ESTIMATING COOL SEASON ANNUAL GRASS FORAGE BIOMASS USING UAV AND GROUND BASED TECHNOLOGY

A Dissertation

by

BRYAN EDWIN SIMONEAUX

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Chair of Committee,	Amir Ibrahim
Co-Chair of Committee,	Clark Neely
Committee Members,	Nithya Rajan
	Sorin Popescu
	Don Renchie
Head of Department,	David Baltensperger

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ABSTRACT

Many research institutions conduct annual forage trials to determine which species and varieties of those species produce the most biomass in specific geographical areas. These trials can be costly, time consuming and labor intensive to conduct. A cool-season annual grass forage trial was conducted at the Texas A&M Research Farm during the 2015-2016 and 2016-2017 growing seasons. This forage trial tests small grains species such as wheat, oat, rye, triticale, and barley in addition to annual ryegrass. The purpose of this research was to determine if Unmanned Aerial Vehicle (UAV) technology or other ground-based sampling methods could replace traditional forage harvesting techniques. The ultimate goal of this project was to develop a low cost and effective method to measure above ground forage biomass.

If researchers are unable to take destructive samples, both visual and plant height measurements were highly correlated for fall and late spring clips. Combining all measurements of the traditional methods into one model to predict forage yield may be a better way to estimate forage yield if the whole plot forage harvest is not possible. In the UAV and ground-based methods no one variable showed a strong relationship across all clip times or species. Models were developed for each species and clip time to better predict forage yield using multiple variables. In some cases, the models did improve the coefficient of determination (R^2) substantially, but in many cases, improvement was marginal. The ground model did as good as or better than the aerial model for wheat, oat, barley and the combined model, whereas the aerial model was far better for rye. There

was little to no difference between the ground and aerial models for ryegrass. Transforming the data also had little effect on model accuracy for wheat, oat and barley. However, transforming the data for rye had a negative effect while ryegrass and the combined species model were improved when the data was transformed.

The best results for evaluating multi species across clip times was the traditional method model created from visual rating, plant height and subsample weight. It seemed far better when evaluating a multi species trial to do so at specific times during the growing season as opposed to throughout the growing season. If only a single species is being evaluated, it seemed that species a specific method and clip time was ideal.

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Nomenclature

ADF	Acid Detergent Fibers
ADG	Average Daily Gain
HT	Plant Height
LAI	Leaf Area Index
NDF	Neutral Detergent Fibers
NDVI	Normalized Difference Vegetation Index
NIR	Near Infra-Red
RCBD	Randomize Complete Block Design
RGB	Red Green Blue
RMSE	Root Mean Square Error
SUB	Sub-Sample
TDM	Total Dry Matter
TVI	Triangular Vegetation Index
UAV	Unmanned Aerial Vehicle
VI	Vegetation Index

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CHAPTER I

INTRODUCTION AND REVIEW OF LITERATURE

There are many methods used to evaluate forage potential, some of the more traditional methods have been utilized for well over 50 years. However, in recent decades there has been an interest in developing more modern and efficient ways to evaluate forage potential. Having a good indication of how much yield potential a particular forage species and cultivar has is useful for livestock managers who rely on good forage production to meet the high demands of their forage systems. It is also important for researchers in forage and small grains breeding programs whom need good forage potential data for cultivar advancement. The United States (U.S.) is one of the largest forage producing nations in the world. According to the 2017 United States Department of Agriculture census, there were 55.7 million acres of land used for forage, which contributed to the sustainable nutritive needs of over 89.9 million cattle and calves. In the southern U.S., Texas is the largest forage producing state with over 5 million acres dedicated to forage production (USDA-NASS, 2018). In a 2016 national forage review, the USDA estimated that total forage production (all dry hay, haylage and green chop) is only down 1% to 90.7 million tons from the previous year (USDA-NASS, 2016).

In the southern U. S., the use of cool season annual grasses for forage is a common practice. Livestock producers have used cool season annual grasses for hay, silage, and pasture, and often times as an accompanying crop for alfalfa (*Medicago* *sativa* L.) establishment. Cool season grasses offer producers valuable production and management alternatives as sources of forage, which utilize similar planting equipment and harvest management techniques as that of alfalfa. For example, small grains used for forage provide ample quality to meet the nutritional needs of most livestock (Maloney et al., 1999). Many livestock managers feed hay, silage or concentrates during the fall to early spring; however, this method can be expensive and greatly reduce any profit realized. One solution to this problem is for livestock managers in the southern U.S. to adapt a more cost effective annual cool-season forage system to feed their livestock (Redmon et al., 1998). The importance of predicting forage production yield can have a great effect on livestock management strategies and stocking rate. Historically this has been achieved through detailed record keeping of forage production, livestock performance, and livestock stocking rates (Redfearn and Bidwell, 2000). In addition, cool season forage researches would benefit greatly from the ability to select experimental lines for high biomass potential, which would aid in variety advancement. For researchers and producers alike, it is important to know which species and varieties of cool season grasses produce the most forage in a given geographical area. For this reason, many universities conduct annual forage trials to determine which species and varieties of those species produce the most biomass in specific geographical areas. The publications that result from these forage trials are aimed at giving unbiased yield and quality data to researchers and forage producers so they can make educated decisions regarding the most appropriate varieties for their geographic region (Neely et al., 2016).

Small grains forage trials can be costly, time consuming and labor intensive to conduct. As noted in past studies, the standard method of determining forage biomass is to clip and weigh the samples, which requires great effort and expense (Sanderson et al 2001). Recently Texas A&M AgriLife Research has been making efforts to incorporate Unmanned Aerial Vehicles (UAV) into determining cool season annual forage biomass. These vehicles utilize remote sensing imagery such as visual spectral images, which can obtain various vegetation indices.

The purpose of this research is to determine if UAV technology or other ground based sampling methods can replace traditional forage harvesting techniques to better assist cool-season annual grass breeders and researchers in estimating forage yield of entries in forage trials and breeding lines for advancement in breeding programs. The specific objectives are 1) to compare mechanically harvested above ground biomass yield of cool-season annual grasses with alternative forms of limited or non-destructive measurements to predict plot yield. 2) Determine at what height UAV measurements should be taken for measuring forage yield, 3) Compare relationships among UAV generated vegetation indices, hand held NDVI, plant height, and visual rating with dry matter forage yield across and between cool-season annual grass species. (4) Develop models to predict forage yield among cool-season annual grasses using UAV generated vegetation indices and ground based measurements. 5) Determine if models generated from UAV data are as reliable as models generated from ground-based measurements. The ultimate goal of this project is to develop a low cost and effective method to measure above ground forage biomass.

Forage Production in the United States: Types and Uses

In the southern U.S., cool-season annual grasses can extend grazing and supply high quality forage for livestock (Bagley et al., 1998). The least costly means of providing livestock winter forage would be perennial grasses, unfortunately there are currently no good cool-season perennial forage grass options well adapted to the south. This makes the use of cool season annual grasses the most common form of winter pasture (Corriher-Olson and Redmon, 2013). Some of the most common cool season annual forage species in the southern U.S. are small grains, which include wheat (*Triticum aestivum*), rye (*Secale cereal*), oats (*Avena sativa*) and triticale (*Triticum secale*) (Bruckner and Raymer, 1990). In addition to these more common southern species, barley (*Hordeum vulgare*) can produce good quality silage and hay similar to that of oats and triticale (Min, D.-H. 2012). Annual ryegrass (*Lolium multiflorum*) is not considered a small grain, but provides a source of high-quality winter and spring forage in the temperate regions of the world (Hill and Gates, 2001)

Winter wheat

Most cultivated winter wheat is an Allohexaploid bread wheat (T*riticum aestivum* L.), which makes up approximately 20% of all calories consumed by humans (Marcussen et al., 2014). In the Southern Great Plains, winter wheat grown for both forage and grain can be an extremely important source of income from both the sale of grain and in terms of weight gained by grazing livestock (Redmon et al., 1995). Wheat forage provides feed for cattle and sheep. The forage is low in fiber but high in protein, energy, and minerals. However, due to the high moisture content it can be difficult to meet the daily dry matter needs of grazing animals (Duncan et al., 2018). Wheat pastures are an excellent feedstock, especially for young growing cattle. While protein and grain supplements are generally not necessary, an additional form of dry matter roughage is sometimes recommended to ensure ruminants receive enough fiber (Heeney and Stanton, 1988). Winter wheat pastures of the Great Plains provide grazing for millions of stocker cattle each year. If only grazed, the cattle will remain on wheat until May; however, dual-purpose wheat is typically grazed from November to mid-March (Mader et al., 1983). Careful considerations should be made on decisions of when to remove cattle from winter wheat as indicated in a study conducted by Dunphy et al. (1982) to determine the effects of wheat forage removal on grain yield. This study indicated that delaying the final forage harvest could significantly reduce grain yield. The study also suggests that grazing should be terminated by the early jointing stage or sooner to obtain maximum grain yields (Dunphy et al., 1982; Taylor et al., 2010).

Rye

Rye is in the same group *Triticeae* as wheat and barley. Of the total rye grown in the U.S. less than 50% is harvested for grain, the rest would be used for pasture, hay or a cover crop (Oelke et al., 1990). It is commonly grown in parts of Europe as a source of bread, alcohol and animal feed. Unlike most grain crops, rye is a cross-pollination cereal, which allows for greater genetic diversity (Monteiro et al., 2016). In the southern U.S. rye is an important forage crop for stocker cattle (Bos pp.) (Newell and Butler, 2013). Of all of the cool-season grasses, rye is the most winter-hardy and productive on poor soils. It matures earlier than most wheat varieties which results in more fall forage than spring forage (Redmon et al., 1998; Snapp et al., 2005). A previous 1983 study by Hay and Abbas (1993) supported this observation and discovered that the earlier forage and higher yields of rye are due to many factors. These factors include rapid germination rates, crop emergence, leaf appearance and expansion in conjunction with a higher leaf area ratio. It was noted that the early initiation of the rye stem extension showed a significant increase in the total assimilation rate (Hay and Abbas, 1983). Rye has also contributed to the improvement of bread wheat by the chromosomal translocation of 1RS [Rye] chromosome. The 1BL/1RS chromosome from rye provided a race-specific disease resistance to a major rust race (Zarco-Hernandez et al., 2005).

Oat

Oats are grown for both human consumption and livestock grazing. Oats are commonly known for their high levels of beta glucan and the hypocholesterolemic properties of oat fiber that are now well-documented (Lia et al., 1995). Oats can be classed as either winter or spring with the main distinctions between the two classes being time of planting and the ability of winter oats to withstand cold temperatures (King and Bacon., 1992). As an annual forage, oats are very versatile and can be planted in both late summer and late winter for livestock grazing and hay production. One disadvantage of oat production is its lack of winter tolerance compared to other small grains (Redmon et. al., 1998). In the Great Plains region of the U.S., especially in the northern states, such as North Dakota, spring planted oats account for 80% of all cereals managed for hay production. South Dakota had almost 90% and Montana 50% of all cereal land, which was devoted to oat hay production (Carr et al., 2004). In the north central U.S., spring sown forage oats are a common companion crop to aid in the establishment of alfalfa. These oats are typically harvested in the early summer at boot stage for silage (Contereras-Govea, F.E. and Albrecht, K.A. 2006). In the southern U.S., oats are grown in the wintertime and provide excellent forage when other high protein sources are scarce (Schrickel et al., 1992). Oat phenotypic categories determine its use and adaptation. Oats can be considered spring or winter types; however, this is not a very clear distinction as they can have a wide range of vernalization and photoperiod dependence. There is some confusion on the terms of "winter type" and "winter hardy". When discussing a "winter type" of oat, the reference would indicate the proper planting time of that variety which would be in the autumn or winter. Many winter types do not have a vernalization requirement and could be planted in the spring. Several studies have attempted to examine the winter tolerance of oats, but it is not easily distinguishable as that of winter and spring wheat. In a study conducted by M. C. Amirshahi and Fred L. Patterson (1956) the researchers had to rely on uniform winter hardiness nursery trials to obtain winter tolerance information on the oat varieties for their trial (Amirshahi and Patterson., 1956). In a 1960, publication by Pfeifer and Kline the major limiting production factor of winter oats is the lack of winterhardiensss for climates with cold winters. The goal of the study was to develop an accurate laboratory technique for selection for winter hardy oats (Pfeifer and Kline., 1960). In the U.S., traditionally winter oat cultivars are grown in the South during the winter, and spring oat cultivars are grown in the Midwest during the spring. Some researchers have noted a possible cold treatment requirement for the induction of flowering (vernalization) which could also be a factor in determining the type of oat. In a publication by King and Bacon 1992, they noted that a reduction in the number of days to heading of winter oat genotypes was attributed to a vernalization treatment and there was no reduction in the spring oat genotypes as a result of vernalization treatments. In the vernalized (24 day treatment) panicle emergence was reduced by 19 days compared to the non-vernalized treatment (King and Bacon., 1992). In addition there can be hulled and hull-less types which would determine their use (Kim et al., 2014). A 1960 published paper by Smith (1960) evaluated the chemical composition of forage oats in relation to their maturity. He found that with maturity the percentage of protein, fat, ash, P, Ca, K, and nitrate declined. He also noted that the early dough stage is when the highest proportion of total yield of the most important nutritional constitutes was produced (Smith, 1960). Collins et al. (1990) noted that nitrogen concentration decreased with later heading.

Triticale

Triticale (*Triticum secale)* is the result of a breeding cross between wheat (*Titicum*) and rye (*Secale*). The species received its name by combining the name of the two parent species involved in the cross. Triticale is similar to wheat in its pollination strategy as it is a self-pollinated crop as opposed to a cross-pollination species like rye (Oelke et al. 1989). Triticale could be a good source of winter grazing in areas of the U.S. that typically have mild winters (Brown and Almodares, 1976). Triticale is said to do well on sandy soils and has a better tolerance to low pH soils than that of wheat, but not quite as good as rye (Min, 2012). Triticale offers good versatility as a cereal grain and can be used as forage, grain and biofuel feedstock (AGMRC, 2017). In arid and semi-arid regions of the world, triticale can out-yield wheat and barley for straw production, which is a major source of animal feed (Bilgili et al., 2009). A study done in Spain evaluated the dual-purpose possibilities of triticale and evaluated the effects of forage removal on grain yield. This study found that on irrigated sites or sites receiving adequate rainfall both good forage and grain yields could be obtained, however, on dryland or low rainfall sites forage cuttings would not be advised (Ramos et al., 1993). A study by A. J. Ciha (1983) evaluated forage production of triticale as compared to other spring grains. He found that in the Pacific Northwest, crude protein yield of triticale was similar to that of other spring grains under most planting dates and harvest stages (Ciha, 1983).

Ryegrass

In the U.S. there are two types of ryegrass: perennial (*Lolum perenne* L.) and annual (*L. multiflorum* La.). The perennial species is not utilized in the Southern U.S. due to poor adaptation; however, annual species are widely utilized in a forage-livestock system (Blount and Prine, 2000). In the southern U.S., annual ryegrass is considered a high-yielding nutritious grass and is the most widely grown of all cool season annual forage grasses. It is very responsive to N inputs and can tolerate wet, poorly drained soils (Vendramini et. al., 2006). Ryegrass tends to be better established under conventional tillage practices as opposed to no-till systems due to small seed size. Cuomo et al. (1999) emphasized this in a 1999 study, where it was concluded that early–season forage production is reduced when establishment took place on a no-till site verses a conventionally tilled site (Cuomo et al., 1999). As with most crops, nutritive value of ryegrass tends to decrease with maturity. This was emphasized in a 2002 publication that evaluated cultivar and environmental effects on annual ryegrass forage yield, yield distribution, and nutritive value. This study noted that crude protein levels decreased as the growing season progressed (Redfearn et al., 2002). In the lower Mid-West, there is an expressed interest in stockpiling annual ryegrass as a source of high-quality winter forage. Kallenbach et al. (2003) noted a decline in forage quality from mid-December to mid-February. However, the acid detergent fiber (ADF) and the neutral detergent fiber (NDF) remained at an acceptable level (Kallenbach et al., 2003)

Barley

Barley (*Hordeum vulgare*) is not as commonly utilized as a winter pasture in comparison to other small grains as it does not have the winter hardiness of wheat and rye. Barley does have its place in some winter pastures, such as those on alkaline soils that are high in saline content (Redmon et al., 1998). In most parts of the U.S., barley is typically used for malting and other feed sources for animals. There is promising research that suggests an expanded use of barley forage in states such as Texas is possible. Its lower preference as a host by Hessian fly (*Mayetiola destructor*), a significant insect pest of wheat, would be one advantage over wheat in certain regions of the state (Ledbetter, K. 2017). A 1986 study by Brink G.E. and Maren G.C evaluated the effects of barley and oat as a companion crop to alfalfa establishment. For spring-seeded alfalfa, both barley and oat were seeded as companion crops to alfalfa to assist in establishment. The study found that barley provided superior quality at various growth stages than that of oat, which is the traditional companion crop to alfalfa (Brink and

Marten, 1986). Barley as a solo crop can also be valued for its higher nutritive value as compared to other small grains cultivars such as wheat, oat and triticale as indicated in a 1982 study on the biological and chemical quality and yield of small grain forage potentials. This study also found that barley has less grass tetany potential as opposed to wheat, oat and triticale, but it did have a greater milk fever potential (Cherney and Marten, 1982).

In conclusion, wheat is the most popular small grain forage species planted in U.S. It has very good cold tolerance; seed is readily available in most geographic locations and it has great flexibility to be used as a dual-purpose crop for both grazing and grain. Rye, which has great early-season forage, particularly in southern climates, generally has more total forage than wheat also. However, one of the major drawbacks to planting rye is that it can become a major weed problem in areas where wheat is grown for grain. Oats, which have a tendency to produce the earliest season forage of any small grains, are also the least winter hardy. Triticale is the result of a cross between rye and wheat and has earlier maturity than wheat, but later than rye. Likewise, triticale generally produces more forage than wheat, but less than rye. Barley is not as commonly grown in the south due to its lower popularity among livestock producers and its seed sources can be difficult to find in some areas. Still, barley has exceptional tolerance of soils high in soluble salts or sodium and has better drought tolerance (Rohila et al., 2002).

