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Evaluation of the botanical composition of kikuyu and fescue grasslands associated with white clover during two seasons in the high valleys of Mexico

Zamora-Juárez, Yessica G.[1]; **Plata-Reyes, Dalia A.**[1*]; **Marín-Santana, María N.**[1]; **López-González, F.**[1]; **Hernández-Mendo, O.**[2]; **Martínez-García, Carlos G.**[1]; **Arriaga-Jordán Carlos, M.**[1]

[1] Universidad Autónoma del Estado de México. Instituto de Ciencias Agropecuarias y Rurales Campus El Cerrillo, El Cerrillo Piedras Blancas, Toluca, Estado de México, México. C. P. 50090. Tels.: 722 296 5552/722 180 6124/722 180 6136 ext. 6500.

[2] Colegio de Postgraduados Campus Montecillo. Programa de Ganadería, Texcoco, México. C. P. 56264. Tel. 55 5804 5900.

* Correspondence: dplatar144@alumno.uaemex.mx

ABSTRACT

Objective. To evaluate the botanical composition of grasslands of kikuyu (*Cenchrus clandestinus*) compared to tall fescue (*Lolium arundinaceum* cv. Cajun II), each one in association with white clover (*Trifolium repens* cv. Ladino), in two independent experiments conducted during two seasons, autumn 2018 and winter 2019.

Methodology. Two independent experiments under small-scale milk production system (SMPS) were established in the municipality of Aculco, State of Mexico, during autumn 2018 and winter 2019. The botanical composition of grasslands under intensive continuous grazing by breeding cows was evaluated. One grassland planted with tall fescue cv. Cajun II and the other invaded by kikuyu; each grassland was associated with white clover cv. Ladino. The botanical composition of both experiments was analyzed using a complete randomized experimental design.

Results. The kikuyu grassland recorded significant differences ($p<0.05$) with a higher proportion of forage during the winter 2018. Whereas the tall fescue cv. Cajun II grassland recorded a proportion of forage ($p<0.05$) higher than its proportion of dead tissue during autumn 2019.

Study Implications: The study of the botanical composition of mixed grasslands destined for livestock grazing allows to identify, propose and define strategies for forage production facing agroclimatic and management conditions in order to generate a better and higher forage yield.

Conclusions: The proportion of kikuyu was higher than that of tall fescue cv. Cajun II during the two seasons and years evaluated. This highlights the adaptability of kikuyu grass under agroecological conditions such as the absence of rains and high temperatures, coupled with the high stocking densities of the milk production systems in the study region.

Keywords: *Cenchrus clandestinus*, winter, *Lolium arundinaceum*, autumn, *Trifolium repens*.

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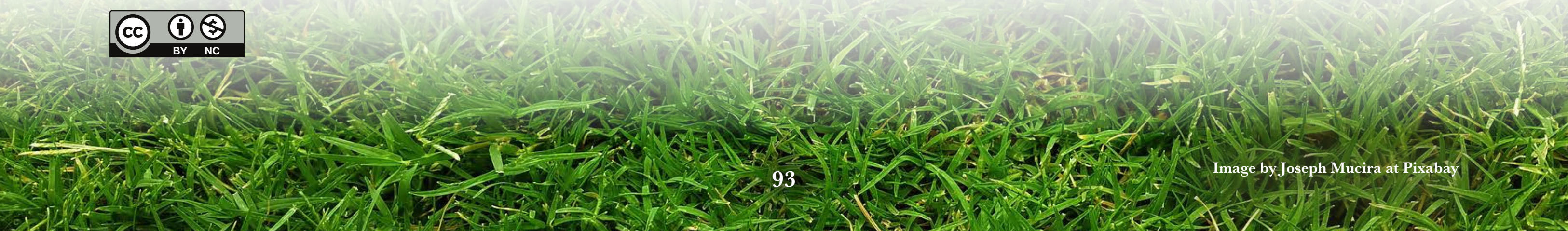
INTRODUCTION

Small-scale milk production systems (SMPS) are characterized by employing family labor; allocating an area of 6.25 ha (hectares) of land for farming activities; and generating income using regional forage resources. This latter is an aspect that has a favorable impact by increasing economic sustainability (Próspero-Bernal *et al.*, 2017).



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SMPSs have a potential for development towards sustainable models based on the improvement of their production processes through the management of their feeding strategies. Such as the implementation of grazing, intensive and continuously, in grasslands which are the basis of livestock feeding (Fadul-Pacheco *et al.*, 2013; Camacho-Vera *et al.*, 2017; Plata-Reyes *et al.*, 2018).

