36.0 ± 7.9	21.01 ± 1.44
Composition of DM		
CP, %	15.5 ± .6	18.1 ± 1.4
ADF, %	36.4 ± .4	30.8 ± .3
NDF, %	60.3 ± .5	51.6 ± .4
EE, %	3.4 ± .7	3.2 ± .3
NSC[b], %	13.3 ± 1.0	17.8 ± .8
Ca, %	.40 ± .04	.69 ± .13
P, %	.34 ± .03	.37 ± .08
Mg, %	.20 ± .01	.34 ± .03
K, %	2.21 ± .12	2.89 ± .07
Na, %	.003 ± .000	.015 ± .005
Fe, ppm	456 ± 82	580 ± 77
Zn, ppm	33 ± 1	38 ± 10
Cu, ppm	26 ± 3	24 ± 1
Mn, ppm	54 ± 1	63 ± 2
Mo, ppm	1.8 ± .5	2.3 ± .3
S, %	.21 ± .02	.26 ± .02
Ash, %	7.61 ± .64	9.45 ± .58
Cr, ppm	35 ± 0	17.5 ± 0
Y, ppm	.88 ± .04	.78 ± .19
Alkanes, ng/g		
C27	7.62 ± .86	10.48 ± 1.61
C29	30.45 ± .88	34.38 ± 1.04
C31	83.99 ± 11.9	92.23 ± .81

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[a] Analysis of CP, EE, and ash (AOAC, 1990), ADF and NDF (Robertson and Van Soest, 1977), Cr (Sandel, 1959)

[b] NSC = 100 - (CP% + EE% + NDF% + ash%)

Table 4a. Digestibilities (%), means ± SE, of DM, CP, ADF, NDF, EE, NSC and Ash in horses (n = 8) grazing bluegrass/white clover (BG/WC) pasture. Predicted either from yttrium (Y) or C27, C29 or C31 n-alkanes.*

Nutrient	Y[+]	C27	C29	C31
DM	47.68 ± 5.47^{c}	71.31 ± 6.42^{d}	46.58 ± 8.94^{c}	69.59 ± 4.11^{d}
CP	66.65 ± 3.47^{a}	81.35 ± 4.27^{b}	76.09 ± .77	80.66 ± 2.43
ADF	35.94 ± 6.66^{a}	65.35 ± 7.51^{b}	54.00 ± 2.45	62.31 ± 5.45
NDF	43.36 ± 6.31^{c}	69.00 ± 6.95^{d}	59.70 ± 1.64	67.24 ± 4.38
EE	-36.53 ± 19.53^{b}	33.51 ± 10.59^{a}	-3.87 ± 13.56^{b}	10.35 ± 18.85
NSC	84.86 ± 2.43^{a}	90.89 ± 2.83	89.20 ± 1.63	91.56 ± 1.21^{b}
Ash	15.71 ± 9.09^{a,c}	56.91 ± 13.53^{b}	57.95 ± 6.25^{b}	70.04 ± 1.73^{d}

[+] For Y digestibilities , n = 7. One horse had a DMD of -6% and was dropped from the equations.

*Values in the same row with different superscripts are different:

^{a,b} $P < .04$

^{c,d} $P < .004$

Table 4b. Digestibilities (%), means ± SE, of DM, CP, ADF, NDF, EE, NSC and Ash in horses (n = 8) grazing bluegrass/white clover fed tall fescue / alfalfa (TF/A) pasture. Predicted either from yttrium (Y) or C27, C29 or C31 n-alkanes.*

Nutrient	Y[+]	C27	C29	C31
DM	81.33 ± 1.55^{b}	75.83 ± 6.00^{b}	47.27 ± 7.73^{a}	62.05 ± 1.65
CP	87.36 ± 1.18^{b}	83.29 ± 4.30^{b}	68.62 ± 4.01^{a}	74.35 ± 2.83
ADF	73.05 ± 2.40^{b}	65.02 ± 8.78^{b}	32.81 ± 9.18^{a}	45.37 ± 2.42
NDF	78.93 ± 1.93^{a,e}	72.38 ± 7.03^{c}	47.59 ± 6.68^{b,d}	57.24 ± 2.30^{f}
EE	55.45 ± 6.07^{g}	54.77 ± 7.28^{g}	-20.43 ± 36.38^{h}	10.61 ± 9.97^{h}
NSC	93.95 ± 1.55^{c,g}	91.48 ± 3.37	86.32 ± 3.85^{d}	88.28 ± 3.03^{h}
Ash	67.64 ± 1.48	62.49 ± 12.33	45.43 ± 7.73	52.40 ± 10.22

[+] For C29 digestibilities, n = 7. One horse had a DMD of -38.4 and was dropped from the equations.

*Values in the same row with different superscripts are different:

^{a,b} P < .007

^{c,d} ^{e,f} P < .02

^{g,h} P < .05

Table 5. Fecal outputs from horses (n = 8) grazing either bluegrass / white clover (BG/WC) or tall fescue / alfalfa pasture (TF/A). Average weight was 560 ± 26 kg. Comparison between TC and predicted outputs*.

Variable	BG / WC	SEM	TF / A	SEM
Fecal DM [a], kg/d, weighed	5.03	.19	4.31	.16
Fecal DM predicted from daily Cr, kg/d	5.34	.66	3.94	.30
Fecal DM predicted from C_a, kg/d	4.81	.59	3.4	.27

[a] TC values from 3 horses on each pasture, first trial.

Table 6. Dry matter intakes, kg/d, of horses (n = 8) grazing either bluegrass / white clover (BG/WC) or tall fescue / alfalfa (TF/A) pasture. Intakes, on a %BW basis, are given in parentheses. Average weight was 560 ± 26 kg. Comparison of predicted intakes.*

Variable	BG / WC	TF / A	SEM
DMI, predicted from daily Cr[+] and Y	10.18 (1.8)[b]	22.62 (4.0)	6.72
DMI, predicted from daily Cr and C27	36.69 (6.6)[a]	32.69 (5.8)[e]	6.72
DMI, predicted from daily Cr and C29	10.51 (1.9)[b]	7.34 (1.3)[f]	6.72
DMI, predicted from daily Cr and C31	17.85 (3.2)	10.58 (1.9)[f]	6.72
DMI, predicted from C_a[+] and Y	9.49 (1.7)[d]	19.71 (3.3)	6.72
DMI, predicted from C_a and C27	35.24 (6.8)[c]	28.75 (5.1)[g]	6.72
DMI, predicted from C_a and C29	10.06 (1.8)[d]	6.36 (1.1)[h]	6.72
DMI, predicted from C_a and C31	16.99 (3.0)	9.23 (1.6)[h]	6.72

[+] Daily Cr is average fecal [Cr] over last 4 d of dosing. C_a is predicted asymptotic fecal [Cr] from monoexponential equation.

*Values in the same column, within a group, with the same superscript are different.

[a,b] [c,d] $P < .01$

[e,f] [g,h] $P < .05$

Table 7. Comparison of 8 d and 12 d dosing. Fits of data to a one-compartment model of fecal kinetics in trials on eight horses grazing bluegrass / white clover (BG/WC) or tall fescue / alfalfa (TF/A) pasture.

BG/WC	12 d Dosing		8 d Dosing	
Horse	R^2	*F*	R^2	*F*
A	.790	41.3	.801	28.2
B	.752	33.4	.796	27.4
C	.874	76.0	.926	87.6
D	.644	19.9	.818	31.4
E	.684	23.8	.706	16.8
F	.750	33.0	.773	23.8
G	.703	26.0	.711	17.2
H	.556	13.8	.659	13.5

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TF/A	12 d Dosing		8 d Dosing	
Horse	R^2	*F*	R^2	*F*
A	.683	23.7	.725	18.4
B	.691	24.6	.726	18.5
C	.739	31.1	.829	33.9
D	.449	9.0	.592	10.2
E	.675	22.9	.885	53.6
F	.639	19.5	.764	22.7
G	.605	18.8	.612	11.0
H	.684	22.8	.690	15.6

Table 8. Estimates of fit of pooled fecal Cr concentration data (Figure 1) to a one-compartment model, model parameters, and calculated variables in horses fed either bluegrass / white clover (BG/WC) or tall fescue / alfalfa (TF/A) pasture. Evaluation with 8 d dosing.

Estimate	BG/WC	TF/A
R^2	.912	.901
F	72.9	63.6
P- value	.0000	.0000
Cr_a, mg/kg[a]	3.83	5.23
SE	.2054	.3181
t-statistic	18.6	16.5
P-value	.0000	.0000
-k, d^{-1}[b]	1.577	1.401
SE	.502	.476
t-statistic	3.14	2.94
P-value	.0069	.0094
TT, h[c]	15.2	17.1
Delay, h[d]	1.80	2.38
FO, kg/d[e]	5.06	3.43
PFM, kg[f]	3.20	2.44

[a] Cr_a is the asymptotic value of fecal Cr concentration over 8 d dosing.

[b] k is the rate constant.

[c] TT is turnover time of the prefecal mass, TT = -1/k

[d] Delay is the time between the administration of Cr and its entry into the prefecal mass

[e] FO is fecal output, FO = Dose of Cr/Cr_a.

[f] PFM is prefecal mass or mixing compartment, PFM = FO x TT.

Table 9a. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses grazing bluegrass / white clover pasture.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.769	23.3	.0005	.801	28.2	.0002
B	.761	22.3	.0005	.796	27.4	.0003
C	.919	79.7	.0000	.926	87.6	.0000
D	.799	27.8	.0003	.818	31.4	.0002
E	.650	13.0	.0031	.706	16.8	.0014
F	.723	18.3	.0010	.773	23.8	.0004
G	.669	14.1	.0024	.711	17.2	.0013
H	.577	9.6	.0075	.659	13.5	.0027
A to H[b]	.898	61.9	.0000	.912	72.9	.0000

[a] Delay was 1.80 h for horses fed bluegrass / white clover pasture.
[b] Pooled data as in Figure 2.

Table 9b. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses grazing tall fescue / alfalfa pasture.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.725	18.4	.0010	.725	18.4	.0010
B	.700	16.3	.0015	.726	18.5	.0010
C	.794	26.9	.0003	.829	33.9	.0001
D	.514	7.4	.0152	.592	10.2	.0063
E	.861	43.2	.0001	.885	53.6	.0000
F	.716	17.6	.0012	.764	22.7	.0005
G	.515	7.4	.0152	.612	11.0	.0051
H	.609	10.9	.0052	.690	15.6	.0017
A to H[b]	.881	51.8	.0000	.901	63.6	.0000

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[a] Delay was 2.38 h for horses fed tall fescue / alfalfa pasture.
[b] Pooled data as in Figure 2.

Table 10. Fecal kinetics in eight horses grazing either bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture estimated with a one-compartment model with a delay[a] between time of administration and entry of marker into the compartment.

Variable	BG/WC	TF/A	SEM	*P*-value
Fecal output, kg DM/d	5.06	3.43	.43	.009
Fecal output g/kg of BW daily	9.04	6.13	1.46	.09
Turnover time, h	15.2	17.1	.95	.09
Prefecal mass, kg DM	3.20	2.44	.38	.09
Prefecal mass, g/kg of BW daily	5.73	4.37	.68	.09

[a] The delays were .075 and .099 d for BG/WC and TF/A, respectively.

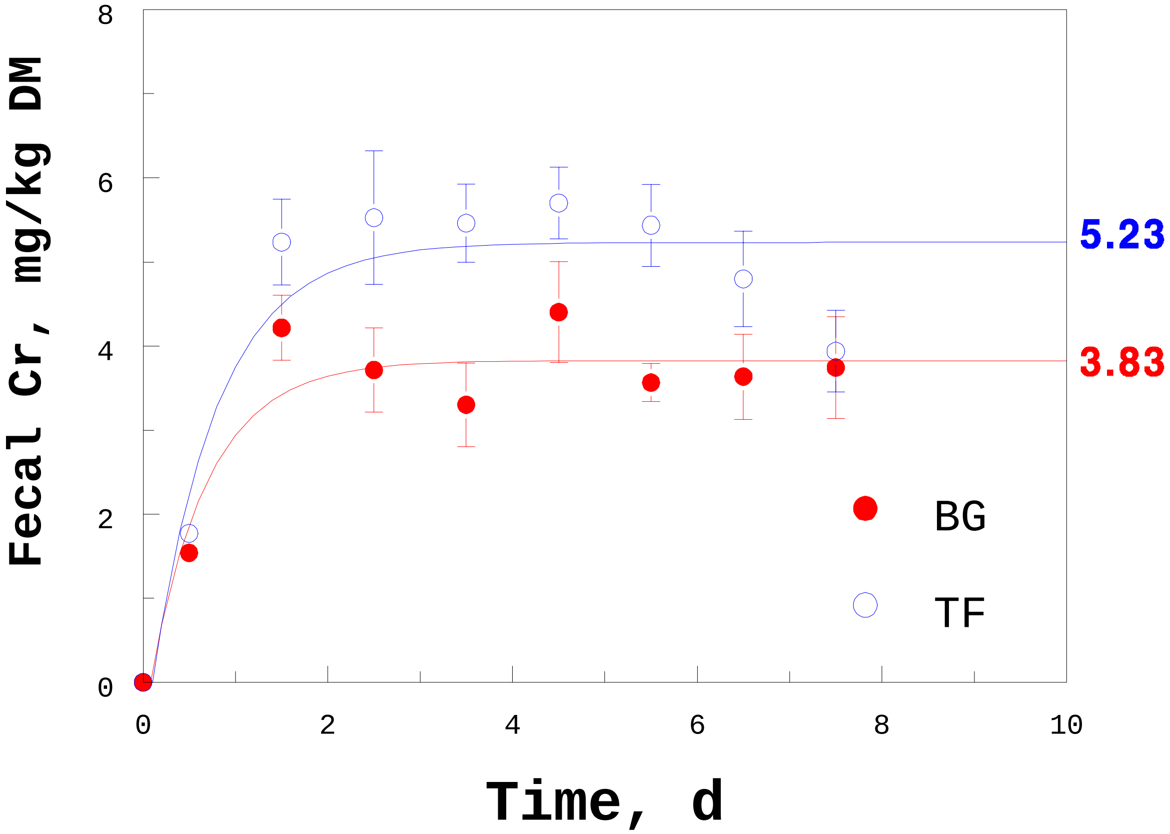

Figure 1. Mean daily concentrations of Cr, C*t*, for horses fed bluegrass/white clover (BG) or tall fescue/alfalfa (TF) pasture are plotted against time, and the data are fit to a one compartment model (see Figure 1, Holland et al., 1998).

Chapter 4. Fecal Output, Digestibility, and Intake of Hay is Predicted by Marker Methods in Horses

ABSTRACT

Marker methods need to be developed in horses that accurately and precisely predict both fecal output and digestibility. Once these methods are compared in confined conditions where intake, fecal output, and digestibility can be measured directly, they can be applied to grazing animals. Methods need to be developed that are simple enough to apply to grazing situations, and inexpensive enough to warrant their continued use. A method of estimating fecal output from fecal kinetics studies was evaluated in 8 mature Thoroughbred geldings. Geldings were divided into two groups, and were fed either tall fescue/alfalfa or orchardgrass/alfalfa hay in a replicated 2x2 Latin square design. Balance marker experiments were conducted over 17-d periods. Chromic oxide was administered 3 times daily in a "greenola bar", which contained 6.68 ± .32 g of Cr per bar. Bars were given for 8 d. Dry matter, Cr, yttrium, n-alkane and nutrient content were analyzed in feed and feces. Daily fecal Cr concentrations (C_t, g/kg DM) were fit to a simple monoexponential model with one rate constant (k), rising to an asymptote (C_a). A delay (d), was added to the model to represent the time needed for Cr to enter the prefecal mass: $C_t = C_a - C_a \cdot e^{-k(t-d)}$. When models were evaluated with pooled data, delays of 4.97 h and 6.70 h gave the best fits (determined by highest estimates of r^2) for tall fescue / alfalfa and orchardgrass / alfalfa hay, respectively. Yttrium and n-alkanes were evaluated as internal markers for digestibility (D) prediction during the balance marker experiments. Yttrium consistently overestimated and n-alkanes consistently underestimated dry matter digestibility, although n-alkanes did give closer estimates to total collection values. Digestibility estimates were combined with fecal output (FO) estimates from C_a: [FO = (Cr dose (d^{-1}) / $C_{a]}$ to predict dry matter intake (DMI): DMI = FO / (100 - D). Combinations of n-alkanes and C_a gave the best estimates of DMI, when compared to weighed intakes. These findings suggest that with proper dosing, Cr would give accurate and precise estimates of FO, and that n-alkanes could be internal markers for digestibility, hence intake, in grazing horses.

Key Words: Fecal Output, Intake, Digestibility, Fecal Kinetics

Introduction

Accurate estimates of fecal output are necessary in studies on grazing livestock so that nutrient digestibility and intake, as well as environmental impact of the animals can be evaluated. The effect of animals on the environment has recently come under public scrutiny due to the possibility of contamination to waterways and the land (Morse, 1996; Van Horn et al., 1996). Therefore, finding way to evaluate nutrient excretion, especially in animals such as horses, which are often housed in close proximity to suburbia, would help in the design of diets that would have less adverse effects (Tamminga, 1996).

The use of a simple monoexponential model, after dosing with an indigestible marker (Cr), has been proposed as an effective way to estimate fecal output (Holland et al., 1998). However, problems with inconsistent fecal output of the external marker need to be addressed (Smith and Reid, 1955; Cuddeford and Hughes, 1990; Galyean, 1993). One possibility of overcoming this would be with several doses of the marker spread throughout the day, or a time-released bolus (Pond et al., 1989; Brandyberry et al., 1991). Time-release boluses were designed with the reticulo-rumen in mind, however, and will not remain in the stomach of simple-stomached animals such as horses.

Internal markers to predict digestibility must be an inherent part of the forage, pass through the digestive tract at a consistent rate, and be indigestible (Kotb and Luckey, 1972). Several markers, such as acid insoluble ash and fibers are somewhat degradable, making them inappropriate markers (Cochran et al., 1987). Other potential markers include rare earth elements, which are not absorbed (Turnbull and Thomas, 1987) and plant wax alkanes (Vulich et al., 1991; Dove and Mayes, 1996). Both have been relatively successful in ruminants, and may prove useful in nonruminants like the horse.

The objective of this study was to evaluate the use of chromium as an external marker for fecal output and yttrium and n-alkanes as internal markers for digestibility in stalled horses fed hay. The model of fecal kinetics (Holland et al, 1998) was further evaluated in preparation for use in grazing horses. These markers were also used, in combination to predict dry matter intake.

Materials and Methods

Eight mature Thoroughbred geldings, average weight 554 ± 23 kg, were used in these studies, which were conducted during the months of February and March 1997. Geldings were housed in individual box stalls, 14 m^2, for the duration of each of 2 trials and fed one of 2 hays during each trial in a 2 x 2 Latin Square design, with periods of 17 d and a 14 d break between trials. The two hays were a tall fescue (*Festuca arundinacea*) / alfalfa (*Medicago sativa*) blend, and an orchardgrass (*Dactylis glomerata*) / alfalfa (*Medicago sativa*) blend. Both hays were harvested at the Middleburg Agricultural Research and Extension Center the previous summer.

Horses were fed hays at 2% of their BW per day initially, and this was adjusted to maintain constant body weight as determined by weekly weighing. The daily ration was divided into three meals per day at 700, 1500 and 2300 h. Horses were fed their respective hay for 7 d prior to the beginning of each trial to allow for diet accommodation.

During the first 3 d of each trial, baseline fecal samples were collected. On d 4, horses began to be dosed three times daily, at 700, 1500, and 2300 h, with a “greenola bar” containing 6.68 ± .32 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for eight d, from d 3 until d 11. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected three times a day, about the same time as when bars were fed. Fecal samples were collected three times daily for the 6 d after dosing ceased. Fecal samples from the same day were combined, weighed, dried in a forced-air oven to a constant weight, and then reweighed for DM determination. On the last day of dosing, a subsample was taken from each fecal grab sample so diurnal excretion of Cr could be determined. Total collection (TC) of feces was made during d 7-11; feces were collected in separate tubs for each horse. Feces from each 24 h period were weighed and a subsample collected for later analysis. Subsamples from individual grab samples and TC samples were weighed, dried to constant weight in a forced air over, and weighed again for DM determination.

Hay samples were collected every other day during the dosing phase of the trial. These samples were weighed, dried in a forced-air oven to constant weight, and weighed again for DM determination.

Samples of hays and feces were ground through a .5 mm screen Thomas-Wiley Laboratory Mill (Model 4, Thomas Scientific, Swedesboro, NJ). Composite samples were made from the hay samples and from the feces from the last 4 d of dosing. These composites were submitted for analysis of DM, CP, EE, NSC, NDF, ADF, Ca, P, K, Mg, Na, Fe, Zn, Cu, Mn, Mo, S and ash by standard methods (Robertson and Van Soest, 1977; AOAC, 1990) at the Northeast DHIA Laboratory (Ithaca, New York). Hay analysis can be found in Table 2.

The twelve reserved "greenola bars" were finely ground in a coffee grinder (Electric Miracle Seed&Coffee Mill, Model MC200, Whatman Scientific, Hillsboro, OR). Cr was analyzed in "greenola bars", hay composites, fecal composites and daily fecal samples using atomic absorption spectrophotometry with an air -acetylene flame after samples were digested with nitric and perchloric acids (Sandel, 1959). Two dilutions of each Cr sample were analyzed in duplicate, and five dilutions of standard solution of potassium dichromate were used for calibration. Yttrium (Y) was analyzed in hay and fecal composite samples by inductively coupled plasma spectroscopy after samples were digested with nitric and perchloric acid. Alkane content was analyzed following preparation methods developed by Ordakowski (1998) and using gas chromatography (Model 5890A, Hewlett Packard, Wilmington, DE).

Dry matter digestibility (D, %) was determined by two methods. The first was calculated from total collection data of both daily DM intake (I, kg) and fecal output (FO, kg):

$$D = (1-FO/I)*100$$

The second method involved using a ratio of the internal markers (M), either Y or alkanes:

$$D = (1-C_I/C_F)*100$$

where C_I is [M] in the feed and C_F is [M] in the feces.

Nutrient digestibilities (ND) were evaluated from the total collection data using the following equation:

$$ND = (1-N_O/N_C)*100$$

where N_C is the total amount of a nutrient consumed (kg/d), and N_O was the total amount of a nutrient found in the feces of an individual animal (kg/d). Nutrient digestibilities were also calculated using the internal markers Y and n-alkanes:

$$ND=[1-(C_I/C_F)(N_I / N_F)]*100$$

where N_I (%DM) is the concentration of a nutrient in the feed samples, and N_F (%DM) is the concentration of a nutrient in the fecal samples.

Pooled daily fecal Cr concentrations (C_t, g/kg DM) for all 8 horses on one diet were fit to a simple, monoexponential model rising to an asymptote (C_a) using a graphics and curve-fitting program (SlideWrite Plus 4.0 for Windows, Advanced Graphics Software, Carlsbad, CA) as previously described (Holland et al., 1998):

$$C_t = C_a - C_a \cdot e^{-kt}$$

where C*t* was fecal Cr concentration (g/kg DM) at time *t* (days). The asymptotic value of fecal Cr determined fecal output (FO):

$$FO = (Cr\ dose/C_a).$$

The rate constant (k, d^{-1}) determined its reciprocal, the turnover time (TT, d). The compartment size, or mass, of the prefecal pool (PFM, kg) was calculated:

$$PFM = FO \times TT = FO/k.$$

Fecal outputs predicted by C_a were compared to total collection values. FO (kg/d) and D (%DM) predicted from markers were used to calculate dry matter intake (DMI, kg/d):

$$DMI = FO/(100 - D)$$

and these predicted intakes were compared to amounts of DM fed.

The data were summarized as means and SEMs, and analyzed by the GLM procedure of SAS (SAS Institute, Inc., Cary, NC) with horse, diet, method and all interactions in the model. Differences in digestibilities predicted by Y and alkanes were compared to total collection values. Differences in predicted fecal outputs from C_t and C_a were compared to TC values. Intake predictions from markers were compared to weighed intakes. Comparisons of means used Fisher's-protected LSD.

Results and Discussion

Digestibility. Dry matter digestibility (D) calculated from weighed intakes and total fecal collection was higher ($P = .013$) by 10% in the OG/A than in the TF/A diet (Table 3). The ADF and NDF digestibilities were also higher in the OG/A diet ($P < .015$). The CP, NSC and ash digestibilities were not different between diets ($P > .2$) (Table 3). The differences between diet digestibilities are supported by the nutrient content of the hays (Table 2). The OG/A hay was higher in CP and NSC, and lower in fiber content, indicating a better quality hay. Although botanical composition analysis was not conducted, the TF/A hay appeared to have a coarse, high stem content, which would also indicate lower quality. Alfalfa hay, in itself, is usually considered a more digestible hay than grass hays (Crozier et al., 1997), so a higher alfalfa content would increase digestibility. Others have also noted a decrease in nutrient digestibilities of diets as alfalfa amount was decreased and fiber content was increased (Pearson et al., 1992). Similar differences in diet digestibilities have been noted between steers and horses; alfalfa had a higher digestibility than grass hays and apparent digestibilities were also higher in steers than horses, indicating that cattle are more efficient at digesting high fiber diets than horses (Vander Noot and Gilbreath, 1970; Cymbaluk, 1990). Level of

NDF has been mentioned as influencing digestibility was well; higher NDF content related to lower diet digestibilities in ruminants, as reviewed by Jung and Allen (1995).

When horses were offered the OG/A diet, only C29 predicted digestibility of all nutrients similarly to TC ($P > .338$). All other internal markers tested had nutrient digestibilities that differed from that of TC ($P < .006$) (Table 3). When horses were fed the TF/A hay, nutrient digestibilities predicted by internal markers were different ($P < .007$) from corresponding TC values. When odd-chain alkanes have been used to predict digestibility in sheep, average dry matter digestibility estimates were near 75%, but errors in recovery of the alkanes were noted as causing problems with these estimates (Casson et al., 1990). Odd chain alkanes have also been tested as internal markers in dairy cows and were found to underestimate dry matter digestibility by 8-15% (Ohajuruka and Palmquist, 1991). This underestimation was attributed to low alkane recoveries in the feces. Incomplete recovery of the alkanes is probably the reason for digestibility underestimates in this study as well. However, alkanes have also given overestimates of digestibility in sheep (Dove et al., 1990). Rare earths, such as yttrium, have been tried as internal markers on the assumption that they meet the stringent requirements of a "marker" (Crooker et al., 1982). Often, the elements are found in minute amounts in the feeds, leading to possible error in analysis. They have also been noted to pass through the digestive tract at a more rapid rate than other diet components. Faster passage rates of theses elements lead to overestimates of digestibility, as observed in this trial (Crooker et al., 1982; Turnbull and Thomas, 1987).

Fecal Output. Fecal outputs was not different between diets ($P = .307$) although it was slightly lower when horses were fed OG/A (Table 4). All methods of predicting FO using Cr differed from that of TC ($P < .002$). The FO predicted from the asymptote Cr value (C_a) was also different from actual TC on either TF or OG hay ($P = .022$ and .017, respectively). Although fecal outputs were overestimated using Cr, the use of the model and an asymptote value (C_a) gave closer estimates of output than the average from the last 4 d of dosing (C_d). These marker and collection estimates were correlated ($r^2 = .78$) however (Figure 2), indicating it might be possible to design calibration equations for fecal output, given a specific diet (Hargreaves, 1998). Chromic oxide has given an overestimation of fecal output in horses (Haenlein et al., 1966), sheep (Moore et al., 1992) and steers (Brandyberry et al., 1991), indicating that methods, either by modeling, as here, or by calibrating will be necessary to improve the prediction.

Intake. Weighed intake was not different between diets (P = .300) although it was slightly higher when horses were fed OG/A. Weighed dry matter intake on a % BW basis was lower in the geldings (1.6 to 1.7%) than has been previously reported of stalled horses consuming hay (Crozier et al., 1997), where horses consumed an average of 2.5% of their BW in DM. However, all horses maintained their body weight, and were within the expected intake of 1.4 to 1.8% BW (NRC, 1989), indicating that they were probably meeting their daily requirements. This lower intake may represent the lower activity level of stalled horses, when compared to free-ranging animals.

When the different D predictions were combined with TC FO to predict intake, several methods provided good results (Table 5). When C_a FO was combined with either C27 or C31, an intake similar to actual intake was estimated ($P > .238$). C_a and C29, C_a and Y and all C_d predictions were different than actual intake ($P < .015$). When horses were OG/A hay, C_a combined with C27 and C31 provided good estimates of intake (P = .868 and .430, respectively). While consuming tall fescue / alfalfa hay, C_a and n-alkanes 27 (P = .341), 29 (P = .329) and 31 (P = .376) all gave intake estimates close to the actual measured DMI. Yttrium did not give satisfactory intake estimates when combined with predicted FO (P = .0001). Intake estimates that combined the C_a value for fecal output were similar to intake estimates using actual TC values with the digestion estimates. This indicates that the C_a was close in predicting FO, and that much of the error associated with the calculations probably falls to the internal markers. Errors in intake predictions have been related to other internal markers; Cr predicted FO and lignin underestimated digestibility when used in lambs and steers (Momont et al., 1994) leading to underestimations in intake predictions. Intake estimates from the NRC (1989) range from 1.4 to 1.8 % of BW for mature horses at maintenance, on a dry matter basis.

Therefore, it is a reasonable assumption that predicted dry matter intakes ranging from 1.4 to around 2% are close estimates.

Fecal Kinetics. The monoexponential equation gave a good fit to fecal Cr data (C_t) from both the TF/A ($r^2 = .742$) and OG/A ($r^2 = .683$) groups. The addition of a delay (*d*, d) into the model, representing a decrease in time (t) in the model:

$$C_t = C_a - C_a \cdot e^{-k(t - d)}$$

of 4.97 h (TF/A) and 6.70 h (OG/A) improved the fit ($r^2 = .810$ and .783 for TF/A and OG/A, respectively). This equation applies calculations used in tracer and indicator-dilution, rather than those often applied in marker studies (Kronfeld and Ramberg, 1981; Merchen, 1988). The curve-fitting program could not solve this equation, so values of *d* had to be inserted until the best fit was found. Daily fecal concentrations of Cr, using this equation, were plotted against time to determine C_a (Figure 1). Model parameters, and estimates of PFM, TT, FO, and the delay are shown in Table 6.

When the equations for each diet were applied to individual horses there was an improvement in both r^2 and F-statistics for all horses when the delay was included (Tables 7 a and b), indicating that the delay did improve the model. Improvements of model fits with the addition of a delay, representing time required for markers to enter into the prefecal mass. Delays have been added to models of passage for ruminants with similar improvements (Quiroz et al., 1988; Thornley et al., 1995). Models for ruminants include two or three exponentials, relating to the compartments of the digestive tract. A two-compartment model, similar to one proposed by Grovum and Williams (1973), has been applied to horses (Corino et al., 1992). This model separated the cecum and colon as two compartments, and the authors believed this model gave good fits to the data. The addition of a second compartment to the model used here did not improve the fit, as was indicated by a decrease in the F-statistic and an increase in standard errors (Holland et al., 1998). The simpler model also reflects the simpler digestive tract of the nonruminant herbivore.

Estimates of FO, TT, and PFM, based on the model with delays of .207 and .279 d for TF/A and OG/A hays are given in Table 8. Fecal output was not different on a kg BW basis, although FO on the OG/A hay was slightly lower. This is probably related to the higher apparent digestibility of the hay. Turnover time was slightly longer when horses were fed the OG/A, as was prefecal mass. The TT and PFM in this study were less than previously reported for horses consuming hay (Holland et al., 1998), possibly indicating that a higher quality hay was used in this trial. The previous study utilized a primarily grass hay, where mixed hays with more alfalfa were used in this study. Legume hays traditionally have lower cell wall content, and higher nutrient digestibilities than grass hays (Jung and Allen, 1995; Crozier et al., 1997), hence, faster turnover rate within the digestive tract.

Implications

The use of a simple, monoexponential model can be used to predict fecal Cr excretion, which can then be used to predict fecal output in horses. Yttrium slightly overestimated and n-alkanes slightly underestimated nutrient digestibilities, but within ranges often observed with other internal markers. The use of the asymptote Cr prediction can be used with digestibility predictions to predict intake in stalled horses. These methods may predict dry matter intake in horses grazing pasture.

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Table 1. Recipe for "greenola bars"[a] for fecal kinetic studies

Ingredients

Oat-based sweet feed	2.16 kg
Cr_2O_3	800 g
Liquid molasses	1 L
Beer	380 mL

Instructions

Mix Cr_2O_3, molasses and beer thoroughly, making sure Cr_2O_3 is well-blended. Add sweet feed slowly, making sure all particles are covered. Divide mixture between 4 well-oiled 12"x15" pans. Put into oven set at low temperature. When mixture begins to set, cut into 20 bars per tray. Put back into oven until completely dry. Remove from pans and place each bar into an individual bag for ease of handling.

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[a] Adapted from Practical Horseman. 1996. 24(4):122. Horse Cookie recipe.