The Importance of estimating forage potential

It is important for both producers and plant breeders to have a good assessment of forage production quality and quantity when evaluating potential forage species and varieties. Producers rely on accurate forage estimates to adjust cattle stocking rates and animal gains. Plant breeders also need accurate forage data to make determinations on genotype advancement for public release of forage cultivars.

Producers

For any producer involved with livestock, forage production is critical. A forage system can assist a livestock manager in some cases with an implementation of a year-round grazing system that utilizes both warm and cool season forages. A key strategy for a successful forage system is to accurately match animal nutrition demand with forage nutrient supply (Redmon, 2003). One important aspect of a good forage system is the maintenance of an accurate stocking rate, which is the specified period of time an expected number of animals can graze on a given acre of land (Hancock, 2011). A Kansas State University forage facts publication illustrated an example of a stocking rate for grazing wheat pastures. This publication stated that the stocking rate for the fall and winter can range from 113 to 226 kilograms of live animal per hectare and increase to 226 to 453 kg per hectare in the spring (Duncan et al., 2018). When comparing forage systems, many factors must be considered to be successful. Four important factors can influence the profitability of the forage system: First, the average daily gain (ADG), which is the expected rate of gain for each animal. Second is the Gain/hectare, which is the expected gain per hectare. Third is the grazing period, which is the number of days when forage can be grazed at a certain stocking rate. Fourth is the stocking rate, which as defined earlier, is the number of animals that can be grazed on a given acre for a certain time. The overall goal is to maintain ADG's approximately 0.68 kg/head/day so that the animals' weight is appropriate to its age. However when stocking rates increase, if an individual animal is unable to select graze for high quality forage it may not be able to meet its nutritional needs (Hancock et al., 2011).

According to Bruckner and Raymer (1990), an important forage management and production decision should be based on the species and cultivar choice for winter forage (Bruckner and Raymer., 1990). In a University of Georgia review on forage systems for stocker cattle, the authors found there are over 60 forage species produced in Georgia alone. Cool season annual performance can vary with location, soil type and management. In addition, forage species vary as to the time they are most productive during the course of a season.

Plant Breeders

Plant variety selection based on phenotypic performance was done long before any type of DNA or molecular markers were available. In order to improve a variety, the best genetic variation would need to be identified. Breeding is a numbers game; the goal is to screen the maximum number of varieties under the maximum number of environments (Araus and Cairns, 2014). Barker and Kalton (1989) stated that there are two important conditions to maximize grassland production: (1) proper management and (2) usage of the proper species and cultivar for a specific climate and purpose (Barker and Kalton, 1989). The latter of the two conditions is where breeding is fundamental.

Selection and improvement of grasses began before humanity itself as prehistoric animals practiced natural selection when grazing on more desired species. It was not until the mid-1930’s that forage grass breeding accelerated (Barker and Kalton, 1989). In the 1960’s, there was a serious effort to genetically improve forage crop nutritive values. Advances in laboratory techniques in analytical chemistry and rumen fermentation technology gave forage breeders the tools to screen thousands of samples in an attempt to improve genetic gains in breeding for forage quality (Casler and Vogel, 1999). There have also been studies conducted to evaluate forage yield. In a 1972 publication that considered genetic variation in yield and quality of oat forage, the authors noted that cultivar variations for forage yield was more consistent than that of forage quality over the course of several years. They concluded that when developing a superior forage oat cultivar, emphasis should be made first on yield and then on forage quality (Stuthman and Marten, 1972). There are direct and indirect economic benefits to forage improvement. The direct benefits would include increased seed and hay sales to the public and the indirect benefit would consist of saleable animal products. The direct and indirect products are a result of improved forages through breeding efforts (Bouton, 2007).

Traditional methods of estimating forage potential

The volume of forage has long been very difficult to accurately measure. Compounding the problem is the fact that the weight of plant material will vary between species. Since all forage in a particular pasture cannot be collected and weighed, sampling is the only way to obtain an estimate of forage yield (Wilm et al., 1944). It is necessary to have some measure of production, availability and consumption of forage in order to gain a better understating of the plant-animal interaction. The plant-animal interaction can determine stocking rates and has a great influence on the pasture output, which in many cases, is the animal product (Bransby et al., 1977).

In 1943, F. A. Coffman first proposed visual forage estimates as a tool evaluating the forage potential of winter oat strains in the U.S. Department of Agriculture Regional Oat Nurseries. His method was to visually rate the experimental lines for leafiness, vigor and overall biomass compared to standard varieties (Atkins et al., 1969). Another study evaluated the accuracy of visual biomass scores and forage yield in red clover (*Trifolium pretense* L.). The study found that visual scores could explain 90% of the variation of actual fresh weights. The evaluators that gave the same plants the same high score was high with an R^2=0.84 (Riday, 1997). Visual rating in space planted red clover seemed very accurate for biomass estimation; however, all forage species may not be so easily evaluated. For this reason, many other methods have been tested throughout the years. For instance, in other crops such as corn (*Zea mays* L.), plant height measurement was a good predictor of plant biomass when collected before reproductive growth. A 2007 study of by-plant prediction of corn forage biomass and nitrogen uptake at various growth stages using remote sensing and plant height concluded that across six site years plant height was a good indicator of plant biomass with an R^2 of 0.81 (Freeman et al., 2007). In the 1970's, a disk meter for measuring yield was used by many researchers. The disk method can either be a simple disk or a weighted disk that is placed on top of the forage and the height of the disk from the ground is measured giving the researcher an indication of forage biomass. This method is effective, yet time consuming and labor intensive; however, the simple disk method has an advantage over the weighted disk, as it is easier to use in the field (Santillan et al., 1979). Many methods can be used to estimate forage biomass in a non-destructive way; however, according to Harmoney et al., (1997) cutting and weighing forage from known areas is the most accurate method for determining forage biomass (Harmoney et al., 1997).

Researchers have discovered adequate ways of measuring biomass; however, these methods can be time consuming and labor intensive. Several studies have indicated that forage cutting to measure yield is costly and destructive; in addition, the demands of personnel and funding did not allow reliable information to be gathered, which could lead to sampling errors in research trials (Wilm et al., 1944; Haydock and Shaw, 1975; Sanderson et al., 2001). To solve some of these problems more recent studies have been conducted to modernize the way forage biomass can be measured (Freeman et al., 2007; Andales et al., 2006; Fricke et al., 2011). The now relatively low cost of remote sensing tools and techniques could be utilized to address many of the problems addressed earlier with the traditional methods of predicting forage yields.

Nondestructive method of estimating forage by use of UAV visual spectrum reflectance and primitive NDVI meters

The introduction of a "vegetation index" came from the first NASA ERTS satellite better known as Landsat 1 in 1972. Landsat 1 was equipped with a multi-spectral scanner to evaluate earth remotely, which could sense such things as spring green-up and fall dry down. Through Landsat 1 research, the development of a ratio that used the difference of the red and infrared radiances over their sums to normalize was developed. This vegetation index, Normalized Difference Vegetation Index (NDVI), is one of the most successful and simple ways to identify live green plant material in a remotely sensed way (LDP, 2015). Precision agriculture systems have been developed as a means to improve profitability and productivity of producers. This system requires high-resolution information, which will enable producers to better manage inputs (Elarab et al., 2015).

Many studies have evaluated the relationship between remotely sensed data and forage biomass (Serrano et al., 2000; Freeman et al., 2007; Tucker, C. J., 1980; Anderson et al., 1993). In a 2002 publication, the relationship between growth traits and spectral vegetation indices in durum wheat (*Triticum turgidum* var.duram) was evaluated. The study aimed at determining if vegetation indices (VI) could identify total dry matter (TDM) and leaf area index (LAI) in durum wheat, and in turn aid selection in a breeding program. The study determined the best growth stage for trait appraisal were Zadoks 65 and 75. The final results were mixed and determined that VI could track changes in the LAI during the growing season at different growth stages, environments and genotypes but was limited due to the lack of predictive ability for the combination of growth stage and environment (Aparicio., et al. 2002). In a 2016 study, Possoch et al. (2016) evaluated the use of multi-temporal crop surface models that were combined with the RGB vegetation index (VI) for forage monitoring in grasslands. The study found that the RGB vegetation index (RGBVI) was not a good estimator of biomass, but if combined with plant heights, medium correlations ($R^2 = 0.50$) could be obtained. In a 2012 study, the relationship between biomass, percent groundcover and remote sensing indices across six winter cover crops was evaluated. A 16-band CROPSCAN sensor was used to obtain ten vegetation indices. The study concluded that there was a strong relationship between NDVI and groundcover with an R^2 of 0.93. The most accurate index for measuring high ranges of biomass was the triangular vegetation index (TVI) with an R^2 of 0.86. In the low to medium biomass range the NDVI did not have a clustering of values; however, there was saturation in the higher ranges (Prabhakara et al., 2015). NDVI has had some criticisms for some of its perceived defects: (1) the differences between "true" NDVI measured at the surface and NDVI measured from space due to atmospheric interference, (2) the sensitivity of NDVI to LAI (leaf area index), which becomes weak with a rising LAI, and (3) soil brightness variations, which can cause variations in NDVI from one image to the next (Carlson and Ripley, 1997). Other problems with NDVI have been attributed to saturation issues in dense vegetation (Mutanga et al., 2004). One possible solution to the saturation issue would be to use the Red Edge peak, which is defined as the point of maximum slope in the vegetation reflectance spectra (680-750nm). This is the point where reflection changes from very low in the red region of the spectra to very high NIR in chlorophyll absorption. This phenomenon is a result of leaf and canopy scattering (Filella and Penuelas., 1994). This idea supports the results of the a study conducted by Mutanga et al (2004) where they indicated during high canopy density the best estimation of biomass can be discovered by using vegetation indices that are based on wavelengths near the red edge rather than using NDVI (Mutanga et al., 2004). Several studies have indicated a good correlation between plant biomass from models obtained between NDVI readings and from primitive handheld devices and plant height of various species (Andersson et al., 2017, Flynn et al., 2008, Freeman et al., 2007).

The use of VI has long been applied to biomass estimation; however, the most popular VI would traditionally use the near infrared region (NIR) with a light reflectance range of 700 to 1300 nm (Bendig et al., 2015). To obtain the NIR band would require the use of a more expensive four band multispectral camera; however, a more cost-effective approach would be the utilization of a UAV mounted three band camera which would only take into account the Red (R), Green (G) and Blue (B) bands (400-700nm) (Bendig et al., 2015, Lessem et al., 2017, 2018). There has been some research into the use of visible band VI that has shown potential to be an effective method of biomass estimation; however; further investigation would be needed (Lussem et al., 2017, 2018, Tilly et al., 2015, Bendig et al., 2015, Possoch et al., 2016). A study conducted by Motohka et al (2010) concluded that visual spectrum VI's can be useful in detecting the early phase of leaf green-up, but may be limited to certain growth stages (Motohka et al., 2010). This may be useful when evaluating a multi-clip forage trial. There has not been a great deal of research on the best flight altitude for biomass screening. In a 2010 study, by Swain et al set out to determine the yield and total biomass of a rice crop by utilizing a low altitude unmanned helicopter, the UAV was flown at 20 meters over the experimental plots and had very good results when compared to satellite images for estimating some biomass parameters (Swain et al., 2010).

CHAPTER II

COMPARING MECHANICAL HARVEST WITH ALTERNTIVE GROUND BASED METHODS FOR ESTIMATING FORAGE YIELDS IN COOL SEASON ANNUAL GRASSES

Introduction

The United States (U.S.) is one of the largest forage producing nations in the world. According to the U.S. Department of Agriculture (USDA-NAAS, 2017) census, there were 23 million hectares of land in the U.S. used for forage, which contributed to the nutritive needs of over 93.6 million beef cattle and calves alone. Texas is the largest forage producing state in the U.S. with over two million hectares dedicated to forage production (USDA-NASS, 2018). Some of the most common cool-season annual forage species in the southern U.S. are small grain crops, which include wheat (*Triticum aestivum*), rye (*Secale cereal*), oats (*Avena sativa*) and triticale (*Triticum secale*) (Bruckner and Raymer, 1990). In addition to these, barley (*Hordeum vulgare*) is another small grains species that is less utilized, but can produce good quality silage and hay similar to that of oats and triticale (Min, D.H., 2012). Annual ryegrass (*Lolium multiflorum*) is not considered a small grain, but is a cool-season annual grass that provides a source of high quality winter and spring forage in the temperate regions of the world (Hill and Gates, 2001). Cool-season grasses offer producers valuable production and management alternatives as sources of forage, which utilize similar planting and harvest equipment as that of alfalfa (*Medicago sativa*).

Wheat is one of the most popular small grains forage species planted in the U.S., due in large part to its excellent cold tolerance and broad adaptation. Furthermore, wheat seed is readily available in most geographic locations and it typically performs well in grain, grazing or dual-purpose cropping systems giving producers flexibility in their operations (Redmon et al., 1995, Hossain et al., 2003). Rye, which has great early-season forage, particularly in southern climates, generally has more total forage than wheat. However, rye can become a major weed problem in areas where wheat is grown for grain (Roberts et. al. 2001), limiting its widespread adoption to certain regions where pasture and hay production dominate the agriculture landscape. Oats tend to produce the earliest season forage of any small grains, but are the least winter hardy, which limits their use as a winter forage to only the most southern states (Stichler and Livingston, 1998). Triticale is the result of a cross between rye and wheat and has earlier maturity than wheat, but later than rye. Triticale generally produces more forage than wheat, but lower forage than rye. Barley is not as commonly grown in the southern U.S. because of limited access to seed sources in some areas and it is more winter tender than wheat. However, barley has exceptional tolerance of soils high in soluble salts or sodium and has better drought tolerance (Rohila et al., 2002, Nevo and Chen, 2010).

The ability to accurately estimate forage yield of experimental lines and varieties without the time and expense of harvesting with and maintaining a plot harvester would aid cool-season forage grass researches and breeders in variety selection and advancement. For researchers and managers alike, it is important to know which species and genotypes of cool-season grasses produce the most forage in a given environment.

To ascertain this information, many universities conduct cool-season annual forage trials to determine which species and genotypes produce the most yield in their respective regions. The publications that result from these forage trials are aimed at giving unbiased yield and quality data to researchers and forage managers so they can make educated decisions regarding the most appropriate varieties for their geographic region (Neely et al., 2016).

In an earlier study on the effectiveness of visual estimations for forge potential in oat and barley species, the researchers compared visual rating of forage potential against actual forage yields. The study found good correlations from a January evaluation when compared to actual yield. Interestingly, when the estimations from January were combined with a second estimation in March, the correlations increased when compared to total yields for the season (Atkins et al., 1969). Another study by Riday (1997), the accuracy of visual biomass scores on estimating forage yield in red clover (*Trifolium pretense* L.) was evaluated. The evaluators that gave the same plants the same high score was high with an R^2=0.84. This study found that visual scores could explain 90% of the variation of actual fresh weights (Riday, 1997).

Although visual rating is a reasonably accurate method for biomass estimation, all forage species may not be so easily evaluated. For this reason, many other non-destructive methods have been tested throughout the years such as plant height (Freeman et al., 2007) and disk meter (Santillan et al., 1979) with success. While these methods were successful, cutting and weighing forage from a known area is the most accurate method for determining forage biomass. Unfortunately, the up-front and routine maintenance costs associated with a mechanical forage harvester can be cost-prohibitive to physically harvest entire test plots (Sanderson et al 2001).

The objective of this research was to compare mechanically harvested aboveground biomass yield of cool-season annual grasses with alternative forms of non-destructive and limited destructive measurements of forage plot yield and to determine which method works best for each species and clip time. A less labor intensive method of estimating plot forage would aid breeders and researchers in evaluating more varieties or experimental lines more efficiently.

Materials and Methods

A cool season annual grass forage trial was conducted at the Texas A&M Research Farm near College Station, TX (30°31'N 96°25'W) during the 2015-2016 (2016) and 2016-2017 (2017) growing seasons. The climate of the location is humid subtropical. The 30-year average annual temperature is 20.6 °C and growing season (October-May) temperature is 19.0°C (College Station 2020). The 30-year long-term average annual precipitation is 1018 mm and growing season precipitation is 435 mm (College Station, 2020). The highest precipitation generally occurs in the months of May, June, and October. Plots for both years were located on a Ships clay (*Udertic Hapluderts*) soil type. The trial was set up in a randomized complete block design (RCBD) with four replications of 37 entries in 2016 and 38 in 2017. Total number of plots in the 2016 was 148 and 2017 152. Each plot measured 6 m in length and 1.5 m in width. The trial included commercial and experimental wheat, oat, rye, triticale, barley and ryegrass cultivars. All plots in the trial were treated with Cruiser Maxx Vibrance for Cereals to protect from seed borne and soil borne diseases and insects (e i. Thiamethoxam, Mefenozam, Fludioxonil and Sedazan) (Syngenta, Basel, Switzerland). All small grains plots were planted at a rate of 106 kg seed ha^{-1}. Ryegrass was planted at 28 kg seed ha^{-1}. Row spacing was 19 cm for both small grains and Ryegrass.

In the first growing season (2016), all plots were planted on October 1, 2015. Treatments included 10 wheat, 12 oat, 3 rye, 2 triticale, 7 ryegrass and 3 barley cultivars (total 37 treatments). In the second growing season (2017), the study was planted on September 29, 2016. The study included 9 wheat, 12 oat, 3 rye, 2 triticale, 9 ryegrass and 3 barley cultivars (total 38 treatments). In both growing seasons, temperatures were normal for the year. However, rainfall was above average in 2016 growing season and the trial received 632 mm of rain from October until May. In 2017, 273 mm of rain was received during the growing season.

Nitrogen fertilizer was applied in both growing season. A total of 101 and 102 kg N ha^{-1} were applied as liquid urea ammonium nitrate (UAN 32-0-0) in the first and second years, respectively, on January 20, 2016 and December 2, 2016. The entire plot was harvested a total of three times per year with a small plot Haldrup 1500 forage harvester (Haldrup, Ilshofen, Germany) which is equipped with an onboard weigh system and 1.5 m header.

