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Review
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INTEGRATION OF RED CLOVER INTO AGRICULTURAL SYSTEMS TO ADDRESS CONTEMPORARY CHALLENGES

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Red clover is a valuable leguminous perennial crop and is considered one of the best predecessors for many agricultural cultures regardless of its usage method. This work compiles the latest scientific developments related to growing red clover as a cover crop, forage, and bioenergy, and analyzes its utilization methods. It's a globally significant crop, as it is cultivated on all continents. Research on the use of red clover in crop rotations carries an alternative and innovative character, especially considering the modern warming conditions that will enhance the intensification of processes of parent rock carbonate migration. Red clover is mainly used as fodder for livestock (and is fed to almost all types of animals and birds in various forms: hay, silage, haylage granulated grass flour, etc.). Its yield can reach 50-87 t green mass ha-1 or 10-15 t dry matter ha-1, and it accumulates up to 500 kg Nitrogen ha-1 in the soil, which improves its fertility. The use of red clover's herbaceous biomass, especially if it is surplus for livestock use, or bioenergy production, is an alternative way of using this agricultural culture. A prospective direction for further research could involve comparing the efficiency of cultivating red clover for use as a cover crop, fodder, and bioenergy purposes, to develop a comprehensive strategy for fully harnessing the potential of red clover and effectively integrating it into modern agricultural practices.

Keywords: red clover, grassland, legumes, environmental impact, forages.

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Конюшина лучна є цінною бобовою багаторічною культурою і вважається одним з найкращих попередників для багатьох сільськогосподарських культур незалежно від способу використання. У цій роботі узагальнено останні наукові розробки, пов'язані з вирощуванням конюшини лучної як покривної, кормової та біоенергетичної культури, а також проаналізовано способи її використання. Конюшина є глобально значущою культурою, оскільки вирощується на всіх континентах. Дослідження щодо використання конюшини лучної в сівозмінах мають альтернативний та інноваційний характер, особливо з огляду на сучасні умови потепління, які сприятимуть інтенсифікації процесів міграції карбонатів материнських порід. Конюшина лучна в основному використовується як корм для худоби (згодовується практично всім видам тварин і птахів у різних формах: сіно, силос, сінаж, гранульоване трав'яне борошно тощо). Її врожайність може досягати 50–87 т/га зеленої маси або 10–15 т/га сухої речовини, а в ґрунті вона накопичує до 500 кг/га азоту, що покращує його родючість. Використання трав'янистої біомаси конюшини лучної, особливо якщо вона є надлишковою для використання у тваринництві або для виробництва біоенергії, є альтернативним способом використання цієї сільськогосподарської культури. Перспективним напрямком подальших досліджень може бути порівняння ефективності вирощування конюшини лучної для використання як покривної культури, кормової та біоенергетичної, з метою розробки комплексної стратегії для повного використання потенціалу конюшини лучної та ефективної інтеграції її в сучасні сільськогосподарські практики.

Ключові слова: конюшина лучна, травостої, бобові, вплив на довкілля, корми.

Introduction. The modern world is facing the challenge of increasing population numbers. According to the UN, the global population is projected to reach 9.5 billion by the year 2050 [117], and the global demand for food, feed, and biofuels will increase by 70 % [42]. This necessitates an increase in production, thus requiring improved efficiency in agricultural cultivation. The application of new technologies based on perennial grasses will help prevent the escalation of a global food crisis. This also aligns with the "green" measures of the Common Agricultural Policy, which stipulates those permanent grasslands should occupy no less than 5 % of the total agricultural land area [39].

In the world, grasslands cover 3.2 × 109 hectares [41], while in Europe, they occupy 5.4 × 107 hectares [39]. The multi-functionality of these agroecosystems is gaining increasing importance [107]. Enhancing plant species diversity through the cultivation of complex mixed stands of grasses and leguminous forage plants in grasslands combines the potential for high dry matter yield [95], high-quality fodder [63], improved dairy cow productivity [76], and reduced greenhouse gas emissions [14].

Alongside permanent grasslands, perennial grasses are widely utilized in crop rotations, where they are grown for 1–2 years. Cultivating red clover in short rotation sequences positively affects soil fertility, particularly by increasing soil nitrogen concentration [9, 10], organic and total carbon content [59], and amino sugars [22] in the soil.

Red clover is often used as fodder for agricultural animals and for energy purposes [38]. Alongside this, this crop plays a significant agroecological role as it contributes to soil fertility improvement. Red clover serves as a potent source of nitrogen accumulation in the soil [67, 81].