Table 2. Nutrient and internal marker content[a] of hays, means ± SE

Nutrient	tall fescue/alfalfa	orchard grass/alfalfa
DM, %	86.79 ± .64	86.48 ± .79
Composition of DM		
CP, %	11.8 ± .1	14.4 ± .2
ADF, %	48.4 ± 1.3	47.5 ± 1.1
NDF, %	66.2 ± 1.0	62.7 ± .8
EE, %	1.1 ± .1	.9 ± .1
NSC[b], %	13.9 ± 1.0	14.2 ± .7
Ca, %	.61 ± .02	.72 ± .02
P, %	.26 ± .01	.30 ± .01
Mg, %	.19 ± .01	.16 ± .01
K, %	2.58 ± .09	2.96 ± .01
Na, %	.019 ± .002	.010 ± .003
Fe, ppm	646 ± 72	605 ± 114
Zn, ppm	37 ± 3	37 ± 3
Cu, ppm	10 ± 1	11 ± 1
Mn, ppm	62 ± 4	59 ± 2
Mo, ppm	1 ± 1	1 ± 0
S, %	.16 ± .01	.17 ± .01
Ash, %	7.21 ± .19	7.97 ± .09
Cr, ppm	667 ± 317	261 ± 261
Y, ppm	.64 ± .06	.93 ± .09
Alkanes, ng/g		
C27	15.95 ± 1.07	15.82 ± .38
C29	48.54 ± 1.52	51.85 ± 1.36
C31	114.12 ± 1.19	121.89 ± 2.38

[a] Analysis of CP, EE, and ash (AOAC, 1990), ADF and NDF (Robertson and Van Soest, 1977), Cr (Sandel, 1959)

[b] NSC = 100 - (CP% + EE% + NDF% + ash%)

Table 3a. Digestibilities (%) of DM, CP, ADF, NDF, EE, NSC, and ash in horses (n = 8) fed tall fescue/alfalfa (TF/A) hay. Predicted either from total collection (TC), yttrium (Y) or C27, C29 or C31 n-alkanes.*

Nutrient	TC	Y	C27	C29	C31	SEM
DM	53.40^{a}	64.13^{b}	46.69^{b}	47.30^{b}	47.15^{b}	1.4
CP	67.89^{a}	75.05^{b}	62.84^{b}	63.41^{b}	63.42^{b}	1.04
ADF	46.31^{a}	58.91^{b}	38.50^{b}	39.24^{b}	39.08^{b}	1.61
NDF	48.99^{a}	60.85^{b}	41.68^{b}	42.32^{b}	42.15^{b}	1.53
EE	6.34^{a}	33.48^{b}	1.81	.20	-1.87	2.99
NSC	$67.50^{a,c}$	74.72^{b}	62.94^{b}	63.41^{d}	63.30^{b}	.97
Ash	$48.87^{a,c}$	59.99^{b}	40.43^{b}	41.13^{d}	40.82^{b}	1.86

*Values in the same row with different superscripts are different:

^{a,b} $P < .005$

^{c,d} $P < .01$

Table 3b. Digestibilities (%) of DM, CP, ADF, NDF, EE, NSC, and ash in horses (n = 8) fed orchardgrass/alfalfa (OG/A) hay. Predicted either from total collection (TC), yttrium (Y) or C27, C29 or C31 n-alkanes.*

Nutrient	TC	Y	C27	C29	C31	SEM
DM	58.62 ^{a}	68.70^{b}	50.79^{b}	57.31	52.52^{b}	1.4
CP	68.45^{a}	76.17^{b}	62.53^{b}	67.48	63.87^{b}	1.04
ADF	52.19^{a}	63.82^{b}	43.25^{b}	50.79	45.27^{b}	1.61
NDF	55.86^{a}	66.62^{b}	47.61^{b}	54.58	49.49^{b}	1.53
EE	-13.96^{a}	3.02^{b}	-35.80^{b}	-18.22	-31.66^{b}	2.99
NSC	68.66^{a}	76.26^{b}	62.48^{b}	67.32	63.60^{b}	.97
Ash	51.88^{a,c}	64.13^{b}	39.86^{b}	50.46	44.84^{d}	1.86

*Values in the same row with different superscripts are different:

^{a,b} $P < .007$

^{c,d} $P < .02$

Table 4. Fecal outputs from horses (n = 8) with an average weight of 554 ± 23 kg, fed either tall fescue/alfalfa (TF/A) or orchardgrass/alfalfa (OG/A) hay. Comparison of TC values against predicted values*.

Variable	TF / A	OG / A	SEM
Fecal DM, weighed, kg/d	4.30^{a,b}	4.00^{a,c}	.20
Fecal DM predicted from daily Cr, kg/d	6.00^{a}	5.65^{a}	.20
Fecal DM predicted from asymptote of Cr (C_a), kg /d	5.01^{b}	4.75^{c}	.20

*Values in the same column with the same superscript are different from TC values

[a] P = .0001
[b] P = .022
[c] P = .017

Table 5. Dry matter intakes of either tall fescue/alfalfa (TF/A) or orchardgrass/alfalfa (OG/A) hay by horses (n = 8) with an average weight of 554 ± 23 kg. Intakes are given on a % BW basis in parentheses. Comparison of weighed intakes versus predicted intakes.*

Variable	TF / A	OG / A	SEM
DMI, kg/d, weighed	8.85 (1.6)^{a}	9.51 (1.7)^{d}	.45
DMI, kg/d, predicted from daily Cr^{a} and Y	16.23 (2.9)^{b}	18.42 (3.3)^{e}	.45
DMI, kg/d, predicted from daily Cr and C27	11.54 (2.1)^{b}	11.45 (2.1)^{f}	.45
DMI, kg/d, predicted from daily Cr and C29	11.58 (2.1)^{b}	13.24 (2.4)^{e}	.45
DMI, kg/d, predicted from daily Cr and C31	11.53 (2.1)^{b}	11.92 (2.2)^{f}	.45
DMI, kg/d, predicted from C_a and Y	14.26 (2.6)^{c}	15.44 (2.8)^{e}	.45
DMI, kg/d, predicted from C_a and C27	9.46 (1.7)	9.61 (1.7)	.45
DMI, kg/d, predicted from C_a and C29	9.47 (1.7)	11.11 (2.0)^{g}	.45
DMI, kg/d, predicted from C_a and C31	9.41 (1.7)	10.01 (1.8)	.45
DMI, kg/d, predicted from TC and Y	12.34 (2.2)^{c}	12.98 (2.3)^{e}	.45
DMI, kg/d, predicted from TC and C27	8.26 (1.5)	11.54 (1.5)	.45
DMI, kg/d, predicted from TC and C29	8.16 (1.5)	11.58 (1.7)	.45
DMI, kg/d, predicted from TC and C31	8.01 (1.4)	11.53 (1.5)	.45

[a] Daily Cr is the average amount of Cr measured in the combined grab samples from the last 4 d of dosing.

*Values in the same column with different superscripts from TC are different from TC values

^{a,b} $P < .009$
^{a,c} $P < .0002$
^{d,e} $P < .0002$
^{d,f} $P < .009$
^{d,g} $P < .013$

Table 6. Estimates of fit of pooled fecal Cr concentration data (Figure 1) to a one-compartment model, model parameters, and calculated variables in horses fed either tall fescue/alfalfa (TF/A) or orchardgrass/alfalfa (OG/A) hay.

Estimate	TF/A	OG/A
R^2	.810	.783
F	29.9	25.3
P- value	.0002	.0004
Cr_a, mg/kg[a]	4.14	4.39
SE	.4304	.5455
t-statistic	9.6	8.1
P-value	.0000	.0002
-k, d^{-1}[b]	1.227	1.082
SE	.590	.548
t-statistic	2.08	1.98
P-value	.0356	.0415
TT, h[c]	19.6	22.2
Delay, h[d]	4.97	6.70
FO, kg/d[e]	5.01	4.75
PFM, kg[f]	4.09	4.39

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[a] Cr_a is the asymptotic value of fecal Cr concentration.
[b] k is the rate constant.
[c] TT is turnover time of the prefecal mass, TT = -1/k
[d] Delay is the time between the administration of Cr and its entry into the prefecal mass
[e] FO is fecal output, FO = Dose of Cr/Cr_a.
[f] PFM is prefecal mass or mixing compartment, PFM = FO x TT.

Table 7a. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses fed tall fescue/alfalfa hay.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.582	9.8	.0071	.700	16.4	.0015
B	.475	6.3	.0228	.807	33.5	.0001
C	.756	21.7	.0006	.821	32.1	.0002
D	.696	16.0	.0016	.775	24.1	.0004
E	.635	12.2	.0037	.736	19.5	.0008
F	.622	11.5	.0044	.727	18.6	.0010
G	.687	15.3	.0019	.781	24.9	.0004
H	.786	25.8	.0002	.832	34.6	.0001
A to H[b]	.742	20.1	.0008	.810	29.9	.0002

[a] Delay was 4.97 h for horses fed tall fescue/alfalfa hay.

[b] Pooled data as in Figure 1.

Table 7b. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses fed orchardgrass/alfalfa hay

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.696	16.0	.0016	.795	27.2	.0003
B	.675	14.5	.0022	.779	24.7	.0004
C	.742	20.1	.0008	.823	32.5	.0001
D	.688	15.5	.0018	.787	25.8	.0003
E	.516	7.5	.0146	.660	13.6	.0027
F	.665	13.9	.0025	.754	21.5	.0006
G	.548	8.5	.0105	.691	15.6	.0017
H	.480	6.5	.0211	.634	12.2	.0037
A to H[b]	.683	15.1	.0018	.783	25.3	.0004

[a] Delay was 6.70 h for horses fed orchardgrass/alfalfa hay.
[b] Pooled data as in Figure 1.

Table 8. Fecal kinetics in horses (n = 8) fed either tall fescue/alfalfa (TF/A) or orchardgrass/alfalfa (OG/A) hay estimated with a one-compartment model with a delay[a] between time of administration and entry of marker into the compartment.

Variable	TF/A	OG/A	SEM	*P*-value
Fecal output, kg DM/d	5.01	4.75	.34	.47
Fecal output g/kg of BW daily	9.07	8.81	.73	.73
Turnover time, h	19.6	22.2	1.3	.09
Prefecal mass, kg DM	4.09	4.39	.15	.09
Prefecal mass, g/kg of BW daily	7.41	8.15	.65	.30

[a] The delays were .207 and .279 d for TF/A and OG/A, respectively.

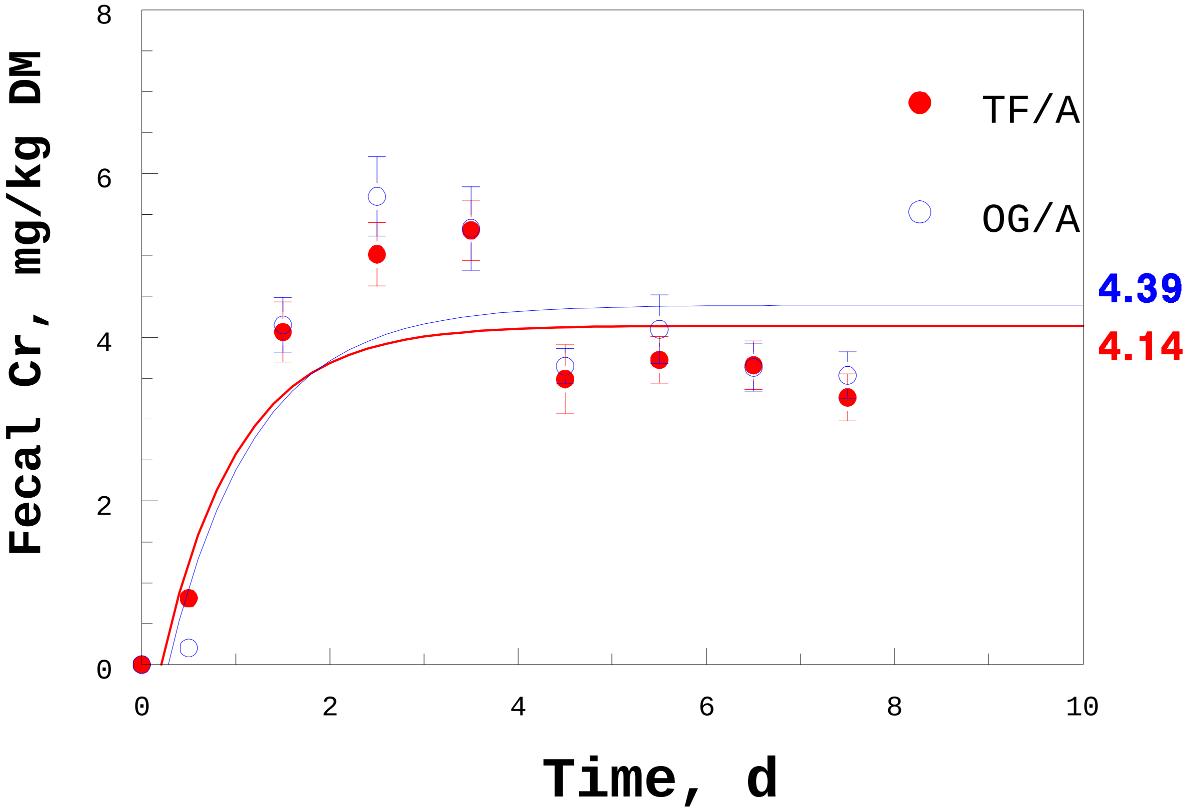

Figure 1. Mean daily fecal concentration of Cr, C*t*, for horses fed tall fescue/alfalfa (TF/A) or orchardgrass/alfalfa (OG/A) hay are plotted against time, and the data are fit to a one compartment model (see Figure 1, Holland et al., 1998).

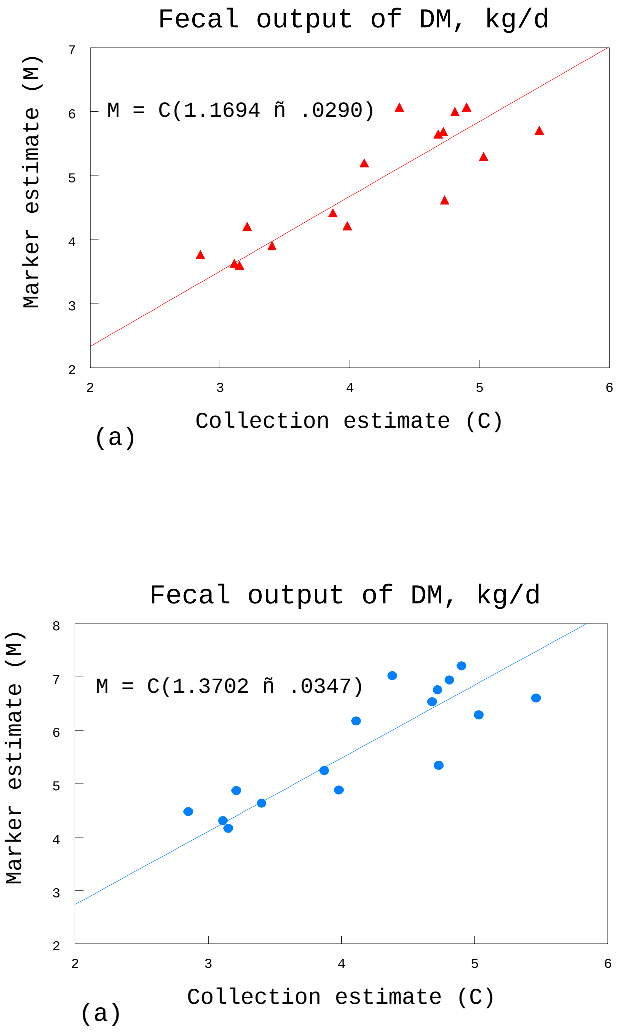

Figure 2. Fecal outputs predicted by marker dilution were correlated with collection values, both unadjusted (a) and adjusted (b) for Cr recoveries in feces. The correlation coefficient was not improved by adjustment (r^2 = .7883 and .7820, respectively).

Chapter 5. Fecal Output, Digestibility, and Intake of Pastures, Estimated by Marker Methods in Stalled and Grazing Horses

ABSTRACT

Marker methods have been developed that accurately estimate fecal output (FO), dry matter digestibility (D), hence, dry matter intake (DMI) in horses fed hays while confined in stalls. These methods were tested in grazing horses, but weighed intake and output were not available for comparison. Chromium (Cr) was used as an external marker and yttrium and n-alkanes as internal markers in 8 mature Thoroughbred geldings consuming either bluegrass/white clover or tall fescue/alfalfa pasture in a replicated 2x2 Latin Square. Horses were divided into two groups and placed on pastures for 14 d prior to the beginning of balance experiments, to allow dietary accommodation. Just prior to the balance experiments, 2 horses from each pasture were placed in stalls, where they were fed fresh-cut pasture 4 times daily, for the duration of the trials. Balance experiments were conducted for 15 d during each trial. Chromic oxide was administered 3 times a day in a "greenola bar" (6.40 ± .48 g Cr/bar) at 700, 1300, and 1900 h. Daily fecal Cr concentration (C_t, g/kg DM), for each diet, were applied to a simple monoexponential model with a single rate constant (k), rising to an asymptote (C_a). Delays (*d*) were added to the models (6.29 h for BG/WC and 5.98 h for TF/A) to simulate the time needed for Cr to enter the prefecal pool: $Ct = C_a - C_a \cdot e^{-k(t-d)}$. C_a was then used to estimate FO: $FO = Cr\ dose^{-d} / C_a$. Yttrium and n-alkanes were used as internal markers to estimate D. These estimates were then used with FO estimates to predict DMI: $DMI = FO / (1-D)$. All estimates were compared to weighed intakes, fecal outputs, and digestibilities from stalled horses. Fecal output estimated by C_a was not different from FO by collection ($P = .365$). Yttrium slightly overestimated and the alkane C27 slightly underestimated D ($P > .08$ for both) on both diets in stalled horses. Since Y and C27 gave similar estimates of D, compared to TC, they combined well with FO C_a to predict DMI ($P > .10$). As in stalled horses fed hay, the use of Cr, when applied to a monoexponential model, gave accurate and precise estimates of FO in horses consuming pasture. Results of internal marker digestibilities, in combination with FO predictions, indicate that these markers may be used successfully in grazing horses.

Key Words: Fecal Output, Digestibility, Intake, Pasture, Fecal Kinetics

Introduction

Fecal output of grazing animals is important in evaluating intake and digestion of various nutrients. Once fecal output can be accurately and precisely determined, it will be possible to determine the output of nutrients, such as N and P, that can have a negative environmental impact (Oldham and Tamminga, 1995; Van Horn et al., 1996).

Multiple compartment models of marker excretion have been the most effective in ruminants for predicting fecal output (Grovum and Williams, 1973; Quiroz et al., 1988) However, only a simple, one-compartment model was defined by marker data in horses, and relates to the simple stomach found in nonruminant herbivores (Holland et al., 1998). This model has not been validated in horses consuming pasture.

Chromic oxide has worked well as an external marker to predict fecal output when it was applied to the simple monoexponential model (Holland et al., 1998). This has been an improvement over the traditional estimate using a plateau value of fecal Cr from the last few days of dosing (Cuddeford and Hughes, 1990) because it gives a more accurate estimate of the asymptotic fecal Cr excretion, or how Cr would be excreted if Cr was dosed for an infinite number of days.

Along with fecal output, it is necessary to find internal markers to predict nutrient digestibility. These two estimates can then be used to predict intake. Various internal markers have been tried with varying success (see review by Kotb and Luckey, 1972). Often, markers that are assumed to be indigestible and unabsorbable have been found to be partially digested or degraded while travelling through the digestive tract (Cochran et al., 1986; Judkins et al., 1990). One type of marker that has been evaluated favorably in ruminants is n-alkanes; the wax component of plants (Dove and Mayes, 1996).

The objective of this study was to evaluate how well the markers chromium, yttrium and n-alkanes estimated fecal output and digestibilities in horses fed fresh-cut pasture in stalls. These values were then combined to estimate intake. Results in stalled animals were compared to results from horses grazing pasture.

Materials and Methods

Eight mature Thoroughbred geldings, average weight 563 ± 22 kg, were used in these studies, which were conducted during the months of June and August, 1997. Geldings were paired by weight and divided into two groups of four. Each group was placed into one of two 8-acre pastures for the first trial and then switched to the other pasture for the second trial in a replicated 2 x 2 Latin Square design, with periods of 15 d. One pasture was predominately bluegrass (*Poa pratensis*) and white clover (*Trifolium repens*) while the second was predominately tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*). Horses were placed on their respective pastures 2-wk prior to the beginning of data collection. Two horses from each group were removed from the pastures before data collection and housed in individual box stalls, 14 m^2, for the duration of each trial.

Pastures were clipped every morning using a Graveley mower (Model 5665, Graveley International, Brillion, WI) so that the length of stems and leaves was not disturbed. Enough pasture was cut to feed the stalled horses for the next 24 h. After clipping, pasture was raked and bagged and stored in a cool room to inhibit fermentation. Stalled horses were offered this fresh-cut pasture in 4 small meals at 700, 1300,1900, and 2200 h to simulate grazing. Horses were fed as much pasture as they could consume so that intake was not limiting.

During the first 3 d of each trial, baseline fecal samples were collected. On d 4, horses began to be dosed three times daily, at 700, 1300, and 1900 h, with a “greenola bar” containing 6.40 ± .48 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for eight d, from d 3 until d 11. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected three times a day, when bars were fed. Fecal samples were collected three times daily for the 4 d after dosing ceased. Fecal samples from the same day were combined, weighed, dried in a forced-air oven to a constant weight, and then reweighed for DM determination. On the last day of dosing, a subsample was taken from each fecal grab sample so diurnal excretion of Cr could be determined. Total collection (TC) of feces was made during d 7 to d 11 from stalled horses; feces were collected in separate tubs for each horse. Feces from each 24 h period were weighed. Subsamples from individual grab samples and TC samples were weighed, dried to constant weight in a forced air over, and weighed again for DM determination.

Pasture samples were subsampled from the daily mowed samples every other day during the dosing phase of the trial. These samples were weighed, dried in a forced-air oven to constant weight, and weighed again for DM determination. These samples were also analyzed for nutrient content as described below.

On one day of each trial pasture samples were collected for botanical composition by methods developed by Blaser et al. (1986). A stick, 1.83 m, was thrown randomly into each acre. The pasture to the left of the stick was clipped using hand-held electric clippers. Total samples for each pasture were hand separated into edible plants and weeds. Samples were weighed wet, to give a total weight and then all samples were dried so that percentages of wet and dry pasture could be determined (Table 2).

Samples of pastures and feces were ground through a .5 mm screen Thomas-Wiley Laboratory Mill (Model 4, Thomas Scientific, Swedesboro, NJ). Composite samples were made from the pasture samples and from the feces from the last 4 d of dosing. These composites were submitted for analysis of DM, CP, EE, NSC, NDF, ADF, Ca, P, K, Mg, Na, Fe, Zn, Cu, Mn, Mo, S and ash by standard methods (Robertson and

Van Soest, 1977; AOAC, 1990) at the Northeast DHIA Laboratory (Ithaca, New York). Pasture analysis can be found in Table 3.

The twelve reserved "greenola bars" were finely ground in a coffee grinder (Electric Miracle Seed&Coffee Mill, Model MC200, Whatman Scientific, Hillsboro, OR). Cr was analyzed in "greenola bars", pasture composites, fecal composites and daily fecal samples by atomic absorption spectrophotometry with an air -acetylene flame after samples were digested with nitric and perchloric acids (Sandel, 1959). Two dilutions of each Cr sample were analyzed in duplicate, and five dilutions of standard solution of potassium dichromate were used for calibration. Yttrium (Y) was analyzed in pasture and fecal composite samples by inductively coupled plasma spectroscopy after samples were digested with nitric and perchloric acid. Alkane content was analyzed following preparation methods developed by Ordakowski (1998) and using gas chromatography (Model 5890A, Hewlett Packard, Wilmington, DE).

Dry matter digestibility (D, %) was determined by two methods. The first was calculated from total collection data of both daily DM intake (I, kg) and fecal output (FO, kg):

$$D = (1-FO/I)*100$$

The second method involved using a ratio of the internal markers (M), either Y or alkanes:

$$D = (1- C_I/C_F)*100$$

where C_I is [M] in the feed samples and C_F is [M] in the fecal samples.

Nutrient digestibilities (ND) were evaluated from the total collection data using the following equation:

$$ND = (1- N_O/N_C)*100$$

where N_C is the total amount of a nutrient consumed (kg/d), and N_O was the total amount of a nutrient found in the feces (kg/d) of an individual animal. Nutrient digestibilities were also calculated using the internal markers Y and n-alkanes:

$$ND=[1-(C_I / C_F)(N_F / N_I)]*100$$

where N_I (%DM) is the concentration of a nutrient in the feed samples and N_F (%DM) is the concentration of a nutrient in the fecal samples.

Pooled daily fecal Cr concentrations (C_t, mg/kg DM) for all 8 horses on one diet was analyzed in terms of a simple, monoexponential model rising to an asymptote (C_a) using a graphics and curve-fitting program (SlideWrite Plus 4.0 for Windows, Advanced Graphics Software, Carlsbad, CA) as previously described (Holland et al., 1998):

$$C_t = C_a - C_a \cdot e^{-kt}$$

Where Ct was fecal Cr concentration (mg/kg DM) at time t (days). The asymptotic value of fecal Cr determined fecal output (FO):

$$FO = (\text{Cr dose}/C_a).$$

The rate constant (k, d^{-1}) determined its reciprocal, the turnover time (TT, d). Therefore, the compartment size, or mass, of the prefecal pool (PFM, kg) can be calculated:

$$PFM = FO \times TT = FO/k.$$

Fecal outputs predicted by C_a were compared to total collection values. FO and D predicted from markers were used to calculate dry matter intake (DMI, kg/d):

$$DMI = FO/(100 - D)$$

and these predicted intakes were compared to amounts of DM fed.

The data were summarized as means and SEMs, and analyzed by the GLM Procedure of SAS (SAS Institute, Inc., Cary, NC) with horse, diet, method and all interactions in the model. Differences in digestibilities predicted by Y and alkanes were compared to total collection values. Differences in predicted fecal outputs from C_t and C_a were compared to TC values. Intake predictions from markers were compared to weighed intakes. Comparison of means used Fisher's protected LSD.

Results and Discussion

Digestibilities. Nutrient digestibilities, in stalled horses, calculated from weighed intake and FO were not different between diets ($P > .082$) except for NSC ($P = .0005$)

which was higher in TF/A (Table 4b) than in BG/WC (Table 4a). Yttrium slightly overestimated nutrient digestibilities ($P > .076$) except for NSC, which was higher than TC ($P = .037$) in stalled horses. The n-alkane C27 slightly underestimated nutrient digestibilities ($P > .074$) in stalled horses, compared to calculated digestibilities. The other two n-alkanes, C29 and C31, also underestimated nutrient digestibilities, compared to calculated values ($P < .007$). When methods were compared in all horses, C27 was different from C29 and C31 ($P = .0001$) and Y ($P < .007$). Digestibility predicted by Y was also different from C29 and C31 ($P = .0001$). Digestibility predictions from C29 and C31 were not different from each other ($P > .070$) except when predicting CP digestibility ($P = .024$). Differences in digestibility estimates, could be due to concentrations of internal markers in various plants. Alkane content varies from plant to plant and in different parts of the same plant, so pasture samples may not have accurately represented pasture consumed (Dove, 1992). Although not documented, a similar occurrence may happen with other internal markers.

When compared on a pasture basis, C27 nutrient digestibilities were not different from calculated values when stalled horse were consuming either BG/WC ($P > .427$) , or TF/A ($P > .075$). Y was not a good predictor of nutrient digestibilities when horse were consuming BG/WC ($P < .029$) but was a good predictor when horses consumed TF/A ($P > .781$). Estimates of D from C29 and C31 were different from the balance estimates ($P < .05$).

When D estimates were compared on a diet basis for all horses, C27 was different than C29 and C31 ($P < .0009$) and Y ($P < .004$). C29 and C31 were not different ($P > .091$) but both were different from Y ($P < .001$). When horses were consuming TF/A, C27 predictions were not different from Y ($P > .068$) but were different from C29 and C31 ($P < .020$). Y predictions were different from C29 and C31 ($P < .01$), but C29 and C31 were not different from each other ($P > .107$).

Nutrient digestibilities for stalled horses were lower than for pastured horses, possibly due to the forage clipped by the investigators. Strips were cut randomly across the pastures, and were probably not indicative of what the geldings would consume when foraging. Pasture samples can be collected using esophageal or ruminal fistulas in grazing ruminants (Olson, 1991), but this is not used in horses. By cutting the pasture and offering it so that refusals were minimal, we removed most of the selectivity that horses use when pasture is available free-choice. Sheep (Cherney et al., 1990), cattle (Burns et al., 1997) and horses (Dulphy et al., 1997a) will consume forages of higher digestibility and lower maturity, if a choice is available. The lower digestibilities in stalled horses was probably related to lack of choice. Noblitt et al. (1963) observed an increase in digestibility in stalled animals, when under a high stress handling procedure. No mention was made of fecal output or quality and it is possible that increases in digestibility were related to faster passage of digesta.

Yttrium gave very high estimates of nutrient digestibilities in the grazing horses when compared to stalled horses. Estimates were often 50% higher. Rare earth elements, in general, are found in minute levels in most forages (Crooker et al., 1982; Turnbull and Thomas, 1987), so it is possible that contamination occurred on pasture, most likely from mineral (soil) consumption. Previous studies in cattle found that n-alkanes underestimated the digestibility of alfalfa and grass hays by 15 to 20% (Ohajuruka and Palmquist, 1991). This might be the case in the stalled horses of this study, but probably not the pastured horses. Except for predictions from C27, the estimates using alkanes are within normal ranges of digestibility for pastures. Some error in using alkanes has been related to the differing levels of alkanes in different plant parts, with leaves containing higher levels than stems (Casson et al., 1990; Dove and Moore, 1995). Estimates from pasture may be more accurate, because animals could choose what was ingested, compared to what horses were offered in stalls.

Fecal Output. Actual fecal output according to diet was different in stalled horses ($P < .005$); higher when fed TF/A (Table 5a). Overall, C_a (the asymptotic level of Cr excretion from prediction equations) estimated FO's similar to TC ($P = .365$) but average daily Cr excretion (C_d) overestimated FO (Table 5a). When all horses were considered, C_a FO predictions were not different from C_d (Table 5b). On a diet basis, in stalled horses, C_a and C_d predicted FO's similar to TC (P = .240 and .064, respectively) when horses were consuming BG/WC. When horses were consuming TF/A, C_a and C_d both predicted TF FO well (P = .921 and .186, respectively). Predictions of FO, on a diet basis, were also compared with all horses. Ca and Cd predictions were not different when horses were consuming BG/WC (P = .321) or TF/A (P = .630) pasture.

Higher fecal output is associated with the higher dry matter intakes of TF/A, both weighed and predicted for stalled and grazing horses. Fecal output predictions for stalled horses were lower than grazing horses and again, was influenced by intake (Table 6). Housing should not have influenced fecal output. According to work conducted in sheep, fecal outputs were similar when sheep were dosed with Cr to predict FO, fit with total collection harnesses, and confined to metabolism crates (Hatfield et al., 1993). The confined animals did have slightly lower FO, similar to the results in this study.

Intake. In stalled horses (Table 6a), there was no difference in weighed intakes, according to diet (P = .325). C_a and Y, in combination with C27, predicted intakes similar to weighed intakes (P = .105 and .185, respectively). C_d, in combination with C27, C29, and C31, gave good predictions of actual intake (P = .749, .269, and .252, respectively). On a diet basis, C_a and C27 predicted actual intakes when horses were consuming BG/WC (P = .244) and TF/A (.251). C_a worked with all internal markers to predict intake when horses were consuming BG/WC (P > .075). This was also observed when horses were consuming TF/A (P > .153 for C29 and C31; P > .87 for Y).

On a diet basis, many methods gave similar intake predictions in all horses (Table 6b). A Ca and C27 combination was not different from Ca and Y on either BG/WC (P > .459) or TF/A (P > .597). Ca and C29 was not different from Ca and C31 on either diet (P > .924 on BG/WC; P > .837 on TF/A). However, intake predictions made with Ca with either Y or C27 were different from predictions from Ca with either C29 or C31 on both BG/WC (P < .022) and TF/A (P < .019).

Weighed dry matter intakes were 1.4 and 1.7 %BW for BG/WC and TF/A, respectively. This falls within the range of expected DMI for mature horses at maintenance, 1.4 to 1.8 %BW (NRC, 1989), and several combinations of markers gave similar estimates (Table 6a). However, these horses were stalled constantly and expended less energy than horses on pasture. Methods that predicted DMI well in stalled horses gave much higher estimates of intake for grazing horses (up to 7% BW), and indicates a trend towards higher intakes in grazing horses. Broodmares in late gestation and early lactation have higher intake requirements than maintenance geldings, and consumed between 1.5 and 2.5 % of BW as estimated using markers (Martin et al., 1989). Mature Arabian geldings consuming different hays while confined in stalls consumed approximately 2.5% of BW (Crozier et al., 1997). A review of studies of intakes has set estimates between 1.1 and 2.5 % of BW, depending on forage type (Dulphy et al., 1997b), indicating that the predictions here are within an acceptable range.

Fecal Kinetics. Mean C_t for the two pastures were fit to the monoexponential equation and plotted against time, rising to an asymptote (Figure 1). The monoexponential equation provided a good fit for both BG/WC ($r^2 = .856$) and TF/A ($r^2 = .862$) without a delay. The delay, representing time for Cr to reach the prefecal mass, can give an estimate of transit time of the digesta as well, working as a total digesta tracer (Titgemeyer, 1997), and would give more information on the total structure of the system, such as turnover time and compartment size (Brownell et al., 1968). This information would be especially useful when supplements are added to the diet of performance horses, as digesta weight or “bowel ballast” could adversely affect performance (Holland et al., 1998).

When a delay (*d,* d) was added to the monoexponential equation as a reduction in time (*t)*:

$$C_t = C_a - C_a \cdot e^{-k(t-d)}$$

the fit was improved (Table 7). Delays were figured to be 6.29 h for BG/WC and 5.98 h for TF/A. The longer delays indicate that the pasture was kept within the digestive tract, indicating that the pasture was of lower quality during this trial. Pastures were higher in

DM and fiber, and lower in CP than when previous studies were conducted (Journal Paper 2). These factors have been associated with longer retention times and lower intakes in steers (Burns et al., 1997; Gunter et al., 1997).

The Ct data for individual horses was fit to the monoexponential equation both with and without the delay. Improvement was observed for each horse on each diet (Tables 8 a and b). Predictions of FO, TT and PFM are given in Table 9. Estimates of retention time, or turnover time, were similar to previously published values for horses consuming hay (Vander Noot et al., 1967) and ruminants (Jung and Allen, 1995); longer TT for BG/WC pasture was due to the higher fiber content of the pasture.

Marker estimates of fecal output were correlated to total collection in stalled horses, although not well (Figure 2). Excretion of Cr in the feces does have diurnal variation, related to time of feeding. Since Cr was dosed to these geldings at times when pasture was offered, there could be a negative influence on recovery.

Implications

The monoexponential equation allowed an improved fecal output prediction. Yttrium may not be a satisfactory internal marker for grazing horses, but n-alkanes may be usable. Time of dosing Cr, in relation to time of forage offer, should be evaluated.

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Table 1. Recipe for "greenola bars"[a] for fecal kinetic studies.