Forage clipping was done three times during the growing season. In 2015-16, clippings were done on January 19, March 28, and April 26, 2016. The clipping dates were November 30 (2016), February 8 and March 3, 2017. Clippings times were categorized into four groups based on time of year: fall (November), mid-winter (January and February), early spring (March) and late spring (April) (Appendix 1). There was only one fall clipping in this study (November 30, 2016) from the 2016-17 growing season due to an early planting date and good moisture conditions at planting. Both the mid-winter and early spring clip times included clips from both growing seasons. Due to above average spring rainfall during the 2015-16 growing season, there was a late spring clipping (April 26, 2016), which did not occur in year two.

Visual rating, plant height and subsample weights (1m linear row) were collected within 24 hrs. immediately prior to mechanical harvest (clipping) of the full plot . Three plant height measurements were recorded per plot using a meter stick. Visual rating was conducted for each plot on a 0-10 scale where 0 indicated no forage potential and 10 indicated high forage potential. The visual rating was based on a visual assessment that took leafiness, height, and overall vigor into consideration to compare treatments. Following plant height and visual scoring, a 1 m linear row random subsample was collected from a single row within each plot. The subsample was cut 5 cm from the soil level to simulate cutting height of the mechanical harvester. Subsamples were weighed immediately following harvest to document fresh (wet) weight prior to drying. Dry weights were recorded after drying in the oven at 50°C for three days. Percent moisture at harvest was then calculated from the two weights (Eq. 2.1) and dry matter percentage was used to calculate dry matter yield from mechanical harvest.

$$\text{\% Dry Matter} = \text{Dry Weight} / \text{Wet Weight} * 100 \qquad \text{[Eq. 2.1]}$$

Statistical analysis was conducted using SAS 9.4 statistical software (SAS Institute, Cary, North Carolina). PROC CORR was used to run Pearson's correlation analyses between plant height, visual rating, subsample dry weight and total plot dry forage yield. This was done for individual species as well as all species combined to assess if certain measurements were better correlated with forage yield for certain species. Correlations were also run for individual species and across species for individual clip times to assess if certain measurements were better correlated with forage yield for certain times of the year. PROC REG was used with a backward stepwise regression (alpha = 0.05) to develop a biomass prediction model using plant height, visual scores, and sub sample weight as possible variables within the model.

Results and Discussion

Visual Rating

Correlation analysis revealed a wide range in relationships between visual rating and dry matter biomass for each specie and clip time combination (Table 1). While number of observations ranged by specie from eight (triticale, fall clip) up to 95 (oats, mid-winter and early spring clips), increasing the number of observations did not always improve correlations as both wheat (n=35) and rye (n=12) were not significant. Instead, the range in biomass played a bigger role. A good example of this is the difference in biomass range between the early spring clip time of Rye (n=24) and Barley (n=20). The correlation between visual rating and forage yield was much greater for Barley (r=0.92, p>0.0001) which had a range in biomass yield between 322 to 1770 kgha-1 compared to

Rye ($r=0.39$, N.S.) which had a smaller range in biomass yield between 320 to 817 kgha-1. Overall, the highest correlation between visual rating and forage yield was achieved for barley ($r=0.92$, $p<0.001$) during the early spring clip. Wheat, rye, ryegrass, and barley all had at least one clip time throughout the season that was not significant, though rye was consistently the poorest.

For individual species, the visual rating correlated the best with yield during the fall clip for triticale ($r=0.84$, $p<0.01$), mid-winter clip for rye ($r=0.43$, $p<0.05$), early spring clip for oats ($r=0.78$, $p<0.001$) and barley ($r=0.92$, $p<0.001$), and late spring clip for wheat ($r=0.60$, $p<0.001$) and ryegrass ($r=0.83$, $p<0.001$). Interestingly, when species were combined by clip, the fall ($r=0.84$, $p<0.001$) visual rating had the highest correlation to yield indicating this was a reliable method to estimate relative forage yield among entries at this time of the year while In fact, correlations between visual scores and forage yields appeared reliable for early and late spring clippings as well, though the mid-winter clipping correlation ($r=0.39$, $p<0.001$) was weak. Intuitively, this makes sense as mid-winter clips are generally lower yielding and with less variation among plots. It is more difficult to detect a relationship when little variation exists. Based on this study, visual scoring would be a viable option for ranking trial entries for forage potential, but not a good method for actually predicting yield since the scoring system is relative to other entries only within a specific clip..

When data were combined across clip times for each specie, ryegrass had the best correlation between visual rating and yield ($r=0.62$, $p<0.0001$) while rye had the lowest correlation ($r=0.39$, $p<0.001$). When all data were combined across species and clip times the visual rating only had a correlation of r=0.50 ($p<0.001$). A scatter plot between visual rating and yield across all species and clip times revealed an exponential relationship between the two measurements (y = 42.659e0.3553x) with an $R^2=0.44$ (Figure 2.1). Still, compared to subsampling, the relationship is not as strong. This weaker relationship could be attributed to the method in which the plots were visually scored at each clipping. When visually rating a plot, its score was standardized based on a reference entry in the trial. In this instance, oat entry ''TAMO 411' (Ibrahim et al., 2018) was given a visual rating and all other plots were scored above or below that rating based on whether they looked better or worse. This may lead to accurate ratings per clip, but would not transcend clip times as a score of eight for TAMO 411 in a fall clipping yielded 4265 kg ha-1 whereas a late spring clipping with a score of eight only yielded 919 kg ha-1. This particular genotype yielded very well in both the fall and late spring clippings in relation to other entries; however, this illustrates the tendency of an observer to standardize the rating based on the surrounding entries in an individual clipping.

In Table 2.1, the visual rating for individual clip times were better in general than the combined species correlation across all clips. Similarly, Riday et al (2009) achieved very good results ($R^2=0.79$, $p<0.0001$) when evaluating red clover plants on an individual basis to compare visual biomass scores with actual forage yield where measurements were taken only once for the growing season at two different locations. In another study by Atkins et al (1969), visual scores were used to estimate forage yield of oat and barley species, which showed a slight increase in coefficient of determination from r=0.40 to r=0.50 for individual species and clip times to r=0.55 to r=0.66 when combining clip times and species.

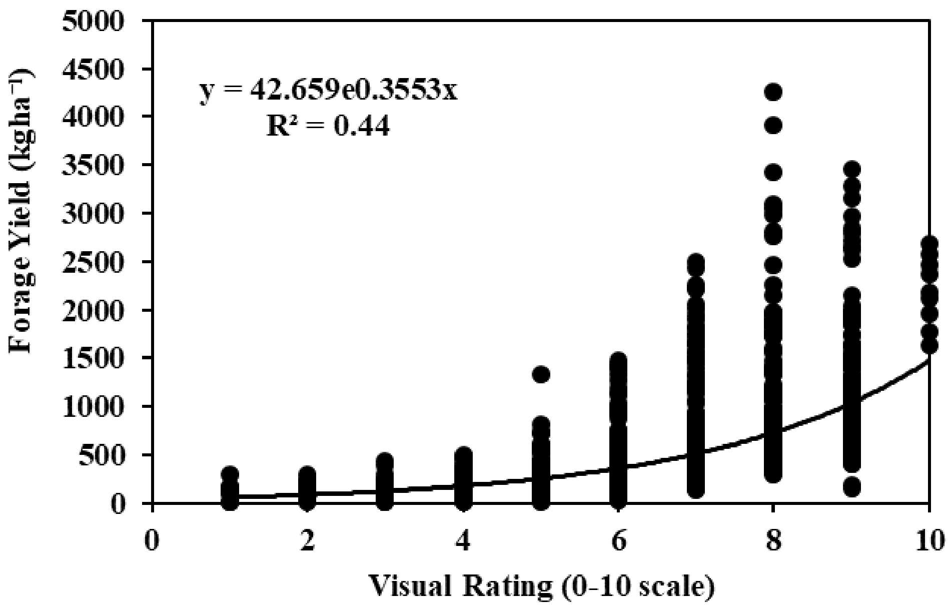
Table 2.1 Pearson Correlation coefficients and number of observations (n) between visual rating and dry matter forage yield for six species and four clip times in a cool season annual forage trial in College Station, TX across the 2016 and the 2017 growing season.

	Clip Time				
	Fall	Mid-winter	Early spring	Late spring	Combined
Wheat	0.32^{NS} n=35	0.50^{***} n=75	0.49^{***} n=75	0.60^{***} n=40	0.50^{***} n=225
Oat	0.53^{***} n=48	0.31^{**} n=95	0.78^{***} n=95	0.42^{**} n=48	0.46^{***} n=288
Rye	0.50^{NS} n=12	0.43^{*} n=25	0.39^{NS} n=24	0.56^{NS} n=12	0.39^{***} n=72
Ryegrass	0.71^{***} n=35	0.20^{NS} n=63	0.53^{***} n=63	0.83^{***} n=28	0.62^{***} n=189
Triticale	0.84^{**} n=8	0.57^{**} n=20	0.78^{***} n=20	0.83^{***} n=12	0.56^{***} n=60
Barley	0.75^{**} n=12	0.61^{**} n=20	0.92^{***} n=20	0.40^{NS} n=8	0.50^{***} n=60
Combined	0.84^{***} n=150	0.39^{***} n=298	0.66^{***} n=298	0.77^{***} n=148	0.50^{***} n=894

*, **, *** Correlation coefficient significant at the 0.05, 0.01, and 0.001 probability levels.

NS, non-significant at $P \geq 0.05$.

Figure 2.1 Relationship between visual rating and total plot dry matter forage yield across clip times and species. Visual Rating was determined using a scale of 0-10, where zero equals no forage yield. Data was collected from a cool-season annual forage trial with six species and taken across four clip times [fall (November), mid-winter (January and February), early spring (March) and Late Spring (April)] during the 2016 and 2017 growing seasons in College Station, TX.



Plant Height

Correlation analysis revealed generally low correlation between plant height and dry matter forage yield for most individual species when combined across clip times; however, coefficient of determination values were much higher for individual clip times when combined across species ranging from r=0.52 (early spring) up to r=0.81 (fall) (Table 2.2).The highest height correlation to yield by species occurred for triticale

($r=0.95$ $p<0.001$) in the fall, wheat ($r=0.69$ $p<0.001$)) and barley ($r=0.87$ $p<0.001$) in mid-winter, oat ($r=0.80$ $p<0.001$) in early spring, and rye ($r=0.77$ $p<0.01$) and ryegrass ($r=0.67$ $p<0.001$) in late spring. As was the case with visual rating, plant height had the best correlation with forage yield for the fall clipping ($r=0.81$ $p<0.001$) (Table 2.2). Plant height could be used to determine forage production rankings for individual clips throughout the growing season, but would not be useful by itself in predicting forage production without the context of growth stage or clip time. Barley had the highest correlation between plant height and yield among the different species when data was combined across all clip times ($r=0.37$, $p<0.01$) while wheat, oat, rye, and triticale had no significant correlation when combined across clip times.

When all data was combined across species and clip times, plant height had a low correlation of $r=0.15$ ($p<0.001$). This poor correlation can likely be attributed to the different growth stages that small grains experience throughout the season. In the fall and winter, cool-season annual grasses remain in the vegetative stage where they tiller and put on only vegetative, leafy growth. Certain varieties also have more prostrate growth than others, the degree of which may be independent of total biomass production. This can reduce correlations among plant height and forage biomass. As noted in studies of other species such as corn, individual plant height measurements were good indicators of biomass during early stages of growth as opposed to the later stages (Freeman et al 2007). Unlike corn, winter small grains species do not begin their reproductive growth cycle until spring, at which time they begin stem elongation and eventually produce reproductive seed heads. During this stage of growth, small grains and ryegrass accumulate biomass rapidly.

Once forage is removed during or after this stem elongation, plants will attempt to recover and shoot up smaller, thinner stems in an effort to produce seed. Hence, many times in late spring plants may be taller than in the fall, but produce less biomass. As a result, height was well correlated with yield at each growth stage or clip time, but led to poor correlation when data was combined across clip times. A good example from this study is based on winter wheat variety 'TAM 401' (Rudd et al., 2012) which had a fall plant height of 31 cm and a yield of 1629 kg h-1 compared to a plant height of 43 cm in the late spring clip with a yield of only 742kg h-1 of dry matter.

Figure 2.2 shows the relationship between plant height and dry matter forage yield across all species separated out by clip time. The highest association with yield was in the fall with an R^2=0.65 across species, followed by late spring (R^2=0.55) and mid-winter (R^2=0.46). The lowest association with yield was in the early spring (R^2=0.27) (Figure 2.2).

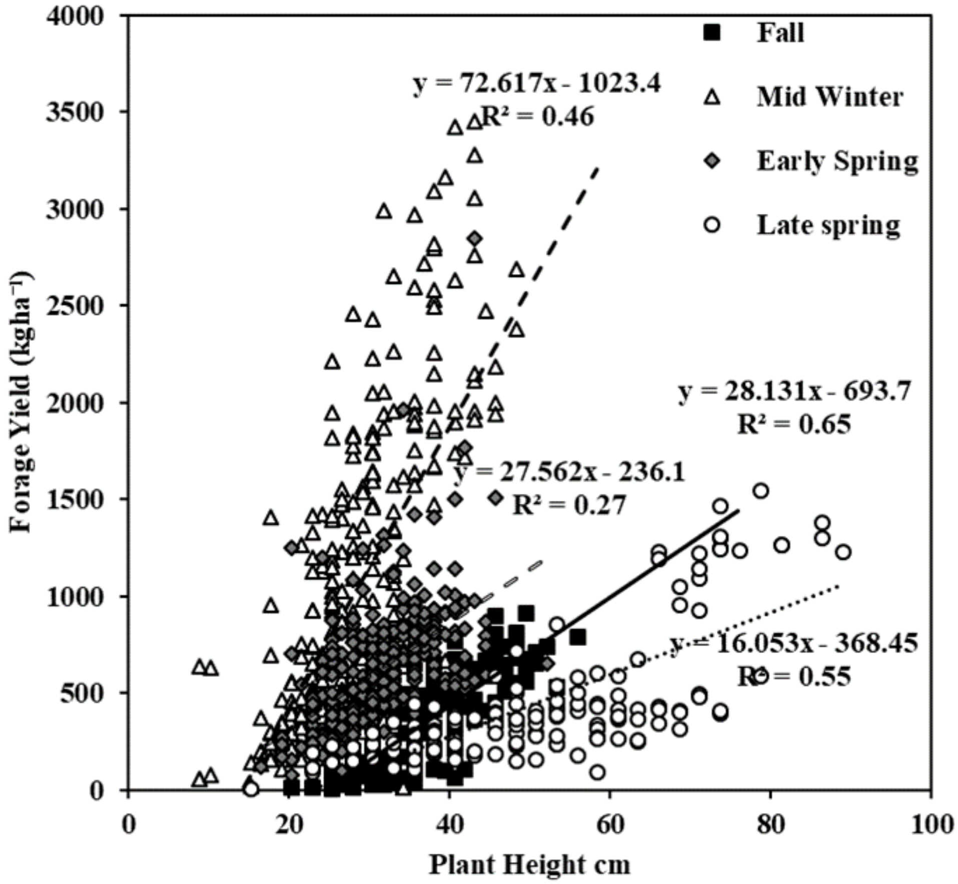
Table 2.2 Pearson Correlation coefficients and number of observations (n) between plant height and dry matter forage yield for six species and four clip times in a cool-season annual

	Clip Time				
	Fall	Mid-winter	Early spring	Late spring	Combined
Wheat	0.21^{NS} n=35	0.69^{***} n=75	0.54^{***} n=75	0.56^{***} n=40	0.05^{NS} n=225
Oat	0.43^{**} n=48	0.64^{***} n=95	0.80^{***} n=95	0.23^{NS} n=48	0.09^{NS} n=288
Rye	0.61^{*} n=12	0.69^{***} n=25	0.64^{***} n=24	0.77^{**} n=12	-0.06^{NS} n=72
Ryegrass	0.53^{**} n=35	0.41^{***} n=63	0.40^{**} n=63	0.67^{***} n=28	0.30^{***} n=189
Triticale	0.95^{***} n=8	0.53^{*} n=20	0.73^{***} n=20	0.65^{*} n=12	0.00^{NS} n=60
Barley	0.37^{NS} n=12	0.87^{***} n=20	0.79^{***} n=20	0.77^{*} n=8	0.37^{**} n=60
Combined	0.81^{***} n=150	0.67^{***} n=298	0.52^{***} n=298	0.74^{***} n=148	0.15^{***} n=894

*, **, *** Correlation coefficient significant at the 0.05, 0.01, and 0.001 probability levels.

NS, non-significant at $P \geq 0.05$.

Figure 2.2 Relationship between plant height and total plot dry matter forage yield for four clip times using data collected from a cool-season annual forage trial with six species and taken across four clip times [fall (November), mid-winter (January and February), early spring (March) and Late Spring (April)] during the 2016 and 2017 growing seasons in College Station, TX.



Sub sample measurement

The subsample method had the highest correlation to yield across all species and clip times of all the methods evaluated in this study (r=0.83 p<0.001). Indeed, an R2=0.68 was achieved when subsample weight was plotted against total plot forage yield, which is better than that of both visual rating and plant height (Figure 2.4). Most species had the highest correlation to yield in the mid-winter clipping with the exception of ryegrass and triticale, which had their highest correlations during the early spring

($r=0.78$ $p<0.001$) and fall clippings ($r=0.79$, $p<0.05$), respectively (Table 2.3). This was expected as ryegrass is slow to establish in the fall and correlations are more difficult to establish when there is little variability among forage yields. Ryegrass produced abundant vegetative growth later in the season leading to the better correlation at that time. Interestingly, the fall clip had the poorest correlation for all but Triticale, but the fall clip time had the highest correlation when all species were combined ($r=0.81$ $p<0.0001$) (Table 2.3).