In the State of Mexico during 2019-2020, the reported area destined for agriculture and pastures was 933 586 ha of which 17 210 ha are mostly represented by areas that changed from agricultural crops to grasslands (SIAP, 2021) and represent an important forage source destined for feeding dairy cattle. The botanical composition is highly variable and susceptible to management and agroclimatic effects, especially in the oldest established grasslands (Sowers *et al.*, 2019).

Therefore, several authors agree on the need to evaluate the different species and varieties of grasses and legumes, in order to obtain knowledge that allows to better face the challenges of seasonal livestock production. Through the inclusion of grasslands characterized by their yield for grazing, adaptation attributes, availability, quality and resistance to different agroecological and management conditions under SMPS (Arango-Gaviria *et al.*, 2017; Marín-Santana *et al.*, 2020; Muciño-Álvarez *et al.*, 2021; Plata-Reyes *et al.*, 2021).

Due to its great adaptation capacity and wide geographical distribution, the kikuyu (*Cenchrus clandestinus* Hochst. ex Chiov Morrone formerly *Pennisetum clandestinum*) has developed in humid subtropical zones and environments different from its natural habitat (Arango-Gaviria *et al.*, 2017), since being a C4 species, it has a greater resistance to high temperatures and to the reduction or absence of rainfall. Its optimum growth temperature ranges between 18-30 °C and it is susceptible to long frost periods. Due to its aggressive growth behavior even in unfavorable agroecological conditions, it shows survival potential; thus, it has been considered a non desirable species (Fraser *et al.*, 2017; Benvenutti *et al.*, 2020).

Whereas the tall fescue (*Lolium arundinaceum* [Schreb] Darbysh formerly *Festuca arundinacea*) cv. Cajun II is a cold-season perennial species recognized for its growth in autumn under favorable temperature and humidity conditions, remaining green for winter grazing after frosts, but its vegetative regrowth decreases during the summer due to stress caused by high temperatures and absence of rainfall (Lee *et al.*, 2013).

The objective of this study was to evaluate the botanical composition of one kikuyu grassland (*Cenchrus clandestinus*) compared to another of tall fescue (*Lolium arundinaceum* cv. Cajun II), each one associated with white clover (*Trifolium repens* cv. Ladino), in two independent experiments carried out during two seasons, autumn 2018 and winter 2019.

MATERIALS AND METHODS

The experiments were conducted in production units (UP) of two collaborating farmers (who jointly manage their UP as one); they follow the rural participatory research guidelines for livestock technology development (Conroy, 2005). Those UP where the experiment was implemented are in Aculco de Espinoza (20° 00' a 20° 17' N, and 99° 40' a 100° 00' W) State of Mexico. Aculco has an area of 453.3 km^2, an altitude of 2440 m and a temperate sub-humid climate; average temperature of 13.2 °C, average annual rainfall of 700 mm, presence of frost from October to February, a rainy season from May to October and a dry season from November to April (Celis-Álvarez *et al*., 2017).

Experimental specifications

In the field, under the specifications of rural participatory research, two experiments were established independently for 42 d, divided into three experimental periods 14 d each. As it is shown in Figure 1. Experiment 1 (Experiment 1): conducted from September 1 to October 12, 2018. Experiment 2 (Experiment 2): conducted from February 15 to March 29, 2019.

Grasslands

Two grasslands of 1.0 ha each were evaluated, delimited with electric fence, both were grazed by four cows for 14 d (experimental period).

Experiment 1. Kikuyu grass (*Cenchrus clandestinus*), naturally established since 2013 by invasion on a plot of untilled land; average grassland height during the experiment, 5.33 cm±0.47.

Experiment 2. Tall fescue (*Lolium arundinaceum* cv. Cajun II) free of endophytes, that was seeded on February 8, 2018; average grassland height during the experiment, 1.87 cm±0.00.

During the time of the experiment, the tall fescue cv. Cajun II grassland was notably invaded by kikuyu. Also, in both experiments each grassland was associated with white clover (*Trifolium repens* cv. Ladino). The seed dose per hectare of tall fescue cv. Cajun II was 22 kg and 3 kg of white clover seed cv. Ladino. It was fertilized at the time of planting with 60 kg of N, 80 kg of P and 60 kg of K per hectare.

The grasslands of the two experiments were fertilized with 50 kg of N approximately every 28±7 d and with 80 kg of P and 60 kg of K per hectare every six months. The results of production, forage chemical composition and animal production were reported in previous studies by Marín-Santana *et al*. (2020) and Plata-Reyes *et al*. (2021).

Treatments

The treatments evaluated were:

EXP1=*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (KY+TB)
EXP2=*Lolium arundinaceum* cv. Cajun II+*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (CJ+KY+TB)

Period 1	**Period 2**	**Period 3**
September 01 to September 14	September 15 to September 28	September 29 to October 12

Experiment 1 (Fall 2018)

Period 1	**Period 2**	**Period 3**
February 15 to March 01	March 02 to May 16	March 17 to March 29

Experiment 2 (Winter 2019)

Figure 1. Distribution of the experimental periods.