Ingredients

Oat-based sweet feed	2.16 kg
Cr_2O_3	800 g
Liquid molasses	1 L
Beer	380 mL

Instructions

Mix Cr_2O_3, molasses and beer thoroughly, making sure Cr_2O_3 is well-blended. Add sweet feed slowly, making sure all particles are covered. Divide mixture between 4 well-oiled 12"x15" pans. Put into oven set at low temperature. When mixture begins to set, cut into 20 bars per tray. Put back into oven until completely dry. Remove from pans and place each bar into an individual bag for ease of handling.

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[a] Adapted from Practical Horseman . 1996. 24(4):122. Horse Cookie recipe

Table 2a. Botanical composition (BC) of bluegrass/white clover (BG/WC) pasture (mean ± SE).

BG/WC[a] Plant	% of wet pasture	% of dry pasture
Bluegrass (*Poa pratensis*)	61.94 ± 15.54	55.34 ± 1.95
White Clover (*Trifolium repens*)	20.44 ± 9.85	21.92 ± .73
Tall Fescue (*Festuca arundinacea*)	.39 ± .39	1.27 ±1 .27
Orchardgrass (*Dactylis glomerata*)	2.23 ± 1.78	2.62 ± .93
Red Clover (*Trifolium pratense*)	.62 ± .62	.59 ± .59
Plantain (*Plantago major*)	9.38 ± 1.11	13.26 ± 7.94
Curly Dock (*rumex crispus*)	4.34 ± 4.34	4.43 ± 4.43
Misc. Weeds	.69 ± .47	.59 ± .59

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[a] Percent DM of total BC sample 27.37 ± 14.21

Table 2b. Botanical composition (BC) of tall fescue/alfalfa (TF/A) pasture (means ± SE).

TF/A[a] Plant	% of wet pasture	% of dry pasture
Tall Fescue (*Festuca arundinacea*)	67.82 ± 7.11	50.29 ± 16.39
Alfalfa (*Medicago sativa*)	9.74 ± 4.7	13.56 ± .85
White Clover (*Trifolium repens*)	10.03 ± 4.56	12.20 ± .52
Bluegrass (*Poa pratensis*)	1.16 ± .56	3.48 ± 2.45
Orchardgrass (*Dactylis glomerata*)	.62 ± .02	1.19 ± .50
Ryegrass (*Lolium perenne*)	.08 ± .08	.17 ± .17
Dandelion (*Taraxacum officinale*)	.81 ± .81	2.12 ± 2.12
Plantain (*Plantago major*)	3.70 ± 3.70	9.32 ± 9.32
Chickory (*Chichorium intybus*)	1.24 ± 1.24	3.39 ± 3.39
Bromegrass (*Bromus secalinus*)	.38 ± .38	.84 ± .84
Misc. Weed	4.47 ± 4.47	3.44 ± 3.44

[a] Percent DM of total BC sample 27.98 ± 15.33

Table 3. Nutrient and internal marker content[a] of pastures, means ± SE.

Nutrient	bluegrass / white clover	tall fescue / alfalfa
DM, %	29.94 ± 2.85	33.19 ± 2.71
Composition of DM		
CP, %	12.6 ± .4	12.2 ± .9
ADF, %	40.5 ± 1.0	42.1 ± .9
NDF, %	66.9 ± 1.5	63.8 ± .5
EE, %	3.1 ± .2	3.1 ± .4
NSC[b], %	10.3 ± 1.1	11.5 ± .6
Ca, %	.67 ± .05	.91 ± .07
P, %	.30 ± .00	.31 ± .00
Mg, %	.18 ± .00	.31 ± .02
K, %	1.78 ± .25	2.12 ± .22
Na, %	.004 ± .001	.010 ± .003
Fe, ppm	481 ± 21	316 ± 46
Zn, ppm	39 ± 2	35 ± 1
Cu, ppm	5 ± 1	5 ± 0
Mn, ppm	58 ± 1	92 ± 5
Mo, ppm	1.7 ± .1	1.7 ± .1
S, %	.18 ± .01	.24 ± .00
Ash, %	7.03 ± .18	9.39 ± .04
Cr, ppm	42 ± 24	51 ± 29
Y, ppm	.39 ± .06	.76 ± .18
Alkanes, ng/g		
C27	15.95 ± 1.07	15.82 ± .38
C29	48.54 ± 1.52	51.85 ± 1.36
C31	114.12 ± 1.19	121.89 ± 2.38

[a] Analysis of CP, EE, and ash (AOAC, 1990), ADF and NDF (Robertson and Van Soest, 1977), Cr (Sandel, 1959)

[b] NSC = 100 - (CP% + EE% + NDF% + ash%)

Table 4a. Digestibilities (%), means ± SE, of DM, CP, ADF, NDF, EE, NSC and Ash in horses fed bluegrass/white clover (BG/WC) pasture. Predicted either from total collection (TC), yttrium (Y) or C27, C29 or C31 n-alkanes.*

BG/WC PASTURED

Nutrient	Y	C27	C29	C31
DM	91.68 ± 2.29^{a}	85.99 ± 4.04^{a}	56.44 ± 2.56^{b}	48.14 ± 2.70^{b}
CP	92.65 ± 1.76^{a}	86.67 ± 4.25^{a}	60.21 ± 2.60^{b}	52.34 ± 4.20^{b}
ADF	90.87 ± 2.36^{a}	83.81 ± 5.23^{a}	51.22 ± 3.26^{b}	42.11 ± 2.59^{b}
NDF	92.72 ± 1.98^{a}	87.58 ± 3.69^{a}	61.72 ± 2.31^{b}	54.49 ± 2.23^{b}
EE	67.40 ± 8.88^{c}	44.22 ± 16.36^{c}	-71.49 ± 9.24^{d}	-104 ± 8.76^{d}
NSC	96.19 ± 1.40^{c}	$94.93 \pm .82^{c}$	81.68 ± 3.47^{d}	82.55 ± 4.98^{d}
Ash	86.22 ± 3.04^{a}	74.12 ± 8.76^{a}	24.50 ± 3.17^{b}	10.17 ± 2.50^{b}

*Values in the same row with different superscripts are different:

^{a,b} $P = .0001$

^{c,d} $P < .01$

BG/WC STALLED

Nutrient	TC	Y	C27	C29	C31
DM	56.88 ± 4.62^{a}	69.25 ± 5.88^{b}	53.03 ± 1.78	43.44 ± 3.31^{b}	43.50 ± 3.91^{b}
CP	$70.14 \pm 3.72^{a,c}$	78.56 ± 4.40^{d}	67.49 ± 1.47	62.08 ± 2.73^{d}	60.82 ± 3.22^{b}
ADF	$47.93 \pm 4.27^{a,c}$	63.16 ± 6.15^{d}	42.94 ± 1.84	31.38 ± 2.95^{b}	31.60 ± 3.14^{b}
NDF	$56.32 \pm 4.00^{a,c}$	68.93 ± 5.55^{d}	52.21 ± 1.96	42.47 ± 3.32^{b}	42.62 ± 3.56^{b}
EE	$27.38 \pm 6.55^{a,c}$	48.79 ± 8.56^{b}	20.57 ± 2.32	4.63 ± 2.35^{d}	4.87 ± 3.43^{d}
NSC	65.38 ± 9.10^{a}	74.72 ± 8.17^{b}	63.64 ± 6.15	56.32 ± 7.09^{b}	55.87 ± 8.43^{b}
Ash	$37.42 \pm 4.16^{a,c}$	55.91 ± 6.82^{d}	31.28 ± 1.24	17.15 ± 4.43^{b}	17.46 ± 4.30^{b}

*Values in the same row with different superscripts are different:

^{a,b} $P <.02$

^{c,d} $P < .05$

Table 4b. Digestibilities (%), means ± SE, of DM, CP, ADF, NDF, EE and NSC in horses fed tall fescue/alfalfa (TF/A) pasture. Predicted either from total collection (TC), yttrium (Y) or C27, C29 or C31 n-alkanes.*

TF/A PASTURED

Nutrient	Y	C27	C29	C31
DM	92.08 ± 1.45^{a}	90.46 ± 2.32^{a}	$71.96 \pm .75^{b}$	64.18 ± 1.31^{b}
CP	91.64 ± 1.56^{a}	89.64 ± 2.89^{a}	70.38 ± 2.00^{b}	62.14 ± 2.87^{b}
ADF	91.62 ± 1.51^{a}	89.96 ± 2.38^{a}	$70.33 \pm .45^{b}$	62.10 ± 1.20^{b}
NDF	92.79 ± 1.32^{a}	91.39 ± 2.03^{a}	$74.51 \pm .34^{b}$	$67.44 \pm .91^{b}$
EE	68.49 ± 2.89^{c}	58.92 ± 11.86^{c}	-18.24 ± 13.15^{d}	-52.26 ± 20.6^{d}
NSC	97.26 ± 1.21^{c}	96.93 ± 1.36^{c}	91.19 ± 2.79	88.95 ± 3.25^{d}
Ash	$88.88 \pm 1.86^{a,c}$	86.31 ± 3.28^{c}	59.98 ± 2.29^{d}	48.82 ± 3.48^{b}

*Values in the same row with the same superscript are different:

^{a,b} P = .002

^{c,d} P < .02

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TF/A STALLED

Nutrient	TC	Y	C27	C29	C31
DM	60.55 ± 2.61^{a}	61.08 ± 6.22	51.31 ± 2.21	49.63 ± 2.28^{b}	49.02 ± 1.68^{b}
CP	72.36 ± 2.45^{a}	72.37 ± 5.35	66.13 ± 1.14	64.83 ± 1.95^{b}	64.40 ± 1.94^{b}
ADF	53.28 ± 1.97^{a}	54.35 ± 5.99	41.92 ± 3.93	40.07 ± 2.87^{b}	39.37 ± 2.64^{b}
NDF	58.44 ± 2.41^{a}	59.13 ± 6.11	48.55 ± 2.99	46.79 ± 2.63^{b}	46.20 ± 2.22^{b}
EE	36.73 ± 3.25^{a}	38.64 ± 7.03	20.99 ± 7.33	18.60 ± 5.87	17.72 ± 5.51^{b}
NSC	77.51 ± 4.45^{a}	76.77 ± 7.04	73.17 ± 3.77	71.89 ± 4.60^{b}	71.45 ± 5.01^{b}
Ash	46.33 ± 4.81^{a}	47.93 ± 7.20	33.41 ± 6.10	31.20 ± 5.87	30.69 ± 4.55^{b}

*Values in the same row with the same superscript are different:

^{a,b} P = .05

Table 5a. Fecal outputs from stalled horses (n = 4) fed fresh-cut bluegrass/white clover (BG/WC) or tall fescue/alfalfa. Average weight was 563 ± 22 kg. Comparison of TC values against predicted values.

Variable	BG / WC	TF / A	SEM
Fecal DM, kg/d, weighed	3.18	3.65	.21
Fecal DM predicted from daily Cr[a], kg/d	3.77	4.06	.21
Fecal DM predicted from C_a, kg/d	2.82	3.62	.21

[a] Daily Cr is the average fecal [Cr] over the last 4 d of dosing.

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Table 5b. Fecal outputs from horses (n = 8) consuming either bluegrass/white clover (BG/WC) or tall fescue/alfalfa pasture. Average weight was 563 ± 22 kg. No difference was found between methods.

Variable	BG / WC	TF / A	SEM
Fecal DM predicted from daily Cr, kg/d	4.33	4.41	.41
Fecal DM predicted from C_a, kg/d	3.75	4.12	.41

Table 6a. Dry matter intakes of fresh-cut bluegrass/white clover or tall fescue/alfalfa by stalled horses (n = 4). Average weight was 563 ± 22 kg. Intakes, on a % BW basis, are given in parentheses. Comparison of actual intakes versus predicted intakes.*

Variable	BG / WC	TF / A	SEM
DMI, kg/d, weighed	7.79 (1.4)^{a,b}	9.31 (1.7)	1.07
DMI, predicted from daily Cr and Y	13.29 (2.4)^{a}	10.88 (1.9)	1.07
DMI, predicted from daily Cr and C27	7.99 (1.4)	8.42 (1.5)	1.07
DMI, predicted from daily Cr and C29	6.61 (1.2)	8.08 (1.4)	1.07
DMI, predicted from daily Cr and C31	6.66 (1.2)	7.95 (1.4)	1.07
DMI, predicted from C_a and Y	10.45 (1.9)	9.55 (1.7)	1.07
DMI, predicted from C_a and C27	6.00 (1.1)	7.54 (1.3)	1.07
DMI, predicted from C_a and C29	5.01 (0.9)	7.21 (1.3)	1.07
DMI, predicted from C_a and C31	5.08 (0.9)	7.09 (1.3)	1.07
DMI, predicted from daily FO (TC) and Y	12.34 (2.2)^{b}	10.06 (1.8)	1.07
DMI, predicted from daily FO (TC) and C27	6.77 (1.2)	7.53 (1.3)	1.07
DMI, predicted from daily FO (TC) and C29	5.62 (1.0)	7.28 (1.3)	1.07
DMI, predicted from daily FO (TC) and C31	5.67 (1.0)	7.14 (1.3)	1.07

*Values in the same column with the same superscript are different from TC values

$^{a}P < .0001$

$^{b}P < .02$

Table 6b. Dry matter intakes of either bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture by horses (n = 8). Average weight was 563 ± 22 kg. Comparison of predicted intakes.*

Variable	BG / WC	TF / A	SEM
DMI, predicted from daily Cr and Y	36.24 (6.4)^{a}	38.84 (6.9)^{i}	1.07
DMI, predicted from daily Cr and C27	31.07 (5.5)^{b}	35.19 (6.3)^{j}	1.07
DMI, predicted from daily Cr and C29	8.56 (1.5)^{c}	12.52 (2.2)^{k}	1.07
DMI, predicted from daily Cr and C31	7.73 (1.4)^{d}	10.63 (1.9)^{l}	1.07
DMI, predicted from C_a and Y	35.21 (6.3)^{e}	37.25 (6.6)^{m}	1.07
DMI, predicted from C_a and C27	29.63 (7.0)^{f}	34.12 (6.1)^{n}	1.07
DMI, predicted from C_a and C29	7.77 (1.4)^{g}	11.89 (2.1)^{o}	1.07
DMI, predicted from C_a and C31	6.93 (1.2)^{h}	10.04 (1.8)^{p}	1.07

*Values in the same column with the same superscript are different

^{a,c} ^{a,d} ^{a,g} ^{a,h} $P < .003$
^{b,c} ^{b,d} ^{b,g} ^{b,h} $P < .02$
^{c,e} $P < .005$
^{c,f} $P < .03$
^{d,e} $P <.004$
^{d,f} $P < .03$
^{e,g} ^{e,h} $P < .004$
^{f,g} ^{f,h} $P < .02$

^{i,k} ^{i,l} ^{i,o} ^{i,p} $P < .005$
^{j,k} ^{j,o} $P < .02$
^{j,l} ^{j,p} $P < .008$
^{k,m} $P < .008$
^{k,n} $P < .02$
^{l,m} $P < .02$
^{l,n} $P < .005$
^{m,o} ^{m,p} $P < .007$
^{n,o} ^{n,p} $P < .02$

Table 7. Estimates of fit of pooled fecal Cr concentration data (Figure 1) to a one-compartment model, model parameters, and calculated variables in horses (n = 8) consuming either bluegrass / white clover (BG/WC) or tall fescue / alfalfa (TF/A) pasture.

Estimate	BG/WC	TF/A
R^2	.885	.897
F	53.8	61.0
P- value	.0000	.0000
Cr_a, mg/kg[a]	5.19	4.57
SE	.5249	.4150
t-statistic	9.6	11.0
P-value	.0000	.0000
-k, d^{-1}[b]	.647	.722
SE	.244	.252
t-statistic	2.65	2.86
P-value	.0146	.0106
TT, h[c]	37.1	33.3
Delay, h[d]	6.29	5.98
FO, kg/d[e]	3.75	4.12
PFM, kg[f]	5.80	5.72

[a] Cr_a is the asymptotic value of fecal Cr concentration.
[b] k is the rate constant.
[c] TT is turnover time of the prefecal mass, TT = -1/k
[d] Delay is the time between the administration of Cr and its entry into the prefecal mass
[e] FO is fecal output, FO = Dose of Cr/Cr_a.
[f] PFM is prefecal mass or mixing compartment, PFM = FO x TT.

Table 8a. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses fed bluegrass/white clover pasture.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.716	17.7	.0012	.795	27.2	.0003
B	.809	29.7	.0002	.845	38.1	.0001
C	.822	32.3	.0002	.857	41.9	.0001
D	.737	19.6	.0008	.816	31.0	.0002
E	.775	24.1	.0004	.788	26.0	.0003
F	.802	28.3	.0002	.812	30.2	.0002
G	.688	15.4	.0018	.764	22.7	.0005
H	.885	53.8	.0000	.897	61.0	.0000
A to H[b]	.856	41.6	.0001	.885	53.8	.0000

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[a] Delay was 6.29 h for horses fed bluegrass/white clover pasture.

[b] Pooled data as in Figure 2.

Table 8b. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses fed tall fescue/alfalfa pasture.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.742	20.1	.0008	.800	27.9	.0003
B	.751	21.2	.0006	.830	34.2	.0001
C	.817	31.3	.0002	.863	44.3	.0001
D	.784	25.3	.0004	.851	40.1	.0001
E	.828	33.7	.0001	.851	40.1	.0001
F	.877	49.8	.0000	.896	60.1	.0000
G	.703	16.6	.0014	.785	25.6	.0003
H	.851	39.9	.0001	.891	57.4	.0000
A to H[b]	.862	43.6	.0001	.897	61.0	.0000

[a] Delay was 5.98 h for horses fed tall fescue/alfalfa pasture.

[b] Pooled data as in Figure 2.

Table 9. Fecal kinetics in 8 horses fed either bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture estimated with a one-compartment model with a delay[a] between time of administration and entry of marker into the compartment.

Variable	BG/WC	TF/A	SEM	*P*-value
Fecal output, kg DM/d	3.75	4.12	.46	.45
Fecal output g/kg of BW daily	6.66	7.32	.33	.09
Turnover time, h	37.1	33.3	1.9	.09
Prefecal mass, kg DM	5.80	5.72	.04	.09
Prefecal mass, g/kg of BW daily	10.30	10.16	.07	.09

[a] The delays were .262 and .249 d for BG/WC and TF/A, respectively.

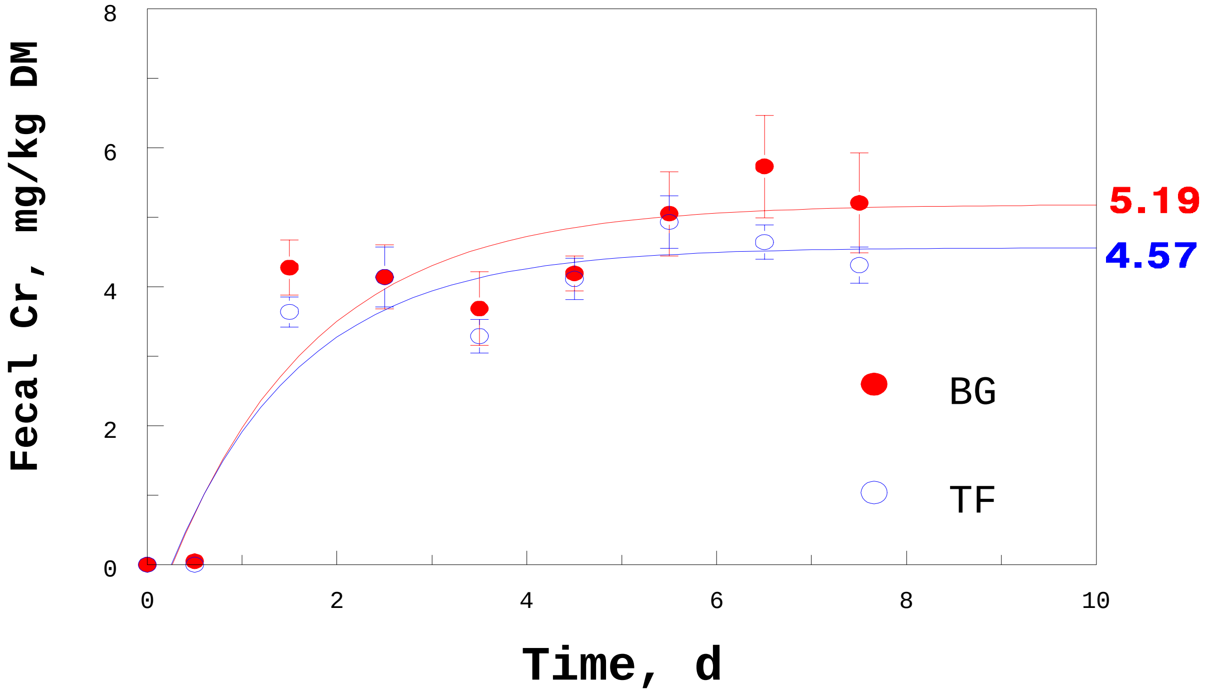

Figure 1. Mean daily fecal concentrations of Cr, C*t*, for horses fed bluegrass / white clover (BG/WC) or tall fescue / alfalfa (TF/A) pasture are plotted against time, and the data are fit to a one compartment model (see Figure 1, Holland et al., 1998).

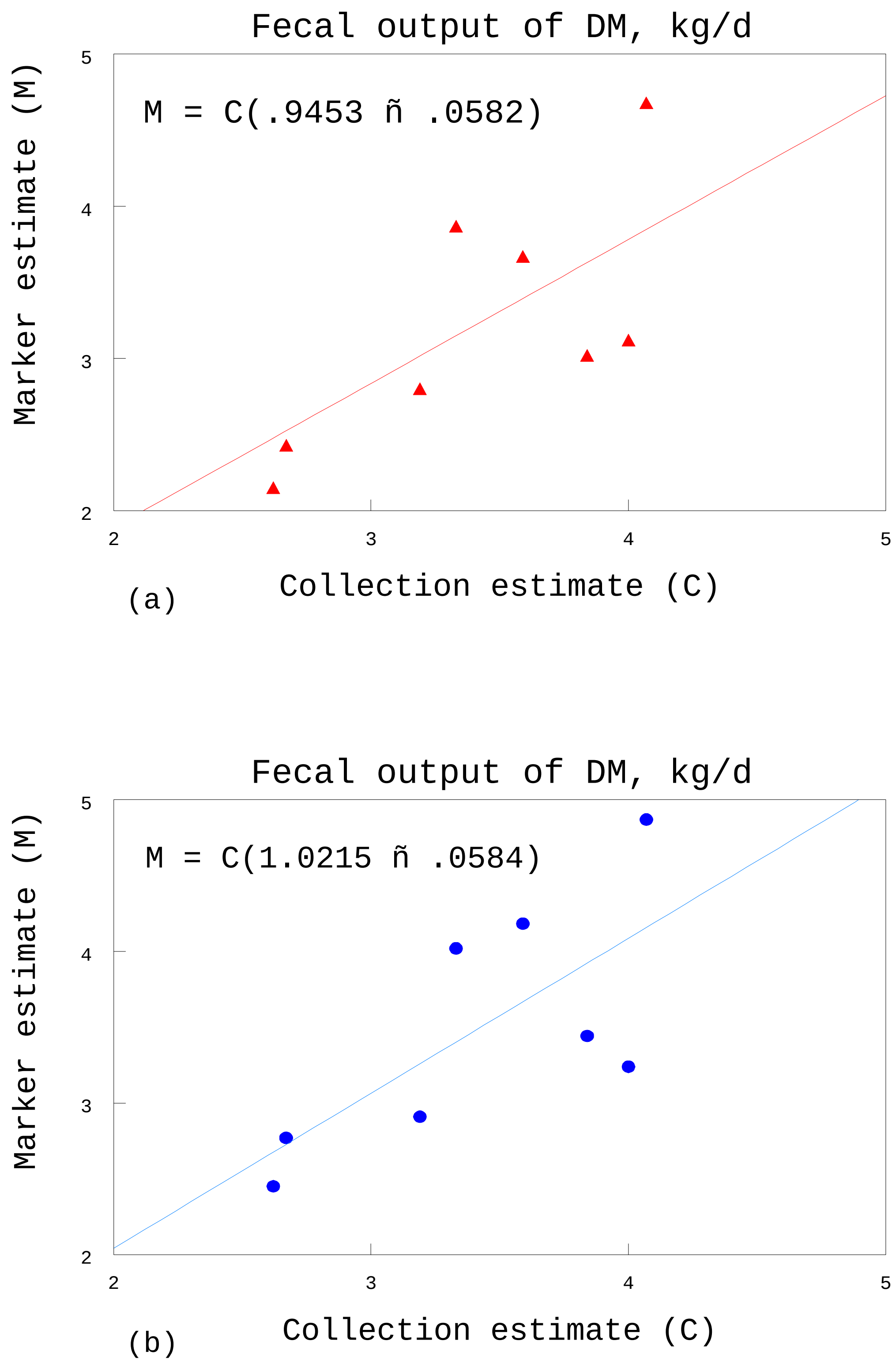

Figure 2. Fecal outputs predicted by marker dilution were correlated with collection values, both unadjusted (a) and adjusted (b) for Cr recoveries in feces. The correlation coefficient was improved slightly by adjustment from .46814 to .50513.

Chapter 6. Fecal Output, Digestibility, and Intake of Pasture is Predicted by Marker Methods in Grazing Horses. Use of Collection Harnesses to Measure Fecal Output.

ABSTRACT

Marker methods have been developed with chromium that provide accurate and precise estimates of fecal output in stalled horses. However, these methods have not been tested in grazing horses because of the difficulty in taking total collections of feces in pasture situations. Although one could assume that methods that work in stalls would be applicable to pastured horses, it would be beneficial to validate the methods. Eight Thoroughbred geldings were divided into two groups and placed on bluegrass/white clover and tall fescue/alfalfa pastures in a replicated 2x2 Latin Square design. Balance experiments were conducted in 15 d periods with a 14 d period between experiments. Horses were placed on pastures 14 d prior to the beginning of balance experiments to allow for dietary accommodation. The external marker, Cr, was administered in "greenola bars" containing 5.98 ± .45 g Cr/bar, at 700, 1300 and 1900 each day for 8 d. Dry matter, Cr, yttrium and nutrients were analyzed in pasture and fecal samples. Daily fecal concentrations of Cr (C_t g/kg DM) were applied to a simple monoexponential equation with one rate constant (k) rising to an asymptote (C_a). Equations included a delay (*d*), which represented the time needed for Cr to enter the prefecal pool. Delays were 4.66 h and 5.35 h for bluegrass/white clover and tall fescue/alfalfa pastures, respectively. Yttrium was evaluated as an internal marker to estimate dry matter digestibility (D). Fecal output (FO) was measured in geldings with total fecal collection harnesses, and was estimated by the following equation: $FO = Cr\ dose\ (d^{-1}) / C_a$. FO was used in combination with D predictions to estimate dry matter intake (DMI): $DMI = FO / (1-D)$. Yttrium gave higher estimates of D than have been previously reported. Fecal output estimated from C_a gave values similar to weighed FO ($P > .20$). When estimated D was used with either TC or estimated FO to predict DMI, the predicted intake was approximately 3.3 % of the horses' BW. These results indicate that Cr can be used to predict FO in grazing horses.

Key Words: Fecal Output, Digestibility, Intake, Pasture, Fecal Kinetics

Introduction

Fecal output estimates are necessary for determination of dry matter intake (DMI). Intake can be measured by weighing, in stalls (Crozier et al., 1997). These measurements, however, may not relate well to DMI in pasture. Factors such as weather, time of year, and activity level all can affect intake, and are difficult to mimic in stalled horses. Fecal output has been estimated using dosed markers (Galyean, 1993; Susmel et al., 1996).

One of the problems with using external markers to predict fecal output in grazing animals has been finding a way to collect all fecal output for comparison to marker predictions (Holechek et al., 1986). Usually this has been accomplished by conducting trials in stalls and feeding hay (Holland et al., 1998). However, these types of trials do not mimic grazing conditions. Recently, a new total fecal and urine collection harness has been developed for use with horses. Although these harnesses have been used on horses with limited turnout time, they have not been used for 24 h collections on pasture (personal communication, Stave Warner, Thoroughbred Maintenance, Los Angeles, CA).

One of the other problems with attempting to predict intake on pasture has been finding an appropriate marker to evaluate digestibility. Many internal markers have been tried with varied levels of success (Cochran et al., 1986; Judkins et al., 1990). Some have been difficult to measure while others have been partially digested or degraded in the digestive tract. One type of internal marker that has been used is n-alkanes (Ohajuruka and Palmquist, 1991). These markers have given favorable results in ruminants and are easy to analyze with gas chromatography (Dove and Mayes, 1996; Ordakowski, 1998).

The objective of this trial was to evaluate the use of a simple monoexponential equation, based on fecal daily Cr concentrations, to predict fecal output, and compare these predictions with total collection of feces in grazing horses. Nutrient digestibilities were also predicted and then dry matter intake was calculated.

Materials and Methods

Eight mature Thoroughbred geldings, average weight 615 ± 25 kg, were used in these studies, which were conducted during the months of May and June, 1998. Geldings were paired by weight and divided into two groups of four. Each group was placed into one of two 8-acre pastures for the first trial and then switched to the other pasture for the second trial in a replicated 2 x 2 Latin Square design. One pasture (BG/WC) was predominately bluegrass (*Poa pratensis*) and white clover (*Trifolium repens*); the second (YF/A) was predominately tall fescue (*Festuca arundinacea*) and alfalfa (*Medicago sativa*). Horses were placed on their respective pastures 2 wk prior to the beginning of data collection.

Each trial lasted for 15 d with a 23 d break between trials. During the first 3 d of each trial, baseline fecal samples were collected. On d 4, horses began to be dosed three times daily, at 700, 1300, and 1900 h, with a "greenola bar" containing 5.98 ± .45 g of Cr per bar. Greenola bars were a mixture of a coarse grain mix, Cr_2O_3, molasses, and beer (Table 1). Ingredients were mixed thoroughly, divided into flat baking pans, and dried in a forced air oven. Thrice daily dosing continued for eight d, from d 3 until d 11. Two "greenola bars" were set aside from each batch for analysis of Cr.

Fecal grab samples were collected three times a day, when bars were fed. Fecal samples were collected three times daily for the 4 d after dosing ceased. Fecal samples from the same day were combined, weighed, dried in a forced-air oven to a constant weight, and then reweighed for DM determination. On the last day of dosing, a small subsample was taken from each fecal grab sample so analysis of the diurnal excretion of Cr could be determined. Total collection (TC) of feces was made during from d 8 to d 10 from all horses. All geldings were fit with a fecal collection harness (Horse Nappy/Diaper, Equisan Pty. Ltd., Melbourne, Victoria, Australia) which were emptied three times daily. Feces were collected in separate tubs for each horse. Feces from each 24 h period, including individual grab samples, were weighed. Subsamples from individual grab samples and TC samples were weighed, dried to constant weight in a forced air over, and weighed again for DM determination.

Pasture samples were collected weekly throughout the trials using hand-held electric clippers with a 10.2 cm wide edge using methods developed by Blaser et al. (1986). Samples were clipped across the pasture in a random fashion until 1 to 2 kg, wet weight, was collected. These samples were weighed, dried in a forced-air oven, weighed again for DM determination, and then used for nutrient analysis. On one day of each trial pasture samples were collected for botanical composition. A stick, 1.83 m, was thrown randomly into each acre. The pasture to the left of the stick was clipped using hand-held electric clippers. Total samples for each pasture were hand separated into edible plants and weeds (Table 2). Samples were weighed wet, to give a total weight and then all samples were dried so that percentages of wet and dry pasture could be determined (Table 2).

Samples of pastures and feces were ground through a .5 mm screen Thomas-Wiley Laboratory Mill (Model 4, Thomas Scientific, Swedesboro, NJ). Composite samples were made from the pasture samples and from the feces from the last 4 d of dosing. These composites were submitted for analysis of DM, CP, EE, NSC, NDF, ADF, Ca, P, K, Mg, Na, Fe, Zn, Cu, Mn, Mo, S and ash by standard methods (Robertson and Van Soest, 1977; AOAC, 1990) at the Northeast DHIA Laboratory (Ithaca, New York). Pasture analysis can be found in Table 3.

The twelve reserved "greenola bars" were finely ground in a coffee grinder (Electric Miracle Seed&Coffee Mill, Model MC200, Whatman Scientific, Hillsboro, OR). Cr was analyzed in "greenola bars", pasture composites, and both fecal composites, daily fecal samples and TC subsamples by atomic absorption spectrophotometry with an air-acetylene flame after samples were digested with nitric and perchloric acids (Sandel, 1959). Two dilutions of each Cr sample were analyzed in duplicate, and five dilutions of standard solution of potassium dichromate were used for calibration. Yttrium (Y) was analyzed in pasture and fecal composite samples by inductively coupled plasma spectroscopy after samples were digested with nitric and perchloric acid.

Dry matter digestibility (D, %) was determined by using a ratio of the internal markers (M), either Y or alkanes:

$$D = (1 - C_I/C_F)*100$$

where C_I is the [M] in the feed samples and C_F is the [M] in the fecal samples.

Nutrient digestibilities (ND) were calculated using the internal marker Y:

$$ND=[1-(C_I / C_F)(N_O / N_I)]*100$$

where N_O (% DM) is the concentration of a nutrient in the fecal samples and N_I (%DM) is the concentration of a nutrient in the feed samples.

Pooled daily fecal Cr concentrations (C_t, mg/kg DM) for all 8 horses on one diet were fit to a simple, monoexponential model rising to an asymptote (C_a) using a graphics and curve-fitting program (SlideWrite Plus 4.0 for Windows, Advanced Graphics Software, Carlsbad, CA) as previously described (Holland et al., 1998):

$$C_t = C_a - C_a \cdot e^{-kt}$$

where C*t* was fecal Cr concentration (mg/kg DM) at time *t* (days). The asymptotic value of fecal Cr determined fecal output (FO):

$$FO = (\text{Cr dose}/C_a).$$

The rate constant (k, d^{-1}) determined its reciprocal, the turnover time (TT, d). The compartment size, or mass, of the prefecal pool (PFM, kg) was calculated:

$$PFM = FO \text{ x } TT = FO/k.$$

Fecal outputs predicted by C_a were compared to total collection values. FO (kg/d) and D (%DM) predicted from markers were used to calculate dry matter intake (DMI, kg/d):

$$DMI = FO/(100 - D)$$

and these predicted intakes were compared.