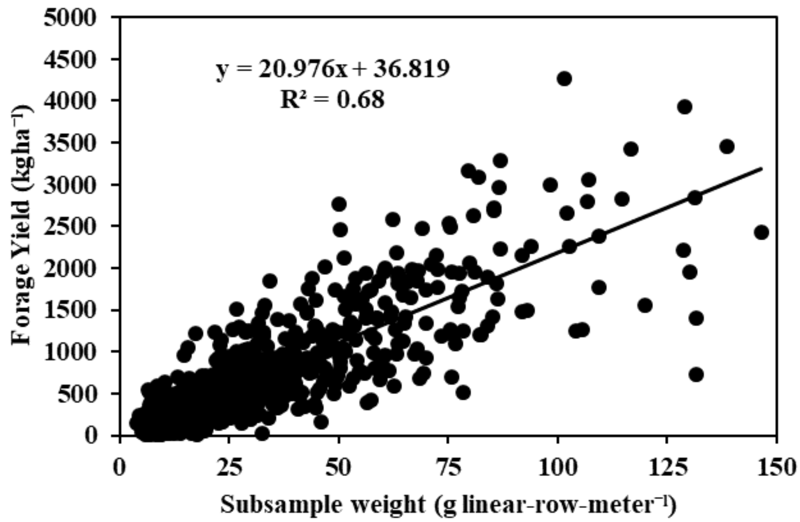
Table 2.3 Pearson Correlation coefficients and number of observations (n) between sub sample dry weight and total plot dry matter forage yield for six species and four clip times in a cool season annual grass forage trial in College Station, TX across the 2016 and 2017 growing seasons.

	Clip Time				
	Fall	Mid-winter	Early Spring	Late Spring	Combined
Wheat	0.41^{*} n=35	0.78^{***} n=75	0.70^{***} n=75	0.62^{***} n=40	0.83^{***} n=225
Oat	0.41^{**} n=48	0.82^{***} n=95	0.72^{***} n=95	0.39^{**} n=48	0.86^{***} n=288
Rye	0.37^{NS} n=12	0.87^{***} n=25	0.54^{**} n=24	0.55^{NS} n=12	0.81^{***} n=72
Ryegrass	0.36^{*} n=35	0.70^{***} n=63	0.78^{***} n=63	0.74^{***} n=28	0.71^{***} n=189
Triticale	0.79^{*} n=8	0.77^{***} n=20	0.35^{NS} n=20	0.76^{***} n=12	0.85^{***} n=60
Barley	0.32^{NS} n=12	0.91^{***} n=20	0.83^{***} n=20	0.58^{NS} n=8	0.91^{***} n=60
Combined	0.81^{***} n=150	0.80^{***} n=298	0.77^{***} n=298	0.69^{***} n=148	0.83^{***} n=894

*, **, *** Correlation coefficient significant at the 0.05, 0.01, and 0.001 probability levels.

NS, non-significant at $P \geq 0.05$.

Figure 2.3 Relationship between subsample dry weight and total plot dry matter forage yield across clip times and species. Data was collected from a cool-season annual forage trial with six species and taken across four clip times [fall (November), mid-winter (January and February), early spring (March) and Late Spring (April)] during the 2016 and 2017 growing seasons in College Station, TX.



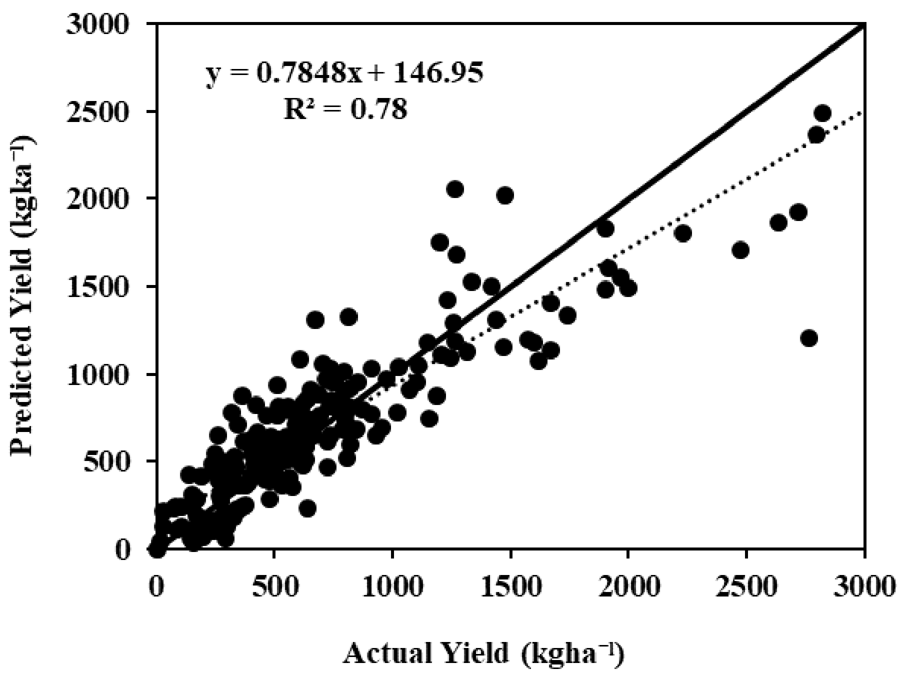
All species achieved good correlations between subsample harvesting and full plot forage yields when combined across clip times and ranged from r=0.71, p<0.001 (ryegrass) to r=0.91, p<0.001 (barley). Correlations were also strong when combined across species by clip and ranged from r=0.69, p<0.001 (late spring) to r=0.81, p<0.001 (fall). Rye showed the most variability in correlations across clip times, which were only significant for the mid-winter (r=0.87, p<0.001) and early spring clippings (r=0.54, p<0.01). Rye matures more rapidly than the other species in the trial, and subsequently, it only produced very sparse growth during the late spring because it had already headed out and was nearing the end of its life cycle. This could explain the poor late spring correlation.

The data strongly indicates that the subsampling method is more strongly correlated to whole plot dry matter forage yield compared to visual scoring or plant height when combining data across species and clip time for cool-season annual grasses. This is supported by the work of Martin et al. (2005) in which they concluded that the standard quadrat harvesting (subsampling) was the most reliable method to estimate forage biomass in a mixed species trial. In fact, this method could be more accurate than the whole plot harvest to which the subsample yields are compared to. While whole plot forage harvest with a forage plot harvester is the industry standard, it can also have its inaccuracies due to factors such as: non-uniform cuts across the plots, unbalanced scales, scales affected by wind and sparse forage in the early fall and late spring that can be difficult to mechanically harvest.

Prediction Model

While subsampling appeared to be the best method for estimating full plot forage yields, correlations could still be stronger and indeed, they were not significant in every instance. Therefore, a prediction model for dry matter forage yield was developed using 669 observations from replications 1, 3 and 4 across all clip times. A backwards stepwise regression was conducted using the following variables: plant height (HT), visual ratings (VR) and subsample weights (SUB). Using this method, all variables were found to be significant and the following prediction model was developed : Yield= (-590.563+51.62051*VR+8.31458*HT+20.42425*SUB). Using this model, predicted yields were plotted against actual yields of replication 2, which produced an adjusted $R^2=0.78$ ($p<0.0001$) and RMSE 258.14 (Figure 2.4). This was an improvement over the stand-alone subsample method with an R^2=0.68 ($p<0.0001$) and RMSE 289.42 (Figure 2.3). A separate model using only plant height and visual rating was created [Yield = (-635.58294+176.44599*VR+3.59274*PH], which generated a much lower adjusted R^2 value (R^2=0.29 $p<0.0001$) and RMSE 468.93. Personal preference would dictate whether the added level of accuracy of taking visual ratings and plant height is worth the extra time and effort beyond utilizing only the subsample method.

Figure 2.4 A graphical representation of predicted forage yield compared to actual forage yield from a model developed using plant height (HT), visual ratings (VR) and subsample dry weights (SUB). Observations used in the model were collected from a cool-season annual forage trial with six species and taken across four clip times [fall (November), mid-winter (January and February), early spring (March) and Late Spring (April)] and the 2016 and 2017 growing seasons in College Station, TX.



It is clear that subsampling was a better method to estimate or rank forage potential of entries in this trial; however, subsampling is still time and labor intensive.

For plant breeders who evaluate thousands of lines, it may not be a practical method for comparing forage yield of breeding lines. In this case, visual scoring or plant height may be useful, but not as accurate as subsampling. Breeders could then utilize this information for the advancement of cultivars from very large observational or preliminary nurseries for forage production.

Conclusion

When evaluating a multi-species forage trial over several clippings throughout the year, the model created in this study using all three measurements was better at predicting forage yield than any individual method alone. However, the subsample method was consistently better than either visual rating or plant height measurements and would be the only acceptable method for comparisons of entries across clip times and species.

If researchers are unable to take destructive samples, both visual and plant height measurements were highly correlated for fall and late spring clips. It is less clear which would be best throughout the entire season though, as visual rating was poorly correlated for the mid-winter clip and plant height was inferior to visual rating for early spring. In general, it would not be advisable to rely solely on visual rating or plant height to make comparisons within a specie for forage yield across all clip times, as correlations were generally low to moderate. Combining all measurements into one model to predict forage yield may be a better way to estimate forage yield if whole plot forage harvest is not possible. A less labor and cost intensive method of estimating plot forage would aid breeders and researchers in evaluating more varieties or lines more efficiently.

CHAPTER III

COMPARING NONDESTRUCTIVE AERIAL AND GROUND METHODS OF ESTIMATING FORAGE YIELD IN COOL SEASON ANNUAL GRASSES USING UAV VISUAL SPECTRUM REFLECTANCE INDICIES, VISUAL EVALUATION AND PRIMATIVE NDVI METERS

Introduction

The United States (U.S.) is one of the largest forage producing nations in the world. In fact, the USDA estimated that total U.S. forage production (all dry hay, haylage and green chop) in 2016 was 82.3 million metric tons (USDA NASS 2016). As a result, there is interest in developing more modern and efficient ways of evaluating breeding lines and cultivars of different forage species for forage biomass. Some of the more traditional methods, such as visual rating and subsampling have been around for over 50 years and are labor intensive. Having a good indication of how much yield potential a particular forage species and cultivar has is useful for livestock managers who rely on good forage production to meet the high demands of their forage systems. It is also important for researchers in forage and small grains breeding programs who need good forage potential data for cultivar advancement.

In the southern U.S., cool-season annual grasses can extend grazing and supply high quality forage for livestock (Bagley et al., 1998). This makes the use of cool season annual grasses the most common form of winter pasture (Corriher-Olson and Redmon, 2013). Some of the most common cool season annual forage species in the southern

U.S. are small grains, which include wheat (*Triticum aestivum*), rye (*Secale cereal*), oats (*Avena sativa*) and triticale (*Triticum secale*) (Bruckner and Raymer, 1990). In addition to these more common southern species, barley (*Hordeum vulgare*) can produce good quality silage and hay similar to that of oats and triticale (Min, D.-H. 2012). Annual ryegrass (*Lolium multiflorum*) is not considered a small grain, but provides a source of high-quality winter and spring forage in the temperate regions of the world (Hill and Gates, 2001). The forage season usually stretches from October to May in Texas where this trial was conducted.

It is important for both livestock managers and plant breeders to have a good assessment of forage production quantity when evaluating potential forage species and varieties. For any livestock manager forage production is critical. A forage system can assist a livestock manager in some cases with an implementation of a year-round grazing system that utilizes both warm and cool season forages (Redmon, 2003). Producers rely on accurate forage estimates to adjust cattle stocking rates and animal gains. Plant breeders also need accurate forage data to make determinations on genotype advancement for public release of forage cultivars. Plant variety selection based on phenotypic performance was done long before any type of DNA or molecular markers were available. In order to improve a variety, the best genetic variation would need to be identified. Breeding is a numbers game; the goal is to screen the maximum number of varieties under the maximum number of environments (Araus and Cairns, 2014).

For these reasons, many universities conduct annual forage trials to determine which species and varieties of those species produce the most biomass in specific geographical areas. The publications that result from these forage trials are aimed at giving unbiased yield and quality data to researchers and forage producers so they can make educated decisions regarding the most appropriate varieties for their geographic region (Neely et al., 2016).

Despite their usefulness, small grains forage trials can be costly, time consuming and labor intensive to conduct. As noted in past studies, the standard method of determining forage biomass is to clip and weigh the samples, which requires great effort and expense (Sanderson et al 2001). Recently, efforts have been made to incorporate unoccupied aerial vehicles (UAV) into estimating cool season annual forage biomass. These vehicles utilize remote sensing imagery such as visual spectral images, which can obtain various vegetation indices (VI). The use of VIs has long been applied to biomass estimation; however, the most popular VI would traditionally use the near infrared region (NIR) with a light reflectance range of 700 to 1300 nm (Bendig et al.,2015). To obtain the NIR band would require the use of a more expensive four band multispectral camera; however, a more cost-effective approach would be the utilization of a UAV mounted three band camera which would only take into account the red (R), green (G) and blue (B) bands (400-700nm) (Bendig et al., 2015, Lessem et al., 2017, 2018). In a 2016 study, Possoch et al. (2016) evaluated the use of multi-temporal crop surface models that were combined with the RGB vegetation index (VI) for forage monitoring in grasslands. The study found that the RGB vegetation index (RGBVI) was not a good estimator of biomass, but if combined with plant height, medium correlations (R square 0.50) could be obtained. A study conducted by Motohka et al (2010) concluded that visual spectrum VIs can be useful in detecting the early phase of leaf green-up, but may be limited to certain growth stages (Motohka et al., 2010). This may be useful when evaluating a multi-clip forage trial. Research is also limited on the best flight altitude for biomass screening. In a 2010 study by Swain et al, investigators set out to determine the yield and total biomass of a rice crop by utilizing a low altitude unmanned rotocopter. The UAV was flown at 20 meters over the experimental plots with positive results when compared to satellite images for estimating some biomass parameters (Swain et al., 2010). Many studies have evaluated the relationship between remote sensing data and forage biomass (Serrano et al., 2000; Freeman et al., 2007; Tucker, C. J., 1980; Anderson et al., 1993). Some research using visible band VIs has shown potential as an effective method of biomass estimation; however, investigators acknowledge further investigation is needed (Lussem et al., 2017, 2018, Tilly et al., 2015, Bendig et al., 2015, Possoch et al., 2016).

The Normalized Difference Vegetation Index (NDVI) is one of the most successful and simple ways to remotely identify live green plant material (LDP, 2015). In a 2015 study Prabhakara et al., a 16-band CROPSCAN sensor was used to obtain ten VIs to determine the relationship between biomass, percent groundcover and remote sensing indices across six winter cover crops The study concluded that there was a strong relationship between NDVI and groundcover with an R^2 value of 0.93 (Prabhakara et al., 2015).

NDVI does have criticisms for some of its perceived defects: (1) the differences between “true” NDVI measured at the surface and NDVI measured from space due to atmospheric interference, (2) the sensitivity of NDVI to LAI (leaf area index) which becomes weak with a rising LAI, and (3) soil brightness variations which can cause variations in NDVI from one image to the next (Carlson and Ripley, 1997). Other problems with NDVI have been attributed to saturation issues in dense vegetation (Mutanga et al., 2004). Still, several studies have indicated a good correlation between plant biomass and NDVI readings from primitive handheld devices or plant height for various species (Andersson et al., 2017, Flynn et al., 2008, Freeman et al., 2007).

These new methods have been recently explored in their application possibilities; however, historically the traditional methods such as visual rating and plant height have been utilized solely or incorporated as a part of the estimation of forage potential in various crops (Ackerman et al 1999, Ud-Din et al 1993, Freeman et al 2007, Fernandez et al 2009). Researchers have utilized visual rating for many applications in the plant science discipline. Several studies have utilized these ratings to evaluate issues such as weed pressure in cover crops, individual space planted clover and winter wheat forage lines (Lawley et. al. 2011, Ud-Din et. al. 1993, Riday, 1997).

The purpose of this research was to compare a ground-based method of forage estimations to an aerial method. The main goal is to determine which method is best to assist cool season annual grass breeders and researchers in estimating forage yield of entries in forage trials and breeding lines for advancement in breeding programs. The specific objectives of this study are to: (1) determine at what height UAV measurements should be taken for measuring forage yield, 2) Compare relationships among UAV generated vegetation indices, hand held NDVI, plant height, and visual rating with dry matter forage yield across and between cool-season annual grass species, (3) Develop models to predict forage yield among cool-season annual grasses using UAV generated VIs and ground based measurements. 4) Determine if models generated from UAV data are as reliable as ones generated from ground based measurements.

Materials and Methods

A cool season annual grass forage trial was conducted at the Texas A&M Research farm near College Station, TX (30°31'N 96°25'W) during the 2016-2017 (2017) growing season. The climate of the location is humid subtropical. The 30-year average annual temperature is 20.6 °C and growing season (October-May) temperature is 19.0°C (College Station., 2020). The 30-year long-term average annual precipitation is 1018 mm and growing season precipitation is 435 mm (College Station, 2020). The highest precipitation generally occurs in the months of May, June, and October. Plots were located on a ships clay (*Udertic Hapluderts*) soil type (USDA-NRCS 2018). The trial was set up in a randomized complete block design (RCBD) with four replications of 38 entries in 2017. Total number of plots in the 2017 growing season was 152. Each plot measured 6 m in length and 1.5 m in width. The trial included commercial and experimental wheat, oat, rye, triticale, barley and ryegrass cultivars. All plots in the trial were treated with Cruiser Maxx Vibrance for Cereals to protect from seed borne and soil borne diseases and insects (e i. Thiamethoxam, Mefenozam, Fludioxonil and Sedazan)

(Syngenta, Basel, Switzerland). The plots were planted at a rate of 75 kg seed ha^{-1} on 19 cm row spacing in plots that were 1.5 meters (m) x 6.0 m in size.

The study was planted on September 29, 2016. Monthly temperature averages were near normal at 18.4°C and there was a below average amount of rainfall (273 mm) (College Station, 2020) during the growing season. The study included 9 wheat, 12 oat, 3 rye, 2 triticale, 9 ryegrass and 3 barley cultivars (38 treatments). The clipping dates were November 30, 2016 (Fall), February 8, 2017 (mid-winter) and March 31, 2017 (early spring). A total of 102 kg N ha^{-1} were applied as liquid UAN (32-0-0) on December 2, 2016. Three plant height measurements were taken per plot just before harvest using a meter stick and averaged to get mean of plot height.

