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CONTEÚDO DE CARBOIDRATOS ESTRUTURAIS E LIGNINA EM GRAMÍNEAS FORRAGEIRAS PERENES DEPENDENDO DA FASE DE CRESCIMENTO

ASSESSMENT OF CONTENTS OF STRUCTURAL CARBOHYDRATES AND LIGNIN OF PERENNIAL FODDER HERBAGES DEPENDING ON VEGETATIVE STAGE GROWTH

СОДЕРЖАНИЕ СТРУКТУРНЫХ УГЛЕВОДОВ И ЛИГНИНА В МНОГОЛЕТНИХ КОРМОВЫХ ТРАВАХ В ЗАВИСИМОСТИ ОТ ФАЗЫ РОСТА

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RESUMO

O objetivo desses estudos é avaliar as gramíneas – *Bromus inermis*, *Festuca pratensis*, *Phleum pratense* e leguminosas – *Trifolium pratense* e *Medicago sativa* (alfafa) de acordo com o teor de fibra em detergente ácido (FDA), fibra em detergente neutro (FDN) e lignina em detergente ácido (LDA), hemicelulose (HC) e celulose (C), dependendo da fase da vegetação: gramíneas – no início da emergência do tubo, nas fases da emergência de cabeça e floração; leguminosas – nas fases da emergência de caule, início da emergência de botão e floração. Verificou-se que à medida que as gramíneas crescem, há um aumento em todas as frações das paredes celulares das leguminosas. As paredes celulares ocupam uma proporção menor de matéria seca do que nas gramíneas, devido ao seu menor conteúdo de HC – 10-12%, em comparação com 25-30% nas gramíneas. Os níveis de FDA, FDN e LDA (% em matéria seca) nelas são – na fase inicial – 25-28, 35-37, 6-7; no início da emergência de botão – 34-38, 41-46, 7-9; no início da floração – 38-41, 49-52, 10-12, respectivamente. Nas leguminosas, observa-se um maior conteúdo de LDA. Conforme a estação de crescimento de gramíneas, a proporção de FDA na FDN aumenta, mas nas gramíneas não excede 50%, enquanto nas leguminosas é de 70-80%, independentemente da fase de crescimento, o que explica a digestibilidade mais baixa das leguminosas em comparação com as gramíneas. Para poder avaliar os níveis de FDA e FDN em gramíneas quanto ao conteúdo de fibra bruta (FB), as equações de regressão correspondentes foram calculadas para gramíneas e forragens preparadas a partir delas. A relação entre FB e FDA foi mais próxima (n = 64, s = 2,4%, r = 0,93) do que entre SC e FDN (n = 64, s = 4,4%, r = 0,87). Com base na pesquisa conduzida e na generalização dos dados da literatura, as normas para o conteúdo de FDA e FDN no padrão para feno e silagem são recomendadas.

Palavras-chave: *gramíneas e leguminosas, fazes de crescimento, fibra bruta, fibra em detergente ácido, fibra em detergente neutro.*

ABSTRACT

The purpose of this study is to evaluate cereal grasses – *Bromus inermis*, *Festuca pratensis*, *Phleum pratense*, and legumes: *Trifolium pratense*, *Medicago varia* in terms of their content of acid-detergent fibre (ADF), neutral detergent fibre (NDF), acid-detergent lignin (ADL) and hemicellulose (HC), cellulose depending on phases of vegetation – grasses: at vegetative, earing and flowering; legumes– vegetative. It was found that as the herbs grow, an increase in all fractions of the cell walls of leguminous herbs is observed, the cell walls occupy a smaller fraction of dry matter than in grasses, due to the lower HC content in them –10-12%, compared with 25-30% in grasses. The contents of ADF, NDF and ADL (% of dry matter) in grasses prior earing are 31-31, 50-55, 4-6: in earing – 32-37, 55-65, 5-6; in flowering – 40-45, 65-70 and 70-72, 7-9, respectively. Legumes have a higher content of ADL. As plants grow, the relative proportion of ADF in NDF increases, but it does not exceed 50% in grasses. In legumes – 70-80% regardless of the growth phase, explains the lower digestibility of legumes than grasses. To judge the levels of ADF and NDF in herbs, depending on the content of crude fibre (CF), the

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corresponding regression equations were calculated for grasses and feed prepared from them. The relationship between CF and ADF was closer (n = 64, s = 2.4%, r = 0.93) than between CF and ADL (n = 64, s = 4.4%, r = 0.87). Based on these studies and generalisation of the literature data, ADF and NDF in hay and haylage standard are recommended.