The data were summarized as means and SEMs, and analyzed by the GLM procedure of SAS (SAS Institute, Inc., Cary, NC) with horse, diet, method and all interactions in the model. Differences in digestibilities predicted by Y and alkanes were compared. Differences in predicted fecal outputs from C_d and C_a were compared to TC values. Intake predictions from markers were compared. Comparisons of means used Fisher's protected LSD.

Results and Discussion

Digestibilities. Although TF/A had slightly higher nutrient digestibility predictions by Y (Table 4), it was not different from BG/WC ($P > .215$). This trend was followed in previous studies on the same pastures (Chapters 3 and 5). Nutrient analysis (Table 3) indicates a slightly lower CP content and higher fiber(s) content in the TF/A pasture, the opposite of what would be expected from the slight digestibility differences. The BG/WC pasture was slightly higher in total DM (25.5% vs. 22.9%), and could have had a slower passage rate, which would have improved digestion. However, this is not supported by fecal kinetics data discussed later. Predicted apparent digestibilities were rather high in this study. Usually, pasture digestibilities are between 60 and 70%. This could indicate a problem with using Y as an internal marker. Fecal recoveries of Y were probably high, possibly due to ingestion of soil by the geldings.

One of the problems with analyzing grazing studies is attempting to predict actual forage intake, not just DMI (Cordova et al., 1978). Studies in ruminants have utilized esophageal and ruminal fistulas to gather samples of consumed pastures, which can be analyzed for individual plants (Holecheck et al., 1982). It is possible that the forage samples collected for nutrient analysis in this study were not truly representative of the forages consumed, leading to errors in digestibility. Studies have been conducted in cattle to evaluate effects of total fecal collection on nutrient digestibilities. One study found slight increases when total fecal collections were made (Noblitt et al., 1963) while no effect was observed in the second study (Phar et al., 1971).

Fecal Output. Total collection data (Table 5) indicated slightly higher fecal output for horses grazing BG/WC (P = .077). C_a and C_d both predicted FO's very close to the TC values (P > .079 for C_a; P > .095 for C_d). When horses were consuming BG/WC, C_a predicted FO better (P = .218) than C_d (P = .030). However, this was the opposite when horses were consuming TF/A. On this pasture, C_a predicted FO well (P = .204), but C_d gave a better prediction (P = .860). Lower fecal output when animals were grazing TF/A is associated with the higher nutrient digestibilities.

One of the concerns with utilizing total collection harnesses and dosing techniques, is that the stress of carrying the excess weight and extra handling may adversely affect fecal output (Cordova et al., 1978). One advantage with using horses is that they are usually more accustomed to handling, and most have experience with wearing blankets, saddles, or driving harnesses. The horses utilized in this study were trained to wear the collection harnesses prior to the study so that it would not be something new to them. The “greenola bars” were also highly palatable to the geldings, as determined by their reaction at daily dosing times. The geldings would literally run across the pasture towards the researchers when called.

A study was conducted in sheep to evaluate the effect of fecal collection harnesses and marker dosing on fecal output, forage intake and weight change, in an attempt to quantify any adverse effects of the harness (Hatfield et al., 1993). Neither the harness nor the dosing technique (twice daily dosing with a balling gun) had an adverse effect on any measurements. Our assumption is that since the horses were accustomed to the fecal collection harnesses there was no stress associated with them. Dantzer and Mormede (1983), who concluded that stress is lessened when situations are predictable support this in a review.

Intake. The summary of intake predictions (Table 6) show that TF/A intake was higher than BG/WC intake (P = .048). The combination of C_a and Y as an intake predictor was lower than that of C_d and Y (P = .006). However, all intakes using Y were higher than expected, indicating that Y either is not a good digestibility indicator or that dry matter intake requirements for grazing horses are higher than predicted by the NRC. Possible errors in intake can be associated with the internal marker, because intake predictions using FO predictions were the same as intake predictions using TC. As a comparison, pregnant and lactating mares had dry matter intakes between 1.5 and 2.5% of BW when grazing a grass/legume pasture (Martin et al., 1989). One would expect intakes of mares in late gestation and early lactation to be higher than that of mature geldings at maintenance, due to the increased demand for fetal growth and milk production.

Fecal Kinetics. The monoexponential equation for the Cr data, without a delay included, provided a good fit to the data for both pastures; $r^2 = .900$ for BG/WC and $r^2 = .902$ for TF/A (Figure 1). Inserting a delay (*d*, d) into the model as a decrease in time (*t*):

$$C_t = C_a - C_a \cdot e^{-k(t-d)}$$

improved the fit of the monoexponential equation for both BG/WC ($r^2=.916$) and TF/A ($r^2=.918$), however, C_a did not change (Table 7). The curve-fitting program used could not solve the above equation, so a series of delays had to be included until the best fit was found. Other model parameters and estimates for TT, FO and PFM are in Table 7. The improvement of model fit with the delay probably gives a good estimate of passage of the marker into the prefecal mass. Th longer delay predicted for the TF/A pasture may be related to the chemical composition of the pastures. The TF/A pasture had a higher grass % than the BG/WC pasture (Tables 2 a and b), and this is usually related to higher fiber content (Jung and Allen, 1995; Gunter et al., 1997). Tall fescue also has a broader and longer leaf than bluegrass, requiring more time to masticate and digest.

When the monoexponential equation with the delay was applied to individual horses, the fit improved on both BG/WC (Table 8a) and TF/A (Table 8b) as determined by increased r^2 values and F-statistics. Estimates of FO, TT and PFM, for data with the delay are given in Table 9. Attempts to use chromic oxide as a marker for either the liquid or particulate phase of the digesta have been unsuccessful (Titgemeyer, 1997). However, in this study, Cr was not attached to either phase, and served as a total digesta tracer, instead. Retention times observed in this trial (36.7 and 42.2 h, for BG/WC and

TF/A, respectively) are similar to times observed in other trials; 38 h for horses consuming alfalfa hay (Vander Noot et al., 1967). Other studies have found retention times between 3 and 36 h (reviewed by Orton et al., 1985). The slightly longer retention time for the TF/A pasture probably is related to the expected higher cell wall content.

Marker estimates and total collection weights of feces (Figure 2) were not strongly correlated in this study ($r^2 = .29$) even though predictions were not different from collections using the equation: FO = Marker dose, d^{-1}/[fecal Cr] (Table 6).

Implications

The Equisan fecal collection harness is an effective way of conducting total collection in grazing geldings. Chromic oxide predicted fecal output well in pasture-kept geldings, but increasing the dosing times per day, or keeping dosing times evenly spread throughout the day, may improve the prediction. Nutrient digestibilities predicted with yttrium were higher than expected, leading to high estimates of dry matter intake.

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Table 1. Recipe for "greenola bars"[a] for fecal kinetic studies.

Ingredients

Oat-based sweet feed	2.16 kg
Cr_2O_3	800 g
Liquid molasses	1 L
Beer	380 mL

Instructions

Mix Cr_2O_3, molasses and beer thoroughly, making sure Cr_2O_3 is well-blended. Add sweet feed slowly, making sure all particles are covered. Divide mixture between 4 well-oiled 12"x15" pans. Put into oven set at low temperature. When mixture begins to set, cut into 20 bars per tray. Put back into oven until completely dry. Remove from pans and place each bar into an individual bag for ease of handling.

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[a] Adapted from Practical Horseman. 1996. 24(4):122. Horse Cookie recipe

Table 2a. Botanical composition (BC) of bluegrass/white clover (BG/WC) pasture (mean ± SE).

BG/WC[a] Plant	% of wet pasture	% of dry pasture
Bluegrass (*Poa pratensis*)	74.67 ± 4.56	83.79 ± .46
White Clover (*Trifolium repens*)	17.66 ± 2.46	11.03 ± .43
Tall Fescue (*Festuca arundinacea*)	.87 ± .87	.87 ± .87
Orchardgrass (*Dactylis glomerata*)	.27 ± .27	.41 ± .41
Ryegrass (*Lolium perenne*)	.11 ± .11	.14 ± .14
Plantain (*Plantago major*)	4.54 ± 2.34	2.33 ± .94
Chickweed (*Stellaria media*)	.29 ± .18	.35 ± .35
Dandelion (*Taraxacum officinale*)	1.11 ± .41	.76 ± .07
Misc. Weeds	.50 ± .04	.35 ± .35

[a] Percent DM of total BC sample 36.49 ± 3.08

Table 2b. Botanical composition (BC) of tall fescue/alfalfa (TF/A) pasture (mean ± SE).

TF/A[a] Plant	% of wet pasture	% of dry pasture
Tall Fescue (*Festuca arundinacea*)	72.64 ± .23	76.28 ± 1.54
Alfalfa (*Medicago sativa*)	5.18 ± .79	4.89 ± .45
White Clover (*Trifolium repens*)	9.57 ± .61	6.52 ± .31
Bluegrass (*Poa pratensis*)	7.55 ± .47	8.62 ± .26
Orchardgrass (*Dactylis glomerata*)	.70 ± .70	.69 ± .69
Red Clover (*Trifolium pratense*)	.20 ± .20	.01 ± .01
Dandelion (*Taraxacum officinale*)	.53 ± .26	.24 ± .07
Plantain (*Plantago major*)	2.79 ± .93	1.87 ± .69
Misc. Weed Chickweed (*Stellaria media*) Buttercup (*Ranunculus muricatus*) Lambsquarters (*Chenopodium album*) Horsenettle (*Solanum elaeagnifolium*)	.40 ± .01	.92 ± .62

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[a] Percent DM of total BC sample 29.66 ± 3.42

Table 3. Nutrient and internal marker content[a] of pastures, means ± SE.

Nutrient	bluegrass/white clover	tall fescue/alfalfa
DM, %	25.46 ± 3.84	22.92 ± 3.39
Composition of DM		
CP, %	15.9 ± .1	15.4 ± .2
ADF, %	35.6 ± .3	37.6 ± .3
NDF, %	58.5 ± .6	60.1 ± 1.1
EE, %	3.9 ± .1	3.5 ± .3
NSC[b], %	14.1 ± .5	13.5 ± .6
Ca, %	.53 ± .04	.56 ± .04
P, %	.32 ± .02	.30 ± .01
Mg, %	.19 ± .00	.23 ± .01
K, %	2.22 ± .17	2.37 ± .17
Na, %	.008 ± .000	.011 ± .002
Fe, ppm	678 ± 43	545 ± 25
Zn, ppm	50 ± 4	55 ± 7
Cu, ppm	7 ± 0	6 ± 0
Mn, ppm	96 ± 7	75 ± 2
Mo, ppm	1.5 ± .1	1.6 ± .1
S, %	.19 ± .00	.20 ± .00
Ash, %	7.58 ± .03	7.58 ± .03
Cr, ppm	156 ± 19	101 ± 59
Y, ppm	.74 ± .04	.78 ± .15

[a] Analysis of CP, EE, and ash (AOAC, 1990), ADF and NDF (Robertson and Van Soest, 1977), Cr (Sandel, 1959)

[b] NSC = 100 - (CP% + EE% + NDF% + ash%)

Table 4. Digestibilities (%), means ± SE, of DM, CP, ADF, NDF, EE, NSC, and ash in horses (n = 8) grazing bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture. Predicted from yttrium (Y).

Nutrient	BG/WC	TF/A
DM	78.80 ± 1.55	82.21 ± 3.44
CP	82.66 ± 1.04	85.39 ± 2.49
ADF	75.09 ± 1.91	78.21 ± 4.32
NDF	77.69 ± 1.91	81.03 ± 4.05
EE	67.88 ± 2.96	73.66 ± 4.56
NSC	100 ± 0	100 ± 0
Ash	44.25 ± 3.58	55.68 ± 8.05

Table 5. Fecal outputs from horses (n = 8) grazing either bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture. Average weight was 615 ± 25 kg. Comparison between TC and predicted outputs*.

Variable	BG / WC	TF / A	SEM
Fecal DM, kg/d, weighed	4.05 [b]	3.43	.24
Fecal DM predicted from daily Cr [a], kg/d	4.82 [b]	3.49	.24
Fecal DM predicted from C_a, kg/d	3.62	2.98	.24

[a] Daily Cr is the average fecal [Cr] over the last 4 days of dosing.

*Values in the same row with the same superscript are different:

[b] P = .030

Table 6. Dry matter intakes of either bluegrass/white clover (BG/WC) or tall fescue/ alfalfa (TF/A) pasture by grazing horses (n = 8). Average weight of horses was 615 ± 25 kg. Comparison of predicted intakes.*

Variable	BG / WC	TF / A	SEM
DMI, predicted from daily Cr and Y	24.28 (3.9)^a	25.47 (4.1)	1.75
DMI, predicted from C_a and Y	17.87 (2.9)^a	20.45 (3.3)	1.75
DMI, predicted from TC and Y	19.29 (3.1)	25.45 (4.1)	1.75

*Values in the same column with the same superscript are different

^{a} $P < .03$

Table 7. Estimates of fit of pooled fecal Cr concentration data (Figure 1) to a one-compartment model, model parameters, and calculated variables in horses grazing either bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture.

Estimate	BG/WC	TF/A
R^2	.916	.918
F	76.2	78.9
P- value	.0000	.0000
Cr_a, mg/kg[a]	4.47	5.72
SE	.3730	.5321
t-statistic	11.98	10.74
P-value	.0000	.0000
-k, d^{-1}[b]	.655	.569
SE	.204	.180
t-statistic	3.21	3.17
P-value	.0062	.0066
TT, h[c]	36.7	42.2
Delay, h[d]	4.66	5.35
FO, kg/d[e]	3.62	2.98
PFM, kg[f]	5.54	5.24

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[a] Cr_a is the asymptotic value of fecal Cr concentration.
[b] k is the rate constant.
[c] TT is turnover time of the prefecal mass, TT = -1/k
[d] Delay is the time between the administration of Cr and its entry into the prefecal mass
[e] FO is fecal output, FO = Dose of Cr/Cr_a.
[f] PFM is prefecal mass or mixing compartment, PFM = FO x TT.

Table 8a. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses grazing bluegrass/white clover pasture.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.890	56.8	.0000	.895	59.9	.0000
B	.622	11.5	.0044	.699	16.2	.0015
C	.854	41.1	.0001	.858	42.3	.0001
D	.618	11.3	.0047	.631	12.0	. 0039
E	.705	16.7	.0014	.775	24.2	.0004
F	.778	24.6	.0004	.830	34.1	.0001
G	.825	33.1	.0001	.872	47.6	.0000
H	.767	23.1	.0005	.812	30.2	.0002
A to H[b]	.900	63.2	.0000	.916	76.2	.0000

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[a] Delay was 4.66 h for horses fed bluegrass/white clover pasture.

[b] Pooled data as in Figure 2.

Table 8b. Fits of data to a one-compartment model of fecal kinetics with and without a delay between administration of an oral dose of Cr and its entry into the compartment in trials on eight horses grazing tall fescue/alfalfa pasture.

	No Delay			Delay[a]		
Horse	R^2	*F*	*P*-value	R^2	*F*	*P*-value
A	.781	24.9	.0004	.846	38.4	.0001
B	.791	26.5	.0003	.846	38.5	.0001
C	.797	27.5	.0003	.849	39.4	.0001
D	.900	63.2	.0000	.902	64.4	. 0000
E	.799	27.8	.0003	.817	31.3	.0002
F	.855	41.4	.0001	.866	45.3	.0000
G	.841	37.0	.0001	.856	41.5	.0001
H	.731	19.0	.0009	.768	23.2	.0005
A to H[b]	.902	64.1	.0000	.918	78.9	.0000

[a] Delay was 5.35 h for horses fed tall fescue/alfalfa pasture.

[b] Pooled data as in Figure 2.

Table 9. Fecal kinetics in eight horses fed either bluegrass/white clover (BG/WC) or tall fescue/alfalfa (TF/A) pasture estimated with a one-compartment model with a delay[a] between time of administration and entry of marker into the compartment.

Variable	BG/WC	TF/A	SEM	*P*-value
Fecal output, kg DM/d	3.62	2.98	.32	.09
Fecal output g/kg of BW daily	5.89	4.85	.52	.09
Turnover time, h	36.7	42.2	2.75	.09
Prefecal mass, kg DM	5.54	5.24	.15	.09
Prefecal mass, g/kg of BW daily	9.01	8.53	.24	.09

[a] The delays were .194 and .223 d for BG/WC and TF/A, respectively.

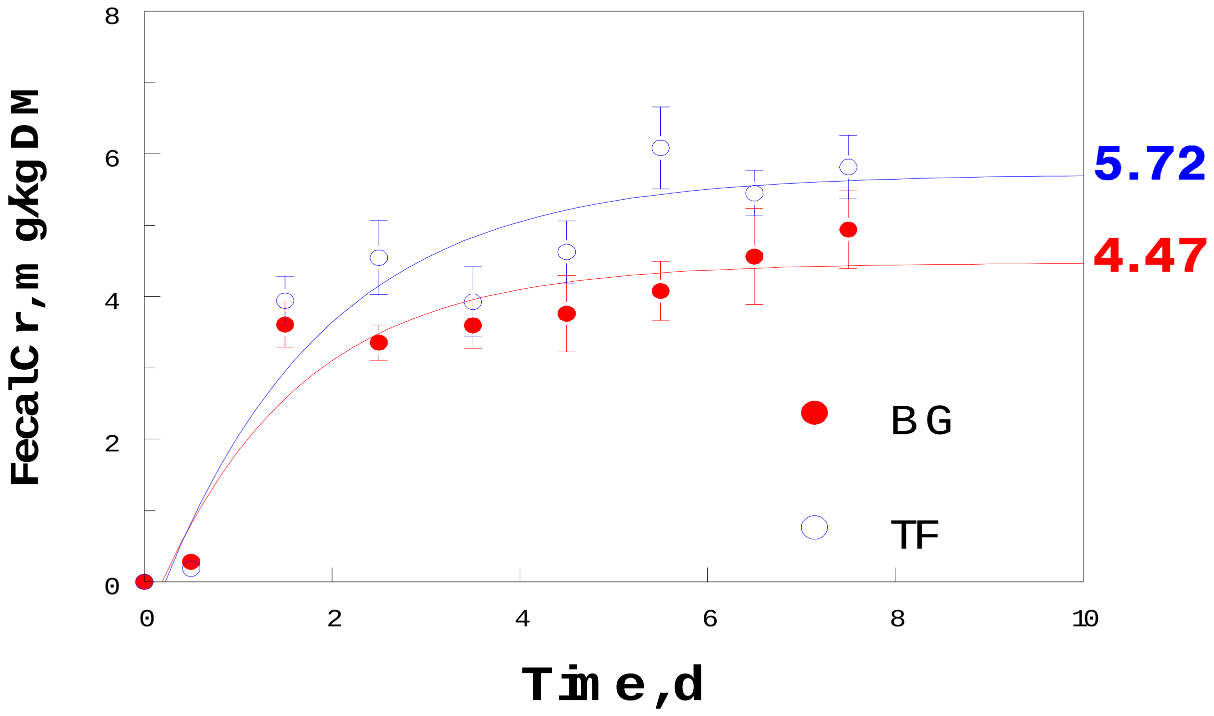

Figure 1. Mean daily fecal concentration of Cr, C*t*, for horses fed bluegrass / white clover (BG/WC) or tall fescue / alfalfa (TF/A) pasture are plotted against time, and the data are fit to a one compartment model (see Figure 1, Holland et al., 1998).

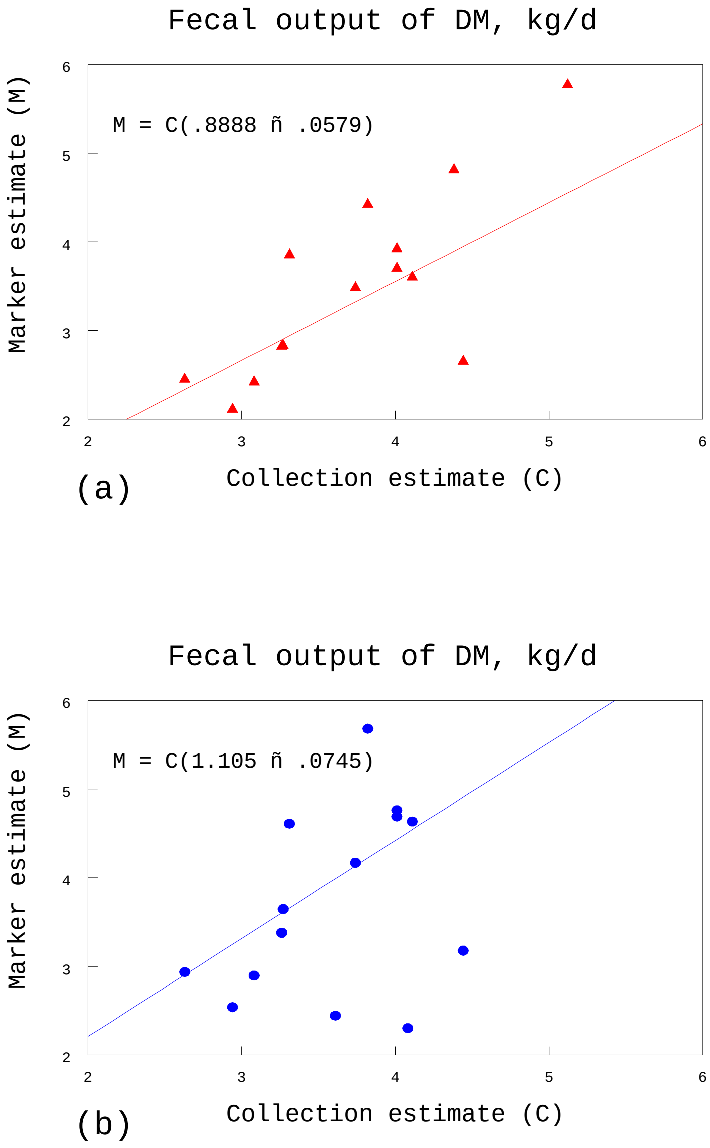

Figure 2. Fecal outputs predicted by marker dilution were correlated with collection values, both unadjusted (a) and adjusted (b) for Cr recoveries in feces. The correlation coefficient was not improved by adjustment (r^2 =.2969 and .2832, respectively).

Summary and Implications

Chromium (Cr) can be used as an effective marker for fecal output predictions in horses consuming hay and pasture, if daily Cr (C_t) values, from initial dose to final dose (8 d), are plotted against time using a monoexponential equation. This equation should include a delay (*d*, d), and fecal output should be predicted using the asymptotic value of chromium excretion (C_a).

$$C_t = C_a - C_a \cdot e^{-k(t-d)}$$

"Greenola bars" are an effective means of dosing Cr to horses on pasture. However, time of dosing needs to be considered. Once daily dosing does not overcome diurnal variation, but dosing three times a day does. This dosing needs to occur at 8 h intervals; dosing every 6 h in a 12 h interval does not effectively overcome diurnal variation. A fourth dose, every 6 h, should be given if fecal samples are going to be collected within a 12 hour time span every day.

Although yttrium (Y) was an accurate internal marker for digestibility predictions in stalled horses, it overestimated digestibility in grazing horses. It is possible that contamination occurred from horses ingesting soil. Alkanes were more effective internal markers, overall. Problems observed with internal markers may be due to the inability to take samples of what horses actually consumed while grazing, as can be done with esophageal and ruminal fistulas in ruminants. Content of internal markers varies with plant type and part of plant consumed. It may be that several internal markers should be used, and the digestibility predictions averaged across these markers.

Estimates of dry matter intake (DMI) were predicted using estimates of fecal output (FO) and dry matter digestibility (D) in the following equation:

$$DMI^{kg/d} = FO^{kg/d} / (100 - D\%)$$

Predicted estimates in stalled horses were similar to what has been reported earlier for mature horses at maintenance. However, DMI estimates in grazing horses were higher than these expected intakes, indicating that grazing animals require more nutrients for maintenance than stalled animals.

Future trials should focus on further development of the Cr dosing procedure. Tests should evaluate dosing four times daily, every 6 h, to determine if this will overcome diurnal variation of fecal Cr excretion. The fecal collection harnesses should be used in grazing horses for comparison to predicted FO. Future studies should also evaluate the use of even-chain alkanes as an external marker. Internal markers also need to be discovered that more accurately predict D in a more consistent basis than the markers used in these trials. If this is not feasible, then several internal markers should be used, and D estimates averaged between the markers. Another facet that should be explored is using a second external marker, placed in a supplement, to predict intake of the supplement in grazing horses.

Appendix A. Nutrient analysis of individual feed samples.

Chapter 3. Pasture Analysis

NUTRIENT	BG/WC	BG/WC	TF/A	TF/A
Cr, ppm	35	0	18	0
Y, ppm	.84	.92	.59	.96
DM	28.10	43.98	19.57	22.44
PROTEIN%	16.00	14.9	19.4	16.7
ADF%	36.8	36.0	31.1	30.5
NDF%	60.7	59.8	51.2	51.9
EE%	4.1	2.7	3.5	2.9
NSCDIFF%	12.3	14.3	17.0	18.5
Ca%	0.36	.44	0.56	.82
P%	0.37	.31	0.44	.29
Mg%	0.19	.20	0.36	.31
K%	2.32	2.09	2.96	2.82
Na%	0.003	.003	0.010	.020
Fe,ppm	374	538	503	657
Zn, ppm	34	33	47	28
Cu, ppm	28	23	24	23
Mn, ppm	47	61	63	62
Mo,ppm	1.5	2.1	2.3	2.3
S%	0.22	.19	0.28	.24
Cl%	0.44	.35	0.45	.46
Ash%	6.97	8.25	8.87	10.02

Chapter 4. Hay Analysis.

NUTRIENT	TF/AG	TF/AG	OG/AG	OG/AG
Cr, ppm	984	350	521	0
Y, ppm	.53±.01	.74±.01	1.09±.01	.77±0
DM	85.32±.53	88.25±.74	85.62±.65	87.34±.93
PROTEIN%	12.7±.1	10.9±.1	14.4±0	14.3±.2
ADF%	48.4±1.1	48.3±1.5	46.5±.9	48.4±1.3
NDF%	64.6±1.2	67.8±.8	62.3±.1	63±1.4
EE%	.7±.1	1.4±.1	.8±0	.9±.1
NSCDIFF%	15.2±1.2	12.5±.7	14.3±.2	14.1±1.1
Ca%	.64±0	.57±0	.75±.01	.68±.01
P%	.26±.01	.25±0	.28±0	.31±0
Mg%	.19±.01	.18±.01	.17±.01	.15±.01
K%	2.43±.01	2.73±.02	2.90±0	3.02±.01
Na%	.023±.001	.015±.001	.013±0	.007±0
Fe,ppm	771±1	521±9	803±11	407±4
Zn, ppm	41±0	32±0	42±1	31±1
Cu, ppm	11±0	9±0	12±1	9±0
Mn, ppm	54±2	69±0	62±1	56±2
Mo,ppm	.6±.6	1.4±.4	1.1±.1	1.0±0
S%	.16±0	.16±.01	.17±0	.16±.01
Ash%	6.88±.01	7.54±.01	8.10±.1	7.84±0

Appendix A (cont'd.). Nutrient analysis of individual feed samples.

Chapter 5. Pasture Analysis.

NUTRIENT	BG/WC	BG/WC	TF/AG	TF/AG
Cr, ppm	83	0	101	0
Y, ppm	.28±.2	.49±.05	.45±.01	1.07±.09
DM	34.2±3.09	25.68±1.65	36.48±2.61	29.9±3.96
PROTEIN%	13.35±.15	11.85±.15	13.75±.25	10.7±.1
ADF%	38.85±.05	42.2±.25	40.55±.15	43.7±.5
NDF%	64.6±1.4	69.3±.95	63.45±.65	64.1±.9
EE%	2.85±.25	3.4±0	2.45±.15	3.8±.05
NSCDIFF%	11.9±1.3	8.7±.6	10.95±1.15	12.1±.7
Ca%	.75±0	.59±0	.8±.02	1.03±.01
P%	.31±.01	.29±0	.31±0	.31±.01
Mg%	.18±0	.18±.01	.28±0	.34±.01
K%	2.21±.01	1.35±.01	2.50±.02	1.75±.02
Na%	.006±0	.003±.001	.015±0	.006±.001
Fe,ppm	502.5±37.5	490±15	238±9.5	395±3
Zn, ppm	44±.5	35±0	34±.5	37±0
Cu, ppm	6±1	5±.5	6±.5	5±.5
Mn, ppm	56±.5	60±0	85±3.5	99±3
Mo,ppm	1.6±.1	1.7±.1	1.5±.15	1.8±.15
S%	.18±0	.18±.03	.24±.01	.25±.01
Ash%	7.31±.01	6.75±.17	9.41±.11	9.38±.01

Chapter 6. Pasture Analysis

NUTRIENT	BG/WC	BG/WC	TF/AG	TF/AG
Cr, ppm	123	189	202	0
Y, ppm	.68±.01	.8±.01	.52±.05	1.04±.04
DM	24.62±8.79	26.29±3.13	20.62±5.62	25.21±5.17
PROTEIN%	16±.5	15.9±.2	15.1±.1	15.7±0
ADF%	36.1±.2	35.1±.4	37.7±.3	37.6±.6
NDF%	59.5±.5	57.6±.3	61.8±.8	58.4±.7
EE%	3.8±.1	4.1±.2	3±0	4±.5
NSCDIFF%	13.3±.3	15±.6	12.7±.8	14.4±.7
Ca%	.46±.01	.61±.01	.49±.01	.63±.01
P%	.36±.01	.29±.01	.30±.01	.29±0
Mg%	.19±.01	.20±.01	.22±.01	.24±0
K%	2.52±.01	1.93±.01	2.66±.05	2.08±.01
Na%	.009±.001	.008±0	.014±.001	.008±0
Fe,ppm	750±20	607±25	533±54.5	557±19.5
Zn, ppm	44±2.5	57±1	66±5	44±3
Cu, ppm	7±0	6±0	6±0	5±0
Mn, ppm	107±8	85±2	77±2	72±1
Mo,ppm	1.7±.1	1.3±0	1.8±.02	1.5±.2
Cl %	.71±.11	.6±0	.78±0	0.84±.09
S%	.20±.01	.19±0	.22±0	.22±0
Ash%	7.4	7.4	7.4	7.5

Appendix B. Data on individual "greenola bars".

Chapter 3.

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar wet (g)	Cr\bar (g)dry
1	11	64	61	53	50	0.943396	88375	4.683875	4.41875
2	11	66	61	55	50	0.909091	112000	6.16	5.6
3	11	60	58	49	47	0.959184	123375	6.045375	5.798625
4	11	71	65	60	54	0.9	122500	7.35	6.615
5	11	54	52	43	41	0.953488	140875	6.057625	5.775875
6	10	50	47	40	37	0.925	136500	5.46	5.0505
7	11	66	61	55	50	0.909091	129500	7.1225	6.475
8	13	65	61	52	48	0.923077	144375	7.5075	6.93
							AVE	6.298359	5.832969

Chapter 4.

Trial 1

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar Wet (g)	Cr\bar Dry (g)
1	11	64	61	53	50	0.943396	88375	4.683875	4.41875
2	11	66	61	55	50	0.909091	112000	6.16	5.6
3	11	60	58	49	47	0.959184	123375	6.045375	5.798625
4	11	71	65	60	54	0.9	122500	7.35	6.615
5	11	54	52	43	41	0.953488	140875	6.057625	5.775875
6	10	50	47	40	37	0.925	136500	5.46	5.0505
7	11	66	61	55	50	0.909091	129500	7.1225	6.475
8	13	65	61	52	48	0.923077	144375	7.5075	6.93
							AVE	6.298359	5.832969

Trial 2

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar Wet (g)	Cr/bar Dry (g)
1	11	47	45	36	34	0.944444	129500	4.662	4.403
2	10	73	70	63	60	0.952381	126000	7.938	7.56
3	10	61	58	51	48	0.941176	112000	5.712	5.376
4	10	51	48	41	38	0.926829	114625	4.699625	4.35575
5	11	60	57	49	46	0.938776	152250	7.46025	7.0035
6	10	51	48	41	38	0.926829	131250	5.38125	4.9875
7	10	58	55	48	45	0.9375	147000	7.056	6.615
8	11	39	37	28	26	0.928571	133000	3.724	3.458
9	11	63	60	52	49	0.942308	181125	9.4185	8.875125
10	10	44	42	34	32	0.941176	153125	5.20625	4.9
11	11	65	63	54	52	0.962963	156625	8.45775	8.1445
							AVE	6.337784	5.970761

Appendix B (cont'd). Data on individual "greenola bars".