The sown area of red clover in Ukraine constitutes over 25 % of the total sown area of perennial grasses or more than 300 thousand ha [64]. In pursuit of sustainable agricultural production and recognizing the benefits of leguminous crops, including red clover, there is encouragement to expand its cultivation area [86]. This crop is considered one of the best precursors for various agricultural cultures regardless of its utilization method [18, 47, 113].

Red clover is a key perennial forage legume of temperate zones, with global utilization. It's recognized as one of the most important leguminous crops worldwide, capable of producing high-protein, digestible fodder [29, 49]. Red clover is known for its ability to rapidly establish and thrive across a wide range of soil types, pH levels, and environmental conditions.

Recent research highlights significant climate changes [19] and their impact on agroecosystems [6, 108, 118]. Key climate change scenarios for Europe predict higher summer and winter temperatures, increased winter precipitation in most regions, and more frequent extreme weather conditions [50]. According to scientists [8], despite advances in science, new technologies, and developments, the global productivity of all agricultural crops has declined by 21 % over the past 60 years due to climate change. Integrating red clover into agricultural systems can enhance their resilience to climate change [58].

According to the findings of Heslop et al. [49], a key survival trait of plants under stress conditions, be it excess moisture or drought, is the dry mass of roots. This trait significantly influences the growth and development of red clover. Red clover can accumulate over 4.08 tons per hectare of root dry matter in the soil, containing more than 80 kg/ha of nitrogen, 24 kg/ha of phosphorus, and 51 kg/ha of potassium [123]. The root system of perennial grasses, located in the plow layer of the soil, is a primary component that facilitates morphological, physiological, physical, and metabolic interactions between above-ground and below-ground plant parts [115].

Tucak et al. [13] investigated twenty-three varieties and populations of red clover from various geographic origins to assess the impact of climatic conditions (precipitation and temperature) on productivity and identify those with high forage potential under ecological stress. It was found that weather conditions with higher precipitation contributed to increased productivity in all varieties and populations, and the yield of the best specimens reached 83.3 tons per hectare of green mass. Due to the biological nitrogen fixation effect, leguminous forages like red and white clover can maintain productivity for up to 30 days during drought [78]. However, according to Stevenson et al.'s research, red clover has a low tolerance to waterlogging, leading to a reduction in chlorophyll content and dry matter yield [101].

Systematizing research related to the cultivation of red clover and conducting an analysis of its utilization methods based on it will help identify promising directions for future research aimed at effective agricultural production management while simultaneously improving soil fertility.

The agroecological significance of red clover

Grasslands, comprising the primary element of various landscapes, play a crucial role in fostering sustainable ecosystem development and have a positive influence on society as a whole [61, 60]. Perennial leguminous grasses act as soil structure formers, serve as excellent predecessors for most crops, and protect the soil from erosion [3, 88].

Cover Crops

Currently, agricultural plants utilize only 30–53 % of applied nitrogen fertilizers [43]. One important approach to nitrogen supply for agricultural plants is the use of organic fertilizers, especially considering the increasing production of nitrogen from biological materials [18]. The value of using organic fertilizers to enhance the productivity of perennial grasses has been demonstrated by various researchers, including a strong positive correlation between the quality of organic fertilizers and biomass accumulation reported by J. K. Nyameasem et al. [31]. Replacing synthetic nitrogen fertilizers could be achieved through the cultivation of red clover, which is used as a cover crop [30, 77, 79], and this could be most feasible if nitrogen fixation in biomass or immobilization in the soil is utilized [32, 112].

The rising costs of mineral fertilizers necessitate the development and refinement of alternative strategies to supply cereal crops with mineral nutrients, particularly nitrogen. Research conducted in England found that reducing fertilizer usage, increasing the use of new red clover varieties, and enhancing farmers' ability to assess botanical composition are potential strategies to improve red clover utilization in grasslands [98]. Perennial leguminous grasses are grown in crop rotations to enhance nitrogen and other nutrient cycling and availability for other crops in the rotation [69].

Studies conducted in the state of Iowa (USA), investigating the use of red clover and alfalfa as cover crops to provide nitrogen for succeeding corn crops, confirmed that the impact of leguminous cover crops on corn yield was associated with nitrogen. Specifically, without nitrogen fertilizer application, corn grain yield following oats with red clover inter-seed was 63 % higher than after oats grown alone, but at the highest fertilizer rate (202 kg/ha of nitrogen), there was no yield difference between legume and oat treatments. These studies also found that inter-seeded red clover produced more biomass and contained more nitrogen than inter-seeded alfalfa. Red clover cover crop can substitute for 87–184 kg/ha of nitrogen for corn, while alfalfa can substitute for 70–121 kg/ha of nitrogen [10].