Keywords: *perennial fodder, phase of growth, crude fibre, acid detergent fibre, neutral detergent fibre.*

АННОТАЦИЯ

Целью настоящих исследований является оценка злаковых трав – костреца безостого, овсяницы луговой, тимофеевки луговой и бобовых – клевера лугового и люцерны посевной по содержанию в них кислотно-детергентной клетчатки (КДК), нейтрально-детергентной клетчатки (НДК) и кислотно-детергентного лигнина (КДЛ), гемицеллюлоз (ГЦ) и целлюлозы (Ц) в зависимости от фазы вегетации: злаков – в начале выхода в трубку, в фазы колошения и цветения; бобовых – в фазы стеблевания, начала бутонизации и цветения. Установлено, что по мере роста трав наблюдается увеличение всех фракций клеточных стенок бобовых трав клеточные стенки занимают меньшую долю сухого вещества, чем в злаковых травах, за счет более низкого содержания в них ГЦ –10-12%, по сравнению с 25-30% в злаковых. Уровни КДК, НДК и КДЛ (% в сухом веществе) в них составляют – в раннюю фазу – 25-28, 35-37,6-7; в начале бутонизации – 34-38, 41-46, 7-9; в начале цветения – 38-41, 49-52, 10-12, соответственно. В бобовых отмечается более высокое содержание КДЛ. По мере вегетации злаковых трав, возрастает доля КДК в НДК, но у злаковых трав она не превышает 50%, в то время как у бобовых трав –70-80% независимо от фазы роста, чем объясняют более низкую переваримость бобовых трав по сравнению со злаковыми. Чтобы иметь возможность судить об уровнях КДК и НДК в травах в зависимости содержания сырой клетчатки (СК) рассчитаны соответствующие уравнения регрессии для злаковых трав и кормов, приготовленных из них. Связь между СК и КДК была более тесной (n=64, s=2.4%, r=0.93), чем между СК и НДК (n=64, s=4.4%, r=0.87). На основании проведенных исследований и обобщения литературных данных рекомендованы нормы содержания КДК и НДК в стандарте на сено и сенаж.

Ключевые слова: *злаковые и бобовые, фазы роста, сырая клетчатка, кислотно-детергентная клетчатка, нейтрально-детергентная клетчатка.*

1. INTRODUCTION

Structural carbohydrates (SC) together with lignin make up the cell walls of plants. These include pectin, hemicellulose (HC) and cellulose (C) (Bailey, 1973). They are of interest due to their value as an energy source for ruminants, which can partially digest them with the help of the microflora of the gastrointestinal tract. It should also be noted the physiological role of structural substances, which consists in ensuring the normal functioning of the rumen and motor function of the gastrointestinal tract (White *et al.,* 2017). Structural carbohydrates together with lignin are combined under the general name “fibre” or “cellulose”. In this case, “cellulose” does not mean “cellular tissue”, but is a term adopted when evaluating feed by the content of structural (fibrous) substances (Kammoun *et al.*, 2020; Rocha-Meneses *et al.*, 2020).

Since the end of the last century, the evaluation of feed and rations for the content of detergent forms of fibre in them has become widespread throughout the world. In the Russian Federation, some forages and forage grasses were also studied by the level of ADF and NDF, for example, in the conditions of the Moscow, Kaluga and Orenburg regions (Vorobyova, 2002; Kharitonov *et al*., 2008; Levakhin *et al*., 2015) and Tatarstan, studies have been conducted on digestion in bull calves and cows depending on the content of structural carbohydrates in the diet. The content of NDF in feed tables is also given (Kirilov, 2009). However, there is still insufficient information on acid-detergent fibre (ADF) levels and neutral detergent fibre (NDF) in different types of forage grasses depending on the growing phase and other conditions.