There were two methods of forage biomass analysis that were evaluated in this study an aerial and ground. The Ground method utilized the hand held NDVI meter, which was a Trimble GreenSeeker Handheld Crop Sensor, Trimble part number 91500-00 (Trimble. GreenSeeker, Sunnyvale, Ca). This sensor produces red and infrared light once the trigger was pulled at an operating height of 80 cm above the plant canopy. The trigger was held while the operator walked the length of the plot as measurements of the reflected light were taken at a continuous pace while the trigger was engaged. Once the trigger was released, an average plot reading was displayed. A numeric reading between 0.00 and 0.99 was produced based upon the NDVI which uses the equation NDVI = (NIR-R)/(NIR + R) (Hansen and Schjoerring., 2003) . In theory, higher values indicate a healthier crop. Three plant height measurements were recorded per plot using a meter stick to get an overall plot average. A visual rating was taken for each plot on a 0-10 scale, where 0 indicates no forage potential and 10 represents very high forage potential. The visual rating was based on a visual assessment that took leafiness, height, and overall vigor into consideration to compare trial entries.

The aerial method utilized UAV data that was collected by using a Phantom 4 multi rotor drone equipped with an RGB camera (DJI, Phantom 4 multi rotor drone, Shenzhen, China). Flights were flown at two different altitudes of 20m and 30m with an 80% overlap just prior to clipping. The resulting images were processed and stitched together using Microsoft: Image Composite Editor (Microsoft. Redmond, Washington). Once the images were stitched, they were uploaded into the ENVI software (Exelis Visual Information Solutions, Boulder, Colorado) and the Region of Interest (ROI) tool was used to draw individual polygons over the entirety of each plot. Once all the polygons were drawn, the ENVI statistics tool was utilized to obtain the reflectance values of the three bands; band 1 red, band 2 green and band 3 blue for each pixel and then averaged across each ROI. Data was compiled in a Microsoft Excel spread sheet and used to calculate the vegetation indices identified in Table 3.1. In order to avoid multicollinearity issues, only the highest correlated VI was used for each species and combined species analysis. In addition to the VI, the same manual plant height was used as described earlier in order to remain consistent across methods. Once all data was taken, the plots were harvested using a Haldrup 1500 forage harvester (Haldrup, Ilshofen, Germany) which has an onboard weigh system and 1.5 m header to give plot total plot yield which was then converted to kg ha -1. In order to better synchronize the yield data and produce a linear relationship with the indices and NDVI results the log 10 of each yield was taken by using the log function in Microsoft Excel.

Table 3.1 List of vegetation indices used to predict forage yield of a cool season annual grass forage trial along with their respective abbreviations, formulas and sources.

Vegetation Indices	Abbreviation	Formula	Reference
Normalized Red	**r**	**Red/ Red+Green+Blue**	Woebbecke et al 1995
Normalized Green	**g**	**Green/Red+Green+Blue**	Woebbecke et al 1995
Normalized Blue	**b**	**Blue/Red+Green+Blue**	Woebbecke et al 1995
Red Green VI	**RG**	**R/G**	Introduced here
Normalized Red Green VI	**rg**	**r/g**	Introduced here
Modified Green Red VI	**MGRVI**	$(G)^2-(R)^2 / (G)^2+(R)^2$	Bendig et al.,2015
Red Green Blue VI	**RGBVI**	$(G)^2-(B*R) / (G)^2+(B*R)$	Bendig et al.,2015
Excessive Green VI	**ExG**	**2g-r-b**	Woebbecke et al 1995
Normalized Green Red VI	**g/r**	**g/r**	Introduced here

Statistical analysis was conducted using SAS 9.4 statistical software (SAS Institute, Cary, North Carolina). PROC CORR was used to run a correlation analyses between plant height, NDVI, R/G, r/g, MGRVI, RGBVI, ExG, g/r and total plot dry forage yield and the log 10 of total plot dry forage yield of all species. PROC REG was used with a backward stepwise regression (alpha = 0.05) to develop the dry forage yield and log yield prediction models using , plant height (PH), Visual Rating (VR) and R/G, r/g, MGRVI, RGBVI, ExG, and g/r for the ground method. A second PROC REG backward stepwise regression (alpha = 0.05) was used to develop the dry forage yield and log yield prediction models by using the best VI R/G, r/g, MGRVI, RGBVI, ExG, and g/r in addition to plant height for the aerial method.

Results and Discussion

Measurements of visual rating, plant heights, hand held NDVI, and six VIs derived from the RGB (UAV) images were taken multiple times across a single growing season in an attempt to estimate forage potential. The UAV data was collected at two flying heights, 20 and 30-meter altitudes just prior to each clipping. . The performance of each species varied based on the time of year as well as temperature and rainfall during the growing season. This is expected as each species performs differently through its life cycle and differs in response to mechanical clipping. In order to capture some of this variability, the study was divided into three seasonal clip times based on the time of year the clippings were performed.

Measurements for VIs are inherently small (between 0 and 1) and the yield data measured in kg ha^{-1} ranged from 20 kg ha^{-1} to 1706 kg ha^{-1} which is comparatively much higher. The yield data was then log transformed in an attempt to create a linear relationship with measurements since VIs are known to produce non-linear relationships with yield (Panda, S.S., et. al. 2010, Dadhwal and Sridhar, 1997). Transforming the data did improve correlations in some cases, but not all. Log transforming the plant height data did not increase the correlation to yield; and in some cases, it decreased it. Several studies have indicated that plant height can be a useful measurement of forage production (Boukerrou, and Rasmusson 1990, Tilly et al 2015, Alheit et al 2014)

UAV Flight Altitude

Each vegetation index was calculated for flight altitudes of 20 and 30 m above soil level to determine which flight altitude produced the best correlations. In every instance for the fall and mid-winter clippings, the correlation coefficient was as good or better at the 20 m altitude flight compared to the 30 m height (Table 3.2). The improvement was quite minimal for the fall clip however, and there was no improvement during the early spring clipping as the vegetative indices showed no correlation at either altitude. Of all the indices, the 20m height showed the greatest and most consistent improvement for the RGBVI index. The correlation coefficients were most improved during the mid-winter clipping when comparing the 20m to the 30m flight heights. Other studies documented a similar improvement at lower UAV flight heights. Liu et. al., noted that, as UAV flight height differed, the image resolution of the same area varied. It was determined that this variation can have an effect on image features which can degrade accuracy of the trait being evaluated (Liu et al 2018). A second study indicated that as the flight height of the UAV increased, the number of detectable targets decreased (Trolove and Shorten 2019). In the current study the effects of flight altitude were minor never the less the accuracy was improved at lower altitudes. This was also seen in several other studies (Torres-Sánchez et al 2014, López-Granados et al 2016, and Peña et al 2015).

Table 3.2 Pearson Correlation coefficients and number of observations (n) among vegetation indices, plant height, and hand held NDVI with dry matter forage yield for a cool-season annual grass forage trial in College Station, TX. Correlation coefficients for vegetative indices are shown at two flight heights (20 and 30 m) and combined across all varieties and species (wheat, oat, barley, rye, triticale, and ryegrass) in the trial.

Season	n	R/G	r/g	MGRVI	RGBVI	ExG	g/r	Plant Height	Hand Held NDVI
20m								**Ground Measurements**	
Fall	n=142	-0.73***	-0.76***	0.74***	0.64***	0.71***	0.76***	0.80***	0.73***
Mid-winter	n=142	-0.45***	-0.48***	0.44***	0.41***	0.42***	0.48***	0.52***	0.26**
Early spring	n=139	-0.14ns	-0.12ns	0.14ns	0.16ns	0.15ns	0.12ns	0.40***	0.47***
Combined	n=423	-0.36***	-0.35***	0.36***	0.25***	0.29***	0.36***	0.38***	0.47***
30m									
Fall	n=142	-0.72***	-0.75***	0.73***	0.53***	0.69***	0.75***	_	_
Mid-winter	n=142	-0.36***	-0.39***	0.36**	0.34***	0.34***	0.39***	_	_
Early spring	n=139	-0.14ns	-0.11ns	0.14ns	0.16ns	0.15ns	0.12ns	_	_
Combined	n=423	-0.35***	-0.33***	0.34***	0.16***	0.25***	0.33***	_	_

UAV and Ground Based Measurement Relationships with Forage Yield

UAV Measurements

A Pearson correlation analysis revealed a wide range in relationships among vegetative indices and plant height with dry matter forage yield for specific species and clip times (Table 3.3). Previous studies have shown non-linear relationships among VIs and biomass, so data was also log transformed to compare correlation coefficients

(Goward et al 1985, Hobbs 1995, and Santin-Janin et al 2009). While number of observations ranged by species from 12 (barley and rye) up to 44 (oats), increasing the number of observations did not necessarily improve correlations. Log transforming the data did improve the correlation coefficient for some species, but not all, and there was not a single VI that was best correlated to all species.

Overall, the strongest correlation obtained between a VI and dry matter forage yield for any species was for R/G, r/g, MGRVI, and g/r ($r=-0.89$, $r=-0.89$, $r=0.89$, $r=0.89$, respectively; $p<0.001$ for all) during the Fall clip for ryegrass when the data was log transformed. In fact, ryegrass had the highest VI correlations across clip times and combined clips (R/G: $r=-0.92$, $p<0.001$; MGRVI: $r=0.92$, $p<0.001$) than any the other species in the study. This could be a result of the naturally shinny, dark green, and smooth leaves that are characteristic of annual ryegrass (Lacefield, G. et al 2003). In general, the correlations between the VIs generated from UAV images and dry matter forage yield was far better for ryegrass in this study than any other small grains species. A study conducted by Golzarian and Frick, 2011 wanted to determine if image classification could distinguish differences between wheat and ryegrass by using color, texture and shape. The study evaluated the digital numbers of red, green and blue on a scale of 0-255. It was determined that ryegrass plants would have more red color than wheat which had more blue color (Golzarian and Frick, 2011). Red (564-580nm) and blue (420-440 nm) light wavelengths are strongly absorbed by chlorophylls (Terashima, I., et. al. 2009). Chlorophyll is the source of the green color that is reflected and found in all plants (Rabinowitch, E.I., 1965) and has a direct relationship with the amounts of nitrogen contained in the plant cells (Wood et al. 1993, Lamb et al .2002, Schepers, J.S, 1996). In this study, it was noted that the ryegrass plots were visibly greener to the naked eye than that of the other species in the trial. This phenomenon could explain why the UAV and hand-held NDVI sensor were able to better detect above ground biomass yields in the ryegrass plots. While coefficients were high for original data, log transformed data improved the coefficient even further for ryegrass. Transforming plant height did improve correlations for fall and mid-winter clips, but early spring plant height was not significant in either case, and VI coefficients were better in every instance.

After ryegrass, rye had the next highest correlations among measurements and dry matter forage yield. Transforming the data had mixed results. Correlation coefficients were only slightly improved by transforming the data for the mid-winter clip, while coefficients were lower for the fall and had no practical impact for early spring. The original data did provide the best correlations when combining data across clips with R/G obtaining the highest correlation ($r=0.75$, $p<0.001$). Rye plant height had strong correlations for each individual clip time, but was much lower than most VIs when combined across clips. The strong plant height correlations reflects the more semi-erect to erect growth pattern of rye (Briggle 1959,Holt 1962, Pfahler et. al., 1986) which has much of its biomass located in the mid to upper canopy throughout the growing season. This is the opposite to ryegrass, which tends to have a semi-prostrate growth pattern (Arnold et. al., 1981,Choi et. al., 2000, Choi et. al., 2011) especially later in the season compared to other annual grass species.

Wheat, oat, and barley had low or no significant correlation in many cases when comparing VIs with dry matter forage yield. Wheat and oat both tended to have stronger correlations during the mid-winter clip time for many of the VIs, while barley had more success in the early spring clip. This could be attributed to when each species initiates spring green up. In this study oat and wheat cultivars tended to mature slower than barley. Interestingly, transforming the data improved mid-winter coefficients for wheat and decreased coefficients for oats. This could be a result of the overall range in yield for each species during the mid-winter clip time. The winter wheat ranged from 212 kg ha^{-1} to 1,043 kg ha^{-1} and the oat had a slightly higher range 264 kg ha^{-1} to 1,260 kg ha^{-1}. It is possible that certain winter wheat cultivars had reached canopy closure before the oats resulting in a plateauing VI response to dry matter forage yield for the higher yields. This often results in a logarithmic relationship and cause an improvement to the correlation coefficient when the data was log transformed for the wheat, but not the oats. Data transformation made no appreciable difference for barley VIs in any clip time, which could be attributed to the data having a smaller range in values for all three clip times (fall 321-770 kg ha^{-1}, Mid-winter 202-649 kg ha^{-1} and early spring 628-1585 kg ha^{-1}). No VI was significant when combining data across clip times for wheat and oats, though R/G produced a coefficient of $r=0.42$ ($p<0.001$) for barley using non-transformed data. Plant height had a much more consistent correlation with dry matter forage yield than VIs for wheat, ranging from $r=0.52$ ($p<0.001$) in the fall to $r=0.36$ ($p<0.05$) in the early spring with a coefficient of $r=0.52$ ($p<0.001$) for combined clip data. Oat plant height correlations were only significant in the fall ($r=0.45$, $p<0.01$), with or without data transformation. This could be attributed to the growth habit of most oats in this study which were noticed to have erect growth early in the season, but as the season progresses and the oats developed a more semi-prostrate growth habit. This would result in much of the oat forage being located in the mid canopy of the plant for which plant height may not be the best determination of forage yield. Non-transformed winter barley plant height produced a high correlation for the mid-winter (r=0.82, p<0.01) and early spring (r=0.80, p<0.01) clips, but was not significant for the fall clip. This is likely an effect of growth habit of the specie, as it tends to maintain an erect growth pattern throughout the year with much of its forage evenly distributed throughout the plant profile, making plant height a reasonable forage assessment tool for most of the growing season. The small sample size and small range in yield likely was the cause of no significant correlation in the fall clip for barley.

Table 3.3 Correlation coefficients of original and log transformed data for plant height, R/G, r/g, MGRVI, RGBVI, ExG, and g/r with dry matter forage yield using visual reflectance data taken with a UAV at a 20m flight altitude. Measurements were taken three times (fall, mid-winter, early spring, and combined species) on a cool-season annual grass forage trial near College Station, TX during the 2017 seasons.

	Original Data				**Log Transformed Data**			
	Winter Wheat							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.52***	0.46**	0.36*	0.52***	0.42**	0.47**	0.39*	0.52***
R/G	-0.35*	-0.30*	-0.04ns	0.07ns	-0.37*	-0.36*	-0.02ns	0.00ns
r/g	-0.34*	-0.32*	-0.07ns	0.02ns	-0.35*	-0.39*	-0.05ns	-0.05ns
MGRVI	0.34*	0.29ns	0.05ns	-0.07ns	0.37*	0.36*	0.02ns	0.00ns
RGBVI	0.10ns	0.47**	0.10ns	-0.08ns	0.17ns	0.54***	0.08ns	0.00ns
ExG	0.23ns	0.38*	0.07ns	-0.06ns	0.28ns	0.45**	0.05ns	0.02ns
g/r	0.34*	0.32*	0.07ns	-0.02ns	0.35*	0.39*	0.05ns	0.05ns
n	n=38	n=38	n=38	n=114	n=38	n=38	n=38	n=114

Table 3.3 Continued.

	Original Data				Log Transformed Data			
	Oat							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.45**	0.08ns	0.16ns	0.17* n=132	0.45**	0.07ns	0.24ns	0.20*
R/G	-0.03ns	-0.51***	0.24ns	0.13ns	-0.03ns	-0.49***	0.26ns	0.06ns
r/g	-0.11ns	-0.54***	0.27ns	0.13ns	-0.12ns	-0.50***	0.28ns	0.07ns
MGRVI	0.03ns	0.51***	-0.24ns	-0.13ns	0.03ns	0.48***	-0.26ns	-0.07ns
RGBVI	0.18ns	0.39**	0.19ns	-0.11ns	0.18ns	0.37*	0.21ns	-0.06ns
ExG	0.12ns	0.46**	-0.09ns	-0.13ns	0.13ns	0.42**	-0.08ns	-0.07ns
g/r	0.11ns	0.54***	-0.27ns	-0.13ns	0.12ns	0.50***	-0.28ns	-0.07ns
n	n=44	n=44	n=44	n=134	n=44	n=44	n=44	n-134
	Rye							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.62**	0.74**	0.77***	0.48**	0.63*	0.71**	0.75**	0.37*
R/G	-0.64*	0.57ns	0.63*	0.75***	-0.60*	-0.62*	0.64*	0.68***
r/g	-0.62**	0.60*	0.62*	0.71***	-0.57ns	-0.65*	0.63*	0.65***
MGRVI	0.64*	0.57ns	-0.63*	-0.74***	0.60*	0.61*	-0.64*	-0.67***
RGBVI	0.64*	0.32ns	-0.39ns	-0.71***	0.58*	0.37ns	-0.41ns	-0.68***
ExG	0.65*	0.46ns	-0.53ns	-0.72***	0.60*	0.51ns	-0.54ns	-0.67***
g/r	0.62*	0.60*	-0.62*	-0.71***	0.57ns	0.65*	-0.63*	-0.65***
n	n=12	n=12	n=12	n=36	n=12	n=12	n=12	n=36
	Ryegrass							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.56***	0.31ns	0.27ns	-0.08ns	0.69***	0.36*	0.28ns	-0.12ns
R/G	-0.82***	-0.68***	-0.79***	-0.83***	-0.89***	-0.76***	-0.83***	-0.92***
r/g	-0.86***	-0.67***	-0.76***	-0.86***	-0.89***	-0.71***	-0.77***	-0.91***
MGRVI	0.83***	0.68***	0.78***	0.85***	0.89***	0.75***	0.81***	0.92***
RGBVI	0.82***	0.68***	0.77***	0.74***	0.87***	0.75***	0.80***	0.85***
ExG	0.84***	0.67***	0.78***	0.81***	0.88***	0.73***	0.80***	0.88***
g/r	0.86***	0.67***	0.76***	0.86***	0.89***	0.71***	0.77***	0.91***
n	n=35	n=35	n=35	n=105	n=35	n=35	n=35	n=105

Table 3.3 Continued.