Research by Sarauskis et al. [37] on different fertilization systems in a crop rotation with winter wheat and winter barley with under-seeded red clover found that using red clover as a cover crop resulted in an energy efficiency coefficient of 8.73 on low-carbon soils and 9.15 on high-carbon soils. These coefficients were the highest among all studied fertilization systems (cover crop + manure, manure + NPK, and cover crop + NPK). Researchers noted that manure application led to a tenfold increase in total greenhouse gas emissions compared to cover crop application. These findings are corroborated by other studies [109].

Rasmussen et al. [77] sowed barley after different cereal-legume mixtures and were surprised to find that, in terms of N_2 fixation, high-yielding alfalfa had the same legacy effect as white clover, which fixed only half the nitrogen contained in the grass-legume mixture. They suggested that this was due to the higher nitrogen content in alfalfa residue, which produced more biomass and, consequently, more carbon input into the soil.

According to data from Lynge et al. [104], using red clover as a cover crop can provide around 500 kg/ha of nitrogen, but this is achievable only if it's cut 4–5 times a year. The second cut of a two-cut system allows for 210 kg/ha of nitrogen. Scientists assert that the amount of nitrogen red clover can contribute as a cover crop depends on the cultivation conditions. This hypothesis is supported by other studies, where nitrogen release from cover crops ranged from 230 to 470 kg/ha [89].

The impact of cover crops from red clover and alfalfa on succeeding crops, according to individual researchers, can exceed the impact of organic fertilizers such as poultry manure when used in vegetable crops [68], or it can be lower than the impact of cattle manure when used in barley cultivation [119].

The positive influence of inter-seeded red clover under winter wheat and its use as a cover crop preceding corn has been confirmed in studies by Schipanski et al. [97] and Gaudin et al. [58].

Red clover is cultivated as a cover crop in the inter-rows of black currants, with the critical factor being the time interval between mowing and incorporating cover crops into cash crops [93]. In these studies, red clover yielded up to 13 tons/ha of dry matter with a nitrogen content of 58 kg ha-1.

Nitrogen Fixation

Grasslands incorporating leguminous crops, particularly red clover, contribute to reduced fertilizer application in pastures, as atmospheric nitrogen fixed by clover can be utilized by grasses, exemplifying niche complementarity between these two species. In other words, the positive effect of mixtures lies in the mutually stimulating impact on nitrogen uptake by grass and legume components, along with increased efficiency in transforming assimilated nitrogen into biomass [48, 111]. The contribution of biological nitrogen fixation to enhancing agricultural efficiency, according to FAO data, approximately doubles the yield of mineral nitrogen fertilizers [11].

Red clover, thanks to biological nitrogen fixation, significantly enhances the productivity of grasslands where it is included as a component of grass mixtures [92]. Specifically, according to Irish researchers W. Burchill et al. [21], white clover can fix 80 kg/ha of nitrogen without the application of nitrogen fertilizers, but this fixation decreases when nitrogen fertilizer is used-under N_{280} application, the amount of biologically fixed nitrogen was 47 kg/ha. The same researchers demonstrate that the introduction of N86 and N_{140} does not significantly affect the amount of fixed nitrogen, which accounted for 64 and 66 kg/ha, respectively.

Research conducted by Kurhak & Gorkusha [65] has shown that incorporating leguminous crops into grass mixtures replaced the need for applying 102 to 200 kg/ha of nitrogen to grasslands. Davydiuk et al. [24] indicate that legume components in late-maturing grass mixtures replaced 98–110 kg/ha of mineral nitrogen in terms of dry matter accumulation and 140–159 kg/ha in terms of digestible protein output. According to the Institute of Animal Feeding (Ukraine) [71], legume-grass mixtures containing two legume components provided almost the same dry matter yield as grass mixtures without legumes but with an input of 180 kg/ha of nitrogen. Even on saline meadows, the inclusion of leguminous grasses in grasslands allows compensation for 60–160 kg/ha of mineral fertilizer nitrogen [70].

According to some researchers [105, 121], the yield of legume-grass mixtures without nitrogen fertilizer application is equivalent to the yield of grasses to which 112–250 kg/ha of effective nitrogen substance is applied. Other researchers assert that N-fixing leguminous grasses exhibit high root phosphatase activity, especially under low phosphorus availability in soils [84].