Currently, a routine evaluation of feed quality is carried out according to two schemes of zootechnical analysis. In both schemes, the feed is divided into crude nutrients (crude protein, crude fat) and crude ash (Kharitonov and Hotmirova, 2009). The differences in these schemes relate to estimates of the carbohydrate nutritional value of feed. Carbohydrates of feeds are usually divided into two main groups: non-structural carbohydrates (NSC), localised inside plant cells and carbohydrates of the cell walls, or structural carbohydrates (SC). From the role of these groups in the body of an animal, the carbohydrates of the first group — mono-, di-, and oligosaccharides, low molecular weight fructosides and starch — are almost completely digestible, and they are also called easily hydrolysable or easily digestible carbohydrates (Bykova and Gibadullina, 2010; Levakhin *et al*., 2007). According to the Weende scheme, which has been effectively used since the end of the 19th century, SC and lignin of feed are included in two groups – crude fibre (CF) and nitrogen-free extracts (NFE). However, their separation by species and digestibility of animals is not achieved. The second scheme, called the Van-Soest scheme, is devoid of these shortcomings, which explains its widespread use since the end of the 20th century (Bykova and Gibadullina, 2010; Pan *et al.,* 2017).

Structural carbohydrates and associated lignin are usually determined empirically during feed analyses. This is a widely used method for determining crude fibre and according to the Weende scheme, which was developed in the 19th century, but is still used in many countries, including the Russian Federation. Over the past decades, another group of empirical methods for analysing plant cell walls, based on the extraction of feed with detergent solutions, first proposed by Van-Soest, has also found widespread use. In both schemes, the feed is analysed for the content of raw nutrients: crude protein, crude fat, and crude ash, and structural carbohydrates along with lignin. They are called crude because they are determined by empirical methods under specified conditions and do not represent pure chemical compounds. Crude protein, determined by the method proposed by the founders of the analysis scheme and bearing the name of their surnames – Hanneberg and Shtoman, is an empirical residue obtained by sequential processing of feed with solutions of sulfuric acid and potassium hydroxide of a strictly defined concentration.

In the non-chernozem zone of Russia, in the crops of perennial grasses, such types of cereal grasses as *Bromus inermis*, *Festuca pratensis*, *Phleum pratense* are widely cultivated, and of legumes – *Trifolium pratense*, *Medicago varia*. These types of grasses and feed from them occupy a high proportion in the feed balance of ruminants (Chateigner-Boutin *et al.*, 2018). The research aims to evaluate the most widely cultivated cereal and legume forage grasses by the content of structural carbohydrates and lignin in them, depending on the vegetation phase (Boswotth and Hudson, 2011). The analysis of the content of structural carbohydrates in forage grasses and the linking of their amount to the phases of plant vegetation is necessary to determine the most rational terms of fodder harvesting, since a balanced feed composition is one of the factors of sustainable development of livestock (Subaeva *et al.*, 2018).

The purpose of this work is to assess the amount of structural carbohydrates and lignin in perennial forage grasses depending on the growth phases.

2. MATERIALS AND METHODS

2.1. Samples

The research target was samples of forage grasses grown on soddy-podzolic soil of the Central experimental base of the V.R. Williams All-Russian Research Institute of Feed. *Bromus inermis* (Morshansky 760 cultivar), *Festuca pratensis* (VIK 5 cultivar), *Phleum pratense* (VIK 7 cultivar) were mown during the phases of leaf-tube formation, ear emergence (heading) and flowering; *Trifolium pratense* (VIK 7 cultivar) and *Medicago varia* (Lugovaya 67 cultivar) – during the shooting, flower-bud formation and flowering phases. Herbal growth phases were determined by visual estimation.