	Original Data				Log Transformed Data			
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.37ns	0.82**	0.80**	0.52**	0.37ns	0.78*	0.77**	0.54***
R/G	-0.20ns	-0.43ns	0.58ns	0.42*	-0.20ns	-0.42ns	0.58**	0.29ns
r/g	-0.24ns	-0.46ns	0.58*	0.31ns	-0.24ns	-0.46ns	0.58**	0.18ns
MGRVI	0.20 ns	0.43ns	-0.57ns	-0.40*	0.20 ns	0.42ns	-0.57ns	-0.26ns
RGBVI	0.27ns	0.53ns	-0.71**	-0.37*	0.27ns	0.54ns	-0.71**	-0.19ns
ExG	0.26ns	0.48ns	-0.62*	-0.34*	0.25ns	0.48ns	-0.62*	-0.19ns
g/r	0.24 ns	0.46ns	-0.58*	-0.46ns	0.24 ns	0.46ns	-0.58**	-0.17ns
n	n=12	n=12	n=12	n=36	n=12	n=12	n=12	n=36
				Combined				
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.80***	0.52***	0.40***	0.39**	0.75***	0.53***	0.41***	0.30**
R/G	-0.73***	-0.44***	-0.15ns	-0.36***	-0.89***	-0.50***	-0.18*	-0.54***
r/g	-0.76***	-0.47***	-0.12ns	-0.36***	-0.90***	-0.51***	-0.16ns	-0.51***
MGRVI	0.74***	0.44***	0.15ns	0.36***	0.90***	0.50***	0.18*	0.52***
RGBVI	0.64***	0.41***	0.17*	0.25***	0.83***	0.48***	0.21**	0.42***
ExG	0.71***	0.42***	0.17ns	0.29***	0.87***	0.48***	0.17*	0.45***
g/r	0.76***	0.47***	0.13ns	0.36***	0.90***	0.51***	0.13ns	0.51***
n	n=141	n=141	n=141	n=423	n=141	n=141	n=141	n=423

*, **, *** Correlation coefficient significant at the 0.05, 0.01, and 0.001 probability levels.

NS, non-significant at $P \geq 0.05$.

Ground-Based Measurements

Correlation analysis in Table 3.5 compares the VR, plant height, and hand held NDVI sensor to dry matter forage yield for each species, clip time and combined clip time for the original and log-transformed yield data. In this study, transforming the data showed little benefit to VR for any specie, except for a slight improvement in ryegrass correlations for each clip time and combined clips (r=0.85, p<0.001) (Table 3.5). Similarly, transforming NDVI data consistently improved correlation coefficients for ryegrass for individual and combined clips ($r=0.88$, $p<0.001$). In fact, log transformed NDVI data had the strongest correlation with dry matter forage yield of any ground based measurement. While transforming plant height data showed some improvement for ryegrass, it generally did more harm than good for correlations with dry matter forage yield in other species.

Outside of ryegrass, NDVI produced few significant correlations for other species with only low to moderate correlations for wheat ($r=0.36$, $p<0.05$) and oats ($r=0.41$, $p<0.01$) in the early spring clip. In general, VR provided a consistently higher correlation to dry matter forage yield than plant height (PH) for all species except rye and the contrast was most striking for oats and ryegrass. Rye PH correlation with dry matter forage yield was better than VR for all individual and combined clips ($r=0.48$, $p<0.01$). Correlations between visual scores and dry matter forage yields appeared reliable for winter wheat, oat and rye during the mid-winter clip time. Intuitively, this makes sense as mid-winter clips during this study took place in February when these species were on the verge of breaking winter dormancy and are generally demonstrating more variability in forage production. It is more difficult to detect a relationship when little variation exists, as was the case in the early spring for these species. Ryegrass and barley demonstrated a wider range in yield throughout the growing season, which made the relationships between dry matter forage yield and VR easier to detect through the season. Based on this study, VR would be a viable option for ranking trial entries for forage potential; however, its use would be limited to within a specific clip and would not be useful to predict yield, especially across clip times.

Table 3.4 Correlation coefficients of original and log transformed data for visual rating, plant height, and hand held NDVI meter with dry matter forage yield. Measurements were taken three times (fall, mid-winter, early spring, and combined species) on a cool-season annual grass forage trial near College Station, TX during the 2017 seasons.

	Original Data				Log Transformed Data			
Winter Wheat								
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
VR	0.49**	0.76***	0.39*	0.33***	0.43**	0.75***	0.41**	0.36***
PH	0.52***	0.46**	0.36*	0.52***	0.42**	0.47**	0.39*	0.55***
NDVI	0.24ns	0.27ns	0.36*	0.06ns	0.20ns	0.36*	0.34*	0.10ns
n	n=38	n=38	n=38	n=144	n=38	n=38	n=38	n=144
Oat								
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
VR	0.65***	0.82***	0.55***	0.63***	0.66***	0.83***	0.55***	0.68***
PH	0.45**	0.08ns	0.16ns	0.18*	0.45**	0.07ns	0.24ns	0.20*
NDVI	0.04ns	0.37*	0.41**	0.22**	0.01ns	0.39**	0.40**	0.25**
n	n=44	n=44	n=44	n=132	n=44	n=44	n=44	n=134
Rye								
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
VR	0.49ns	0.71**	0.10ns	0.11ns	0.42ns	0.63*	0.10ns	0.14ns
PH	0.62*	0.74**	0.77**	0.48**	0.63*	0.71**	0.75**	0.37*
NDVI	0.56ns	0.05ns	-0.20NS	-0.26ns	0.60*	0.06ns	-0.24ns	-0.14ns
n	n=12	n=12	n=12	n=36	n=12	n=12	n=12	n=36
Ryegrass								
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
VR	0.72***	0.69***	0.76***	0.79***	0.78***	0.78***	0.79***	0.85***
PH	0.56***	0.31ns	0.27ns	-0.08ns	0.69***	0.37*	0.28ns	-0.12ns
NDVI	0.83***	0.65***	0.83***	0.78***	0.90***	0.74***	0.88***	0.88***
n	n=35	n=35	n=35	n=105	n=35	n=35	n=35	n=105
Winter Barley								
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
VR	0.75***	0.73*	0.87***	0.46*	0.74***	0.70*	0.85***	0.49**
PH	0.37ns	0.82**	0.80**	0.52**	0.37ns	0.79**	0.77**	0.54***
NDVI	0.48ns	0.20ns	-0.11ns	-0.02ns	0.50ns	0.20ns	-0.14ns	0.03ns
n	n=12	n=12	n=12	n=36	n=12	n=12	n=12	n=36

Table 3.4 Continued.

	Original Data				Log Transformed Data			
	Combined							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
VR	0.83***	0.73***	0.68***	0.68***	0.89***	0.74***	0.70***	0.74***
PH	0.80***	0.52***	0.40***	0.39***	0.75***	0.53***	0.41***	0.30***
NDVI	0.73***	0.26**	0.47***	0.47***	0.87***	0.33***	0.52***	0.61***
n	n=141	n=141	n=141	n=423	n=141	n=141	n=141	n=423

*, **, *** Correlation coefficient significant at the 0.05, 0.01, and 0.001 probability levels.

NS, non-significant at $P \geq 0.05$.

Prediction Models Aerial Method

Since no one variable showed a strong relationship across all clip times or species, models were developed to predict dry matter forage yield. The best VIs of the six evaluated here (R/G, r/g, MGRVI, RGBVI, ExG, and g/r) and plant height were used in a backward stepwise regression to develop individual models for each individual species, clip time, and combinations of species and clip times. In addition, models were created across species and clip times for both the original yield data and the log transformed yield data (Table 3.4). Using this data, 48 models were attempted, though five were not significant and thus not used.

Of the 43 models that were created, eight used only plant height, 12 used only a VI and 23 utilized both plant height and a VI (Table 3.4). Models for winter wheat and ryegrass had slightly higher adjusted R^2 values when yield data was log transformed whereas the oat, cereal rye and barley did not. The winter wheat model with the highest correlation to log transformed yield was the mid-winter clip time [(Y=1.40.019* HT+21713*RGBVI) Adj R^2=0.54, RMSE 142]. The ryegrass model for the fall clipping

[(Y=1.3+0.018* HT+2.1*MGRVI) Adj R^2=0.82 ($p<0.0001$), RMSE 27] had the highest coefficient of determination for the log transformed yield data (R^2=0.82, ($p<0.0001$)) of any individual clip, though the developed models fit the data well for all clips. In fact, the combined model across clips was the best (R^2=0.84 $p<0.0001$) for ryegrass

For oat, only the fall and mid-winter clip time models were significant, and even then, adjusted R^2 values were low, regardless of whether the yield data was transformed or not. The fall clip time for barley, for both the normal and log transformed forage data, did not produce a significant model; however; moderately high R^2 values were produced for both the mid-winter and early spring clip time models without transforming the data. Interestingly the early spring model [(Y=123.4+35.3*HT) Adj R^2=0.65($p<0.0001$), RMSE 174 utilized only plant height while the log transformed early spring model [(Y=3.4-1.1*RGBVI) Adj R^2=0.28 ($p<0.0001$), RMSE 167] only utilized RGBVI, but produced a similar adjusted R^2. The mid-winter model had a slightly lower adjusted R^2 value, but had a much lower RMSE compared to the early spring clip. Despite the models fitting the data moderately well for mid-winter and early spring, combining data across clip times produced a model that explained very little of the variability in yield.

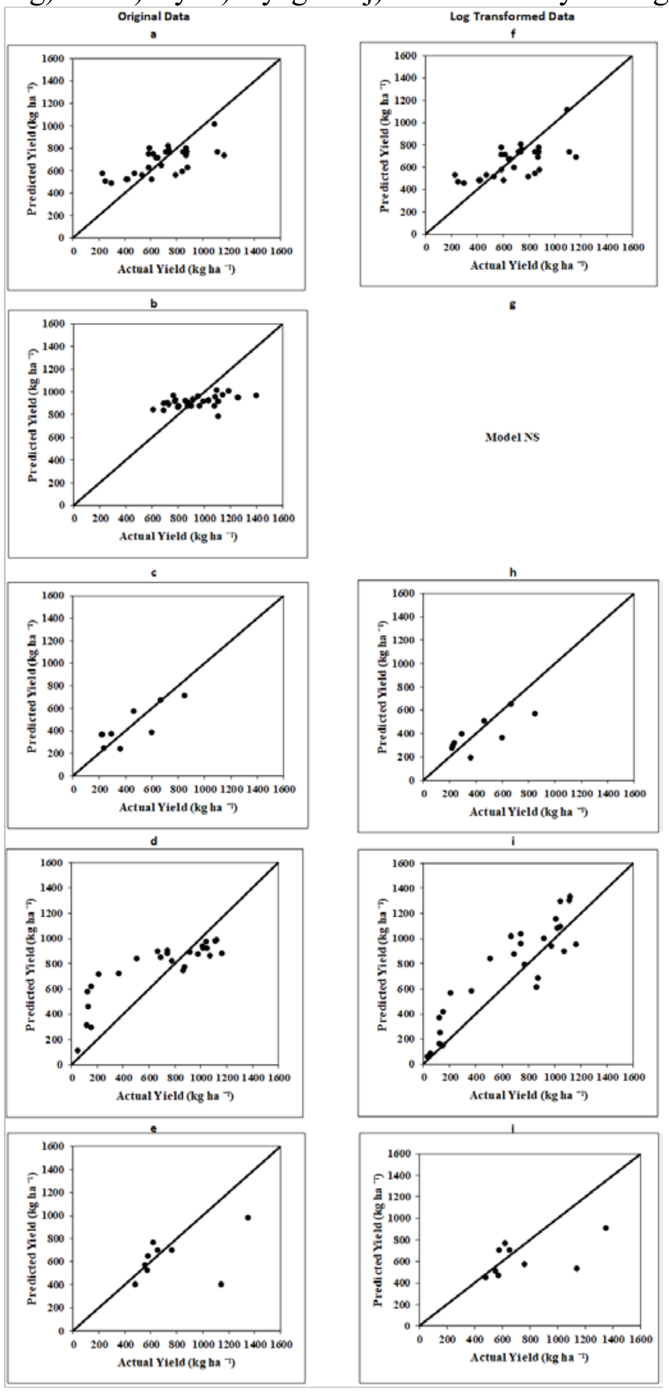
For cereal rye, the developed models explained over half of the variability for all three clip times and was best using the original yield data, though each model used a different parameter. The best rye model was for the mid-winter clip time [(Y=751638+751650*gr) Adj R^2=0.70 ($p<0.0001$), RMSE 63]; however, the combined model did nearly as well in terms of the R^2. Of the combined species models, the fall clip time had the highest R^2 for both the original and log transformed forage data with an adjusted $R^2=0.76 (p<0.0001)$ and $R^2=0.89 (p<0.0001)$, respectively. In every case, transforming the data improved the adjusted R^2 value, but also increased the RMSE. The combined clip time and species was poor for both the original ($R^2=0.23 (p<0.0001)$) and log transformed ($R^2=0.36 (p<0.0001)$) forage data (Table 3.4), indicating there was not a "one size fits all" model. However, combining plant height with the gr VI in a model shows promise that a single model could be used across species to predict dry matter forage yield in the fall for a multi-species cool-season annual grass forage trial. Unfortunately, the models were less accurate for the mid-winter clip and rather poor in the early spring.

Table 3.5 Original vs log transformed data of the aerial method for each species and clip time.

		Original Data				Log Transfomed Data			
Species		**Fall**	**Mid-Winter**	**Early Spring**	**Combined**	**Fall**	**Mid-Winter**	**Early Spring**	**Combined**
Wheat	Model	Y=1610.4+14.2*HT-1786.6*RG	Y=-792.3+21.2*HT+2235.0*RGBVI	Y=-262.0+20.4*HT+1513.7*RGBVI	Y=243.2+13.7*HT	Y=3.6+0.008*HT-1.4*RG	Y=1.4+0.019*HT+2.1*RGBVI	Y=2.3+0.010*HT+0.7*RGBVI	Y=2.4+0.010*HT
	Adj R^2	0.27	0.43	0.28	0.21	0.23	0.54	0.29	0.25
	RMSE	120	137	153	213	120	142	152	218
	n	28	28	28	84	28	28	28	84
Oat	Model	Y=-1088.7+26.0*HT+1837.2*RGBVI	Y=-363298+363804*gr	n.s.	Y=970.4+6.5*HT-463.6*ExG	Y=1.8+0.014*HT+0.981*RGBVI	Y=-233.7+236.4*gr	n.s.	n.s.
	Adj R^2	0.27	0.28		0.04	0.26	0.25		
	RMSE	149	167		234	151	169		
	n	33	33		99	33	33		
Rye	Model	Y=-447.4+907.0*ExG	Y=-751638+751650*gr	Y=-81.1+25.6*HT	Y=-3382.3+15.2*HT+3695.3*RG	Y=0.683+2.2*ExG	Y=-979.7+981.8*gr	Y=2.3+0.015*HT	Y=-1.1+4.0*RG
	Adj R^2	0.57	0.70	0.51	0.68	0.48	0.70	0.49	0.44
	RMSE	37	63	93	137	40	60	90	159
	n	9	9	9	27	9	9	9	27
Ryegrass	Model	Y=122.1+600.5*MGRVI	Y=383.4+1629.0*MGRVI	Y=-84.2+16.1*HT+2998.8*MGRVI	Y=310.2+2722.2*MGRVI	Y=1.3+0.018*HT+2.1*MGRVI	Y=2.0+0.019*HT+1.7*MGRVI	Y=2.2+0.010*HT+2.0*MGRVI	Y=2.1+3.7*MGRVI
	Adj R^2	0.70	0.45	0.67	0.73	0.82	0.62	0.71	0.84
	RMSE	35	154	136	210	27	149	163	354
	n	26	26	26	78	26	26	26	78
Barley	Model	n.s.	Y=-209.9+30.2*HT	Y=-123.4+35.3*HT	Y=-1373.1+17.0*HT+1739.6*RG	n.s.	Y=1.8+0.033*HT	Y=3.4-1.1*RGBVI	Y=2.3+0.014*HT
	Adj R^2		0.62	0.65	0.37		0.56	0.63	0.27
	RMSE		84	174	245		103	178	393
	n		9	9	27		9	9	27
Combined	Model	Y=-267903+23.1*HT+267352*gr	Y=-281022+19.3*HT+280943*gr	Y=-309.5+24.6*HT+1354.9*RGBVI	Y=1409.2+13.7*HT-1288.5*RG	Y=-531.3+0.022*HT+532.7*gr	Y=-261.0+0.017*HT+263.2*gr	Y=2.2+0.013*HT+0.808*RBGVI	Y=4.34+0.010*HT-2.1*RG
	Adj R^2	0.76	0.43	0.27	0.23	0.89	0.48	0.32	0.36
	RMSE	156	174	203	294	208	178	205	342
	n	105	105	105	315	105	105	105	315

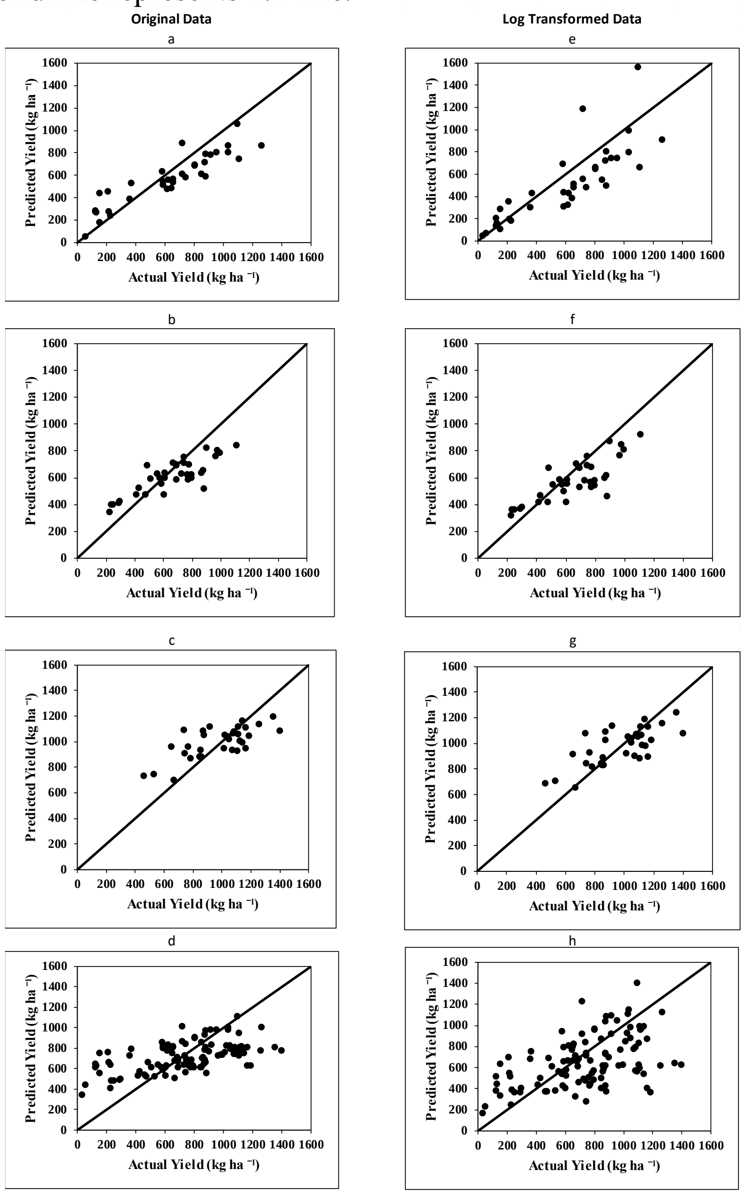
After validating the prediction models using a subset of the data from replication two, Figure 3.1 illustrates the accuracy of the models for the original and log transformed forage yield data. The effectiveness of the log transformation is most noticeable in the scatter plots for ryegrass (Figure 3.1, d and i). The log transformation of the ryegrass forage yield data distributed the data points closer to the 1 to 1 line. The data for the original rye forage yield (plot c; R^2=0.68 ($p<0.0001$) have a more uniform distribution around the 1 to 1 line than that of the transformed rye yield (plot h; R^2=0.44($p<0.0001$). The predicted yields for both the original and log transformed forage yield data for winter wheat, oat and barely were not very well correlated with actual yield. The wheat model overpredicted low yields and underpredicted high yields, while the oat model showed no discernible relationship. The barley models did well at predicting low yields but underpredicted high yielding plots.