According to various scientists [5, 56, 72, 94] depending on soil-climatic conditions, red clover can accumulate from 45 to 340 kg/ha of nitrogen annually.

Moreover, the multifunctional nature of red clover as a melliferous plant, providing essential pollen for bees during a substantial period, highlights its ecological value beyond its agricultural benefits [23, 81].

Ensuring food security

Forages

The primary purpose of red clover is its utilization as animal feed. This perennial leguminous crop plays a crucial role in sustainable livestock development [106]. It is fed to ruminants [90], pigs [34], sheep [83], poultry [53, 102], and even fish [116] in various forms, such as hay, silage, haylage, chopped green material, and pellets [44, 51, 54, 55, 80, 82]. Compared to annual cereal and seed crops, perennial pastures have significantly lower nutrient losses and reduced pesticide requirements, while also contributing to soil carbon accumulation. Studies on feed bio-rectification [62] have produced protein of comparable quality to soybean meal, suitable as a poultry and swine substitute, without compromising animal productivity.

Grazing dairy cattle on clover pastures enriches milk with polyunsaturated fatty acids, especially α-linolenic acid, while starch-rich concentrates alter biohydrogenation pathways [35]. Balanced feeding systems enhance animal health, productivity, and their ability to meet growing global food demand. This has positive implications for the global economy and the livelihoods of those dependent on livestock.

In Europe, the utilization of grasslands by farmers has evolved significantly in recent decades to address ecological concerns. The production of grass feeds is closely linked to preserving ecosystem biodiversity [52], and the inclusion of leguminous grasses in pasture communities is a vital foundation for sustainable and competitive ruminant production, emphasizing their growing role in livestock development [99].

Red Clover's Yield and Nutritional Value

Red clover's yield can reach 9–15 t ha-1 of dry matter [1], and it accumulates sufficient nitrogen in the soil to support subsequent crops in the rotation with reduced nitrogen fertilizer rates of 10–12 % without compromising economic efficiency [12]. Red clover stands provide a yield of 12–13 t ha-1 of feed units and 1.7 t ha-1 of digestible protein, with up to 142 g of digestible protein per kg of feed unit [103, 124]. Lagush's research [66] revealed that, under an organo-mineral fertilization system, red clover could yield 10.5 t ha-1 of dry matter, containing 0.93 feed units, 105.6 g of digestible protein, and 12.4 MJ of metabolizable energy per kg of dry matter.

Livestock farms, especially those focused on dairy production, are increasingly interested in perennial legume-grass pastures due to their high productivity and protein concentration. The incorporation of such mixtures is limited by the low persistence of many legume varieties. Red clover *(Trifolium pratense)* and alfalfa *(Medicago sativa)* are among the most common legume species used in ruminant feeding systems. Feeds from red clover have lower crude protein concentrations than most alfalfa varieties but exhibit higher organic matter digestibility [2]. Notably, high protein concentrations in legume feeds that are easily degradable can lead to bloat issues [45].

Research by N. J. Hoekstra et al. [91] demonstrates that the resilience and nutritional value of different red clover varieties correlate closely with morphological characteristics, particularly stem length, suggesting stem length could be crucial in plant selection.

Research conducted in the state of Minnesota (USA) [122] has established that leguminous crops, including red clover studied in the research, have the potential to provide high-quality forage. However, the digestible energy of all leguminous crops in monoculture or mixture exceeds even the energy requirements of adult horses for maintenance (2.29 Mcal/kg). Horses preferred red and white clover over alfalfa, and this preference positively correlated with the content of crude protein and nutritional energy for the horses.

Important avenues of utilizing red clover include silage production [27, 100], its use in mixtures with other leguminous crops, particularly with white clover, red fescue, and sainfoin [45], and its application as a cover crop [87]. Indeed, intercropping leguminous crops can enhance feed productivity at the system level and help preserve biodiversity while concurrently reducing nitrogen losses through leaching and gas emissions.

The use of mixed grass-legume pastures, both for grazing livestock and for silage production, is widely practised in European agricultural systems with moderate climates, especially in the United Kingdom, where 70 % of grasslands on dairy farms contain red clover [25].

Usage of Red Clover in Various Sectors

Forage production from red clover could provide an alternative source of high-quality dietary protein. Research aiming to enhance protein solubility explored strategies to inhibit oxidative-reductive processes in red clover and white clover plants [57]. Using sulfites was found to slow the activity of polyphenol oxidase and peroxidase enzymes while enhancing the solubility of Rubisco enzyme forms. Increased sulfite content correlated with increased protein digestibility in red clover, suggesting sulfite inhibits oxidative enzymes and enhances red clover protein quality.