2.2. Determination of the content of structural carbohydrates

To determine the content of structural carbohydrates, empirical methods were used to ensure the separation of carbohydrates according to their digestibility in animal nutrition. Weende scheme assumes the use of empirical methods that imply the definition of not clearly defined chemical compounds, but groups of compounds: crude protein, crude fibre, crude ash, crude fat, neutral detergent fibre, acid detergent fibre. These groups are determined under strictly defined conditions. For example, crude fibre is determined by sequential processing of a sample with solutions of sulfuric acid and potassium hydroxide of specified concentrations. Crude fat is determined by an extract of organic solvents. Crude ash is empirically defined as the residue after combustion of a sample of feed in a muffle furnace. According to the Weende scheme, crude fibre is determined analytically, and nitrogen-free extract (NFE) is determined by a calculation method by the formula (Equation 1). According to Weende scheme, structural carbohydrates and lignin are included in two groups – (crude fibre) CF and nitrogen-free extracts.

In the framework of the analysis of feed by the Weende scheme, the level of structural substances is estimated by the content of crude fibre (CF). The sample is prepared in the following way: samples of freshly cut plants are crushed on sample grinders, then dried at a temperature of 60ºC in a drying cabinet with forced ventilation. The dry sample is ground in a mill until it passes through a 1 mm sieve, for this, the international standard "GOST 31675-2012. Stern. Methods for Determining Crude Fibre Content Using Intermediate Filtration". At the same time, the following equipment was used: laboratory scales of high accuracy class, a plant sample grinder, a laboratory mill, an electric drying cabinet with a temperature range in the working chamber from 0°C to 160°C, an electric muffle furnace, an electric vacuum pump, a water jet, an electric stove. However, CF does not represent the sum of indigestible substances, as well as the entire amount of structural carbohydrates, some of which, as well as part of lignin, are removed during its determination and enter, along with non-structural carbohydrates, in the group of NFE. Simultaneously, knowledge of the entire amount of structural carbohydrates and its components is necessary for more accurate feed intake and digestibility prediction.

In the 1960s Van-Soest proposed a method for analysing carbohydrates in cell walls of feed grass using detergents (Van-Soest, 1963a; 1963b). When processing a feed sample with a neutral detergent, the contents of the cell are removed. The resulting residue, called neutral detergent fibre (NDF), is the sum of cellulose (C), hemicellulose (HC) and acid-detergent lignin (ADL), and the acid detergent dissolves the HC, leaving C and ADL – acid-detergent fibre (ADF). NDF, representing the entire amount of SC and lignin, is one of the most important feed quality indicators. The feed intake (Mertens, 2010), its nutritional value (Weiss, 1998) and, ultimately, the productive effect, depend on its level and digestibility. Current models for determining the digestibility of organic matter. Consequently, the energy content in feeds is based on data on the NDF content and its digestion rate in animals. NDF is also necessary to calculate the content of carbohydrates soluble in a neutral detergent (Van-Soest *et al.*, 1991; Beth de Ondarza, 2000) normalised in the diet of cows. The content of ADF correlates with digestibility. On its basis a regression equation is proposed for determining the digestibility of nutrients (Rohweder *et al.*, 1978), which is used to evaluate the energy value of feed (Bogomolov and Malinin, 2008).