Chapter 5

Trial 1

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar Wet (g)	Cr/bar Dry (g)
1	12	53	53	41	41	1	121625	4.986625	4.986625
2	12	88	87	76	75	0.986842	121625	9.2435	9.121875
3	12	55	55	43	43	1	138250	5.94475	5.94475
4	12	50	50	38	38	1	132125	5.02075	5.02075
5	12	64	63	52	51	0.980769	126875	6.5975	6.470625
6	12	52	52	40	40	1	124250	4.97	4.97
							AVE	6.127188	6.085771

Trial 2

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar Wet (g)	Cr/bar dry (g)
1	8	64	60	56	52	0.928571	125125	7.007	6.5065
2	8	65	61	57	53	0.929825	124250	7.08225	6.58525
3	8	60	57	52	49	0.942308	152250	7.917	7.46025
4	8	71	68	63	60	0.952381	131250	8.26875	7.875
5	8	51	48	43	40	0.930233	84875	3.649625	3.395
6	8	64	61	56	53	0.946429	158375	8.869	8.393875
							AVE	7.132271	6.702646

Chapter 6

Trial 1

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar wet (g)	Cr/bar dry (g)
1	11	71	69	60	58	0.966667	140875	8.4525	8.17075
2	11	60	58	49	47	0.959184	84875	4.158875	3.989125
3	11	56	54	45	43	0.955556	112000	5.04	4.816
4	11	62	59	51	48	0.941176	140875	7.184625	6.762
5	11	53	51	42	40	0.952381	121625	5.10825	4.865
6	11	63	61	52	50	0.961538	126875	6.5975	6.34375
							AVE	6.090292	5.824438

Trial 2

Cr Bar	Dry Bag (g)	Wet Wt (g)	Dry Wt (g)	Wet bar (g)	Dry bar (g)	%DM	Cr, ppm	Cr/bar Wet (g)	Cr/bar Dry (g)
1	8	57	56	49	48	0.979592	132125	6.474125	6.342
2	8	53	52	45	44	0.977778	119875	5.394375	5.2745
3	8	72	67	64	59	0.921875	151375	9.688	8.931125
4	8	51	49	43	41	0.953488	169750	7.29925	6.95975
5	8	43	40	35	32	0.914286	119875	4.195625	3.836
6	8	57	55	49	47	0.959184	116375	5.702375	5.469625
							AVE	6.458958	6.1355

Appendix C. Individual fecal data

Chapter 3

Trial 1

NUTRIENT	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
Cr, ppm	4436±236	4139±26	4926±44	5338±280	5495±123	5775±18	5005±140	3868±105
Y, ppm	2.11±.01	1.95±.08	1.12±.01	3.92±.04	5.25±.08	2.92±.01	1.81±.02	4.08±.01
DM	28.3±2.0	29.0±4.0	31.5±2.2	27.6±1.3	27.5±1.8	29.0±1.8	28.3±1.8	29.4±3.0
PROTEIN%	9.9±.05	10.3±.05	10.0±.05	13±.1	12.4±0	12.9±.2	10.4±.15	12.2±.1
ADF%	42.9±.4	43.3±1.2	43.6±.05	45±.7	43.9±1.7	46.5±.6	44.1±2.3	43.3±.3
NDF%	65.1±3.3	61.8±2.8	67.1±.7	54.7±.4	56.8±2	60.7±2.2	67.6±.05	65.7±.7
EE%	6.6±0	7±.05	6.9±.25	4.1±.05	4.5±0	6.1±.2	7.3±0	5.5±.05
NSCDIFF%	5±3.4	7.5±3	3.1±.9	11.2±.3	7.0±2	6.8±2.1	2±.3	.7±.7
Ca%	.63±.01	.61±.02	.54±.02	.99±.02	.97±.02	1.03±.01	.64±.03	.81±0
P%	.83±0	.88±.01	.84±.01	.86±.03	.76±.02	.88±0	.92±.01	.76±.02
Mg%	.34±.01	.40±.01	.3±0	.54±.01	.49±.01	.41±.01	.38±0	.48±.01
K%	1.68±.01	1.64±.02	2.28±.05	1.36±.02	1.74±.04	1.6±.01	1.74±0	1.47±.04
Na%	.368±.01	.483±.01	.162±.02	.598±.01	.182±.02	.323±.01	.116±.00	.378±.01
Fe,ppm	1545±5	1645±35	987±14	2950±70	4035±135	1785±25	1235±25	3070±80
Zn, ppm	109±5	114±2.5	85±2	154±2	97±3.5	120±4.5	76±5	136±6
Cu, ppm	26±.5	26±1	23±0	36±.5	30±0	29±1.5	17±1.5	32±.5
Mn, ppm	185±7	190±5.5	136±2	256±4	209±1	197±5.5	134±13.5	216±1
Mo,ppm	4.1±.55	4.6±.55	3.3±.15	4.8±.45	5.5±.35	4.1±.05	3.9±0	4.9±.25
S%	.16±.01	.16±.01	.15±.01	.2±0	.2±0	.22±.01	.16±0	.21±.01
Ash%	13.5±.11	13.5±.11	13±.12	17.04±.04	19.34±.01	13.52±.05	12.8±.09	16.04±0

Trial 2

NUTRIENT	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
Cr, ppm	4332	6169	4270	2827	2249	3002	4034	2389
Y, ppm	4.59	4.52	3.54	1.37	2.41	0.83	6.24	1.71
DM	20.5±.2	21.2±.3	18.9±.3	19.5±.4	20.3±.4	19.8±.2	21.5±.9	21.3±.4
PROTEIN%	11.8±.05	11.4±.05	11.8±.15	9.5±.05	10.0±.2	10.3±0	11.8±0	9.0±.1
ADF%	43.6±1.6	43±2.8	44±1.9	45.5±.1	45.7±3.5	45.4±.7	45.5±1.6	47.0±.7
NDF%	57.5±1.9	54±1.1	55.9±.15	64.4±.25	63.5±2.4	65.4±.75	58.3±.8	65.8±.75
EE%	9.7±.05	11.9±.1	9.8±.35	11.8±.35	11±.1	11.5±0	11±..05	11.7±.15
NSCDIFF%	3.8±2	5.6±1.4	7.6±.2	4.0±.2	3.6±2.5	3.1±.9	.75±.75	2.6±1.2
Ca%	1.08±.02	.93±.02	.96±.01	.55±.01	.58±.01	.55±0	.95±.08	.48±.02
P%	.93±.02	1.07±.03	1.04±0	.75±.01	.77±0	.81±0	.80±.07	.73±.01
Mg%	.59±.02	.56±.01	.55±.01	.32±0	.28±.01	.25±0	.53±.05	.30±.01
K%	1.46±.03	1.46±.01	1.84±.02	1.46±.05	1.78±.01	1.74±.02	1.32±.06	1.36±.01
Na%	.248±.003	.322±.01	.102±.01	.188±.002	.074±.002	.123±.01	.089±.003	.158±.004
Fe,ppm	3055±75	3275±55	2130±50	954±5	1675±15	568±12.5	3970±270	1325±55
Zn, ppm	92±3	92±1	99±.5	67±1	72±1	65±1	89±6.5	70±.5
Cu, ppm	17±.5	20±1.5	19±0	10±0	11±0	10±0	18±1	9±.5
Mn, ppm	180±3.5	192±.5	174±2.5	107±1.5	134±5	102±1.5	190±10.5	115±4.5
Mo,ppm	6.1±.1	6.6±0	5.4±.6	4.0±.1	4.1±.15	2.4±.1	6.4±.8	3.3±.1
S%	.2±.05	.18±.05	.21±.01	.14±.01	.15±0	.15±0	.19±.01	.14±0
Ash%	17.31±.03	17.23±.27	14.99±.15	10.47±.01	11.97±.07	9.83±.05	18.4±3.0	10.98±.18

Appendix C (cont'd.). Individual fecal data

Chapter 4.

Trial 1

NUTRIENT	93TF	94OG	98TF	99OG	100OG	101TF	102TF	103OG
Cr, ppm	3658	4568	5031	3124	3019	3981	4559	2774
Y, ppm	1.99±.01	3.05±.01	1.9±.01	3.19±.01	3.38±.04	1.42±.08	1.95±.04	2.67±.01
DM	21.49±1.37	20.87±.42	20.69±.27	22.68±.88	20.76±.53	21.85±1.24	19.53±1.00	21.42±.8
PROTEIN%	7.7±0	9.8±.3	8.9±.1	10.7±0	11.5±.2	9.1±0	7.8±.1	11.3±.2
ADF%	57.5±2.8	54.1±1.2	57.2±.4	51.8±1.5	61.5±.8	53.1±1.3	55.1±1.8	54.2±.7
NDF%	73.2±.7	65.6±.5	67.5±.4	62.9±.1	70.1±.1	70.6±.8	72.9±.3	67±.8
EE%	2.1±0	2.9±.1	2.4±.1	2.6±0	2.4±.1	2.1±0	1.5±.1	1.9±.1
NSCDIFF%	9.1±.9	13.8±1.5	12.8±.4	14.8±.2	7.1±.2	11.3±.8	9.9±.1	10.7±.6
Ca%	.84±.01	1.02±.03	.86±0	1.18±.01	1.18±.1	.71±.02	.78±.01	1.15±.02
P%	.49±.01	.64±.02	.54±0	.62±0	.56±.05	.51±.01	.55±.01	.61±.01
Mg%	.31±.01	.3±.01	.28±0	.31±0	.25±.02	.26±.01	.30±0	.29±0
K%	1.24±.01	1.46±.02	1.02±.01	1.46±0	1.40±.12	1.06±.02	1.27±.04	1.37±.02
Na%	.028±.001	.038±.001	.055±.001	.021±0	.02±.002	.033±.002	.049±.004	.036±.002
Fe,ppm	985±6	1215±35	1130±0	1105±5	957±73	780±5	881±16	1205±55
Zn, ppm	44±1	57±1	47±0	50±0	48±4	39±0	43±1	55±1
Cu, ppm	21±1	20±1	17±0	22±1	20±2	13±1	15±2	25±1
Mn, ppm	96±4	155±2	103±0	134±1	107±9	90±0	104±1	139±7
Mo,ppm	2.2±.1	2.9±.4	2.3±.02	2.5±0	2.3±.2	2±.5	1.9±.3	2.4±.9
S%	.13±0	.15±0	.14±0	.16±0	.16±0	.13±0	.12±0	.16±.01
Ash%	7.94±.11	7.97±1.79	8.5±.05	9.08±.01	9.06±.13	6.96±.06	8.04±.12	9.33±.12

Trial 2

NUTRIENT	93OG	94TF	98OG	99TF	100TF	101OG	102OG	103TF
Cr, ppm	4130	2861	4926	2861	2870	3658	4699	2966
Y, ppm	2.96±.03	1.48±.01	3.27±.01	2.01±.01	2.09±0	2.07±0	3.73±.01	1.62±.04
DM	19.39±.44	18.52±.16	17.90±.53	20.80±.24	19.40±.64	20.57±.25	19.31±.97	19.62±.37
PROTEIN%	10.8±.1	7.6±.1	10.4±0	7.7±.1	8.3±.1	11.6±.1	11.3±.1	7.8±.1
ADF%	52.8±2.1	51±.4	55.4±2.1	56.6±.1	57.8±2	54.2±.9	53.4±1.4	57.1±1
NDF%	67.2±.8	71.5±1.7	66±1.6	73.9±1	74.3±1.2	70.3±.9	65.1±.2	75.4±2.2
EE%	2.1±.1	1.5±.1	2.1±.1	1.6±.1	1.4±.1	2.1±0	2.6±0	1.7±0
NSCDIFF%	10.1±.7	10.6±1.9	11.5±1.8	8.6±1.2	7.6±1.2	7.3±.9	11.3±.3	7.7±2.2
Ca%	1.13±.03	.70±.01	1.06±.03	.74±.01	.79±.02	.88±.01	1.18±.01	.62±0
P%	.66±.01	.51±.01	.67±.02	.45±.01	.45±.01	.62±.02	.71±0	.44±.01
Mg%	.3±.01	.34±0	.26±0	.31±0	.27±0	.24±0	.30±.01	.28±0
K%	1.82±.06	1.45±.02	2.01±.04	1.27±.02	1.45±.01	1.98±.01	1.64±.01	1.12±.01
Na%	.024±.001	.049±.002	.021±0	.022±.001	.023±.001	.023±.002	.052±.002	.054±.005
Fe,ppm	1095±45	951±17	1350±160	1065±5	1000±0	915±40	1145±5	809±25
Zn, ppm	56±1	41±2	63±1	37±0	45±1	56±2	64±1	38±1
Cu, ppm	22±1	17±1	22±1	16±1	20±2	20±1	22±0	15±1
Mn, ppm	137±4	103±3	137±0	121±2	107±1	106±2	145±1	92±5
Mo,ppm	2.4±.4	2.8±.3	2.9±.2	2.1±0	2.7±.1	2.2±.2	2.8±.2	1.8±.3
S%	.16±0	.13±0	.16±0	.13±0	.12±0	.16±0	.17±.01	.13±.01
Ash%	9.9±.07	8.89±.02	10.1±.14	8.35±.13	8.51±.06	8.83±.06	9.69±.04	7.55±.03

Appendix C (cont'd.) Individual fecal data

Chapter 5

Trial 1

NUTRIENT	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
Cr, ppm	5102	6169	6467	3947	4944	5880	5163	3894
Y, ppm	6.25±.03	1.54±.01	6.49±.02	7.13±.09	1.47±.02	1.73±.04	1.18±.02	9.66±.03
DM	21.09±.84	21.43±.74	21.66±.25	16.83±.69	16.88±.7	20.38±.60	21.96±.77	21.21±.26
PROTEIN%	15.0±.2	9.1±.05	11.8±.1	16.1±.2	9.1±.15	9.0±.2	8.9±.05	13.3±.25
ADF%	41.9±1.4	49.8±.5	49.3±1.4	43±.9	50.8±.8	51±1.8	49.1±2.1	43±.4
NDF%	55.45±.55	65.9±1.1	59.2±1.05	57.4±2.4	69.5±.8	67.2±.3	68.5±1.2	57.5±2.8
EE%	11.7±.05	5.3±.15	11.2±.1	12.4±0	4.4±.6	4.2±.1	4.9±0	12.1±0
NSCDIFF%	4.2±.05	7.5±.95	3.3±.45	1.8±1.8	3.5±.8	6.1±.4	7.3±.9	2.6±2.6
Ca%	1.05±.01	.98±.03	1.02±.01	.97±0	.98±.01	.87±.01	.92±.04	1.11±.01
P%	.86±0	.98±.03	.89±0	.81±0	.71±.01	.76±.01	.8±.01	.82±.01
Mg%	.40±0	.40±.01	.35±0	.4±0	.40±.01	.37±.01	.35±.01	.43±0
K%	1.86±.01	1.73±0	1.98±.01	2.01±.03	1.64±.01	1.34±0	1.73±.01	1.51±.01
Na%	.122±.01	.048±.002	.039±.001	.241±.001	.048±0	.040±.002	.018±0	.097±.002
Fe,ppm	1940±60	962±58	2395±45	2100±130	901±75	972±4	240±6.5	3080±0
Zn, ppm	77±1.5	59±2.5	82±1.5	79±.5	50±.5	55±.5	54±.5	92±1.5
Cu, ppm	22±.5	9±0	19±1.5	23±.5	10±1	8±.5	11±.5	19±1.5
Mn, ppm	176±2.5	149±4	215±1	148±.5	155±0	186±4	144±1.5	194±.5
Mo,ppm	5.1±.3	3.7±.35	5.5±.15	4.8±.1	3.9±.35	4±.5	3.2±.35	5.2±0
S%	.23±.01	.17±0	.18±0	.24±.01	.18±0	.18±.01	.17±.01	.22±.01
Ash%	13.78±.22	12.41±.08	14.61±.37	13.06±.08	13.54±.7	13.52±.21	10.45±.18	14.92±.21

Trial 2

NUTRIENT	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
Cr, ppm	3877	4866	4489	3947	4349	6196	4209	2625
Y, ppm	9.9±.09	2.21±.17	10.78±.14	3.74±.04	1.3±.01	1.13±.1	2.11±.32	4.31±.08
DM	22.03±.32	22.54±.49	20.81±.5	20.83±.52	21.67±1	23.17±.54	20.57±.38	22.99±.56
PROTEIN%	11.7±.35	7.5±0	10.5±.2	11.9±.25	8.3±.1	8.5±.1	8.3±.1	11.8±.15
ADF%	44.7±1.55	49.2±1.35	47.7±1.6	46.5±1.25	49.5±1.9	48.5±.2	49.2±2.45	45.2±.75
NDF%	56.5±0	66.3±1.2	60.7±1.35	60.5±.4	70.4±.1	67.5±.3	66.2±.1	60±2.3
EE%	12.4±.1	5.8±.1	13.7±.1	13.4±0	5.3±.15	5.6±0	5.4±.1	12.6±0
NSCDIFF%	6.95±.65	6.9±1.3	3.2±.95	4.1±.3	6.4±.3	9.5±.6	9.8±.1	4.8±2.7
Ca%	1.16±.01	.85±.01	1.03±.01	.83±.01	.69±.02	.52±.01	.8±.03	.86±.03
P%	.57±.03	.80±.01	.60±0	.57±0	.6±.01	.63±0	.64±.05	.58±.01
Mg%	.50±.01	.57±.01	.44±0	.37±0	.30±.01	.24±.01	.42±0.03	.44±0
K%	1.30±.04	1.13±.02	1.57±.02	1.32±.1	1.42±.04	1.15±.02	1.26±.06	1.32±.01
Na%	.05±.01	.071±.004	.046±.001	.115±.005	.019±.001	.055±.006	.02±.01	.044±0
Fe,ppm	2125±115	903±11	1850±40	1130±0	601±58	662±81	563±9	1495±115
Zn, ppm	59.5±1.5	42±0	61±2	62±0	44±2	43±1	46±1	52±0
Cu, ppm	16±1.5	6±0	17±0	13±1	7±0	6±.5	8±.5	14±1.5
Mn, ppm	170±3.5	164±1	163±2	147±2	127±6.5	101±3	176±5	152±4
Mo,ppm	4.7±.1	3.7±.3	4.9±.1	3.8±.6	2.7±.2	2.6±.6	3±0	3.9±0
S%	.23±.01	.19±.01	.21±.01	.21±0	.18±.01	.17±0	.18±0	.20±0
Ash%	12.48±.15	13.54±.01	12.04±.1	10.16±.16	9.7±.19	8.93±.22	10.55±.09	10.85±.23

Appendix C (cont'd.). Individual fecal data

Chapter 6

Trial 1

NUTRIENT	93BG	94BG	99BG	100BG	102TF	131TF	132TF	133TF
DM IN								
DM OUT	3.61±.23	4.38±.4	3.27±.16	4.08±.51	2.94±.21	4.44±.21	3.08±.65	2.63±.15
Cr, ppm	6064	4060	5172	5049	7175	5531	6957	7132
Y, ppm	2.49±.06	3.84±.13	3.74±.09	3.99±.03	7.17±.17	5.48±.05	6.31±.05	4.63±.07
DM	15.66±.94	16.48±1.4	14.47±1.15	15.54±1.48	17±.83	15.43±.85	17.11±.95	14.54±.82
PROTEIN%	12.0±.1	12.0±.2	15.9±.1	13.6±.4	14.4±.1	13.9±.2	14.2±.1	14.2±0
ADF%	41.5±.2	39.3±.2	42.7±1.9	40.0±.4	43.1±1	46±.9	45.5±1.9	45.9±.6
NDF%	61.7±.15	60.8±2.3	59.4±.1	59.5±.2	59.8±2.1	62.8±1.3	58.8±.8	61.2±.9
EE%	5.4±0	6.1±.1	5.4±.1	5.5±.1	6.1±0	4.4±.1	4.9±.1	4.9±.1
NSCDIFF%	0	0	0	0	0	0	0	0
Ca%	.64±.02	.48±0	.65±0	.66±.01	.81±.07	.71±.05	.65±.03	.58±0
P%	.93±0	1.0±0	1.04±0	1.04±0	.95±.09	.84±.02	.80±.06	.92±.04
Mg%	.28±0	.31±0	.31±0	.29±.01	.35±.02	.35±0	.35±.02	.29±.01
K%	2.88±.02	2.93±.06	2.61±.05	3.22±.01	2.31±.14	2.36±.05	2.31±.07	2.84±.1
Na%	.096±.008	.219±.02	.635±.032	.276±.014	.186±.009	.257±.084	.040±.002	.375±.012
Fe,ppm	1135±35	1695±195	1685±45	1970±40	3940±340	3160±360	3385±35	2475±5
Zn, ppm	79±3	84±2	107±5	87±1	91±7	79±5	107±1	117±9
Cu, ppm	20±0	18±1	22±1	22±0	16±1	17±2	14±0	20±1
Mn, ppm	181±4.5	250±14	236±4	233±1	203±14	181±5	201±6	188±5
Mo,ppm	3.6±0	4.6±.1	3.9±.1	4.6±.1	7.1±.7	7.2±.1	5.9±.3	5.3±.2
Cl %	.36±.01	.49±.07	.48±.02	.45±.01	.34±.01	.39±.05	.26±.01	.52±0
S%	.21±0	.22±.01	.27±.01	.24±.01	.28±.03	.25±0	.27±.01	.26±.01
Ash%	20.9	21.1	19.3	21.4	19.7	18.9	22.1	19.7

Trial 2

NUTRIENT	93TF	94TF	99TF	100TF	102BG	131BG	132BG	133BG
Cr, ppm	6161	4445	4857	4822	4270	2949	4892	4927
Y, ppm	3.62±.09	4.62±.18	3.38±.06	4.32±.10	3.41±.09	4.45±.06	3.95±.11	2.89±.04
DM	16.25±2.33	17.86±2.16	14.41±1.44	15.55±1.4	18.11±2.18	17.68±1.30	17.39±1.47	16.81±1.08
PROTEIN%	10.9±.1	11.3±0	12.7±.2	14.3±.3	14.1±0	13.2±.1	13±0	11.5±.1
ADF%	46.3±.3	47.7±.2	47±.9	44.4±2.3	39.7±1	46.2±.4	42±.2	42.7±.1
NDF%	62.3±3.1	64.5±.2	69.1±.8	58.7±.1	58.8±3.1	61.1±.2	62.3±1.3	66.3±.5
EE%	6.3±0	5.5±0	5.4±.5	5.4±0	6.8±0	5.6±.1	5.8±0	6.9±.5
NSCDIFF%	0	0	0	0	0	0	0	0
Ca%	.71±.02	.61±.08	.77±.02	.8±.04	.77±.03	.67±.02	.85±0	.58±.03
P%	.79±.02	.73±.1	.88±.03	.83±.05	.76±.03	.70±0	.83±.02	.84±.04
Mg%	.41±.01	.41±.05	.46±.02	.42±.02	.36±.01	.31±.01	.39±.01	.3±.02
K%	2.52±.02	2.40±.2	2.59±.01	3.03±.05	2.54±0	2.21±.04	2.63±.04	3.01±.13
Na%	.042±.001	.056±.005	.162±.001	.108±.002	.049±.003	.088±.001	.031±.002	.127±.006
Fe,ppm	2025±55	2605±355	1885±95	2660±160	1400±30	1840±0	1760±60	1155±65
Zn, ppm	64±2	70±8	68±1	75±3.5	62±2	65±1	71±0	74±4
Cu, ppm	14±1	13±2	17±1	17±0	16±1	13±0	14±1	18±0
Mn, ppm	169±2.5	191±24	157±4	161±8	190±3	225±1.5	233±5	196±10
Mo,ppm	5.7±.3	5.7±1	5.4±.3	7±.3	4.5±.2	5.1±.4	4.9±.1	3.9±.4
Cl %	.47±.02	.48±.02	.39±.01	.60±.01	.35±.04	.31±0	.41±.02	.42±.01
S%	.21±0	.23±0	.22±.01	.29±.01	.25±.01	.22±.01	.28±.02	.2±.02
Ash%	20.5	18.7	12.8	21.6	20	20.1	18.9	15.3

Appendix D. Yttrium digestibilities for individual horses

Chapter 3

Trial 1

Nutrient	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
DM	0.582938	0.548718	0.214286	0.8010204	0.851429	0.732877	0.513812	0.808824
PROTEIN	0.732796	0.699333	0.491262	0.8566759	0.89764	0.80918	0.672854	0.870903
ADF	0.508536	0.463508	0.058527	0.7092532	0.787627	0.596899	0.411192	0.731484
NDF	0.549806	0.537781	0.125363	0.788635	0.835666	0.685303	0.455164	0.756336
EE	0.190529	0.071471	-0.59512	0.7450313	0.790469	0.491031	-0.04347	0.671719
NSCDIFF	0.843233	0.745677	0.816797	0.8747865	0.941404	0.898	0.926917	0.992489
Ca	0.343225	0.312225	-0.0611	0.7144783	0.790536	0.601435	0.554095	0.775783
P	-0.01797	-0.16729	-0.94188	0.5374595	0.693946	0.364973	-0.31506	0.607676
Mg	0.2911	0.098	-0.179	0.6839412	0.785265	0.678029	0.0766	0.730353
K	0.683005	0.665321	0.189104	0.9063529	0.910291	0.85218	0.617357	0.902848
Na	-50.152	-71.611	-41.444	-6.933467	-0.80787	-4.7494	-17.792	-3.8132
Fe	-0.41286	-0.62696	-0.70128	-0.012155	-0.03658	0.178284	-0.31625	-0.01098
Zn	-0.37736	-0.558	-1.02455	0.1827733	0.614587	0.1456	-0.11927	0.307307
Cu	0.583	0.595655	0.304692	0.7015	0.81375	0.677375	0.682231	0.745333
Mn	-0.42861	-0.58685	-0.97956	0.1913651	0.505698	0.165095	-0.206	0.345143
Mo	0.050167	-0.15256	-0.441	0.5846957	0.643696	0.524043	-0.053	0.640038
S	0.682286	0.656381	0.438571	0.8469231	0.885385	0.774077	0.629714	0.845731
Ash	0.26025	0.199934	-0.34271	0.6411683	0.695062	0.618006	0.182549	0.675805

Trial 2

Nutrient	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
DM	0.83	0.827	0.78	0.358	0.635	-0.06	0.875	0.485
PROTEIN	0.888864	0.890737	0.856177	0.605243	0.763754	0.293333	0.918283	0.7
ADF	0.759351	0.758474	0.685714	0.1975	0.541745	-0.32209	0.815341	0.335027
NDF	0.810194	0.818602	0.761204	0.313779	0.615311	-0.15061	0.858495	0.43756
EE	0.484688	0.356656	0.32625	-1.22812	-0.18088	-2.58529	0.570313	-0.77221
NSCDIFF	0.963606	0.94542	0.905803	0.806917	0.901203	0.752932	0.994718	0.899323
Ca	0.733913	0.766826	0.693913	0.11725	0.47075	-0.4575	0.827899	0.382
P	0.566849	0.492849	0.373151	-0.41618	0.173382	-1.52529	0.726027	-0.10574
Mg	0.705	0.715059	0.644118	-0.0272	0.489	-0.325	0.805147	0.2275
K	0.914118	0.912602	0.859931	0.575873	0.706018	0.16543	0.942907	0.683077
Na	-1.81067	-2.71373	-0.496	-39.232	-8.00333	-42.46	0.258333	-26.1233
Fe	0.104569	0.023147	0.192069	-0.34313	-0.34073	-0.32035	0.144397	-0.49644
Zn	0.582933	0.575573	0.4192	-0.30345	0.203636	-1.08788	0.703333	-0.09242
Cu	0.879583	0.855833	0.825833	0.753077	0.845577	0.592308	0.90625	0.821731
Mn	0.514286	0.472762	0.392381	-0.27211	0.094259	-1.00222	0.623016	-0.09676
Mo	0.54913	0.503565	0.483478	-0.42667	0.168611	-0.41333	0.652174	0.055833
S	0.869231	0.880231	0.822308	0.561561	0.732927	0.22439	0.908654	0.648293
Ash	0.888864	0.890737	0.856177	0.605243	0.763754	0.293333	0.918283	0.7

Appendix D (cont'd.). Yttrium digestibilities for individual horses

Chapter 4

Trial 1

Nutrient	93TF	94OG	98TF	99OG	100OG	101TF	102TF	103OG
DM	0.733668	0.642623	0.721053	0.658307	0.677515	0.626761	0.728205	0.59176
PROTEIN	0.838724	0.757042	0.80448	0.745875	0.742847	0.732732	0.832945	0.679833
ADF	0.683988	0.584652	0.670273	0.619019	0.574129	0.590779	0.690347	0.524439
NDF	0.698588	0.62409	0.708475	0.658	0.637685	0.592356	0.693053	0.56122
EE	0.202	-0.29413	0.043429	-0.1115	0.034	-0.119	0.417143	0.031
NSCDIFF	0.84075	0.655483	0.765053	0.646042	0.840126	0.722704	0.822842	0.694713
Ca	0.650875	0.51448	0.625094	0.46192	0.493387	0.586203	0.6685	0.3744
P	0.498692	0.184	0.420538	0.242714	0.356	0.268346	0.424615	0.111143
Mg	0.566	0.37	0.588842	0.376353	0.526471	0.489579	0.570526	0.304
K	0.864263	0.820269	0.882889	0.827821	0.844552	0.837292	0.857844	0.807255
Na	0.676174	-0.04354	0.332826	0.447538	0.504615	0.464826	0.420522	-0.12985
Fe	0.660169	0.459832	0.591089	0.529377	0.616247	0.622646	0.689193	0.387746
Zn	0.714537	0.5155	0.680171	0.592857	0.632	0.645195	0.714732	0.465714
Cu	0.492182	0.405	0.568818	0.373	0.463333	0.559182	0.629091	0.15
Mn	0.527111	0.1075	0.467833	0.260839	0.44429	0.378333	0.476148	0.08529
Mo	0.024667	0.058818	-0.0695	0.222727	0.326727	-0.24333	0.138667	0.109818
S	0.783875	0.685	0.755875	0.678118	0.678	0.696938	0.796	0.616
Ash	0.693017	0.64873	0.655305	0.616622	0.639837	0.622663	0.68214	0.530044

Trial 2

Nutrient	93OG	94TF	98OG	99TF	100TF	101OG	102OG	103TF
DM	0.739865	0.5	0.764526	0.631841	0.645933	0.628019	0.793566	0.54321
PROTEIN	0.803636	0.651376	0.829091	0.740037	0.73044	0.698238	0.837217	0.672972
ADF	0.716364	0.47205	0.731012	0.568762	0.576373	0.583421	0.772719	0.459737
NDF	0.722667	0.472714	0.75381	0.598891	0.612062	0.584895	0.787133	0.491773
EE	0.393333	0.464286	0.451667	0.579429	0.646	0.132	0.404889	0.445071
NSCDIFF	0.813759	0.576	0.808333	0.746816	0.784768	0.807404	0.834908	0.718488
Ca	0.567941	0.385965	0.633676	0.522246	0.509368	0.518588	0.642529	0.502912
P	0.446452	-0.02	0.492097	0.3376	0.3628	0.256	0.528194	0.19568
Mg	0.48	0.055556	0.592667	0.239467	0.469	0.4048	0.588	0.289111
K	0.843311	0.734432	0.843593	0.828806	0.811978	0.756106	0.888132	0.812513
Na	0.108571	-0.63333	0.295	0.460267	0.4572	-0.22229	-0.53029	-0.6452
Fe	0.300491	0.087332	0.220516	0.247754	0.320537	0.163686	0.420467	0.290378
Zn	0.530323	0.359375	0.522419	0.5745	0.502188	0.328	0.57471	0.457313
Cu	0.364444	0.055556	0.425556	0.345778	0.213333	0.173333	0.496444	0.238333
Mn	0.363929	0.253623	0.425089	0.354667	0.451043	0.295857	0.466607	0.390667
Mo	0.376	0	0.3185	0.448	0.317286	0.1816	0.4232	0.412429
S	0.74	0.59375	0.765	0.701	0.7345	0.628	0.781125	0.628688
Ash	0.671684	0.410477	0.697258	0.592467	0.600459	0.581026	0.74539	0.542394

Appendix D (cont'd.). Yttrium digestibilities for individual horses

Chapter 5

Trial 1

Nutrient	93BG P	94BG S	98BG P	99TF P	100TF S	101TF S	102BG S	103TF P
DM	0.955	0.818	0.957	0.937	0.694	0.74	0.763	0.953
PROTEIN	0.949438	0.87594	0.961993	0.926233	0.797484	0.829818	0.842	0.954538
ADF	0.951467	0.766703	0.945434	0.933194	0.616651	0.672996	0.700471	0.95016
NDF	0.961374	0.814337	0.960594	0.943007	0.664823	0.724634	0.748692	0.957407
EE	0.815263	0.661544	0.831018	0.681143	0.450449	0.554286	0.592526	0.767878
NSCDIFF	0.984118	0.885294	0.988076	0.989644	0.902192	0.85516	0.854613	0.98884
Ca	0.937	0.762187	0.94152	0.923613	0.62515	0.71725	0.70928	0.934788
P	0.875161	0.424645	0.876548	0.835387	0.299161	0.362581	0.388387	0.875677
Mg	0.9	0.595556	0.916389	0.91	0.562857	0.656429	0.539167	0.927821
K	0.962127	0.857529	0.961475	0.949348	0.799264	0.86064	0.814475	0.971612
Na	0.085	-0.456	0.7205	-0.0122	0.0208	0.306667	0.289	0.696067
Fe	0.826441	0.65192	0.795258	0.444118	-0.15843	-0.06185	0.886918	0.391765
Zn	0.92125	0.755955	0.919864	0.853618	0.55	0.579412	0.709136	0.872824
Cu	0.835	0.727	0.863833	0.7585	0.49	0.653333	0.5655	0.851167
Mn	0.858571	0.51575	0.834911	0.890306	0.442	0.431059	0.390571	0.892729
Mo	0.856563	0.579125	0.852188	0.7984	0.2044	0.306667	0.526	0.837067
S	0.9425	0.828111	0.957	0.937	0.7705	0.805	0.776167	0.956917
Ash	0.915171	0.691023	0.914059	0.912563	0.559698	0.62644	0.661197	0.925479

Trial 2

Nutrient	93TF P	94TF S	98TF P	99BG P	100BG S	101BG S	102TF S	103BG P
DM	0.892	0.516	0.901	0.869	0.623	0.566	0.493	0.886
PROTEIN	0.881907	0.660748	0.90285	0.908245	0.735941	0.688692	0.60672	0.886481
ADF	0.889529	0.455085	0.891938	0.855652	0.557784	0.501209	0.42919	0.882087
NDF	0.904805	0.499388	0.906251	0.885635	0.617016	0.577273	0.47639	0.901299
EE	0.647579	0.261263	0.643079	0.472147	0.412324	0.285176	0.279526	0.577529
NSCDIFF	0.937967	0.724	0.973818	0.938264	0.722667	0.526092	0.589372	0.937103
Ca	0.878369	0.600583	0.901	0.815712	0.559102	0.617492	0.606214	0.833831
P	0.801419	-0.24903	0.808387	0.742517	0.22	0.057172	-0.04671	0.772
Mg	0.841176	0.188588	0.871882	0.730722	0.371667	0.421333	0.373706	0.721333
K	0.919771	0.687474	0.911183	0.871911	0.603452	0.630296	0.63496	0.888533
Na	0.1	-4.72733	0.241	-4.02167	-1.38767	-6.95667	-0.69	-0.672
Fe	0.418987	-0.10646	0.536329	0.697898	0.537598	0.413657	0.277365	0.652184
Zn	0.824865	0.450595	0.836784	0.767943	0.526057	0.4668	0.369676	0.830629
Cu	0.6544	0.4192	0.6634	0.6594	0.4722	0.4792	0.1888	0.6808
Mn	0.814545	0.198222	0.837	0.67905	0.202017	0.269433	0.098667	0.7112
Mo	0.718	0.005111	0.7305	0.707176	0.401235	0.336235	0.155	0.738471
S	0.90064	0.63216	0.91684	0.847167	0.623	0.590111	0.63496	0.873333
Ash	0.844102	0.301348	0.872925	0.802821	0.458237	0.425834	0.42976	0.816756