Figure 3.5 Aerial method original and log transformed data Predicted vs actual yield data on a 1:1 line a) Wheat b) Oat c) Rye d) Ryegrass e) Winter Barley for original data, f) Wheat g) Oat h) Rye i) Ryegrass j) Winter Barley for log-transformed data.



The log transformed forage data for the fall clip time (R^2=0.89(p<0.0001); plot e) was an improvement over the original forage yield data (R^2=0.76 (p<0.0001); plot a) (Figure 3.2). While the predicted yields are more evenly distributed along the 1 to 1 line for lower yields when transforming the data, they appear to be more scatter as yield increases. Similar to the fall clip, the mid-winter clip model tended to underpredict low forage yields and overpredict the higher yields in that particular clip using the original data (plot b; R^2=0.43 (p<0.0001)). Transforming the mid-winter clip yield data only had a minimal improvement on over or under predicting forage yield (plot f; R^2=0.48 (p<0.0001)). The ability of the model to predict forage yield during the early spring clip was quite poor for both original (plot d; R^2=0.27 (p<0.0001)) and transformed (plot h; R^2=0.32 (p<0.0001)) data, though there was again a slight improvement using the transformed data. In the model where species and clip times were combined, low yields were significantly underpredicted and higher yields were overpredicted using the original data (plot d; R^2=0.23 (p<0.0001)). The log transformation helped with over or under predicting to some degree, but caused more scatter in the predicted values and R^2 values were quite low in either case. (plot h; R^2=0.36 (p<0.0001)). It is noteworthy that the log transformed forage yield data had better R^2 values for all clip times and combined clip times than that of the original data.

Figure 3.6 Validation of models for original and log transformed data using vegetative indices and plant height to predict actual yield based on clip time. Plots are assigned as a) Fall clip b) Mid-winter c) early Spring d) combined for original forage yield data and e) Fall clip f) mid-winter g) early spring h) combined for log transformed forage yield data. Solid line represents 1:1 line.



Prediction Models Ground Based Method

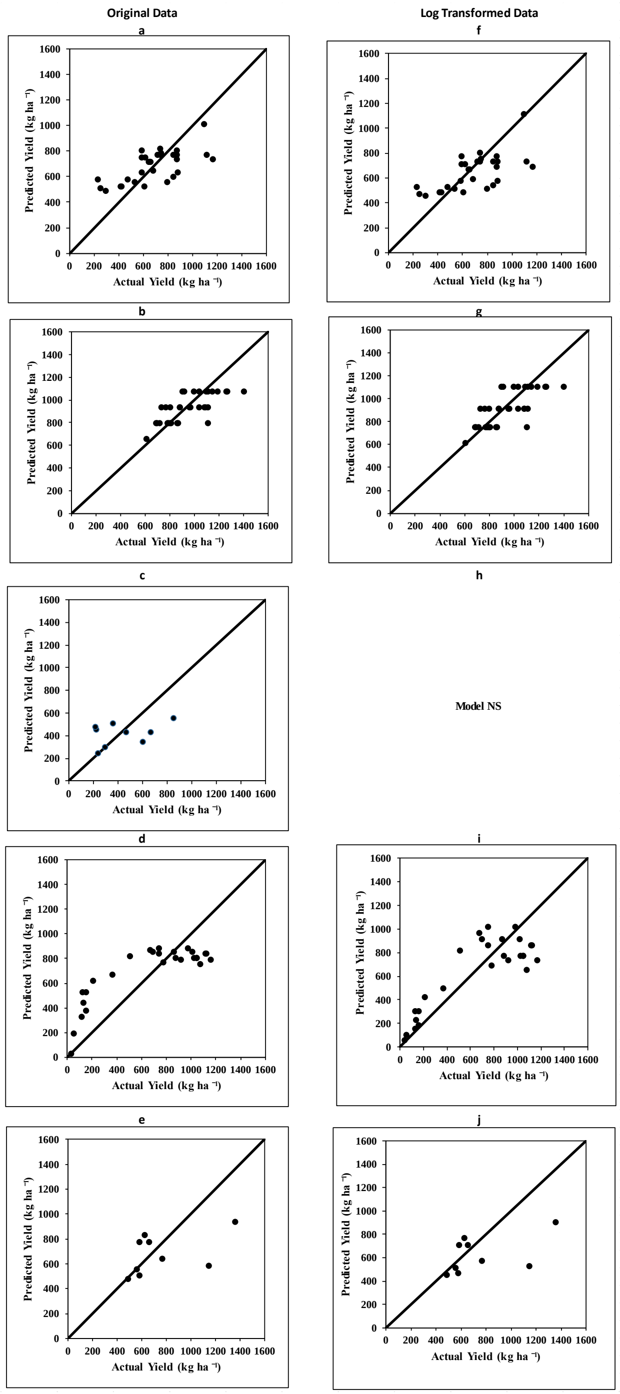
Since no one variable showed a strong relationship across all clip times or species, models were developed to predict dry forage yield using visual rating, plant height and handheld NDVI. Of the 48 models attempted, three were not significant. Of the 45 models that were created, 11 used only plant height, 15 used only a visual rating and five used only hand held NDVI. Some models for species and clip times were more accurate when all measurements were combined. Five models used plant height and visual rating, four used plant height and handheld NDVI, two used visual rating and handheld NDVI, and three models utilized all three measurements. Using the original data for the cereal rye was superior in every clip time including the combined data across clips, though the combined model was much less accurate than the individual clip models. Similar to cereal rye, predicted winter barley yields were more highly correlated to actual yield for each individual clip using original data compared to log transformed data and individual clip times were much better at predicting yield than combining all three clips together There were no appreciable differences between the models using original and transformed data for winter wheat. The models for mid-winter and early spring produced moderately high R^2 values, but the fall clip and combined models were quite poor. For the oat models, the only appreciable difference between the original data and transformed data occurred for the combined model where transforming improved the R^2, but increased the RMSE. In general the adjusted R^2 values were low except for the mid-winter clip [(Y=2.2+0.082*VR) Adj $R^2=0.67$ ($p<0.0001$), RMSE 198], which had the lowest RMSE as well. Transforming the forage data improved individual clip models and the combined model for ryegrass, though the fall clip produced the most accurate model [(Y=0.637+0.019*HT+1.3*NDVI) $R^2=0.84$ ($p<0.0001$), RMSE 23]. The winter barley models were better in every case using the original data, except when combining clip data. In fact, the barley models rivalled the accuracy of the ryegrass models using transformed data, except when combined across clips. When combining data across species, transforming the data improved R^2 values, but also increased RMSE values slightly. The combined species fall clip time produced the best R^2 value ($R^2=0.87$ ($p<0.0001$)), compared to the mid-winter and early spring clips, but again, the RMSE was not as low (RMSE=187).

Table 3.6 Original vs log transformed data of the ground method for each species and clip time.

		Original Data				Log Transformed Data			
Species		**Fall**	**Mid-Winter**	**Early Spring**	**Combined**	**Fall**	**Mid-Winter**	**Early Spring**	**Combined**
Wheat	Model	Y=-113.3+100.0*VR	Y=-699.2+174.1*VR	Y=-1063.4+23.1*HT+1891.1*NDVI	Y=243.9+13.7*HT	Y=2.2+0.068*VR	Y=1.6+0.149*VR	Y=1.9+0.019*HT+0.946*NDVI	Y=2.4+0.010*HT
	Adj R^2	0.19	0.59	0.48	0.21	0.15	0.60	0.50	0.25
	RMSE	129	118	129	213	129	119	130	218
	n	28	28	28	84	28	28	28	84
Oat	Model	Y=-173.8+128.1*VR	Y=-121.5+117.6*VR	Y=-670.1+114.7*VR+1264.4*NDVI	Y=-180.9+139.3*VR	Y=2.3+0.072*VR	Y=2.2+0.082*VR	Y=2.3+0.045*VR+0.503*NDVI	Y=2.2+0.084*VR
	Adj R^2	0.38	0.66	0.27	0.36	0.41	0.67	0.26	0.45
	RMSE	139	115	153	193	140	119	153	198
	n	33	33	33	99	33	33	33	99
Rye	Model	Y=-227.1+14.6*HT	Y=-222.1+29.3*HT	Y=-81.1+25.6*HT	Y=-119.7+20.4*HT	n.s	n.s.	Y=2.3+0.015*HT	n.s.
	Adj R^2	0.42	0.45	0.51	0.22			0.49	
	RMSE	43	85	93	217			90	
	n	9	9	9	27			9	
Ryegrass	Model	Y=-78.2+379.7*NDVI	Y=-63.6+83.8*VR	Y=-165.2+1540.5*NDVI	Y=-514.2+1645.6*NDVI	Y=0.637+0.019*HT+1.3*NDVI	Y=1.9+0.098*VR	Y=2.1+1.0*NDVI	Y=0.991+2.3*NDVI
	Adj R^2	0.69	0.47	0.69	0.59	0.84	0.60	0.76	0.75
	RMSE	35	152	135	256	23	152	126	257
	n	26	26	26	78	26	26	26	78
Barley	Model	Y=-295.7+109.9*VR	Y=-209.9+30.2*HT	Y=-692.3+159.7*VR+18.4*HT	Y=17.2+21.3*HT	Y=2.0+0.093*VR	Y=1.8+0.033*HT	Y=2.3+0.095*VR	Y=2.3+0.014*HT
	Adj R^2	0.67	0.62	0.87	0.25	0.67	0.56	0.74	0.27
	RMSE	67	84	96	273	71.16	103	122	275
	n	9	9	9	27	9	9	9	27
Combined	Model	Y=-923.6+105.8*VR+19.0*HT	Y=-453.4+104.6*VR+11.4*HT	Y=-642.2+75.7*VR+13.9*HT+888.2*NDVI	Y=-533.7+137.1*VR+8.0*HT	Y=0.455+0.121*VR+0.013*HT+1.17*NDVI	Y=1.7+0.094*VR+0.010*HT	Y=2.0+0.035*VR+0.008*HT+0.582*NDVI	Y=1.1+0.113*VR+0.005*HT+0.884*NDVI
	Adj R^2	0.77	0.57	0.56	0.47	0.87	0.61	0.63	0.58
	RMSE	154	150	156	244	187	153	158	279
	n	105	105	105	315	105	105	105	315

The effectiveness of the log transformation is most noticeable in the scatter plots for ryegrass (Figure 3.3, d and i), which also happened to be the best model for any species combined across clip times. The relationship between predicted and actual forage yield for cereal rye were among the poorest of all the species. The combined models for winter oat show no improvement from transforming the forage yield data as the RMSE increased slightly, despite the R^2 value improving (Figure 3.3, b and h). The combined models developed for both winter wheat and barley clearly did not predict forage yield with a high level of confidence, regardless of using original or log transformed forage yield data.

Figure 3.7 Models developed to predict forage yield of cool-season annual grass species using ground measurements and comparing model performance using original data or log transformed data. Predicted forage yield is compared to actual yield data using a 1:1 (black) line for the following species: a) Wheat b) Oat c) Rye d) Ryegrass e) Winter Barley for original data and f) Wheat g) Oat h) Rye i) Ryegrass j) Winter Barley for log transformed data.



When running validations for models using ground-based measurements to predict forage yield across species for specific clip times, there was a consistent trend. For every individual clip time, including the combined model, the models using the original forage data overpredicted lower forage yields and underpredicted higher forage yield. Using log transformed forage data improved the R^2 values for the models in every case, but also increased the RMSE. Still, using the transformed data was an improvement as it reduced the amount of over or underprediction, though improvements were only modest for the mid-winter and early spring clips. The fall clip time of the log transformed forage data had the highest $R^2=0.87 (p<0.0001)$ (RMSE=187), while the mid-winter and early clip times only achieved a $R^2=0.61 (p<0.0001)$ (RMSE=153) and $R^2=0.63 (p<0.0001)$ (RMSE=158), respectively. The individual clip times were better at predicting forage yield compared to the combined model, which had a lower R^2 and nearly double the RMSE.

Figure 3.8 Models developed to predict forage yield of cool-season annual grass species using ground measurements and comparing model performance using original data or log transformed data. Predicted forage yield is compared to actual yield data using a 1:1 (black) line for the following clip times: a) Fall clip b) Mid winter c) early Spring d) combined for original data, e) Fall clip f) mid winter g) early spring h) combined for log transformed data.

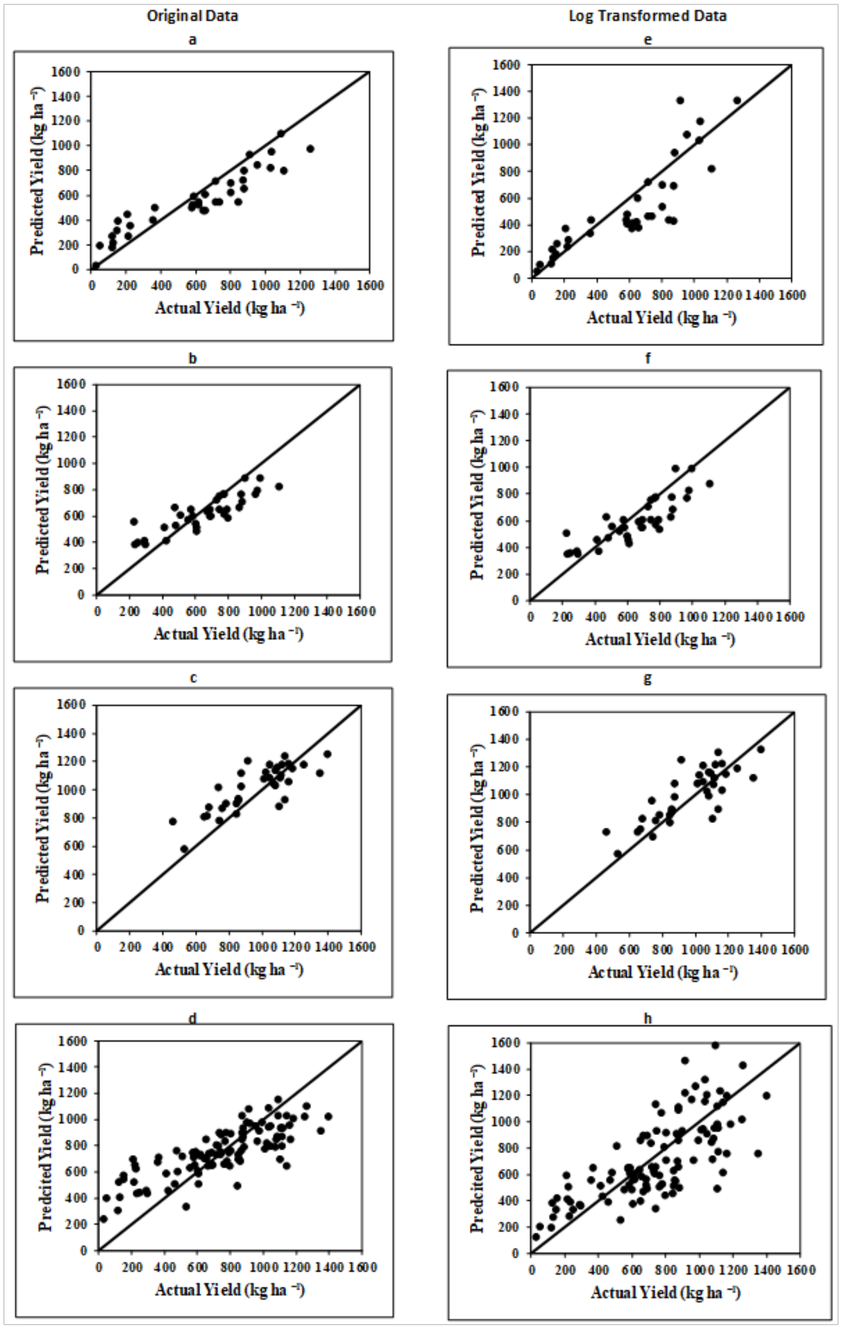


Table 3.7 compares the R^2 for all measured variables in the study which include PH visual rating, handheld NDVI, R/G, r/g, MGRVI, RGBVI, ExG, and g/r alongside the aerial and ground models and using both original or log transformed forage yield data. For each species, the variables that correlated well to forage yield varied widely among species and clip times. In many cases, the aerial or ground models made little or no improvement to the cR^2 values; however, there were some instances where it made a significant improvement such as with the early spring clip of wheat and oat, fall and mid-winter clips for rye, fall and early spring clips for barley, and early spring clip for the combined data. The early spring clip benefitted the most often from the ground-based model. The ground-based model in general did better than aerial-based models for wheat, oat, and barley; generally did worse for rye, and was not much different for ryegrass. In general, combining data across clip times for specific species did not achieve good results, though there were some exception such as with rye (aerial model) and ryegrass (aerial model).