Samples of feed dried at a temperature of 60-65°C in an oven with forced ventilation were ground before passing through a sieve with 1 mm openings. Samples of fresh green grasses were taken – *Bromus inermis*, *Festuca pratensis* and *Phleum pratense* on the Central experimental base of the V.R. Williams All-Russian Research Institute of Feed. For this, an area of 5 by 5 meters was allocated on the crops of each culture. After mowing the grass in this area, point samples were taken diagonally, which, after combining, amounted to a mass of 2 kg. The samples were transported to the laboratory within 30 min after taking, and were crushed on a plant grinder into sections of 1-3 cm. After thorough mixing, an average sample weighing 0.5 kg was isolated from the combined sample by dividing the sample by the diagonal, spread on a flat solid surface. The average sample was immediately dried in an oven with forced ventilation in trays with a perforated bottom with a layer of 2 cm at 60°C until constant weight. Then grinded on a mill until passing through a sieve with holes of 1 mm. Analyses for the content of NDF, ADF and ADL were carried out according to (Goering and Van-Soest, 1970). The results of analysis for the content of NDF, ADF and ADL are shown in Figure 1. ADF is the sum of cellulose and lignin, and NDF is the sum of ADF and hemicellulose. Both the ADF and NDF are higher in the flowering phase for all three analysed crops – *Bromus inermis*, *Festuca pratensis* and *Phleum pratense*. Using paper filters instead of glass filters (Vorobiev *et al.*, 1984), hemicelluloses (HC) – by the difference between NDF-ADF, cellulose (C) – ADF-ADL. ADF and NDF were determined in separate subsamples of the sample, ADL – treatment of ADF 72% sulfuric acid. In the samples, the content of NDF, ADF and ADF-lignin was determined. Since NDF is the sum of hemicellulose, cellulose, and lignin, and the ADL is the sum of only cellulose and lignin, hemicellulose was determined by calculation – by subtracting the ADL value from the ADF content. Similarly, the ADF is the sum of cellulose and lignin, so the cellulose content was found by subtracting lignin from the value of the ADL (Table 1). Statistical data processing was performed using the "Excel" program using "Statistica 6.0", (Stat Soft Inc.), USA.

3. RESULTS AND DISCUSSION:

As the herbs grow, the proportion of cell walls in all types of cereal grass increases (Figure 1), although they differ somewhat depending on the growth phase in the level of accumulation of structural substances. So, *Phleum pratense* already in the booting phase was characterised by a higher level of NDF, and in the *Bromus inermis* in the phases of heading and flowering, it remains lower than the other two grasses. In *Festuca* *pratensis* and *Phleum pratense* NDF content to flowering reaches >70%. Types of grasses also differ in the dynamics of the accumulation of structural substances. During the period from shooting to flowering, the increase in NDF in the relatively early ripening *Festuca pratensis* occurred at a higher rate (0.8% per day) than in the *Bromus inermis* (0.45% per day) and the later ripe form – *Phleum pratense* (0.38% per day). In all phases of growth, *Phleum pratense* has a higher ADL content, but its accumulation rate was higher in *Festuca pratensis*.

In legumes, the accumulation of cell walls also increases with growth (Table 2). However, they occupy a smaller fraction of dry matter than in cereal grasses due to the lower hemicellulose (HC) content in them, which did not exceed 10-12%. The increase in the NDF level from the shooting phase to flowering occurs mainly due to cellulose, which is the main part of the ADF. Legumes are also characterised by a higher content of ADL *Bromus inermis*, *Festuca pratensis*, and *Phleum pratense* in terms of lignification in the flowering phase to *Medicago varia*. By the time of flower-bud formation and flowering, there was a more intensive accumulation of cell walls in *Trifolium pratense* than in *Medicago varia*.

As the herbs grow simultaneously as the cell walls accumulate, their composition changes (Table 3). The increase in NDF is mainly due to cellulose and, to a lesser extent, lignin. In this regard, the share of ADF in the composition of the NDF increases, including C and ADL, while there is a significant decrease in the proportion of HC. So, the NDF of young grasses is composed of HC by half. In the future, the proportion of HC decreases, which is especially typical for *Festuca pratensis*.

In NDF of leguminous herbs in all phases of growth, cellulose prevails, and a higher proportion of ADL is noted than in NDF of cereals. In this regard, the proportion of ADF in the NDF in the budding and flowering phases of legumes exceeds 80%, while in cereal grasses, depending on the growth phase, it was in the range 44-64%. Lower digestibility of legume fibre may be associated with the composition of NDF compared to cereal. However, this is offset by a higher rate of legumes digestion, which contributes to an increase of their consumption (West, 1998).

The fact that structural carbohydrates accumulate as herbs grow has been reported in many studies. Such information is most fully contained in the tables on the composition of feeds in the USA and Canada, later data refer to climatic conditions of the US states: North Dakota (Schroeder, 2008); Wisconsin (Mertens, 2002); Colorado (Stanton, 2010). There are results of studies conducted in several other countries, such as, for example, Finland (Rinne *et al*., 2002), Serbia (Vasiljević *et al*., 2009), Czech Republic (Homolka *et al*., 2012), Romania (Leto *et al*., 2013) and others.