Appendix D (cont'd.). Yttrium digestibilities for individual horses

Chapter 6

Trial 1

Nutrient	93BG	94BG	99BG	100BG	102TF	131TF	132TF	133TF
DM	0.726908	0.822917	0.818182	0.829574	0.927476	0.905109	0.917591	0.887689
PROTEIN	0.79525	0.86725	0.819138	0.8555	0.930384	0.91255	0.922887	0.894675
ADF	0.686163	0.80731	0.784726	0.811634	0.916544	0.884085	0.901034	0.863639
NDF	0.716906	0.819133	0.818306	0.83	0.929362	0.903463	0.921981	0.889087
EE	0.612053	0.715868	0.741368	0.753947	0.851567	0.860667	0.866067	0.817067
NSCDIFF	1	1	1	1	1	1	1	1
Ca	0.620174	0.815304	0.746783	0.756087	0.879327	0.862347	0.891224	0.867429
P	0.29475	0.508333	0.474222	0.508889	0.768833	0.734	0.781333	0.656533
Mg	0.597684	0.711211	0.703053	0.740526	0.883864	0.848864	0.869545	0.852364
K	0.688	0.794202	0.8115	0.782778	0.936605	0.915714	0.928789	0.880421
Na	-1.912	-3.307	-11.8411	-4.21333	0.030143	-0.74393	0.765714	-2
Fe	0.58686	0.59998	0.591107	0.553467	0.460375	0.436773	0.479231	0.479925
Zn	0.509841	0.662091	0.557409	0.663864	0.899348	0.886288	0.867061	0.801455
Cu	0.22	0.544857	0.428	0.465714	0.805333	0.730833	0.808667	0.626667
Mn	0.538196	0.586449	0.598579	0.629813	0.807545	0.776688	0.785948	0.726545
Mo	0.421882	0.521059	0.582471	0.54	0.712056	0.62	0.731222	0.670222
Cl	0.861577	0.877845	0.876958	0.892254	0.968179	0.9525	0.972667	0.925333
S	0.71335	0.8053	0.7543	0.796	0.907091	0.892045	0.899364	0.867636
Ash	0.228959	0.495311	0.522716	0.508378	0.805662	0.757365	0.755108	0.701838

Trial 2

Nutrient	93TF	94TF	99TF	100TF	102BG	131BG	132BG	133BG
DM	0.712707	0.774892	0.692308	0.759259	0.765396	0.820225	0.797468	0.723183
PROTEIN	0.800745	0.838057	0.750854	0.78049	0.791604	0.850566	0.834025	0.799654
ADF	0.646593	0.714561	0.615	0.715415	0.734202	0.763077	0.757094	0.663023
NDF	0.693834	0.751498	0.635568	0.757762	0.760104	0.809063	0.780436	0.681161
EE	0.547975	0.690625	0.5842	0.67465	0.610244	0.754146	0.712829	0.529776
NSCDIFF	1	1	1	1	1	1	1	1
Ca	0.676556	0.782143	0.623556	0.693968	0.703361	0.802295	0.717131	0.736623
P	0.218172	0.433621	0.065379	0.310241	0.384138	0.565517	0.419	0.197655
Mg	0.509708	0.615625	0.409667	0.57825	0.577	0.721	0.60415	0.5845
K	0.652288	0.740385	0.616481	0.648928	0.690725	0.793886	0.723373	0.567995
Na	-0.50675	-0.575	-5.237	-2.2535	-0.43938	0.82	0.213375	-3.39738
Fe	-0.0434	-0.05229	-0.04233	-0.15092	0.45799	0.454366	0.4114	0.472924
Zn	0.582545	0.642045	0.524	0.589205	0.744386	0.794737	0.74714	0.640386
Cu	0.1964	0.415	-0.0472	0.1806	0.373333	0.61	0.526333	0.169
Mn	0.326347	0.403125	0.328389	0.461097	0.474706	0.523529	0.443541	0.361271
Mo	-0.0906	0.145	-0.1088	-0.12467	0.186538	0.293846	0.234846	0.109
Cl	0.839417	0.871429	0.857	0.827857	0.862917	0.907	0.861283	0.8061
S	0.726045	0.764773	0.692	0.682318	0.690789	0.791579	0.700842	0.708421
Ash	0.215533	0.439	0.474347	0.30592	0.364865	0.511081	0.481527	0.427284

Appendix E. TC digestibilities for individual horses

Chapter 4

Trial 1

NUTRIENT	93TF	94OG	98TF	99OG	100OG	101TF	102TF	103OG
DM IN (kg)	8.03±.62	8.78±.20	6.32±.38	10.96±.44	10.73±.42	7.59±.43	6.78±.39	10.43±.53
DM OUT (kg)	4.06±.34	3.27±.12	2.89±.17	4.54±.25	4.85±.10	3.48±.26	3.17±.12	4.77±.32
DM	0.494396	0.627563	0.542722	0.585766	0.547996	0.541502	0.532448	0.542665
PROTEIN	0.693453	0.746536	0.679545	0.692201	0.639025	0.67147	0.712842	0.641119
ADF	0.399334	0.566691	0.45958	0.538553	0.402189	0.496978	0.467725	0.466935
NDF	0.427087	0.607835	0.522194	0.581777	0.491405	0.498917	0.472376	0.508163
EE	-0.51681	-0.35009	-0.56781	-0.34626	-0.35601	-0.37549	-0.0019	-0.08617
NSCDIFF	0.697303	0.640585	0.614923	0.571283	0.775579	0.659143	0.695476	0.657799
Ca	0.336395	0.493485	0.385532	0.348273	0.288847	0.491354	0.430171	0.298754
P	0.050678	0.148715	0.050268	0.100952	0.095993	0.100638	0.010948	0.003664
Mg	0.178138	0.342758	0.326116	0.244633	0.335289	0.372582	0.261761	0.219841
K	0.741996	0.812497	0.808056	0.791455	0.781791	0.799997	0.755642	0.783949
Na	0.384482	-0.08866	-0.09349	0.330853	0.30461	0.342155	0.003912	-0.26647
Fe	0.35406	0.436474	0.329799	0.429977	0.461311	0.53615	0.465742	0.313713
Zn	0.457401	0.494549	0.475803	0.506865	0.483424	0.563868	0.509641	0.401109
Cu	0.034756	0.379271	0.293297	0.240572	0.24666	0.458139	0.36243	0.04722
Mn	0.101148	0.068907	0.127784	0.104721	0.219929	0.235837	0.09953	-0.02531
Mo	-0.85388	0.01812	-0.7529	0.05856	0.054901	-0.52833	-0.48058	0.002179
S	0.589197	0.671379	0.599881	0.610133	0.574585	0.62747	0.649336	0.569567
Ash	0.416498	0.63354	0.435048	0.535649	0.494425	0.536171	0.453617	0.473218

Trial 2

NUTRIENT	93OG	94TF	98OG	99TF	100TF	101OG	102OG	103TF
DM IN (kg)	9.05±.45	10.25±.53	7.74±.48	10.00±.42	11.07±.39	11.24±.24	7.14±.44	10.77±.4
DM OUT (kg)	3.95±.14	4.7±.24	3.18±.2	4.38±.25	5.45±.31	4.34±.26	2.72±.17	4.85±.31
DM	0.563536	0.541463	0.589147	0.562	0.507678	0.613879	0.619048	0.549675
PROTEIN	0.670363	0.680286	0.701198	0.690587	0.625113	0.686783	0.698968	0.677749
ADF	0.523857	0.515831	0.529726	0.486733	0.410845	0.567608	0.579693	0.467628
NDF	0.534438	0.51644	0.569583	0.522593	0.460479	0.569138	0.606349	0.499196
EE	-0.01842	0.508711	0.041344	0.499429	0.507678	0.099051	-0.10053	0.453177
NSCDIFF	0.687356	0.611161	0.664907	0.698656	0.700668	0.800093	0.694698	0.7226
Ca	0.274699	0.436885	0.359553	0.431368	0.31766	0.500314	0.338936	0.510173
P	0.070754	0.064585	0.112028	0.2116	0.113821	0.227758	0.127496	0.207428
Mg	0.127072	0.133875	0.287855	0.245667	0.261518	0.382206	0.238095	0.299494
K	0.736965	0.756455	0.726552	0.796242	0.738511	0.746848	0.793125	0.815251
Na	-0.49645	-0.49789	-0.23256	0.3576	0.245107	-0.26868	-1.82993	-0.62117
Fe	-0.17427	0.163017	-0.36278	0.104664	0.055045	0.131939	-0.07172	0.300743
Zn	0.211549	0.4125	0.165041	0.493563	0.307673	0.302491	0.213518	0.465239
Cu	-0.06691	0.133875	-0.00431	0.221333	-0.09405	0.145117	0.068783	0.249458
Mn	-0.06778	0.315518	-0.00512	0.231913	0.236545	0.269128	0.013605	0.399567
Mo	0.053416	0.082927	-0.19147	0.343	0.050523	0.150534	-0.06667	0.421011
S	0.563536	0.627439	0.589147	0.644125	0.630759	0.613879	0.595238	0.634111
Ash	0.448853	0.459365	0.470713	0.514947	54.45192	0.565121	0.529155	0.549078

Appendix E (cont'd.). TC digestibilities for individual horses

Chapter 5 (STALLED HORSES ONLY)

Trial 1

NUTRIENT	94BG	100TF	101TF	102BG
DM IN (kg)	7.83±.64	10.87±.58	9.01±.43	9.81±.72
DM OUT (kg)	2.74±.20	3.99±.12	3.12±.12	3.74±.19
DM	0.650064	0.632935	0.653718	0.618756
PROTEIN	0.761467	0.757069	0.773343	0.749589
ADF	0.551433	0.54015	0.564479	0.518171
NDF	0.643022	0.597935	0.633252	0.59574
EE	0.349242	0.340781	0.406374	0.344528
NSCDIFF	0.779452	0.881948	0.807094	0.766128
Ca	0.54275	0.550345	0.623418	0.532341
P	-0.10625	0.159302	0.151051	0.016145
Mg	0.222364	0.475621	0.542413	0.258693
K	0.726068	0.759205	0.814393	0.70156
Na	-1.79949	-0.17461	0.076582	-0.14373
Fe	0.330738	-0.3896	-0.41423	0.818094
Zn	0.530767	0.460198	0.439838	0.53211
Cu	0.475096	0.388224	0.538291	0.301053
Mn	0.06892	0.330646	0.242254	0.019659
Mo	0.190773	0.04563	0.076582	0.237513
S	0.669505	0.724701	0.740289	0.639937
Ash	0.405922	0.471832	0.502473	0.454994

Trial 2

NUTRIENT	94TF	100BG	101BG	102TF
DM IN (kg)	7.15±.58	8.85±.6	4.68±.39	10.20±1.42
DM OUT (kg)	3.33±.18	3.84±.42	2.62±.09	4.07±.58
DM	0.534266	0.566102	0.440171	0.6009804
PROTEIN	0.673551	0.696088	0.598435	0.6904801
ADF	0.475649	0.491043	0.356595	0.5507605
NDF	0.518281	0.559214	0.454712	0.587908
EE	0.289142	0.323629	0.077929	0.4329721
NSCDIFF	0.734416	0.68081	0.388692	0.6768271
Ca	0.615656	0.49256	0.506591	0.6900819
P	-0.20189	0.102279	-0.21618	0.1762176
Mg	0.21921	0.276836	0.253561	0.5070934
K	0.699269	0.543603	0.523109	0.7127059
Na	-4.51119	-1.74802	-9.26353	-0.330065
Fe	-0.0647	0.46781	0.24366	0.4312708
Zn	0.471329	0.454528	0.31221	0.5039216
Cu	0.441119	0.392542	0.328205	0.3615686
Mn	0.228481	0.081582	0.057621	0.2906318
Mo	0.042657	0.310867	0.143791	0.3349673
S	0.646042	0.566102	0.471273	0.7127059
Ash	0.327714	0.376472	0.259367	0.5512093

Appendix F. C27 digestibilities for individual horses

Chapter 3

Trial 1

Nutrient	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
DM	0.520548	0.529716	0.561657	0.573037	0.619007	0.596411	0.576744	0.614132
PROTEIN	0.693068	0.686667	0.716505	0.692465	0.73826	0.711269	0.715262	0.739102
ADF	0.435464	0.440907	0.475363	0.376136	0.456951	0.390065	0.487519	0.457344
NDF	0.482871	0.518308	0.512607	0.546468	0.57979	0.523829	0.525791	0.507569
EE	0.070176	0.032353	0.111118	0.452906	0.464219	0.229875	0.091794	0.336563
NSCDIFF	0.819925	0.734962	0.89791	0.731326	0.850169	0.845663	0.936391	0.98482
Ca	0.245575	0.28325	0.4087	0.387348	0.464391	0.396928	0.611898	0.54687
P	-0.16932	-0.21647	-0.08212	0.007514	0.217405	0.039135	-0.14459	0.207135
Mg	0.1857	0.06	0.343	0.321824	0.450912	0.512824	0.1963	0.455059
K	0.635873	0.651222	0.548127	0.799059	0.770609	0.776332	0.666959	0.803661
Na	-57.7573	-74.67	-22.652	-16.0231	-3.6228	-7.69947	-15.356	-8.7272
Fe	-0.62293	-0.6955	0.051961	-1.17181	-1.65058	-0.24334	-0.14563	-1.04314
Zn	-0.58215	-0.62364	-0.12818	-0.75355	0.01448	-0.2928	0.025818	-0.39989
Cu	0.521	0.578621	0.612538	0.3595	0.52375	0.511833	0.723423	0.485333
Mn	-0.64102	-0.6537	-0.10311	-0.73511	-0.26395	-0.2633	-0.04967	-0.32343
Mo	-0.09106	-0.20111	0.197	0.10887	0.088913	0.279826	0.0835	0.272538
S	0.635048	0.641905	0.687143	0.671538	0.706923	0.658154	0.677714	0.688231
Ash	0.150263	0.166229	0.251774	0.230044	0.22026	0.422002	0.288515	0.344821

Trial 2

Nutrient	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
DM	0.910101	0.91568	0.893608	0.941001	0.81944	0.902745	0.944372	0.85275
PROTEIN%	0.941163	0.946947	0.930704	0.963722	0.882848	0.935333	0.963391	0.914369
ADF%	0.872597	0.882727	0.848571	0.92625	0.772755	0.879016	0.917273	0.810192
NDF%	0.899515	0.911922	0.884944	0.936936	0.809237	0.894709	0.936606	0.839459
EE%	0.727188	0.687625	0.675375	0.795235	0.414412	0.671912	0.8075	0.494147
NSCDIFF%	0.980732	0.973499	0.954614	0.982256	0.951008	0.977391	0.997634	0.971263
Ca%	0.85913	0.886783	0.852522	0.918875	0.73755	0.866625	0.922899	0.8236
P%	0.770685	0.753753	0.697973	0.869853	0.590088	0.768912	0.87726	0.684382
Mg%	0.843824	0.861647	0.828529	0.9056	0.7466	0.87875	0.912706	0.7795
K%	0.954533	0.957564	0.932512	0.961023	0.854217	0.923629	0.974422	0.909538
Na%	-0.488	-0.8032	0.2792	-2.69733	-3.46467	-2.977	0.667733	-6.742
Fe,ppm	0.525948	0.52569	0.610724	0.876566	0.335143	0.879175	0.61669	0.572862
Zn, ppm	0.7792	0.79392	0.72016	0.880212	0.605091	0.808939	0.867093	0.688182
Cu, ppm	0.93625	0.93	0.916083	0.977308	0.923423	0.962692	0.958	0.949115
Mn, ppm	0.742857	0.744	0.707238	0.883093	0.550852	0.816778	0.831111	0.686944
Mo,ppm	0.761304	0.758957	0.75113	0.868889	0.587722	0.870667	0.844174	0.7305
S%	0.930769	0.941846	0.914385	0.959707	0.867561	0.929024	0.959077	0.89961
Ash%	0.941163	0.946947	0.930704	0.963722	0.882848	0.935333	0.963391	0.914369

Appendix F (cont'd.). C27 digestibilities for individual horses

Chapter 4

Trial 1

NUTRIENT	93TF	94OG	98TF	99OG	100OG	101TF	102TF	103OG
DM	0.5573	0.53462	0.565351	0.468223	0.514362	0.542356	0.60147	0.484277
PROTEIN	0.731409	0.683542	0.695157	0.604694	0.611875	0.671827	0.754945	0.595083
ADF	0.473709	0.459	0.485909	0.407363	0.357226	0.497525	0.545767	0.398555
NDF	0.498025	0.510369	0.545472	0.468	0.453152	0.499461	0.549735	0.445072
EE	-0.329	-0.68563	-0.49143	-0.729	-0.458	-0.374	0.145	-0.2255
NSCDIFF	0.734783	0.551259	0.633684	0.449399	0.758699	0.659513	0.740125	0.613902
Ca	0.418563	0.3676	0.415469	0.162987	0.23536	0.491906	0.513719	0.2088
P	0.165115	-0.06286	0.096538	-0.178	0.028	0.101615	0.155962	-0.12414
Mg	0.277211	0.179412	0.358947	0.029882	0.285294	0.373263	0.37	0.119765
K	0.773942	0.765897	0.817407	0.732166	0.765379	0.800214	0.791469	0.756234
Na	0.460696	-0.35923	-0.04022	0.140615	0.252308	0.34287	0.149957	-0.42892
Fe	0.43404	0.29642	0.362451	0.26792	0.420795	0.536654	0.544074	0.225679
Zn	0.524585	0.368929	0.501341	0.366667	0.444571	0.564341	0.581537	0.324286
Cu	0.154273	0.225	0.327727	0.024667	0.19	0.458727	0.455909	-0.075
Mn	0.212444	-0.1625	0.170278	-0.14981	0.161258	0.236667	0.231556	-0.15684
Mo	-0.62433	-0.22591	-0.6675	-0.20909	-0.01618	-0.52667	-0.2635	-0.12582
S	0.640063	0.589706	0.619375	0.499294	0.514	0.627875	0.70075	0.514353
Ash	0.488747	0.542463	0.462573	0.403635	0.4564	0.536674	0.533727	0.405644

Trial 2

NUTRIENT	93OG	94TF	98OG	99TF	100TF	101OG	102OG	103TF
DM	0.521671	0.420436	0.489098	0.389022	0.33841	0.489782	0.561095	0.320748
PROTEIN%	0.638993	0.595596	0.628364	0.568376	0.495908	0.586294	0.653098	0.51411
ADF%	0.478545	0.387578	0.415095	0.284004	0.207793	0.428884	0.515649	0.19729
NDF%	0.490133	0.388348	0.464667	0.334028	0.274534	0.430905	0.546367	0.244888
EE%	-0.11533	0.378571	-0.19233	0.301714	0.338	-0.19	-0.26822	0.1755
NSCDIFF%	0.657603	0.50816	0.583227	0.579632	0.597504	0.735957	0.648177	0.581736
Ca%	0.205676	0.287719	0.203441	0.206772	0.082491	0.34	0.238206	0.261439
P%	-0.01768	-0.1832	-0.10442	-0.0998	-0.1916	-0.02	-0.00545	-0.19504
Mg%	0.044	-0.09556	0.114267	-0.26273	0.007	0.184	0.122	-0.05622
K%	0.711934	0.691941	0.659897	0.715762	0.648388	0.665629	0.761603	0.721436
Na%	-0.63886	-0.89467	-0.533	0.103867	-0.01507	-0.67571	-2.26114	-1.4444
Fe,ppm	-0.28602	-0.05869	-0.69496	-0.24897	-0.27063	-0.14656	-0.23502	-0.05434
Zn, ppm	0.136516	0.256875	-0.03848	0.293531	0.069062	0.07871	0.093677	0.193688
Cu, ppm	-0.16844	-0.09556	-0.24911	-0.08622	-0.47111	-0.13333	-0.07311	-0.13167
Mn, ppm	-0.16939	0.134203	-0.25013	-0.07146	-0.02658	0.034643	-0.1367	0.094667
Mo,ppm	-0.1472	-0.16	-0.4819	0.0835	-0.27671	-0.122	-0.2292	0.127
S%	0.522	0.52875	0.489	0.503563	0.5035	0.49	0.533563	0.448313
Ash%	0.396403	0.316154	0.341696	0.323362	0.252836	0.425599	0.457409	0.320099

Appendix F (cont'd.). C27 digestibilities for individual horses

Chapter 5

Trial 1

Nutrient	93BG P	94BG S	98BG P	99TF P	100TF S	101TF S	102BG S	103TF P
DM	0.812474	0.581801	0.770858	0.856601	0.45357	0.531161	0.508462	0.925282
PROTEIN	0.788764	0.715071	0.797588	0.83256	0.638647	0.693018	0.672	0.927455
ADF	0.797241	0.464185	0.709403	0.84836	0.315985	0.410136	0.378193	0.920469
NDF	0.838628	0.573588	0.790142	0.870635	0.401939	0.503281	0.478297	0.932033
EE	0.228211	0.222667	0.10007	0.276245	0.019429	0.196	0.154105	0.629592
NSCDIFF	0.933647	0.736555	0.936496	0.976493	0.825479	0.738731	0.698185	0.982192
Ca	0.7368	0.453813	0.68856	0.826613	0.33115	0.489963	0.39648	0.895938
P	0.478452	-0.32142	0.342548	0.626355	-0.25052	-0.14981	-0.26968	0.801613
Mg	0.582222	0.071111	0.554722	0.795714	0.22	0.38025	0.043333	0.884821
K	0.841774	0.672787	0.794833	0.885028	0.641824	0.748616	0.61486	0.9547
Na	-2.82267	-2.344	-0.4885	-1.29753	-0.7472	-0.25067	-0.476	0.515
Fe	0.274911	0.200565	-0.09037	-0.26176	-1.067	-0.91541	0.765249	0.029412
Zn	0.671	0.4395	0.573227	0.667735	0.197059	0.241324	0.396182	0.797059
Cu	0.310667	0.373	0.274833	0.451833	0.09	0.374667	0.098	0.7625
Mn	0.409143	-0.11218	0.120804	0.751012	0.004353	-0.02628	-0.26514	0.828824
Mo	0.40075	0.033375	0.212813	0.5424	-0.4196	-0.25067	0.016	0.74
S	0.759778	0.605222	0.771	0.857	0.5905	0.64825	0.535333	0.93125
Ash	0.645603	0.290372	0.542313	0.801532	0.214363	0.326155	0.296662	0.881084

Trial 2

NUTRIENT	93TF P	94TF S	98TF P	99BG P	100BG S	101BG S	102TF S	103BG P
DM	0.877444	0.509672	0.958964	0.919662	0.525782	0.504854	0.557663	0.936366
PROTEIN%	0.865505	0.656542	0.959766	0.943966	0.66751	0.644937	0.65714	0.93627
ADF%	0.874185	0.44833	0.955247	0.911848	0.444005	0.431102	0.502371	0.933803
NDF%	0.891583	0.493183	0.961175	0.930159	0.518476	0.517857	0.54352	0.944242
EE%	0.598632	0.252105	0.852184	0.677647	0.261118	0.184706	0.371895	0.762824
NSCDIFF%	0.929351	0.720579	0.989157	0.962299	0.65131	0.459483	0.642017	0.96469
Ca%	0.861476	0.595631	0.959	0.887458	0.445661	0.563729	0.656699	0.906712
P%	0.773839	-0.26452	0.920645	0.842759	0.01931	-0.07534	0.087484	0.872
Mg%	0.819118	0.178529	0.946941	0.835556	0.21	0.34	0.454	0.843556
K%	0.908629	0.6836	0.963217	0.921778	0.501422	0.578333	0.68176	0.937422
Na%	-0.025	-4.79833	0.685667	-2.06667	-2.002	-8.075	-0.47333	0.061333
Fe,ppm	0.338291	-0.12018	0.807975	0.813204	0.418624	0.331245	0.37001	0.804735
Zn, ppm	0.800541	0.443784	0.932405	0.858286	0.404114	0.391366	0.450486	0.904914
Cu, ppm	0.6064	0.412	0.8606	0.792	0.3364	0.406	0.2928	0.8208
Mn, ppm	0.788788	0.188283	0.932495	0.804	-0.0033	0.16675	0.214222	0.837867
Mo,ppm	0.678833	-0.00722	0.888389	0.821176	0.247176	0.242941	0.263333	0.853176
S%	0.88684	0.6276	0.96556	0.906667	0.526	0.5325	0.68176	0.928889
Ash%	0.82245	0.292687	0.947373	0.879585	0.318844	0.345133	0.502868	0.897126

Appendix G. C29 digestibilities for individual horses

Chapter 3

Trial 1

Nutrient	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
DM	0.659334	0.643262	0.67262	0.596673	0.608352	0.64249	0.680106	0.610728
PROTEIN	0.781495	0.762	0.78835	0.709751	0.730704	0.744144	0.784595	0.737075
ADF	0.598107	0.575327	0.608319	0.411201	0.441273	0.459513	0.612308	0.453127
NDF	0.631856	0.634119	0.636124	0.571959	0.567658	0.578047	0.64126	0.503742
EE	0.338059	0.265	0.336382	0.483656	0.44875	0.317563	0.312941	0.331406
NSCDIFF	0.871805	0.798684	0.923782	0.746427	0.845843	0.863236	0.95188	0.984702
Ca	0.462925	0.455575	0.55855	0.421783	0.448928	0.465594	0.7064	0.543348
P	0.167559	0.076	0.192118	0.063297	0.194811	0.148541	0.134118	0.200973
Mg	0.4203	0.286	0.5095	0.359941	0.435059	0.568294	0.392	0.450824
K	0.740778	0.735077	0.662643	0.810353	0.763986	0.801799	0.748054	0.802135
Na	-40.8293	-56.477	-16.658	-15.0663	-3.75627	-6.70893	-11.3733	-8.8028
Fe	-0.15536	-0.28786	0.292217	-1.04974	-1.7271	-0.10178	0.133333	-1.05902
Zn	-0.12633	-0.23327	0.157727	-0.65499	-0.01397	-0.1456	0.26303	-0.41077
Cu	0.659	0.679931	0.710731	0.3955	0.51	0.567417	0.790769	0.481333
Mn	-0.16824	-0.25611	0.176444	-0.63759	-0.30044	-0.11946	0.205926	-0.33371
Mo	0.223278	0.087667	0.4005	0.158957	0.062609	0.361826	0.306667	0.266885
S	0.74019	0.728	0.766429	0.69	0.698462	0.697077	0.75619	0.685808
Ash	0.395072	0.366689	0.441393	0.273321	0.197748	0.487814	0.461761	0.339729

Trial 2

Nutrient	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
DM	0.480317	0.141087	-0.38402	0.274483	0.187945	0.553427	0.229421	0.055325
PROTEIN	0.660055	0.457474	0.097839	0.746052	0.72945	0.752	0.764	0.743107
ADF	0.263896	-0.19925	-0.97143	0.48375	0.475203	0.536022	0.466705	0.430577
NDF	0.419417	0.099301	0.659173	0.558553	0.559452	0.596202	0.591334	0.518377
EE	-0.57625	-2.19441	0.038375	-0.43335	-0.35235	-0.25824	-0.24094	-0.51756
NSCDIFF	0.888676	0.728992	0.865555	0.875789	0.886857	0.913293	0.984746	0.913789
Ca	0.186087	-0.15778	0.56313	0.432125	0.3939	0.4885	0.502971	0.4708
P	-0.32493	-1.51816	0.105315	0.088971	0.053353	0.113765	0.208767	0.053147
Mg	0.097647	-0.41482	0.492059	0.3392	0.4148	0.535	0.437265	0.3385
K	0.737301	0.566042	0.800083	0.727158	0.66333	0.707113	0.835114	0.728615
Na	-7.59733	-17.4399	-1.1352	-24.8813	-9.31067	-14.252	-1.14193	-22.226
Fe	-1.73897	-3.85039	-0.15314	0.135961	-0.53542	0.536632	-1.47098	-0.28141
Zn	-0.27573	-1.10741	0.17104	0.161485	0.088	0.267273	0.143227	0.064545
Cu	0.631667	0.284167	0.751417	0.841154	0.823154	0.856923	0.72925	0.847346
Mn	-0.48571	-1.6179	0.132762	0.181648	-0.03726	0.297333	-0.08873	0.060833
Mo	-0.37913	-1.46496	0.262783	0.082222	0.047889	0.504	-0.00452	0.1915
S	0.6	0.405308	0.746385	0.717951	0.694146	0.727805	0.736192	0.698829
Ash	0.660055	0.457474	0.794726	0.746052	0.72945	0.752	0.764	0.743107

Appendix G (cont'd.). C29 digestibilities for individual horses

Chapter 4

Trial 1

NUTRIENT	93TF	94OG	98TF	99OG	100OG	101TF	102TF	103OG
DM	0.509768	0.569656	0.572956	0.55028	0.585667	0.499291	0.570721	0.560896
PROTEIN	0.702913	0.707361	0.700764	0.665625	0.669375	0.641016	0.73652	0.655507
ADF	0.417872	0.49972	0.495364	0.49871	0.452452	0.450349	0.511614	0.488305
NDF	0.444768	0.547223	0.553831	0.55	0.534167	0.452467	0.515881	0.527881
EE	-0.47	-0.55875	-0.464	-0.4625	-0.242	-0.503	0.080714	-0.04262
NSCDIFF	0.706645	0.585035	0.640421	0.534266	0.794448	0.627546	0.720586	0.671517
Ca	0.356875	0.4152	0.426219	0.292	0.34864	0.444203	0.477156	0.326867
P	0.076538	0.017143	0.113154	0.003571	0.172	0.017269	0.0925	0.043607
Mg	0.200526	0.241176	0.370737	0.179412	0.391176	0.314421	0.322632	0.251118
K	0.749959	0.783517	0.820765	0.773448	0.800138	0.781457	0.77579	0.79261
Na	0.403478	-0.25692	-0.02109	0.273077	0.363077	0.281174	0.086043	-0.21569
Fe	0.373995	0.349377	0.374176	0.38076	0.506603	0.493152	0.509794	0.341227
Zn	0.474146	0.416429	0.510512	0.464286	0.526857	0.523439	0.550073	0.425119
Cu	0.064545	0.283333	0.340091	0.175	0.31	0.407909	0.415	0.085417
Mn	0.128889	-0.075	0.185537	0.027419	0.285516	0.165	0.173778	0.01579
Mo	-0.79667	-0.13364	-0.63683	-0.02273	0.134364	-0.67	-0.3585	0.042182
S	0.601875	0.620588	0.626375	0.576471	0.586	0.592938	0.67825	0.586824
Ash	0.434506	0.576901	0.472456	0.495556	0.536933	0.493174	0.498669	0.494337

Trial 2

NUTRIENT	93OG	94TF	98OG	99TF	100TF	101OG	102OG	103TF
DM	0.589108	0.458404	0.558851	0.421481	0.386671	0.578116	0.592117	0.364269
PROTEIN	0.689594	0.622092	0.679273	0.590982	0.53322	0.657678	0.677594	0.544881
ADF	0.551636	0.427702	0.495219	0.321503	0.266431	0.52743	0.549851	0.248124
NDF	0.5616	0.428422	0.538	0.368907	0.328232	0.529102	0.5784	0.292708
EE	0.041	0.419286	-0.029	0.338286	0.387	0.015333	-0.17867	0.227714
NSCDIFF	0.705596	0.540384	0.640319	0.601648	0.627296	0.781518	0.673021	0.608224
Ca	0.317015	0.334386	0.312559	0.248316	0.150404	0.453882	0.292	0.308211
P	0.124968	-0.10568	0.046871	-0.0422	-0.1034	0.156	0.065548	-0.11936
Mg	0.178	-0.02378	0.2356	-0.1966	0.0805	0.3248	0.184	0.010667
K	0.752311	0.712125	0.706487	0.730648	0.674414	0.723325	0.778437	0.739077
Na	-0.40914	-0.77053	-0.323	0.1508	0.060067	-0.38657	-2.03086	-1.2896
Fe	-0.10576	0.010668	-0.46278	-0.18356	-0.17658	0.051278	-0.14781	0.01243
Zn	0.257548	0.305563	0.103774	0.330531	0.137969	0.237677	0.157677	0.24475
Cu	-0.00467	-0.02378	-0.078	-0.02933	-0.36222	0.062222	0.002667	-0.06
Mn	-0.00548	0.190928	-0.07888	-0.01535	0.049406	0.201214	-0.05643	0.152
Mo	0.0136	-0.084	-0.2789	0.1315	-0.18221	0.0716	-0.1424	0.182286
S	0.589	0.559625	0.559	0.529563	0.54025	0.578	0.5665	0.48325
Ash	0.481008	0.360958	0.431875	0.3588	0.308139	0.524712	0.495724	0.363156

Appendix G (cont'd.). C29 digestibilities for individual horses

Chapter 5

Trial 1

Nutrient	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
DM	0.630084	0.499147	0.576858	0.712798	0.465326	0.547291	0.460394	0.723521
PROTEIN	0.58427	0.658494	0.626112	0.663949	0.645927	0.703491	0.64	0.733033
ADF	0.600952	0.357792	0.46322	0.69566	0.329766	0.430259	0.317529	0.707324
NDF	0.682407	0.488918	0.612359	0.740366	0.413987	0.520227	0.427399	0.749882
EE	-0.51895	0.068316	-0.66232	-0.45257	0.039184	0.223429	0.071579	-0.3631
NSCDIFF	0.869412	0.684244	0.882697	0.952822	0.828995	0.747644	0.668739	0.934466
Ca	0.482	0.34536	0.42472	0.652013	0.344625	0.507363	0.3376	0.61705
P	-0.02645	-0.58381	-0.21442	0.250097	-0.22532	-0.11058	-0.39355	0.269935
Mg	0.177778	-0.11333	0.1775	0.59	0.235714	0.401393	-0.05	0.576143
K	0.688597	0.607814	0.621023	0.769252	0.64904	0.757192	0.577285	0.833296
Na	-6.52333	-3.008	-1.7495	-3.61113	-0.712	-0.208	-0.62	-0.7848
Fe	-0.42704	0.041825	-1.01409	-1.53235	-1.02536	-0.85007	0.742346	-2.57176
Zn	0.3525	0.328205	0.211682	0.333147	0.213235	0.267206	0.337273	0.253176
Cu	-0.35667	0.2485	-0.3395	-0.10017	0.108333	0.396	0.01	0.126
Mn	-0.16286	-0.33302	-0.62402	0.500282	0.024412	0.008729	-0.38857	0.370071
Mo	-0.17938	-0.15856	-0.45406	0.0816	-0.391	-0.208	-0.08	0.0432
S	0.527222	0.526833	0.577	0.713	0.59875	0.66025	0.49	0.747
Ash	0.302517	0.149465	0.154579	0.601677	0.230191	0.349143	0.228044	0.562389