Food Industry and Pharmaceuticals

Red clover, a perennial forage crop rich in isoflavones (such as formononetin and biochanin A), can serve as a source of bioactive compounds with potential health benefits for both humans and animals [110], including the treatment of neurodegenerative disorders [7].

Red clover could become a new source of high-quality protein for producing dietary protein with enhanced solubility. Research investigating the mitigation of oxidative-reductive processes in red clover plants suggests that sulfite treatment could improve protein solubility and quality [57].

Incorporating red clover into feeding systems and exploring its potential in various sectors highlights its importance in addressing food security and promoting sustainable agriculture practices.

Growing for energy purposes

The increasing global population poses significant challenges not only for sustainable food and feed production but also for fuel, while simultaneously reducing anthropogenic impact on climate, the environment, and biodiversity [4]. This has led to a growing replacement of fossil fuels with renewable energy sources, such as bioenergy. The global production of bioenergy is projected to continue growing, with estimates from the International Energy Agency suggesting that biofuels could account for up to 27 % of the world's transport fuel by 2050 [73]. However, this transition must occur in a manner that integrates bioenergy production into land use for food and feed production. Furthermore, bioenergy production should significantly reduce greenhouse gas emissions, with the European Union's Renewable Energy Directive aiming for at least a 60 % reduction in GHG emissions compared to fossil fuels [28]. Therefore, urgent development of low-carbon emission and high-production biomass crops is essential to address these challenges.

Researchers have shown increased interest in using perennial grasses, including red clover, for bioenergy purposes [20, 40, 95]. Utilizing grass biomass as an energy source, particularly when there's excess for animal feeding, serves as an alternative use for this agricultural crop [120]. Numerous studies have investigated the bioenergy potential of both permanent and temporary grasslands [17, 33, 74, 114, 36]. Biomass regularly harvested and used as an energy source without reducing green cover in the region is considered a renewable resource and ecologically neutral, as it has a net-zero carbon emission balance.

The most common method for using grassland biomass for bioenergy production in Europe involves converting harvested biomass into methane through anaerobic [15]. Management practices like fertilizer application usually increase biomass production on managed pastures and subsequently enhance methane output [75]. According to W.-F. Cong et al. [46], fertilizer application increased methane output and net energy balance by 10 %, reaching 82–110 GJ ha-1 per year. These researchers also noted that, among all the tasks associated with grassland biofuel production, the most energy was expended in biogas production processes (average of 58 %), followed by biogas purification and compression (26 %), and crop cultivation and harvesting (13 %).

Danish researchers [16] investigated the biogas potential of residual fractions of four organically grown crops, including red clover, after protein extraction. According to these findings, 65 % of the methane potential from fresh red clover can be extracted through co-fermentation of residual fractions with green biogas after protein extraction. In subsequent studies [26, 85], the same researchers outlined a concept for a green biorefinery that divides green mass from red clover into various fractions produced during processing: protein concentrate, press cake, and "brown juice". The mixture of press cake and brown juice remains rich in nutrients and maintains an appropriate C:N ratio, making it suitable for co-fermentation within the biogas process as part of the biorefinery concept; thus, it's independent of other co-substrates.

Conclusions

The study underscores the intricate interplay between agriculture, population growth, sustainability, and climate change. The integration of innovative practices, such as cultivating red clover and perennial grasses, stands as a strategy to navigate the challenges posed by population growth, climate change, and the global demand for food security. By fostering greater productivity, resilience, and environmental consciousness, these practices present a potential path forward for a more sustainable agricultural future.

The use of red clover as a cover crop contributes to increasing agricultural land productivity and, consequently, enhancing the economic efficiency of agricultural production. By replacing organic fertilizers with animals, it reduces greenhouse gas emissions. As a forage crop, red clover enhances the efficiency of the livestock sector and ensures food security. Utilizing red clover for bioenergy purposes contributes to environmental conservation.

The importance of all the aforementioned outcomes of red clover cultivation is difficult to overestimate and cannot be compared, as they are all highly significant and ultimately contribute to improving soil fertility and mitigating the negative impact of climate change.

For future research, it could be promising to compare the effectiveness of cultivating red clover for cover cropping, fodder, and energy purposes. Given the versatility of the crop, studying combined methods of its utilization is worthwhile, as this will lead to new insights and align with the green course of the European Union's Common Agricultural Policy.

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