The generalisation of this data is somewhat difficult because it was obtained under different soil, climatic and weather conditions. The phases of vegetation are often interpreted and fixed in different ways, mostly by visual estimation. For example, for different researchers, the shooting stage is referred to as the phase of "vegetative growth" or "early stage of growth", or "before heading", "before flower-bud formation". At the same time, this phase in cereal grasses consists of 7 stages, depending on the length of the stem from the tillering node and can last 10-20 days (Zadoks *et al*., 1974). As for the earing and flowering phases, each of them has the duration up to 7-10 days, so in some studies the beginning, middle and end of these phases are noted, and in other cases there is no such separation. At the same time, the composition of growing herbs changes daily. Nevertheless, it is possible to reveal a certain agreement between the data on the content of ADF and NDF, depending on the phase of growth of herbs with the given results.

Achieving optimal ADF and NDF in herbs cannot always be established by the growth phase, especially since the growth phases are somewhat stretched over time. Therefore, it is advisable to determine grass harvesting timing based on actual analysis data by daily sampling throughout the entire period of fodder harvesting, as suggested in (Instruction of the Ministry..., 2009). In the last decade, digital technologies, technologies for remote sensing of the state of crops, and the Internet (Khudyakova *et al*., 2019), have been more active in collecting information on the content of nutrients in the grass. However, these methods require further improvement.

As already noted, in the analysis of feed according to the Weende scheme, the content of fibrous substances and the degree of digestibility of the feed are estimated by the level of crude fibre. To this day, a lot of data on the level of CF was accumulated. Therefore, it is of interest to study the relationship between CF and ADF and NDF to have some opportunity to assess the levels of detergent forms of fibre, based on the content of CF. Correlation analysis (the sample includes the results of the analysis of samples of five types of cereal grasses in three growth phases each, as well as silage and haylage prepared from these herbs) showed a fairly close relationship between CF and ADF (Figure 2, Equation 1). Between CF and NDF, it was less close (Figure 3, Equation 2), which is related to the composition of these fibre types. CF and ADF consist of the same compounds – cellulose and lignin, although in quantitative terms differ from each other, while the NDF, in addition to them, includes hemicellulose (Equations 2-3).

As is known, the adequacy of the regression models depends on the size and composition of the sample; these models describe the relationship rather well for herbs of one family, although of different growth phases within the same soil and climatic zone, but worse for herbs of different families grown under different conditions. However, with a large sample size including samples of herbs obtained in a wide range of growing conditions, it becomes possible to develop more universal equations. For example, in (Pan *et al.*, 2017), it is possible to determine the concentration of ADF in herbs by the regression equations derived from a large sample: for cereal grass (Equation 4), for legumes (Equation 5).

4. CONCLUSIONS:

Based on the purpose of the article, the study evaluated the structural carbohydrates of cereals and legumes (alfalfa and meadow clover) depending on the growth phases. It was found that as the herbs grow, an increase in all fractions of the cell walls of legumes is observed, the cell walls form a smaller fraction of dry matter than in cereal herbs, due to the lower HC content of – 10-12%, compared with 25-30% in grasses. The ADF levels, NDF and ADL (% in dry matter) in them are – in the early phase – 25-28, 35-37.6-7; at the beginning of budding – 34-38, 41-46, 7-9; at the beginning of flowering – 38-41, 49-52, 10-12, respectively. In legumes, a higher content of ADL was noted. As the vegetation of cereal grasses, the proportion of ADF in the NDF increases, but in cereal grass it does not exceed 50%. In comparison, in leguminous grasses it is 70-80% regardless of the growth phase, explaining the lower digestibility of legumes compared to cereal grasses. To assess the levels of ADF and NDF in grasses, depending on the content of crude fibre (CF), the corresponding regression equations were calculated for cereal grasses and feed prepared from them. The relationship between CF and ADF turned out to be closer (n=64, s=2.4%, r=0.93) than between CF and NDF (n=64, s=4.4%, r=0.87).