Trial 2

NUTRIENT	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
DM	0.703451	0.451182	0.738437	0.513075	0.343236	0.434756	0.521322	0.537313
PROTEIN	0.675243	0.615187	0.742897	0.658895	0.539823	0.644937	0.628439	0.538954
ADF	0.696204	0.381904	0.714018	0.463377	0.229348	0.350652	0.460714	0.521108
NDF	0.738214	0.432158	0.751897	0.574841	0.332571	0.449675	0.505307	0.599134
EE	0.030842	0.162053	0.055421	-0.96232	-0.02415	0.069412	0.319316	-0.71582
NSCDIFF	0.829409	0.686934	0.930711	0.770494	0.51669	0.383046	0.61205	0.744552
Ca	0.665515	0.546942	0.738	0.314898	0.231644	0.502034	0.627961	0.325119
P	0.453903	-0.41677	0.492903	0.042793	-0.35931	-0.22741	0.011097	0.074
Mg	0.563235	0.079618	0.660941	-0.00106	-0.095	0.246667	0.408294	-0.13178
K	0.779371	0.645503	0.764949	0.523822	0.308933	0.518704	0.65512	0.547289
Na	-1.475	-5.4965	-1.00867	-17.6683	-3.161	-9.35833	-0.59667	-5.79067
Fe	-0.59778	-0.25506	-0.22709	-0.12308	0.194169	0.236673	0.317273	-0.41262
Zn	0.518378	0.376811	0.568054	0.137314	0.174057	0.305857	0.404486	0.312114
Cu	0.0496	0.3412	0.1092	-0.2662	0.0802	0.322	0.2336	-0.2964
Mn	0.49	0.090545	0.568626	-0.19315	-0.39065	0.048917	0.148444	-0.17293
Mo	0.2245	-0.1285	0.286778	-0.08859	-0.04347	0.135882	0.201667	-0.06218
S	0.72676	0.58276	0.77992	0.431833	0.343	0.466389	0.65512	0.485556
Ash	0.571281	0.20752	0.663701	0.266975	0.055867	0.252526	0.461253	0.25577

Appendix H. C31 digestibilities for individual horses

Chapter 3

Trial 1

Nutrient	93BG	94BG	98BG	99TF	100TF	101TF	102BG	103TF
DM	0.795544	0.793956	0.817772	0.560146	0.565701	0.634373	0.804338	0.580612
PROTEIN	0.869282	0.862667	0.882201	0.683102	0.70185	0.739141	0.868065	0.716798
ADF	0.759571	0.754951	0.782	0.357143	0.381409	0.448945	0.762538	0.410951
NDF	0.779761	0.788876	0.797476	0.53266	0.521336	0.569796	0.780272	0.46547
EE	0.604	0.575882	0.630647	0.43625	0.389688	0.304219	0.579176	0.279844
NSCDIFF	0.923308	0.883835	0.957579	0.723146	0.829326	0.860562	0.970526	0.983522
Ca	0.6787	0.68585	0.7543	0.368696	0.389884	0.455145	0.82017	0.50813
P	0.502	0.466824	0.550353	-0.0227	0.108541	0.131892	0.469647	0.139351
Mg	0.6532	0.588	0.727	0.301176	0.374529	0.559853	0.6276	0.408471
K	0.844923	0.847131	0.812235	0.792941	0.738699	0.797924	0.845683	0.786875
Na	-24.024	-32.166	-8.828	-16.5413	-4.26587	-6.85967	-6.57867	-9.5588
Fe	0.308816	0.256864	0.606066	-1.23793	-2.01929	-0.12332	0.469167	-1.21781
Zn	0.326182	0.288364	0.531212	-0.80693	-0.12261	-0.168	0.548606	-0.51957
Cu	0.796	0.81531	0.839	0.34	0.4575	0.558958	0.871846	0.441333
Mn	0.301111	0.275185	0.54163	-0.78794	-0.43978	-0.14135	0.51363	-0.43657
Mo	0.535333	0.473556	0.666333	0.081739	-0.03783	0.349348	0.575333	0.210346
S	0.844571	0.843048	0.87	0.661538	0.666154	0.691154	0.850667	0.661577
Ash	0.638108	0.63456	0.689093	0.206603	0.111793	0.477799	0.670329	0.288808

Trial 2

Nutrient	93TF	94TF	98TF	99BG	100BG	101BG	102TF	103BG
DM	0.634014	0.664051	0.686084	0.587062	0.581542	0.628429	0.638691	0.558725
PROTEIN	0.760731	0.787789	0.794726	0.746052	0.72945	0.752	0.764	0.743107
ADF	0.481896	0.530909	0.551429	0.48375	0.475203	0.536022	0.466705	0.430577
NDF	0.591359	0.647689	0.659173	0.558553	0.559452	0.596202	0.591334	0.518377
EE	-0.10944	-0.2495	0.038375	-0.43335	-0.35235	-0.25824	-0.24094	-0.51756
NSCDIFF	0.921645	0.893994	0.865555	0.875789	0.886857	0.913293	0.984746	0.913789
Ca	0.42713	0.54713	0.56313	0.432125	0.3939	0.4885	0.502971	0.4708
P	0.067452	0.015014	0.105315	0.088971	0.053353	0.113765	0.208767	0.053147
Mg	0.364882	0.446588	0.492059	0.3392	0.4148	0.535	0.437265	0.3385
K	0.8151	0.830256	0.800083	0.727158	0.66333	0.707113	0.835114	0.728615
Na	-5.0512	-6.2128	-1.1352	-24.8813	-9.31067	-14.252	-1.14193	-22.226
Fe	-0.92781	-0.89724	-0.15314	0.135961	-0.53542	0.536632	-1.47098	-0.28141
Zn	0.10208	0.17568	0.17104	0.161485	0.088	0.267273	0.143227	0.064545
Cu	0.74075	0.72	0.751417	0.841154	0.823154	0.856923	0.72925	0.847346
Mn	-0.04571	-0.024	0.132762	0.181648	-0.03726	0.297333	-0.08873	0.060833
Mo	0.029304	0.035826	0.262783	0.082222	0.047889	0.504	-0.00452	0.1915
S	0.718462	0.767385	0.746385	0.717951	0.694146	0.727805	0.736192	0.698829
Ash	0.760731	0.787789	0.794726	0.746052	0.72945	0.752	0.764	0.743107

Appendix H (cont'd.). C31 digestibilities for individual horses

Chapter 4

Trial 1

NUTRIENT	93TF	94OG	98TF	99OG	100OG	101TF	102TF	103OG
DM	0.482003	0.527166	0.547868	0.490416	0.536742	0.502594	0.518714	0.529058
PROTEIN	0.685937	0.678097	0.683244	0.621042	0.630243	0.643882	0.704583	0.630396
ADF	0.384607	0.449692	0.465818	0.431871	0.387645	0.454738	0.452415	0.451006
NDF	0.41304	0.501945	0.527709	0.49	0.479032	0.456839	0.4572	0.493467
EE	-0.554	-0.71463	-0.54971	-0.6575	-0.389	-0.491	-0.03071	-0.11863
NSCDIFF	0.689882	0.543538	0.619368	0.472168	0.770119	0.63052	0.686717	0.647573
Ca	0.320125	0.35672	0.392625	0.1976	0.271547	0.448641	0.413781	0.2778
P	0.023769	-0.08114	0.061231	-0.12929	0.074	0.025115	-0.0175	-0.02611
Mg	0.154842	0.165294	0.333895	0.07	0.319118	0.319895	0.240526	0.196529
K	0.735671	0.761869	0.810272	0.743241	0.776483	0.783202	0.748613	0.777493
Na	0.369391	-0.38262	-0.08087	0.176154	0.287692	0.286913	-0.02474	-0.30431
Fe	0.338223	0.284315	0.337536	0.298194	0.448205	0.497198	0.450375	0.293207
Zn	0.444098	0.358071	0.481854	0.392857	0.470857	0.527244	0.495537	0.383214
Cu	0.011091	0.211667	0.301455	0.065	0.228333	0.412636	0.344091	0.01875
Mn	0.079111	-0.1825	0.137852	-0.10226	0.200952	0.171667	0.07363	-0.05595
Mo	-0.89933	-0.247	-0.73267	-0.15909	0.031909	-0.65667	-0.52317	-0.02764
S	0.579125	0.582647	0.6045	0.52	0.537	0.596188	0.63925	0.556706
Ash	0.402192	0.534591	0.44157	0.428296	0.482126	0.497221	0.437901	0.457478

Trial 2

NUTRIENT	93OG	94TF	98OG	99TF	100TF	101OG	102OG	103TF
DM	0.537145	0.470194	0.511481	0.441153	0.399379	0.540547	0.528654	0.409614
PROTEIN%	0.650322	0.630459	0.644364	0.605039	0.542358	0.627664	0.627811	0.577798
ADF%	0.494909	0.440373	0.440277	0.34494	0.280791	0.485996	0.480343	0.302505
NDF%	0.506133	0.441077	0.487714	0.390706	0.341382	0.487814	0.5133	0.343864
EE%	-0.08033	0.432143	-0.141	0.361143	0.399	-0.071	-0.36067	0.283571
NSCDIFF%	0.668348	0.55056	0.60117	0.615408	0.634592	0.762362	0.622532	0.63656
Ca%	0.230603	0.349123	0.237735	0.274281	0.167035	0.406	0.182676	0.358246
P%	0.014258	-0.0812	-0.05687	-0.0062	-0.0818	0.082	-0.07874	-0.0384
Mg%	0.074	-0.00111	0.1524	-0.15527	0.0985	0.2656	0.058	0.082222
K%	0.720974	0.718498	0.67454	0.739952	0.680788	0.699066	0.744225	0.757949
Na%	-0.58743	-0.73133	-0.467	0.180133	0.078467	-0.50814	-2.49886	-1.124
Fe,ppm	-0.24566	0.032572	-0.62199	-0.14268	-0.15355	-0.03213	-0.32505	0.083858
Zn, ppm	0.163613	0.320938	0.006226	0.353656	0.154844	0.170839	0.027613	0.299375
Cu, ppm	-0.13178	-0.00111	-0.19533	0.006222	-0.33556	-0.02	-0.15133	0.016667
Mn, ppm	-0.1327	0.208841	-0.1963	0.019725	0.068014	0.131179	-0.22188	0.213333
Mo,ppm	-0.1112	-0.06	-0.4181	0.1615	-0.15907	-0.0098	-0.3188	0.241429
S%	0.537	0.569375	0.511	0.545813	0.54925	0.541	0.499563	0.520625
Ash%	0.415344	0.375106	0.370038	0.380948	0.321683	0.48304	0.416746	0.409218

Appendix H (cont'd.). C31 digestibilities for individual horses

Chapter 5

Trial 1

Nutrient	93BG P	94BG S	98BG P	99TF P	100TF S	101TF S	102BG S	103TF P
DM	0.533562	0.511357	0.521874	0.614591	0.453042	0.534519	0.489409	0.635006
PROTEIN%	0.476404	0.667356	0.577498	0.5492	0.637985	0.695636	0.659333	0.646945
ADF%	0.497416	0.374456	0.393426	0.591739	0.314732	0.415166	0.35418	0.612947
NDF%	0.600005	0.50218	0.561957	0.65171	0.400843	0.507518	0.45815	0.669228
EE%	-0.91305	0.092491	-0.87846	-0.94857	0.017633	0.202857	0.121439	-0.80265
NSCDIFF%	0.835529	0.692437	0.867445	0.936712	0.82516	0.740959	0.686529	0.913333
Ca%	0.3476	0.362347	0.34992	0.533188	0.329925	0.494313	0.373173	0.493563
P%	-0.29277	-0.54271	-0.37232	-0.00597	-0.25281	-0.14	-0.31871	0.034516
Mg%	-0.03556	-0.08444	0.070556	0.45	0.218571	0.385536	0.006389	0.439464
K%	0.607801	0.617991	0.571747	0.69046	0.641168	0.75076	0.599986	0.77954
Na%	-8.47533	-2.904	-2.107	-5.18567	-0.7504	-0.24	-0.533	-1.36033
Fe,ppm	-0.7973	0.066688	-1.27596	-2.39706	-1.07079	-0.89908	0.756183	-3.72353
Zn, ppm	0.1845	0.345636	0.109182	0.105441	0.195588	0.247794	0.372864	0.012353
Cu, ppm	-0.70867	0.268	-0.51367	-0.47583	0.088333	0.38	0.063167	-0.15583
Mn, ppm	-0.46457	-0.29843	-0.83518	0.329647	0.002529	-0.01753	-0.314	0.166941
Mo,ppm	-0.48538	-0.1285	-0.64313	-0.232	-0.4222	-0.24	-0.022	-0.26533
S%	0.404556	0.539111	0.522	0.615	0.58975	0.65125	0.517389	0.665417
Ash%	0.121549	0.171535	0.044654	0.465664	0.212925	0.331902	0.269501	0.421275

Trial 2

NUTRIENT	93TF P	94TF S	98TF P	99BG P	100BG S	101BG S	102TF S	103BG P
DM	0.640186	0.487655	0.677429	0.441996	0.34745	0.391555	0.48574	0.42803
PROTEIN%	0.606355	0.641121	0.683037	0.609165	0.542134	0.563882	0.60129	0.430414
ADF%	0.631762	0.423561	0.647435	0.385142	0.23404	0.301232	0.421309	0.408366
NDF%	0.682683	0.470427	0.694133	0.512857	0.336635	0.407792	0.469161	0.504762
EE%	-0.17474	0.218526	-0.1645	-1.24841	-0.01791	-0.00141	0.269579	-1.11976
NSCDIFF%	0.793223	0.708033	0.914579	0.737034	0.519632	0.336092	0.583702	0.684414
Ca%	0.594563	0.577476	0.677	0.215017	0.236322	0.464136	0.600777	0.166237
P%	0.338065	-0.32129	0.374839	-0.09676	-0.35103	-0.32083	-0.06116	-0.144
Mg%	0.470588	0.141647	0.582	-0.147	-0.08833	0.189333	0.365059	-0.39822
K%	0.732571	0.669394	0.710223	0.4544	0.313141	0.482074	0.62992	0.440711
Na%	-2	-5.05867	-1.47633	-20.39	-3.13567	-10.1467	-0.71333	-7.38933
Fe,ppm	-0.93671	-0.17047	-0.51278	-0.28682	0.199076	0.17858	0.267387	-0.74518
Zn, ppm	0.416216	0.418811	0.467486	0.011543	0.179086	0.253029	0.360973	0.150171
Cu, ppm	-0.152	0.3856	-0.0982	-0.4508	0.0858	0.2704	0.1776	-0.6016
Mn, ppm	0.381818	0.151838	0.468192	-0.3671	-0.38218	-0.02347	0.086222	-0.44907
Mo,ppm	0.06	-0.05244	0.120722	-0.24729	-0.03775	0.070118	0.143	-0.31224
S%	0.6688	0.61088	0.72868	0.349	0.347	0.425778	0.62992	0.364444
Ash%	0.480341	0.26093	0.585403	0.160107	0.061615	0.195639	0.421887	0.080563

Appendix I. Fecal outputs for individual horses

Chapter 3

Trial 1

AVERAGE Cr/BAR (g): 5.83 AVERAGE Cr DOSE/DAY (g): 17.49

			predicted FO	
HORSE	DAY	TC	from days	from asym.
93	D5	5.02	3.4463054	
	D6	4.8	5.4014824	
	D7	4.94	3.5191147	
	D8		3.3647557	12d
AVE		4.92	3.9329145	3.7196937
stdev		0.111355	0.9810734	8d
SE		0.06	0.49	3.6017298
94	D5	4.9	3.9347582	
	D6	6.23	5.9489796	
	D7	5.1	3.1627486	
	D8		3.5693878	12 d
AVE		5.41	4.1539685	3.9022758
stdev		0.717147	1.2375203	8 d
SE		0.41	0.62	3.6728265
98	D5		3.1327243	
	D6		3.7292111	
	D7		3.7015873	
	D8		3.0846561	12 d
AVE			3.4120447	3.733191
stdev			0.3510141	8 d
SE			0.17	3.4952038
99	D5	4.48	2.3625557	
	D6	4.64	2.9568893	
	D7	4.34	2.6722689	
	D8		5.431677	12 d
AVE		4.486667	3.3558477	2.946925
stdev		0.150111	1.4050082	8 d
SE		0.09	0.7	2.4773371
100	D5	4.44	3.0748945	
	D6	4.78	3.9977143	
	D7	4.13	2.7082688	
	D8		2.7082688	12 d
AVE		4.45	3.1222866	3.4314303
stdev		0.325115	0.608671	8 d
SE		0.19	0.3	2.8832839
101	D5	3.06	2.3909774	
	D6	4.91	3.1725014	
	D7		3.3200456	
	D8		3.2239631	12 d
AVE		3.985	3.0268719	2.9912776
stdev		1.308148	0.4283166	8 d
SE		0.93	0.21	2.6743119

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 3

Trial 1 (cont'd.)

			predicted FO	
HORSE	DAY	TC	from days	from asym.
102	D5	5.17	2.3242525	
	D6	4.36	3.9347582	
	D7		3.9659864	
	D8		3.2340976	12 d
AVE		4.765	3.3647737	3.4626807
stdev		0.572756	0.7716	8 d
SE		0.41	0.39	3.3770998
103	D5		4.4617347	
	D6		4.851595	
	D7		4.4617347	
	D8		4.6269841	12 d
AVE			4.6005121	3.9922392
stdev			0.1846273	8 d
SE			0.09	3.857521

Trial 2

AVERAGE Cr/BAR (g): 5.97 AVERAGE Cr DOSE/DAY (g): 17.91

		predicted FO	
HORSE	DAY	from days	from asym.
93	D5	3.578422	
	D6	3.377334	
	D7	4.849716	
	D8	4.507929	12 d
AVE		4.07835	3.5585138
stdev		0.712015	8 d
SE		0.36	3.945803
94	D5	2.966214	
	D6	2.154457	
	D7	2.728935	
	D8	5.950166	12 d
AVE		3.449943	3.3246705
stdev		1.701298	8 d
SE		0.85	3.01059
98	D5	3.290465	
	D6	3.590617	
	D7	5.472044	
	D8	5.171816	12 d
AVE		4.381236	3.6498879
stdev		1.09996	8 d
SE		0.55	3.9121887

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 3
Trial 2 (cont'd.)

		predicted FO	
HORSE	DAY	from days	from asym.
99	D5	4.568878	
	D6	5.016807	
	D7	5.950166	
	D8	8.598176	12 d
AVE		6.033507	5.4141475
stdev		1.804004	8 d
SE		0.9	5.9030982
100	D5	4.804185	
	D6	5.591633	
	D7	9.054601	
	D8	10.44315	12 d
AVE		7.473392	6.5460526
stdev		2.707112	8 d
SE		1.35	6.6505756
101	D5	6.020168	
	D6	5.562112	
	D7	7.256888	
	D8	5.881773	12 d
AVE		6.180235	5.9285005
stdev		0.742958	8 d
SE		0.37	6.3174603
102	D5	3.735141	
	D6	3.259327	
	D7	6.733083	
	D8	8.185558	12 d
AVE		5.478277	4.473027
stdev		2.371088	8 d
SE		1.19	4.6519481
103	D5	8.977444	
	D6	5.531192	
	D7	8.977444	
	D8	9.137755	12 d
AVE		8.155959	6.7103784
stdev		1.751475	8 d
SE		0.88	7.4192212

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 4

Trial 1

AVERAGE Cr/BAR (g): 6.68   AVERAGE Cr DOSE/D (g): 20.04

			predicted FO		
HORSE	DAY	TC	from days	from diurn mix	from asym.
93	D5	3.89	5.812065		
	D6	4.8	6.010798		
	D7	5.93	5.228281		
	D8	4.28	5.453061	4.914173615	4.6153846
AVE		4.725	5.626051		
stdev		0.885607	0.351557		
SE		0.38	0.18		
94	D5	3.31	4.508436		
	D6	2.87	3.717996		
	D7	3.53	4.80115		
	D8	3.88	5.022556	4.978881988	3.9079563
AVE		3.3975	4.512535		
SE		0.21	0.28		
stdev		0.422798	0.570008		
98	D5	2.61	3.309661		
	D6	3.19	3.62387		
	D7	3.67	4.243066		
	D8	3.11	4.63567	4.455313473	3.6004312
AVE		3.145	3.953067		
stdev		0.434012	0.597882		
SE		0.22	0.3		
99	D5	4.32	6.325758		
	D6	4.56	5.871667		
	D7	5.39	6.506494		
	D8	4.62	7.6082	6.898450947	5.6883338
AVE		4.7225	6.57803		
stdev		0.463492	0.736876		
SE		0.23	0.37		
100	D5	5.02	6.506494		
	D6	4.83	6.256634		
	D7	5.3	7.004544		
	D8	4.97	8.119935	8.875110717	5.3029902
AVE		5.03	6.971902		
stdev		0.197146	0.826087		
SE		5.03	0.41		
101	D5	3.84	5.725714		
	D6	3.11	5.375536		
	D7	4.61	4.404396		
	D8	4.36	5.351135	5.558945908	4.2189474
AVE		3.98	5.214195		
stdev		0.662772	0.566336		
SE		0.33	0.29		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 4

Trial 1 (cont'd.)

HORSE	DAY	TC	predicted FO from days	from diurn mix	from asym.
102	D5	2.58	4.883041		
	D6	3.34	4.914174		
	D7	3.7	4.627107		
	D8	3.22	4.751067	4.194223524	4.2118537
AVE		3.21	4.793847		
stdev		0.466905	0.13174		
SE		0.23	0.06		
103	D5	6.24	6.774848		
	D6	4.57	7.386657		
	D7	4.75	7.386657		
	D8	4.04	7.24774	7.737451737	6.0745681
AVE		4.9	7.198975		
stdev		0.942797	0.290236		
SE		0.47	0.15		

Trial 2

AVERAGE Cr/BAR (g): 6.68 AVERAGE Cr DOSE/DAY (g): 20.04

HORSE	DAY	TC	predicted FO from days	from diurn mix	from asym.
93	D5	3.62	5.228281		
	D6	3.97	4.63567		
	D7	3.76	5.414753		
	D8	4.13	5.170279	4.017642342	4.4218888
AVE		3.87	5.112245		
stdev		0.225241	0.334399		
SE		0.11	0.17		
94	D5	4.25	8.875111		
	D6	4.47	6.561886		
	D7	4.73	6.91989		
	D8	5.27	8.0937	6.618229855	5.6498449
AVE		4.68	7.612646		
stdev		0.439545	1.066027		
SE		0.22	0.53		
98	D5	2.96	4.8523		
	D6	3.13	3.766917		
	D7	3.13	3.778993		
	D8	3.21	4.187213	3.501659969	3.6284628
AVE		3.1075	4.146356		
Stdev		0.105317	0.509561		
SE		0.05	0.26		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 4
Trial 2 (cont'd.)

HORSE	DAY	TC	predicted FO from days	from diurn mix	from asym.
99	D5	3.78	7.134211		
	D6	4.84	6.600791		
	D7	4.69	7.024185		
	D8	4.22	8.462838	6.736134454	6.0672116
AVE		4.3825	7.305506		
Stdev		0.48072	0.805093		
SE		0.24	0.4		
100	D5	5.59	7.460908		
	D6	5.6	5.753661		
	D7	5.63	5.696418		
	D8	5.02	7.61109	7.111426544	5.7142857
AVE		5.46	6.630519		
stdev		0.293825	1.047615		
SE		0.15	0.52		
101	D5	4.07	6.189006		
	D6	4.52	6.898451		
	D7	3.48	5.519141		
	D8	4.36	5.240586	3.842761266	5.2038432
AVE		4.1075	5.961796		
stdev		0.457921	0.740503		
SE		0.23	0.37		
102	D5	2.25	4.63567		
	D6	3.59	3.629777		
	D7	2.91	5.572859		
	D8	2.65	5.100534	3.907956318	3.770461
AVE		2.85	4.73471		
stdev		0.563087	0.830062		
SE		0.28	0.41		
103	D5	4.51	6.542605		
	D6	3.84	5.627633		
	D7	5.63	7.843444		
	D8	5.25	7.483196	5.783549784	5.9982041
AVE		4.8075	6.87422		
stdev		0.795168	0.99569		
SE		0.4	0.5		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 5

Trial 1

AVERAGE Cr/BAR (g): 6.09   AVERAGE Cr DOSE/day (g): 18.27

HORSE	DAY	TC	predicted FO from days	from diurn mix	from asym.
93	D5		4.209677		
	D6		3.491305		
	D7		2.957268		
	D8		2.509615	2.813799	3.1082001
AVE			3.291966		
stdev			0.731671		
SE			0.37		
94	D5	2.09	3.491305		
	D6	2.48	2.71875		
	D7	3.67	1.965996		
	D8	2.45	3.242236	3.163636	2.4282297
AVE		2.6725	2.854572		
stdev		0.688204	0.674222		
SE		0.34	0.34		
98	D5		3.999562		
	D6		3.153806		
	D7		2.479642		
	D8		2.603306	2.891738	2.7606528
AVE			3.059079		
stdev			0.692088		
SE			0.35		
99	D5		4.810427		
	D6		4.061805		
	D7		4.788991		
	D8		4.72093	5.091973	4.4173114
AVE			4.595538		
stdev			0.357862		
SE			0.18		
100	D5	3.59	5.22		
	D6	3.97	2.821622		
	D7	4.45	3.782609		
	D8	3.98	3.939198	4.558383	3.1214762
AVE		3.9975	3.940857		
stdev		0.352077	0.985552		
SE		0.18	0.49		
101	D5	2.99	3.53864		
	D6	2.61	2.783788		
	D7	3.77	2.965909		
	D8	3.39	3.562793	3.53864	2.7965713
AVE		3.19	3.212782		
stdev		0.500932	0.397355		
SE		0.25	0.2		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 5

Trial 1 (cont'd.)

HORSE	DAY	TC	from days	from diurn mix	from asym.
102	D5	3.06	4.386555		
	D6	2.84	2.509615		
	D7	4.2	3.866667		
	D8	4.26	5.272727	5.245478	3.6686747
AVE		3.59	4.008891		
stdev		0.744849	1.15585		
SE		0.37	0.58		
103	D5		4.5		
	D6		5.381443		
	D7		4.176		
	D8		4.878505	4.855169	4.6008562
AVE			4.733987		
stdev			0.51839		
SE			0.26		

Trial 2

AVERAGE Cr/BAR (g): 6.7

AVERAGE Cr DOSE/DAY (g): 20.1

			predicted FO		
HORSE	DAY	TC	from days	from diurn mix	from asym.
93	D5		5.761589		
	D6		4.282046		
	D7		4.448644		
	D8		6.179741	6.92867	5.0125251
AVE			5.168005		
stdev			0.944873		
SE			0.47		
94	D5	3	4.054711		
	D6	3.14	4.573714		
	D7	3.81	4.142857		
	D8	3.37	3.786187	3.849558	3.874153
AVE		3.33	4.139367		
stdev		0.354495	0.326891		
SE		0.18	0.16		
98	D5		3.942857		
	D6		4.298604		
	D7		4.783648		
	D8		5.036496	6.082067	4.489567
AVE			4.515401		
stdev			0.489321		
SE			0.25		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 5

Trial 2 (cont'd.)

HORSE	DAY	TC	from days	from diurn mix	from asym.
99	D5		5.015038		
	D6		4.824012		
	D7		4.992515		
	D8		7.570942	8.595361	5.3445513
AVE			5.600627		
stdev			1.316306		
SE			0.66		
100	D5	4.83	5.08126		
	D6	4.22	6.423756		
	D7	3.18	3.928922		
	D8	3.12	3.798405	3.998002	3.0153707
AVE		3.8375	4.808086		
stdev		0.832361	1.221665		
SE		0.42	0.61		
101	D5	2.87	4.397802		
	D6	2.6	3.475165		
	D7	2.58	3.041033		
	D8	2.43	2.640538	3.057296	2.1518443
AVE		2.62	3.388635		
stdev		0.183121	0.754184		
SE		0.09	0.38		
102	D5	4.76	6.888124		
	D6	2.39	4.142857		
	D7	4.96	4.483531		
	D8	4.15	4.250212	4.906817	4.678513
AVE		4.065	4.941181		
stdev		1.168603	1.30573		
SE		0.58	0.65		
103	D5		7.281659		
	D6		8.407563		
	D7		7.672546		
	D8		7.23689	8.10778	7.5
AVE			7.649665		
stdev			0.541832		
SE			0.27		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 6

Trial 1

AVERAGE Cr/BAR (g): 5.82 AVERAGE Cr DOSE/DAY (g): 17.46

HORSE	DAY	TC	predicted FO from days	from diurn mix	from asym.
93	D5	3.52	3.722814		
	D6	4.05	3.260504		
	D7	3.25	2.386876		
	D8		2.660369	2.866995	1.9134247
AVE		3.606667	3.007641		
stdev		0.406981	0.600378		
SE		0.23	0.3		
94	D5	3.64	8.827098		
	D6	4.52	3.906789		
	D7	4.99	3.750806		
	D8		5.700294	4.006425	4.831212
AVE		4.383333	5.546247		
stdev		0.685298	2.359318		
SE		0.4	1.18		
99	D5	3.3	9.238095		
	D6	3.53	5.142857		
	D7	2.97	3.270888		
	D8		2.660369	2.952816	2.8450383
AVE		3.266667	5.078052		
stdev		0.281484	2.96767		
SE		0.16	1.48		
100	D5	4.86	7.281068		
	D6	3.12	9.875566		
	D7	4.26	2.475542		
	D8		3.5889	3.627675	1.8039054
AVE		4.08	5.805269		
stdev		0.883855	3.403168		
SE		0.51	1.7		
102	D5	2.91	3.107868		
	D6	2.58	2.044496		
	D7	3.32	2.858546		
	D8		2.439229	2.37551	2.1274522
AVE		2.936667	2.612535		
stdev		0.37072	0.468536		
SE		0.21	0.24		
131	D5	4.59	7.445629		
	D6	4.7	3.668067		
	D7	4.02	3.30369		
	D8		3.197802	3.575671	2.6729945
AVE		4.436667	4.403797		
stdev		0.365011	2.037867		
SE		0.21	1.02		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 6

Trial 1 (cont'd.).

HORSE	DAY	TC	from days	from diurn mix	from asym.
132	D5	2.89	4.597156		
	D6	2.07	2.646256		
	D7	4.28	3.14708		
	D8		2.632293	2.54519	2.4354861
AVE		3.08	3.255696		
stdev		1.117184	0.925808		
SE		0.65	0.46		
133	D5	2.39	3.668067		
	D6	2.91	2.057022		
	D7	2.59	2.987169		
	D8		2.386876	2.433449	2.4661017
AVE		2.63	2.774783		
stdev		0.262298	0.709158		
SE		0.15	0.35		

Trial 2

AVERAGE Cr/BAR (g): 6.14

AVERAGE Cr DOSE/DAY (g): 18.42

			predicted FO		
HORSE	DAY	TC	from days	from diurn mix	from asym.
93	D5	3.35	3.898413		
	D6	3.92	3.248677		
	D7	2.52	2.598758		
	D8		2.956661	2.536142	2.8377754
AVE		3.263333	3.175627		
stdev		0.704012	0.550297		
SE		0.41	0.28		
94	D5	5.14	2.981547		
	D6	4.16	4.032399		
	D7	2.74	4.001738		
	D8		5.10957	4.541948	3.9414226
AVE		4.013333	4.031314		
stdev		1.206703	0.869007		
SE		0.7	0.43		
99	D5	3.67	4.616541		
	D6	3.14	3.654762		
	D7	3.12	3.912489		
	D8		3.629557	3.732523	3.8664987
AVE		3.31	3.953337		
stdev		0.311929	0.46025		
SE		0.18	0.23		

Appendix I (cont'd.). Fecal outputs for individual horses

Chapter 6

Trial 2 (cont'd.).