Winter wheat had the highest $R^2=0.61 (p<0.0001)$ for both the original (Y=-699.2+174.1*VR) and log transformed (Y=1.6+0.149*VR) forage yield data during the mid-winter clip time with the ground method which only utilized VR in the model. This was only a slight increase from using the visual rating alone for both the original ($R^2=0.58 (p<0.0001)$) and log transformed ($R^2=0.56 (p<0.0001)$) forage data. The VR produced the highest $R^2=0.69 (p<0.0001)$ for the oat mid-winter clip using the log transformed forage yield data. Neither model produced a better R^2 value. The best R^2 for cereal rye ($R^2=0.73$ ($p<0.0001$)) was generated by both the original (Y=-

751638+751650*gr) and log transformed (Y=-979.7+981.8*gr) models during the mid-winter clip using the aerial method which only utilized the gr VI in the model. Ryegrass had the highest $R^2=0.85$ ($p<0.0001$) for log transformed (Y=0.637+0.019*HT+1.3*NDVI) forage yield data during the fall clip time with the ground model. In this case, the model utilized plant height and hand held NDVI. For the combined clip time for ryegrass the aerial model (Y=2.1+3.7*MGRVI) had the highest correlation to yield of the log transformed forage yield data with a $R^2=0.84$ ($p<0.0001$). Winter barley had the highest $R^2=0.90$ ($p<0.0001$) for the original forage yield data (Y=-692.3+159.7*VR+18.4*HT) during the late spring clip time with the ground method which utilized the VR and HT in the model. When combining species, the best correlation occurred using log transformed forage yield data for the fall clip time. Both the ground model (Y=0.455+0.121*VR+0.013 *HT+1.17*NDVI) with an $R^2=0.88$ ($p<0.0001$) which used the visual rating, NDVI and plant height, and aerial model (Y=-531.3+ 0.022 *HT+ 532.7*gr) with a $R^2=0.89$ ($p<0.0001$) and utilized plant height and the gr VI were very highly correlated to forage yield. When combining clip times across species, the highest correlations occurred with the log transformed forage yield data and the ground model (Y=1.1+0.113*VR +0.005*HT+0.884*NDVI; $R^2=0.59$ ($p<0.0001$)). The ground model included all three variables: visual rating, plant height and hand held NDVI.

Table 3.7 Coefficient of determination of original vs log transformed data for plant height, visual rating, hand held NDVI, R/G, r/g, MGRVI, RGBVI, ExG, and g/r individual variables compared to aerial and ground models.

	Original Data				Log Transformed Data			
	Winter Wheat							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.27	0.21	0.13	0.27	0.18	0.22	0.15	0.27
VR	0.24	0.58	0.15	0.11	0.18	0.56	0.17	0.13
NDVI	0.06	0.07	0.13	0.00	0.04	0.13	0.12	0.01
R/G	0.12	0.09	0.00	0.00	0.02	0.13	0.00	0.00
r/g	0.12	0.10	0.00	0.00	0.12	0.15	0.00	0.00
MGRVI	0.12	0.08	0.00	0.00	0.14	0.13	0.00	0.00
RGBVI	0.01	0.08	0.01	0.00	0.03	0.29	0.00	0.00
ExG	0.05	0.14	0.00	0.00	0.08	0.20	0.00	0.00
g/r	0.12	0.10	0.00	0.00	0.12	0.15	0.00	0.00
n	n=38	n=38	n=38	n=114	n=38	n=38	n=38	n=114
Aerial Model	0.32	0.47	0.33	0.22	0.29	0.58	0.34	0.26
Ground Model	0.22	0.61	0.52	0.22	0.18	0.61	0.54	0.26
n	n=28	n=28	n=28	n=84	n=28	n=28	n=28	n=84
	Oat							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.20	0.00	0.03	0.03	0.20	0.00	0.06	0.04
VR	0.42	0.67	0.30	0.40	0.40	0.69	0.09	0.46
NDVI	0.00	0.14	0.17	0.05	0.00	0.15	0.16	0.06
R/G	0.00	0.26	0.06	0.02	0.00	0.24	0.07	0.00
r/g	0.01	0.29	0.07	0.02	0.01	0.25	0.08	0.00
MGRVI	0.00	0.26	0.06	0.02	0.00	0.23	0.07	0.00
RGBVI	0.03	0.15	0.04	0.01	0.03	0.14	0.04	0.00
ExG	0.01	0.21	0.00	0.02	0.02	0.18	0.00	0.00
g/r	0.01	0.29	0.07	0.02	0.01	0.25	0.08	0.00
n	n=44	n=44	n=44	n=134	n=44	n=44	n=44	n-134
Aerial Model	0.31	0.31	0.00	0.06	0.31	0.27	0.00	0.00
Ground Model	0.40	0.67	0.32	0.36	0.42	0.69	0.31	0.45
n	n=33	n=33	n=33	n=99	n=33	n=33	n=33	n=99

Table 3.7 Continued.

	Original Data				Log Transformed Data			
	Rye							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.38	0.55	0.59	0.23	0.4	0.5	0.56	0.14
VR	0.24	0.5	0.01	0.01	0.18	0.4	0.01	0.02
NDVI	0.31	0	0.04	0.06	0.36	0	0.06	0.02
R/G	0.41	0.32	0.4	0.56	0.36	0.38	0.17	0.46
r/g	0.38	0.36	0.38	0.5	0.32	0.42	0.4	0.42
MGRVI	0.41	0.32	0.4	0.55	0.36	0.37	0.17	0.45
RGBVI	0.41	0.1	0.15	0.5	0.34	0.14	0.17	0.46
ExG	0.42	0.21	0.28	0.52	0.36	0.26	0.29	0.45
g/r	0.38	0.36	0.38	0.5	0.32	0.42	0.4	0.42
n	n=12	n=12	n=12	n=36	n=12	n=12	n=12	n=36
Aerial Model	0.62	0.73	0.57	0.7	0.54	0.73	0.55	0.46
Ground Model	0.49	0.52	0.57	0.25	0	0	0.55	0
n	n=9	n=9	n=9	n=27	n=9	n=9	n=9	n=27
	Ryegrass							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.31	0.1	0.07	0	0.48	0.13	0.08	0.01
VR	0.52	0.48	0.58	0.62	0.61	0.61	0.62	0.72
NDVI	0.69	0.33	0.69	0.61	0.81	0.55	0.77	0.77
R/G	0.67	0.46	0.62	0.48	0.79	0.58	0.69	0.85
r/g	0.74	0.45	0.58	0.55	0.79	0.5	0.59	0.83
MGRVI	0.69	0.46	0.61	0.72	0.79	0.56	0.66	0.85
RGBVI	0.67	0.46	0.59	0.55	0.76	0.56	0.64	0.72
ExG	0.71	0.45	0.61	0.66	0.77	0.53	0.64	0.77
g/r	0.74	0.45	0.58	0.74	0.79	0.5	0.59	0.83
n	n=35	n=35	n=35	n=105	n=35	n=35	n=35	n=105
Aerial Model	0.71	0.47	0.7	0.73	0.84	0.65	0.74	0.84
Ground Model	0.71	0.49	0.7	0.6	0.85	0.61	0.77	0.75
n	n=26	n=26	n=26	n=78	n=26	n=26	n=26	n=78

Table 3.7 Continued.

	Original Data				Log Transformed Data			
	Winter Barley							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.14	0.67	0.64	0.27	0.14	0.61	0.59	0.29
VR	0.56	0.53	0.76	0.21	0.55	0.49	0.72	0.24
NDVI	0.23	0.04	0.01	0	0.25	0.04	0.02	0
R/G	0.04	0.18	0.34	0.18	0.04	0.18	0.34	0.08
r/g	0.06	0.21	0.34	0.1	0.06	0.21	0.34	0.03
MGRVI	0.04	0.18	0.32	0.16	0.04	0.18	0.32	0.07
RGBVI	0.07	0.28	0.5	0.14	0.07	0.29	0.5	0.04
ExG	0.07	0.23	0.38	0.12	0.06	0.23	0.38	0.04
g/r	0.06	0.21	0.34	0.21	0.06	0.21	0.34	0.03
n	n=12	n=12	n=12	n=36	n=12	n=12	n=12	n=36
Aerial Model	0	0.67	0.69	0.42	0	0.61	0.68	0.3
Ground Model	0.71	0.67	0.9	0.28	0.71	0.61	0.78	0.3
n	n=9	n=9	n=9	n=27	n=9	n=9	n=9	n=27
	Combined							
	Fall	**Mid-winter**	**Early spring**	**Combined**	**Fall**	**Mid-winter**	**Early spring**	**Combined**
Plant Height	0.64	0.27	0.16	0.15	0.57	0.28	0.17	0.09
VR	0.69	0.53	0.46	0.46	0.8	0.56	0.49	0.55
NDVI	0.54	0.07	0.22	0.22	0.76	0.11	0.27	0.37
R/G	0.53	0.2	0.02	0.13	0.79	0.25	0.03	0.29
r/g	0.58	0.23	0.02	0.13	0.8	0.26	0.03	0.26
MGRVI	0.55	0.2	0.02	0.13	0.8	0.25	0.03	0.27
RGBVI	0.41	0.17	0.03	0.06	0.69	0.23	0.03	0.17
ExG	0.51	0.18	0.03	0.09	0.76	0.23	0.04	0.21
g/r	0.58	0.23	0.02	0.13	0.8	0.26	0.03	0.26
n	n=141	n=141	n=141	n=423	n=141	n=141	n=141	n=423
Aerial Model	0.77	0.44	0.28	0.24	0.89	0.49	0.33	0.36
Ground Model	0.78	0.58	0.57	0.47	0.88	0.62	0.64	0.59
n	n=105	n=105	n=105	n=315	n=105	n=105	n=105	n=315

Conclusions

In this study, no one variable showed a strong relationship across all clip times or species. This was expected due to differences in physiological appearance and growth stage. Models were developed for each species and clip time to better predict forage yield using multiple variables. In some cases, the models did improve the R^2 substantially, but in many cases, improvement was marginal. The ground model did as good as or better than the aerial model for wheat, oat, barley and the combined model, whereas the aerial model was far better for rye. There was little to no difference between the ground and aerial models for ryegrass. Transforming the data also had little effect on model accuracy for wheat, oat and barley. However, transforming the data for rye had a negative effect while ryegrass and the combined species model were improved when the data was transformed.

When individual species were combined across clip time wheat, oat, and barley had very poor results with both the ground and aerial models. This is a good indication that it would not be beneficial to evaluate wheat, oat and barley over the course of a growing season, but rather select the best clip time to conduct an evaluation for each species. When the original data for rye was evaluated across clip times, the aerial model produced good results with a significant improvement over the ground model. The best results for a combined clip time was the log transformed ryegrass aerial model. Interestingly there were very good results when all the species were combined across clip times, especially when using the transformed data for the ground model. This could be misleading as the rye and ryegrass may overcompensate for the poor combined results of the wheat, oat and barley. Due to the scatter and large RMSE of the overall combined model across clip time and species, it would be more useful to utilize a combined species model for a specific clip time than across clip times. The best-combined species clip time was in the fall when the transformed data was used for either the ground or aerial model. This would indicate that a combined species forage model would be fairly reliable in estimating forage yield early in the growing season, perhaps 60 days post emergence.

Both winter wheat and oat provided the best estimation of forage yield during the mid-winter (February) clip, which occurred after the first clipping and prior to stem elongation, but even then, results suggest it may not be good enough for reliable prediction of actual yield if trying to select the top performing line or entry in a forage trial. For both species, the ground model, which utilized only the visual rating, was best and log transforming the data only made a slight improvement over the original data. Forage biomass analysis of rye had good results throughout the season with the original data for the aerial method. Although correlations between yield and plant height alone were good for individual clip times throughout the season, the combined clip was poor. This could be attributed to the plants growth pattern progression throughout the growing season. Early in the season the plant growth focuses on vegetative stage and as the season progresses the plant growth moves into a reproductive growth pattern. Plant height is affected by this occurrence in that most of the forage for a plant during the vegetative growth period would be located throughout the plant in which plant height would be a good indicator of forage potential. However, as the plant moves into the reproductive phase more energy is focused on seed production in which the plant would remain tall but not have a tremendous amount of forage in the lower to mid canopy. Interestingly when rye plant height was combined with RG VI the result produced a model with a relatively low RMSE and high R^2 value across clips.

Ryegrass had the most noticeable improvement when the data was log transformed, which made the data more linear. Ryegrass is slow to establish in the fall and does not produce a large amount of forage until later in the winter or spring. Consequently, the best time of the year to evaluate ryegrass was in the fall before canopy closure, which is ideal for reflectance measurements such as NDVI and other VIs, before images become saturated. However, the ryegrass models continued to perform well in later cuts likely due to higher amounts of red band reflectance compared to other species like wheat and oats. Combining plant height with MGRVI consistently improved aerial models for ryegrass as well. In fact, plant height showed up consistently in many of the models regardless of clip time or species. For barley, plant height and visual rating were most useful in estimating forage yield, particularly when combined in the late spring clip.

In general, rye and ryegrass showed much greater potential for producing reliable single species models that would work well across clip times. A single model using UAV spectral sensors could likely be used to estimate forage in the fall for all species if combined with plant height, but reliability will likely decline as the season progresses into the winter, and especially spring, using the techniques described in this study. The ground models using plant height and visual rating in general did a better job than the aerial models for individual species and clip times, particularly visual rating, indicating that a trained eye can reliability select high yielding lines or entries. However, this measurement is subjective, and will inevitably vary from person to person based on experience.

CHAPTER IV

CONCLUSIONS

Knowing the yield potential a particular forage species or cultivar has is useful for livestock managers who rely on good forage production to meet the high demands of their forage systems. It is also important for researchers in forage and small grains breeding programs to accurately measure forage potential for cultivar advancement. Some of the more traditional methods have been utilized for well over 50 years. Recently Texas A&M AgriLife Research has been making efforts to incorporate Unmanned Aerial Vehicles (UAV) into determining cool season annual forage biomass. These vehicles utilize remote sensing imagery to measure reflectance in order to obtain various vegetation indices. The purpose of this research was to determine if UAV technology or other ground-based sampling methods could replace traditional forage harvesting techniques to better assist cool-season annual grass breeders and researchers in estimating forage yield of entries in forage trials and breeding lines for advancement in breeding programs. Ideally, this would provide a low cost and effective method to measure above ground forage biomass.

The traditional method of evaluating a multi-species cool-season annual grass forage trial over several clippings throughout the year was completed over a two-year period. The individual methods used were plant height, visual score and a subsample method. The model created in this study using all three measurements was better at predicting forage yield across species and clip times than any individual method alone. However, the subsample method was consistently better than either visual rating or plant height measurements and would be the only acceptable stand-alone method for comparisons of entries across clip times and species. If researchers are unable to take destructive samples, both visual and plant height measurements were highly correlated for fall and late spring clips. Combining all measurements into one model to predict forage yield may be a better way to estimate forage yield if whole plot forage harvest is not possible.

Remote sensing methods of evaluating forage potential were applied to the 2017 forage trial through the use of UAVs (aerial method). No one variable showed a strong relationship across all clip times or species using the aerial method, so models were developed for each species and clip time to better predict forage yield using multiple variables. In some cases, the models did improve the R^2 values substantially, but in many cases, improvement was marginal. The ground model did as good or better than the aerial model for some species, but not all. Log transforming the data had mixed results and proved to be more useful to ryegrass and the combined model than to wheat, oat, barley and ultimately having a negative effect on rye.

This study has demonstrated that both ground and aerial methods can be useful in predicting forage yield of cool-season annual grasses, but may be specific to certain species or clip times. The best results for evaluating multi-species across clip times was the model produced using the ground measurements of visual rating, plant height and sub sample weight. This method is labor intensive; however; it does not require expensive plot harvesting equipment, which can be a major limitation to smaller forage breeding operations. The aerial method was equally as good at producing a combined model for the fall clip, but was inferior to the ground model in subsequent clips. It seemed far better when evaluating multiple species in a forage trial to do so at specific times during the growing season as opposed to throughout the entire growing season. Regardless of ground or aerial models, the best time to evaluate a combined multi-species trial was during the fall clip, which is generally 45-60 days post emergence. If only a single species is being evaluated, researchers may want to use models on a case-by-case basis since correlations among measurements varied widely by clip time and species. If the main goal is to remain non-destructive, the best ground model provided modest results for winter wheat in the mid-winter and early spring clip times, mid-winter only for oats, and all three clip times for rye. The best ground model showed good potential for barley and ryegrass for all three clip times. The best aerial models rarely exceeded the potential of ground base models except for rye, and performed similarly for ryegrass (all clips), wheat (mid-winter only), and barley (mid-winter only). The best results from a remote sensing platform were obtained for ryegrass. Depending on the researches resources and goals, forage evaluation on either a multi or single species forage trial could be obtained at a low cost and with minimal labor.

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APPENDIX A

Appendix 2.1. Timing and grouping of forage clips from a cool-season annual grass forage trial conducted in College Station, TX across the 2015-2016 and 2016-2017 growing seasons.

Time of Year

Clipping Date	**FALL**	**MID WINTER**	**EARLY SPRING**	**LATE SPRING**
January 19, 2016		X		
March 28, 2016			X	
April 26, 2016				X
November 30, 2016	X			
February 8, 2017		X		
March 3, 2017			X	