The results obtained will contribute to making optimal decisions regarding the timing of mowing herbs and their use in feeding livestock.

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NFE (by Veende), % = 100 – (crude protein + crude fibre + crude fat + crude ash) (Eq. 1)

ADF = 1.0764CF+6.3265 (n =64, R^2 =86.5, s =2.43) (Eq. 2)

NDF = 1.5986CF+7.9237 (n =64, R^2 =75.7, s =4.36) (Eq. 3)

ADF= 6.89+0.5NDF, (n=722, R^2=0.62, s =3.1) (Eq. 4)

ADF= -0.73+ 0.82NDF (n= 2899, R^2= 0.84) (Eq. 5)

Table 1. *Results of the analysis for the content of NDF, ADF and ADL, % in dry matter*

Herbs	Growth phases	NDF	ADF	ADF-lignin
Bromus inermis	shooting	45.24	26.97	5.47
	earing	52.50	32.09	6.32
	flowering	65.24	40.14	7.53
Festuca pratensis	shooting	42.70	26.50	4.26
	earing	62.54	36.01	6.32
	flowering	72.00	45.76	7.86
Phleum pratense	shooting	48.84	30.08	6.94
	earing	59.02	36.40	7.80
	flowering	72.20	42.70	10.62

***Table 2**. Contents of structural carbohydrates and lignin in fresh herbage of Medicago varia and Trifolium pratense, % in dry matter*

Structural substances	***Medicago varia***			***Trifolium pratense***		
	shooting	**budding**	**flowering**	**shooting**	**budding**	**flowering**
ADF	28.9	34.3	41.0	25.4	38.2	45.1
NDF	37.0	40.6	48.8	35.7	46.7	52.3
CF	21.7	26.4	32.9	15.8	26.1	32.1
HC	8.1	6.3	7.8	10.3	8.6	7.2
C	22.8	27.4	31.1	19.3	29.4	32.6
L	6.1	6.9	10.0	6.1	8.8	12.5

***Table 3.** The composition of cell walls, % in NDF*

Crop cultures	**The proportion of cell wall components in % of NDF**			
	ADF	**HC**	**C**	**ADL**
Bromus inermis				
– shooting	52.6	47.4	41.6	11.1
– earing	55.8	39.2	44.8	11.0
– flowering	61.4	38.6	49.8	11.5
Festuca pratensis				
– shooting	43.6	56.4	34.5	9.1
– earing	57.6	42.4	47.5	10.1
– flowering	63.6	36.4	52.6	10.9
Phleum pratens				
– shooting	51.1	48.9	39.3	11.8
– earing	52.7	47.3	41.4	11.3
– flowering	59.1	40.8	44.4	14.7
Trifolium pratense				
– shooting	71.1	28.9	54.2	17.0
– budding	83.0	18.3	62.8	18.9
– flowering	86.2	13.4	62.4	23.8
Medicago varia				
– shooting	78.1	22.1	61.7	16.4
– budding	84.5	15.5	67.6	16.9
– flowering	84.1	15.9	63.6	20.4