HORSE	DAY	TC	from days	from diurn mix	from asym.
100	D5	3.89	3.299301		
	D6	3.87	3.667131		
	D7	3.46	4.176871		
	D8		3.641756	3.912489	3.4965831
AVE		3.74	3.696265		
stdev		0.242693	0.361655		
SE		0.14	0.18		
102	D5	4.25	3.987013		
	D6	3.55	4.636295		
	D7	3.65	6.497354		
	D8		4.032399	3.209059	4.4406943
AVE		3.816667	4.788265		
stdev		0.378594	1.177203		
SE		0.22	0.59		
131	D5	5.34	5.159664		
	D6	4.76	4.96496		
	D7	5.26	8.843015		
	D8		7.158958	6.418118	5.7869934
AVE		5.12	6.531649		
stdev		0.314325	1.832374		
SE		0.18	0.92		
132	D5	4.5	3.160604		
	D6	3.88	3.580175		
	D7	3.94	4.657396		
	D8		3.68032	3.927505	3.6195716
AVE		4.106667	3.769624		
stdev		0.341955	0.633222		
SE		0.2	0.32		
133	D5	3.67	3.616729		
	D6	3.62	3.827135		
	D7	4.74	5.658986		
	D8		2.92381	3.268855	3.7227162
AVE		4.01	4.006665		
stdev		0.632693	1.167194		
SE		0.37	0.58		

Appendix J. Dry matter intakes for individual horses

Chapter 3

Trial 1

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
93	Cr & Y	11.79573	9.422201	10.04555	8.918725	8.631024	0.5829
	Cr & C27	10.26069	8.196038	8.738269	7.758081	7.507821	0.5205
	Cr &C29	14.44086	11.53507	12.29821	10.9187	10.56648	0.6593
	Cr & C31	24.05868	19.2176	20.489	18.19071	17.60391	0.7955

94		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
	Cr & Y	11.98759	9.195657	10.14846	8.641702	8.132063	0.5487
	Cr & C27	11.5033	8.824155	9.738465	8.292579	7.80353	0.5297
	Cr &C29	15.16256	11.63117	12.83632	10.93049	10.28587	0.6432
	Cr & C31	26.26214	20.14563	22.23301	18.93204	17.81553	0.794

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
98	Cr & Y		4.339527		4.746755	4.45406	0.2142
	Cr & C27		6.070856		6.640555	6.231084	0.4383
	Cr &C29		10.41539		11.39279	10.69029	0.6726
	Cr & C31		18.7157		20.47201	19.20966	0.8178

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
99	Cr & Y	22.56281	16.88442	12.66332	14.82412	12.46231	0.801
	Cr & C27	10.51522	7.868852	5.901639	6.908665	5.807963	0.573
	Cr &C29	11.13315	8.331267	6.24845	7.314654	6.149269	0.5967
	Cr & C31	10.20687	7.6381	5.728575	6.70607	5.637645	0.5601

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
100	Cr & Y	29.94616	20.99596	22.74563	23.0821	19.38089	0.8514
	Cr & C27	11.67979	8.188976	8.871391	9.002625	7.559055	0.619
	Cr &C29	11.36364	7.967314	8.631256	8.758938	7.354443	0.6084
	Cr & C31	10.24637	7.183974	7.782639	7.897767	6.631361	0.5657

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
101	Cr & Y	14.93823	11.34407	10.0337	11.19431	9.996256	0.7329
	Cr & C27	9.886026	7.507433	6.640238	7.408325	6.615461	0.5964
	Cr &C29	11.16084	8.475524	7.496503	8.363636	7.468531	0.6425
	Cr & C31	10.91357	8.287746	7.330416	8.178337	7.303063	0.6344

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
102	Cr & Y	9.810777	6.910736	6.931304	7.116413	6.951872	0.5138
	Cr & C27	11.2686	7.937633	7.961257	8.173872	7.984881	0.5767
	Cr &C29	14.91091	10.50328	10.53454	10.81588	10.5658	0.6801
	Cr & C31	24.37404	17.16914	17.22024	17.68012	17.27133	0.8043

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 3

Trial 1 (cont'd)

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr TC	Cr ASYM.	crasym8	DMD
103	Cr & Y		24.05858		20.8682	20.18828	0.8088
	Cr & C27		11.92019		10.33947	10.00259	0.6141
	Cr &C29		11.81608		10.24917	9.915232	0.6107
	Cr & C31		10.96805		9.513591	9.203624	0.5806

Trial 2

INTAKE = FO/(1-DMD)

		Cr DAILY	Cr ASYM.	crasym8	DMD
93	Cr & Y	24	20.94118	23.23529	0.83
	Cr & C27	45.38376	39.59956	43.93771	0.9101
	Cr &C29	7.850683	6.850106	7.600539	0.4803
	Cr & C31	11.14754	9.726776	10.79235	0.634

		Cr DAILY	Cr ASYM.	crasym8	DMD
94	Cr & Y	19.9422	19.19075	17.39884	0.827
	Cr & C27	40.92527	39.38316	35.70581	0.9157
	Cr &C29	4.016766	3.865409	3.504482	0.1411
	Cr & C31	10.27091	9.883894	8.961	0.6641

		Cr DAILY	Cr ASYM.	crasym8	DMD
98	Cr & Y	19.90909	16.59091	17.77273	0.78
	Cr & C27	41.16541	34.30451	36.74812	0.8936
	Cr &C29	3.16474	2.637283	2.825145	-0.384
	Cr & C31	13.95349	11.62791	12.4562	0.6861

		Cr DAILY	Cr ASYM.	crasym8	DMD
99	Cr & Y	9.392523	8.426791	9.190031	0.358
	Cr & C27	102.2034	91.69492	100	0.941
	Cr &C29	8.314948	7.460011	8.135687	0.2748
	Cr & C31	14.60402	13.10245	14.28917	0.5871

		Cr DAILY	Cr ASYM.	crasym8	DMD
100	Cr & Y	20.46575	17.94521	18.21918	0.635
	Cr & C27	41.36213	36.268	36.82171	0.8194
	Cr &C29	9.198375	8.065509	8.188647	0.1879
	Cr & C31	17.84946	15.65114	15.89008	0.5815

		Cr DAILY	Cr ASYM.	crasym8	DMD
101	Cr & Y	5.830189	5.59434	5.962264	-0.06
	Cr & C27	63.5149	60.94553	64.95375	0.9027
	Cr &C29	13.83789	13.2781	14.15137	0.5534
	Cr & C31	16.63079	15.95802	17.00753	0.6284

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 3

Trial 2 (cont'd)

INTAKE = FO/(1-DMD)

		Cr DAILY	Cr ASYM.	crasym8	DMD
102	Cr & Y	43.84	35.76	37.2	0.875
	Cr & C27	98.56115	80.39568	83.63309	0.9444
	Cr &C29	7.111342	5.800675	6.034259	0.2294
	Cr & C31	15.16745	12.37199	12.87019	0.6387

		Cr DAILY	Cr ASYM.	crasym8	DMD
103	Cr & Y	15.84466	13.02913	14.40777	0.485
	Cr & C27	55.43478	45.58424	50.40761	0.8528
	Cr &C29	8.637663	7.102784	7.854345	0.0553
	Cr & C31	18.49082	15.20508	16.81396	0.5587

Chapter 4

Trial 1

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
93	ACTUAL		8.03				
	Cr & Y	17.76192	21.14157	18.43785	18.73827	17.34885	0.7337
	Cr & C27	10.68444	12.71742	11.09103	11.27174	10.43596	0.5573
	Cr &C29	9.649123	11.48511	10.01632	10.17952	9.424725	0.5098
	Cr & C31	9.131274	10.86873	9.478764	9.633205	8.918919	0.482

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
94	ACTUAL		8.78				
	Cr & Y	9.513151	12.61891	13.96195	16.22832	10.94012	0.6426
	Cr & C27	7.305544	9.690589	10.72196	12.4624	8.401375	0.5346
	Cr &C29	7.901464	10.48106	11.59656	13.47897	9.086684	0.5697
	Cr & C31	7.191201	9.538917	10.55415	12.26734	8.269882	0.5272

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
98	ACTUAL		6.32				
	Cr & Y	11.29437	14.16278	15.99139	13.23055	12.90785	0.7211
	Cr & C27	8.076392	9.088817	10.26231	8.490566	8.283479	0.5654
	Cr &C29	7.377049	9.250585	10.44496	8.641686	8.430913	0.573
	Cr & C31	6.967485	8.737005	9.865074	8.161911	7.96284	0.5479

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
99	ACTUAL		10.96				
	Cr & Y	13.81329	19.25666	20.19315	16.35938	16.65203	0.6583
	Cr & C27	8.875517	12.37307	12.9748	10.51147	10.69951	0.4682
	Cr &C29	10.49589	14.63198	15.34356	12.43051	12.65288	0.5503
	Cr & C31	9.262166	12.91209	13.54003	10.96939	11.16562	0.4904

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 4

Trial 1 (cont'd.)

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
100	ACTUAL		10.73				
	Cr & Y	15.5969	21.6124	27.53488	20.27907	16.43411	0.6775
	Cr & C27	10.35832	14.35338	18.28666	13.46787	10.91433	0.5144
	Cr &C29	12.14096	16.82356	21.43374	15.78566	12.79266	0.5857
	Cr & C31	10.8569	15.04425	19.16685	14.11612	11.43967	0.5367

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
101	ACTUAL		7.59				
	Cr & Y	10.66452	13.96034	14.89818	9.833869	11.30761	0.6268
	Cr & C27	8.697552	11.38549	12.15035	8.020105	9.222028	0.5424
	Cr &C29	7.948872	10.40543	11.10445	7.329738	8.428201	0.4993
	Cr & C31	8.001608	10.47447	11.17813	7.378368	8.484117	0.5026

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
102	ACTUAL		6.78				
	Cr & Y	11.81015	17.62325	15.41575	17.47609	15.48933	0.7282
	Cr & C27	8.055207	12.02008	10.51443	11.9197	10.56462	0.6015
	Cr &C29	7.477289	11.1577	9.760075	11.06452	9.806662	0.5707
	Cr & C31	6.669437	9.952213	8.705589	9.869105	8.747143	0.5187

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
103	ACTUAL		10.43				
	Cr & Y	12.00392	17.63841	18.96129	14.77217	14.87016	0.5918
	Cr & C27	9.501648	13.96161	15.00873	11.69284	11.77041	0.4843
	Cr &C29	11.15919	16.39718	17.62696	13.73263	13.82373	0.5609
	Cr & C31	10.40561	15.28987	16.43661	12.80527	12.89021	0.5291

Trial 2

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
93	ACTUAL		9.05				
	Cr & Y	14.87889	19.64629	15.45559	14.91734	16.99346	0.7399
	Cr & C27	8.091156	10.68367	8.404767	8.112064	9.241062	0.5217
	Cr &C29	9.41835	12.43612	9.783402	9.442687	10.75688	0.5891
	Cr & C31	8.360337	11.0391	8.684381	8.38194	9.548499	0.5371

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
94	ACTUAL		10.25				
	Cr & Y	9.36	15.22	13.24	9.54	11.3	0.5
	Cr & C27	8.074534	13.12974	11.42167	8.229814	9.748102	0.4204
	Cr &C29	8.641064	14.05096	12.22304	8.807238	10.43205	0.4584
	Cr & C31	8.833522	14.36391	12.49528	9.003398	10.6644	0.4702

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 4

Trial 2 (cont'd.)

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
98	ACTUAL		7.74				
	Cr & Y	13.20594	17.62208	14.862	13.29087	15.41401	0.7645
	Cr & C27	6.087297	8.12292	6.850656	6.126444	7.105109	0.4891
	Cr &C29	7.050555	9.408297	7.934709	7.095897	8.229426	0.5589
	Cr & C31	6.366428	8.495394	7.16479	6.407369	7.430911	0.5115

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
99	ACTUAL		10				
	Cr & Y	11.89571	22.97664	18.30527	14.63878	16.48561	0.6318
	Cr & C27	7.168576	13.84615	11.0311	8.821604	9.934534	0.389
	Cr &C29	7.571305	14.62403	11.65082	9.3172	10.49265	0.4215
	Cr & C31	7.838225	15.13958	12.06156	9.645669	10.86256	0.4412

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
100	ACTUAL		11.07				
	Cr & Y	15.41937	18.72352	20.07907	15.19345	16.12539	0.6459
	Cr & C27	8.252721	10.02116	10.74667	8.131802	8.630593	0.3384
	Cr &C29	8.902658	10.81037	11.59302	8.772216	9.310289	0.3867
	Cr & C31	9.090909	11.03896	11.83816	8.957709	9.50716	0.3994

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
101	ACTUAL		11.24				
	Cr & Y	11.04839	16.02151	10.32258	11.53226	13.97849	0.628
	Cr & C27	8.055664	11.68169	7.52646	8.408467	10.19208	0.4898
	Cr &C29	9.741645	14.12657	9.101683	10.16829	12.3252	0.5781
	Cr & C31	8.944505	12.97062	8.35691	9.336235	11.31665	0.5405

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
102	ACTUAL		7.14				
	Cr & Y	13.80814	22.91667	18.9438	18.79845	18.2655	0.7936
	Cr & C27	6.493506	10.77694	8.908635	8.840283	8.589656	0.5611
	Cr &C29	6.987007	11.59598	9.585683	9.512135	9.242461	0.5921
	Cr & C31	6.047104	10.03607	8.296202	8.232548	7.999151	0.5287

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
103	ACTUAL		10.77				
	Cr & Y	10.52977	15.0394	12.65324	10.94571	13.13485	0.5432
	Cr & C27	7.080818	10.11335	8.508759	7.360518	8.832622	0.3207
	Cr &C29	7.566462	10.80698	9.092339	7.865345	9.438414	0.3643
	Cr & C31	8.147019	11.63618	9.789973	8.468835	10.1626	0.4096

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 5

Trial 1

INTAKE = FO/(1-DMD)

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
93	Cr & Y	55.77778	62.44444	69.11111	0.955
	Cr & C27	13.38667	14.98667	16.58667	0.8125
	Cr &C29	6.785618	7.596648	8.407678	0.6301
	Cr & C31	5.381647	6.024871	6.668096	0.5336

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
94	ACTUAL		7.83			
	Cr & Y	14.67033	15.65934	17.36264	13.35165	0.818
	Cr & C27	6.384505	6.814921	7.556193	5.810617	0.5818
	Cr &C29	5.330405	5.689758	6.308644	4.851268	0.4991
	Cr & C31	5.464593	5.832992	6.467458	4.973393	0.5114

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
98	Cr & Y	71.16279	67.2093	64.18605	0.957
	Cr & C27	13.35661	12.61458	12.04714	0.7709
	Cr &C29	7.232333	6.830537	6.523281	0.5769
	Cr & C31	6.400335	6.044761	5.772851	0.5219

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
99	Cr & Y	73.01587	80.79365	70.15873	0.937
	Cr & C27	32.0781	35.49512	30.82287	0.8566
	Cr &C29	16.01671	17.72284	15.38997	0.7128
	Cr & C31	11.93565	13.20706	11.4686	0.6146

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
100	ACTUAL		10.87			
	Cr & Y	13.0719	12.87582	14.90196	10.19608	0.694
	Cr & C27	7.320644	7.210835	8.345534	5.710102	0.4536
	Cr &C29	7.48083	7.368618	8.528147	5.835048	0.4653
	Cr & C31	7.312614	7.202925	8.33638	5.703839	0.453

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
101	ACTUAL		9.01			
	Cr & Y	12.26923	12.34615	13.61538	10.76923	0.74
	Cr & C27	6.804608	6.84727	7.551195	5.972696	0.5312
	Cr &C29	7.046609	7.090789	7.819748	6.185112	0.5473
	Cr & C31	6.852846	6.895811	7.604726	6.015038	0.5345

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
102	ACTUAL		9.81			
	Cr & Y	15.14768	16.91983	22.1519	15.48523	0.763
	Cr & C27	7.302685	8.157038	10.67941	7.465419	0.5084
	Cr &C29	6.653076	7.431431	9.729429	6.801334	0.4604
	Cr & C31	7.030944	7.853506	10.28202	7.187622	0.4894

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 5

Trial 1 (cont'd.)

INTAKE = FO/(1-DMD)

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
103	Cr & Y	100.6383	103.4043	97.87234	0.953
	Cr & C27	63.31995	65.06024	61.57965	0.9253
	Cr &C29	17.10669	17.57685	16.63653	0.7235
	Cr & C31	12.9589	13.31507	12.60274	0.635

Trial 2

INTAKE = FO/(1-DMD)

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
93	Cr & Y	47.87037	64.16667	46.38889	0.892
	Cr & C27	42.16966	56.52529	40.8646	0.8774
	Cr &C29	17.43676	23.37268	16.89713	0.7035
	Cr & C31	14.36909	19.2607	13.9244	0.6402

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
94	ACTUAL		7.15			
	Cr & Y	6.880165	8.553719	7.954545	7.995868	0.516
	Cr & C27	6.79176	8.44381	7.852335	7.893127	0.5097
	Cr &C29	6.067784	7.543732	7.015306	7.051749	0.4512
	Cr & C31	6.500098	8.081202	7.515128	7.554167	0.4877

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
98	Cr & Y	45.65657	61.41414	45.35354	0.901
	Cr & C27	110.2439	148.2927	109.5122	0.959
	Cr &C29	17.27829	23.24159	17.16361	0.7384
	Cr & C31	14.01116	18.84687	13.91816	0.6774

		Cr DAILY	Cr MIX	Cr ASYM.	DMD
99	Cr & Y	42.74809	65.64885	40.76336	0.869
	Cr & C27	69.73848	107.0984	66.50062	0.9197
	Cr &C29	11.50133	17.66276	10.96734	0.5131
	Cr & C31	10.03584	15.41219	9.569892	0.442

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
100	ACTUAL		8.85			
	Cr & Y	10.18568	12.75862	10.61008	8.01061	0.623
	Cr & C27	8.097849	10.1434	8.435259	6.368621	0.5258
	Cr &C29	5.846529	7.323386	6.090134	4.598051	0.3432
	Cr & C31	5.885057	7.371648	6.130268	4.628352	0.3475

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
101	ACTUAL		4.68			
	Cr & Y	6.036866	7.81106	7.050691	4.953917	0.566
	Cr & C27	5.29186	6.847102	6.18057	4.342557	0.5049
	Cr &C29	4.635527	5.997877	5.414013	3.803963	0.4348
	Cr & C31	4.306377	5.571992	5.029586	3.533859	0.3916

Appendix J (cont'd.). Dry matter intakes for individual horses

Chapter 5

Trial 2 (cont'd.)

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr ASYM.	DMD
102	ACTUAL		10.2			
	Cr & Y	8.027613	9.74359	9.684418	9.230769	0.493
	Cr & C27	9.201899	11.16889	11.10106	10.58105	0.5577
	Cr &C29	8.502193	10.31962	10.25695	9.776478	0.5213
	Cr & C31	7.913669	9.605289	9.546957	9.099747	0.4857

103		Cr DAILY	Cr MIX	Cr ASYM.	DMD
	Cr & Y	67.10526	71.14035	65.78947	0.886
	Cr & C27	120.283	127.5157	117.9245	0.9364
	Cr &C29	16.53339	17.52756	16.20921	0.5373
	Cr & C31	13.37413	14.17832	13.11189	0.428

Chapter 6

Trial 1

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
93	Cr & Y	13.2186	11.0216	10.50897	25.44855	6.993775	0.7269
94	Cr & Y	24.73179	31.33823	22.64257	31.67702	27.27273	0.8229
99	Cr & Y	17.9868	27.94279	16.22662	15.0165	2.85	0.8182
100	Cr & Y	23.94366	34.09624	21.30282	27.23005	10.56338	0.8296
102	Cr & Y	40.55172	36	32.82759	39.58621	29.37931	0.9275
131	Cr & Y	46.78609	46.36459	37.72392	95.78504	28.13488	0.9051
132	Cr & Y	37.37864	39.56311	30.9466	41.99029	29.61165	0.9176
133	Cr & Y	23.41941	24.66607	21.63847	28.22796	21.99466	0.8877

Trial 2

INTAKE = FO/(1-DMD)

		TC(fecal)	Cr DAILY	Cr MIX	Cr TC	Cr ASYM	DMD
93	Cr & Y	11.34702	11.06857	8.840933	12.18239	9.885137	0.7127
94	Cr & Y	17.8143	17.90315	20.16881	20.03554	17.50333	0.7749
99	Cr & Y	10.75723	12.83718	12.1222	11.04972	12.57719	0.6923
100	Cr & Y	15.53801	15.37183	16.24429	15.78729	14.54092	0.7593
102	Cr & Y	16.28303	20.41773	13.68286	17.2208	18.92583	0.7654
131	Cr & Y	28.47608	36.31813	35.70634	30.47831	32.20245	0.8202
132	Cr & Y	20.2963	18.61728	19.40741	19.40741	17.87654	0.7975
133	Cr & Y	14.48699	14.48699	11.81358	16.97977	13.43931	0.7232

Appendix K. Cr data for individual horses

Chapter 3

Trial 1 - Daily Fecal Cr, g/kg fecal DM

DAY	93 BG	94 BG	98 BG	99 TF	100 TF	101 TF	102 BG	103 TF
0	0	0	0	0	0	0	0	0
1	1.068	1.348	2.188	1.698	1.348	1.715	1.566	1.645
2	5.233	4.690	4.918	7.630	4.848	5.093	5.583	7.018
3	3.868	5.862	4.673	11.585	6.843	9.415	5.285	5.530
4	5.775	4.603	4.113	7.508	6.125	6.860	3.150	4.603
5	5.075	4.445	5.583	7.403	5.688	7.315	7.525	3.920
6	3.238	2.940	4.690	5.915	4.375	5.513	4.445	3.605
7	4.970	5.530	4.725	6.545	6.458	5.268	4.410	3.920
8	5.198	4.900	5.670	3.220	6.458	5.425	5.408	3.780
9	4.690	4.393	4.743	5.145	4.410	6.178	4.568	3.833
10	4.620	3.763	4.078	3.605	3.395	5.670	4.988	4.480
11	4.848	4.585	4.095	3.728	3.588	3.535	5.688	4.655
12	3.815	3.518	3.990	3.833	3.658	3.850	4.235	3.518

Trial 1. Comparison of combined grab sample and TC sample Cr g/kg fecal DM

	93 BG	94 BG	99 TF	100 TF	101 TF	102 BG
GRAB	4.970	5.530	6.545	6.458	5.268	4.410
TC	4.795	5.250	7.525	5.425	6.423	4.848

Trial 2 - Daily Fecal Cr, g/kg fecal DM

DAY	93 TF	94 TF	98 TF	99 BG	100 BG	101 BG	102 TF	103 BG
0	2.238	0	0	0	0	0	0	0
1	2.310	1.768	1.330	1.453	1.330	1.558	2.380	1.811
2	3.885	3.728	4.095	2.293	3.308	3.885	5.600	3.833
3	4.988	4.393	4.690	2.923	2.398	2.520	2.820	2.188
4	4.288	5.338	5.355	2.520	2.573	1.890	3.623	1.803
5	5.005	6.038	5.443	3.920	3.728	2.975	4.795	1.995
6	5.303	8.313	4.988	3.570	3.203	3.220	5.495	3.238
7	3.693	6.563	3.273	3.010	1.978	2.468	2.660	1.995
8	3.973	3.010	3.463	2.083	1.715	3.045	2.188	1.960
9	6.808	5.548	6.440	4.830	3.378	3.080	5.373	2.293
10	4.270	4.410	3.325	1.785	2.293	3.570	4.778	4.515
11	5.355	4.620	5.723	3.728	3.185	3.798	3.220	2.975
12	6.213	4.795	5.845	4.025	2.363	2.870	3.710	2.695

Appendix K (cont'd.). Cr data for individual horses

Chapter 4

Trial 1 - Daily Fecal Cr, g/kg fecal DM

DAY	93 TF	94 OG	98 TF	99 OG	100 OG	101 TF	102 TF	103 OG
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
1	0	0	0	0	0.543	0.604	0	0.376
2	5.123	3.710	2.380	2.835	4.025	4.988	4.343	3.973
3	4.813	6.860	6.143	3.623	6.003	6.038	6.545	5.810
4	6.335	6.169	6.108	5.023	5.040	6.694	5.250	2.555
5	3.448	4.445	6.055	3.168	3.080	3.500	4.104	2.958
6	3.334	5.390	5.530	3.413	3.203	3.728	4.078	2.713
7	3.833	4.174	4.723	3.080	2.861	4.550	4.331	2.713
8	3.675	3.990	4.323	2.634	2.468	3.745	4.218	2.765
9	3.203	3.881	3.999	2.800	2.354	2.170	3.920	2.424
10	0.945	0.726	1.619	0.464	0.324	0.158	1.208	0.245
11	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0
14	0	0	0	0	0	0	0	0

Trial 1. Comparison of samples g/kg fecal DM

	93 TF	94 OG	98 TF	99 OG	100 OG	101 TF	102 TF	103 OG
GRAB	3.675	3.990	4.323	2.634	2.468	3.745	4.218	2.765
TC	4.734	3.728	8.278	2.511	2.888	6.545	6.195	6.230
DIURN MIX	4.078	4.025	4.498	2.905	2.258	3.605	4.778	2.590

Trial 1. Diurnal variation g/kg fecal DM

TIME	93 TF	94 OG	98 TF	99 OG	100 OG	101 TF	102 TF	103 OG
700	4.725	4.743	5.548	3.395	2.888	4.655	4.813	2.853
1500	5.355	4.743	5.530	2.861	2.223	3.001	4.874	2.310
2300	3.631	4.095	4.183	2.853	3.238	4.235	4.323	3.351

Appendix K (cont'd.). Cr data for individual horses

Chapter 4

Trial 2 - Daily Fecal Cr, g/kg fecal DM

DAY	93 OG	94 TF	98 OG	99 TF	100 TF	101 TF	102 OG	103 TF
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
1	0	0	0	0.853	0.805	0.718	0	0.984
2	3.920	4.183	3.666	3.360	5.110	5.355	5.731	3.036
3	5.670	4.786	5.941	4.130	3.605	4.008	7.849	4.052
4	5.743	5.504	7.193	4.489	3.955	4.401	6.528	4.113
5	3.833	2.258	4.130	2.809	2.686	3.238	4.323	3.063
6	4.323	3.054	5.320	3.036	3.483	2.905	5.521	3.561
7	3.701	2.896	5.303	2.853	3.518	3.631	3.596	2.555
8	3.876	2.476	4.786	2.368	2.633	3.824	3.929	2.678
9	5.618	4.209	6.230	2.888	2.853	2.870	6.930	2.511
10	1.838	0.464	2.166	0.284	1.278	0	0.993	0.354
11	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0
13	0	0	0	0	0	0	0	0
14	0	0	0	0	0	0	0	0

Trial 2. Comparison of samples g/kg fecal DM

	93 OG	94 TF	98 OG	99 TF	100 TF	101 OG	102 OG	103 TF
GRAB	3.876	2.476	4.786	2.368	2.633	3.824	3.929	2.678
TC	4.340	3.798	6.545	3.719	3.106	5.058	5.163	3.553
DIURN MIX	4.988	3.028	5.723	2.975	2.818	5.215	5.128	3.465

Trial 2. Diurnal variation g/kg fecal DM

TIME	93 OG	94 TF	98 OG	99 TF	100 TF	101 OG	102 OG	103 TF
700	6.991	4.332	4.804	3.789	2.625	3.308	5.513	3.316
1500	3.973	4.288	6.379	2.450	2.433	4.944	3.920	2.748
2300	3.264	2.258	5.740	2.730	2.993	5.854	4.148	3.290

Appendix K (cont'd.). Cr data for individual horses

Chapter 5

Trial 1 - Daily Fecal Cr, g/kg fecal DM

DAY	93 BG	94 BG	98 BG	99 TF	100 TF	101 TF	102 BG	103 TF
0	0.683	88	0	0	0	44	0	0
0	0	0	123	0	0	0	0	0
0	0	0	0	0	0	0	0	0
1	0	0	0	0	0	0	0	0
2	6.195	4.585	5.128	3.658	2.958	3.658	4.113	3.483
3	5.793	4.340	5.618	4.918	2.695	3.238	4.165	3.360
4	4.533	6.423	4.655	3.448	3.360	3.605	4.095	3.325
5	4.340	5.233	4.568	3.798	3.500	5.163	4.165	4.060
6	5.233	6.720	5.793	4.498	6.475	6.563	7.280	3.395
7	6.178	9.293	7.368	3.815	4.830	6.160	4.725	4.375
8	7.280	5.635	7.018	3.780	4.638	5.128	3.465	3.745
9	4.988	5.950	5.215	2.363	4.253	5.495	3.658	3.185
10	0	1.733	0.280	0	0	0	0.280	0.245
11	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0

Trial 1. Comparison of samples g/kg fecal DM

	93 BG	94 BG	98 BG	99 TF	100 TF	101 TF	102 BG	103 TF
GRAB	7.280	5.635	7.018	3.780	4.638	5.128	3.465	3.745
DIURN MIX	6.493	5.775	6.318	3.588	4.008	5.163	3.483	3.763

Trial 1. Diurnal variation g/kg fecal DM

TIME	93 BG	94 BG	98 BG	99 TF	100 TF	101 TF	102 BG	103 TF
700	7.910	5.163	6.388	5.950	5.303	4.165	3.133	6.370
1300	7.298	6.178	6.843	2.030	3.605	6.685	4.358	2.328
1900	4.235	5.775	4.935	2.275	2.275	4.095	2.625	2.520

Appendix K (cont'd.). Cr data for individual horses

Chapter 5

Trial 2 - Daily Fecal Cr, g/kg fecal DM

DAY	93 TF	94 TF	98 TF	99 BG	100 BG	101 BG	102 TF	103 BG
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
1	0	0	0	0	0	0	0	0.403
2	3.010	4.480	3.255	3.238	3.675	4.638	4.603	2.653
3	3.973	6.650	4.025	4.270	2.013	4.323	4.270	2.608
4	2.153	4.533	2.763	2.783	2.240	2.380	3.133	2.380
5	3.473	4.935	5.075	3.990	3.938	4.550	2.905	2.748
6	4.673	4.375	4.655	4.148	3.115	5.758	4.830	2.380
7	4.498	4.830	4.183	4.008	5.093	6.580	4.463	2.608
8	3.238	5.285	3.973	2.643	5.268	7.578	4.708	2.765
9	2.625	5.338	3.448	2.415	4.760	6.720	3.640	2.345
10	0	0.700	0	0	0.569	0.376	0.516	0
11	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0

Trial 2. Comparison of samples g/kg fecal DM

	93 TF	94 TF	98 TF	99 BG	100 BG	101 BG	102 TF	103 BG
GRAB	3.238	5.285	3.973	2.643	5.268	7.578	4.708	2.765
DIURN MIX	2.888	5.198	3.290	2.328	5.005	6.545	4.078	2.468

Trial 2. Diurnal variation g/kg fecal DM

TIME	93 TF	94 TF	98 TF	99 BG	100 BG	101 BG	102 TF	103 BG
700	4.813	4.043	4.725	2.258	3.920	5.968	4.515	2.380
1300	2.783	4.988	3.080	2.345	5.548	7.700	5.163	2.520
1900	1.680	5.863	2.258	2.258	5.005	7.053	3.203	2.590

Appendix K (cont'd.). Cr data for individual horses

Chapter 6

Trial 1 - Daily Fecal Cr, g/kg fecal DM

DAY	93 BG	94 BG	99 BG	100 BG	102 TF	131 TF	132 TF	133 TF
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
1	0	0	0.438	0.201	0.324	0	0	0
2	3.903	3.535	2.275	3.185	4.148	2.748	3.395	4.865
3	3.325	3.763	2.485	2.625	4.795	2.905	4.655	5.985
4	3.378	2.5753	2.888	2.748	2.730	4.025	3.150	3.255
5	4.690	1.978	1.890	2.398	5.618	2.345	3.798	4.760
6	5.355	4.463	3.395	1.768	8.540	4.760	6.598	8.488
7	7.315	4.655	5.338	7.053	6.108	5.285	5.548	5.845
8	6.563	3.063	6.563	4.865	7.158	5.460	6.633	7.315
9	6.073	4.620	6.108	1.663	5.443	3.395	5.600	6.930
10	0	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0

Trial 1. Comparison of samples g/kg fecal DM

	93 BG	94 BG	99 BG	100 BG	102 TF	131 TF	132 TF	133 TF
GRAB	6.563	3.063	6.563	4.865	7.158	5.460	6.633	7.315
TC	3.973	4.774	6.948	6.440	7.053	2.100	6.913	5.985
DIURN MIX	6.090	4.358	5.913	4.813	7.350	4.883	6.860	7.175

Trial 1. Diurnal variation g/kg fecal DM

TIME	93 BG	94 BG	99 BG	100 BG	102 TF	131 TF	132 TF	133 TF
700	2.135	3.168	9.538	8.663	8.943	7.175	6.003	5.425
1300	9.170	6.930	6.475	3.798	7.595	5.985	8.558	12.163
1900	5.635	2.730	2.153	1.768	4.480	2.293	5.460	7.840

Appendix K (cont'd.). Cr data for individual horses

Chapter 6

Trial 2 - Daily Fecal Cr, g/kg fecal DM

DAY	93 TF	94 TF	99 TF	100 TF	102 BG	131 BG	132 BG	133 BG
0	1.216	0.114	0.210	0	0.105	0	0	0
0	0.420	0.140	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0
1	0	0	0	1.146	0.324	0.726	0.341	0.193
2	5.145	3.605	4.883	2.748	4.428	2.590	4.900	4.043
3	7.228	3.745	3.850	3.203	2.713	3.483	4.340	4.095
4	7.088	4.305	3.903	2.958	4.900	3.570	5.005	3.710
5	4.725	6.178	3.990	5.583	4.620	3.570	5.828	5.093
6	5.670	4.568	5.040	5.023	3.973	3.710	5.145	4.813
7	7.088	4.603	4.708	4.410	2.835	2.083	3.955	3.255
8	6.230	3.605	5.075	5.058	4.568	2.573	5.005	6.300
9	6.528	5.880	4.375	2.398	2.765	2.170	3.885	4.165
10	0.534	0.359	0	0	0	0	0	0.088
11	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0

Trial 2. Comparison of samples g/kg fecal DM

	93 TF	94 TF	99 TF	100 TF	102 BG	131 BG	132 BG	133 BG
GRAB	6.230	3.605	5.075	5.058	4.568	2.573	5.005	6.300
TC	6.178	5.268	5.093	3.938	4.305	3.378	4.288	3.710
DIURN MIX	7.263	4.148	4.935	4.708	5.740	2.870	4.690	5.635

Trial 2. Diurnal variation g/kg fecal DM

TIME	93 TF	94 TF	99 TF	100 TF	102 BG	131 BG	132 BG	133 BG
700	4.830	5.005	8.488	7.088	6.790	3.255	5.338	7.070
1300	9.608	4.375	4.480	4.638	4.900	3.343	4.970	6.738
1900	8.068	2.835	1.768	2.485	4.655	1.890	2.975	3.588

Vita

Janice Lee Holland, daughter of Richard L. and Mary A. Holland, was born December 13, 1968 in Ridgewood, New Jersey. She graduated from Cedar Cliff High School, Camp Hill, Pennsylvania, in June, 1986. She entered The Pennsylvania State University, State College, Pennsylvania, in August, 1986 and received her Bachelor of Science degree (Animal Production) in May, 1990. In August 1990 the author began her graduate studies at Virginia Polytechnic Institute and State University, Blacksburg, Virginia. She received a Master of Science degree in Animal Science (Equine Nutrition) in December, 1994. The author then continued her graduate studies in Animal and Poultry Science (Equine Nutrition) at Virginia Tech beginning in January, 1995. She married James Warren Rowland, III on June 29, 1996.

Janice Lee Holland