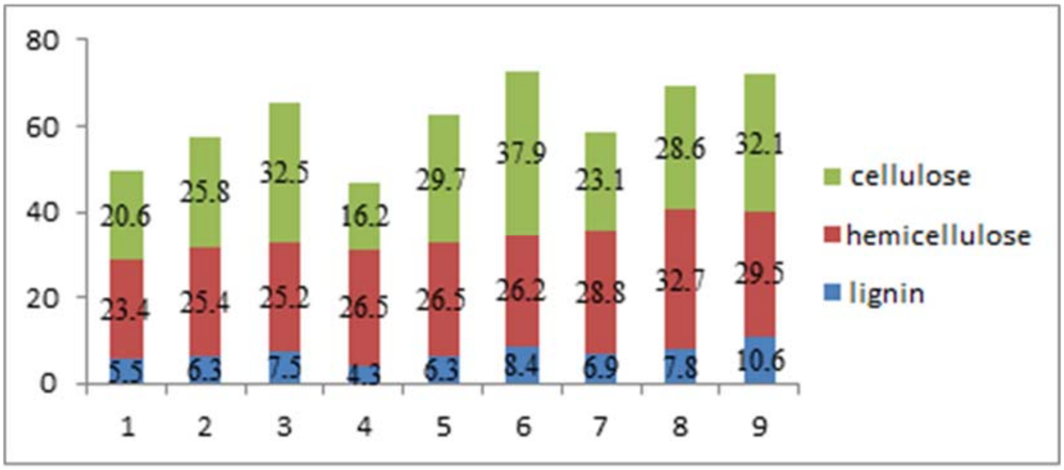

Figure 1. *Contents of structural carbohydrates and lignin in grasses at different vegetation phases, % dry matter: 1, 2, 3 – Bromus inermis at vegetative, earing and flowering, respectively; 4, 5, 6 – the same for festuca pratense; 7, 8, 9 – the same for Phleum pratense*

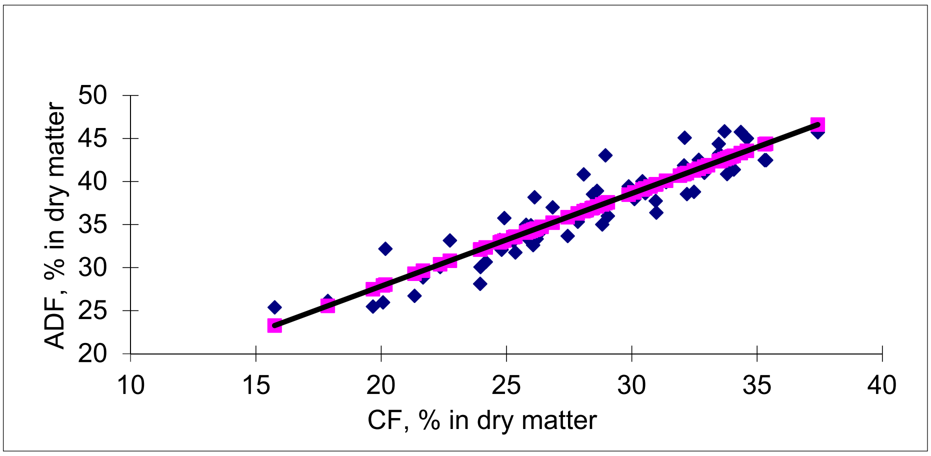

Figure 2. *The relationship CF to ADF*

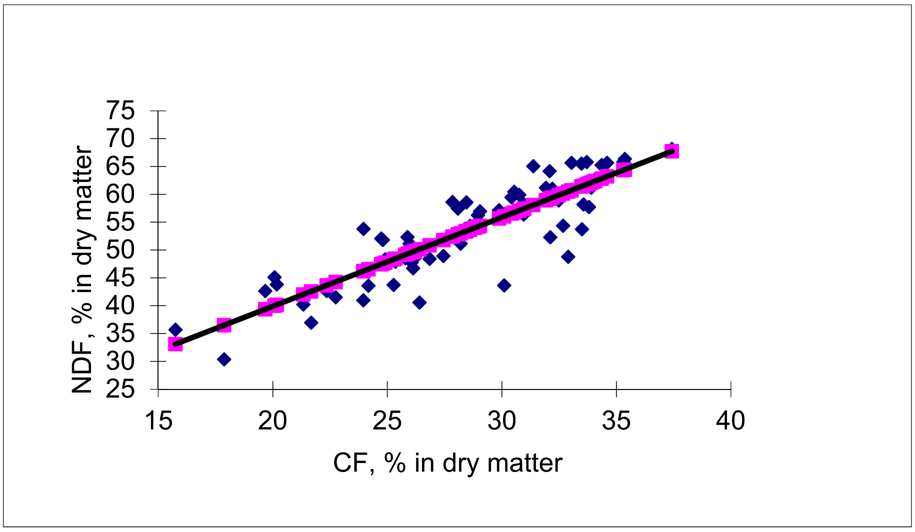

Figure 3. *The relationship CF to NDF*