Evapotranspiration

The estimation of potential evapotranspiration is important as an indicator in water use with agricultural, ecological, and other activities that require planning. To calculate the monthly potential evapotranspiration, the model described by Segura-Castruita and Ortiz-Solorio (2017) was used from maximum-minimum temperatures and altitude recorded for both experiments.

Botanical composition of grasslands

It was determined, from five subsamples per grassland and experimental period of 50 g each, cut randomly with scissors at ground level delimiting the cutting area with a metal quadrant of 0.5×0.5 m, the cuts were made during three experimental periods with a frequency of 14 d. Subsequently, the samples were separated manually in the laboratory, depending on the species present, grasses (Kikuyu or tall fescue cv. Cajun II), white clover, living, dead and other components (wild plants); samples were dried in a forced-air stove at 55 °C up to constant weight for 48 h and results were expressed as g per 100 g DM (Plata-Reyes *et al.*, 2021).

Statistical analysis

The experimental design was of repeated measures in time according to the following model of analysis of variance (Gutiérrez and Manrique, 1996, Kaps and Lamberson, 2004):

$$Y_{ijk} = \mu + \tau_i + \delta_{ij} + \rho_k + (\tau\rho)_{ik} + \varepsilon_{ijk}$$

where: Y_{ijk}=response variable; μ=overall mean; τ_i=treatment effect; δ_{ij}=error associated with experimental units; ρ_k=period effect; $(\tau\rho)_{ik}$=interaction between period and treatment; and ε_{ijk}=errors associated with periods.

The analysis of variance and Tukey test were performed on all variables (all were normally distributed) to identify significant differences ($p<0.05$) between the type of prairie, time of year and period. The results were analyzed with Minitab statistical software (v.14, Minitab Inc., State College, PA, USA).

RESULTS AND DISCUSSION

Figure 2 shows the temperature, minimum, average and maximum recorded during experiment 1. A lower temperature is observed compared to the winter 2019 (Figure 3).

Table 1 presents the climatic conditions during the experiments. Rainfall was only recorded at the beginning of the first period of experiment 1 with 10.7 mm (Figure 2). Whereas the estimated potential evapotranspiration was lower in the third period of autumn 2018 (Figure 4), and higher in the second period of winter 2019 (Figure 5). This was the result of precipitation disruption, limited irrigation management, and high stocking density on grazing (Plata-Reyes *et al.*, 2021). In addition to decreasing annual rainfall in the

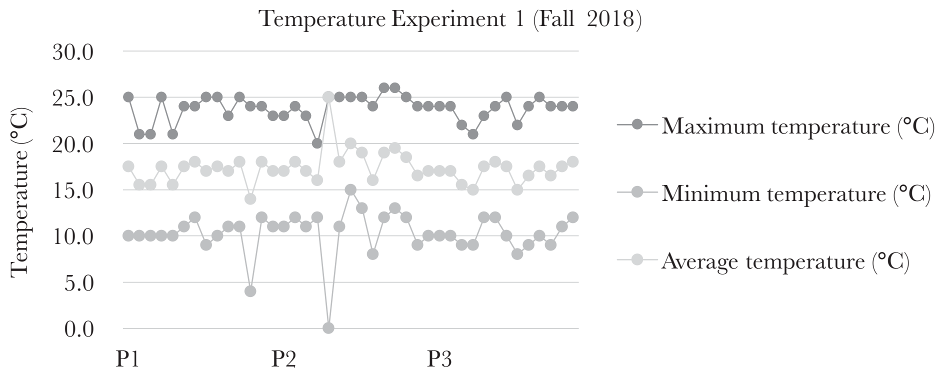

Figure 2. Maximum, average, and minimum temperatures recorded during experiment 1.

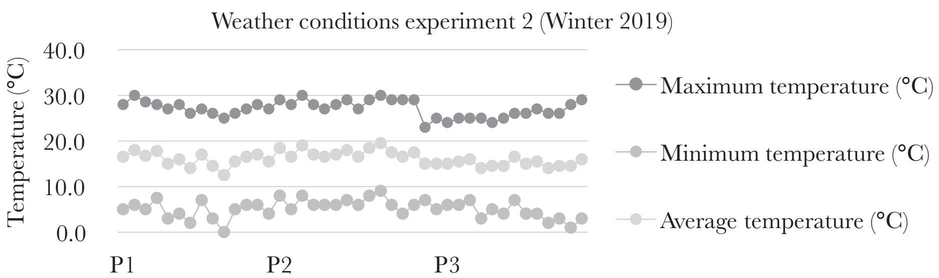

Figure 3. Maximum, average, and minimum temperatures recorded during experiment 2.

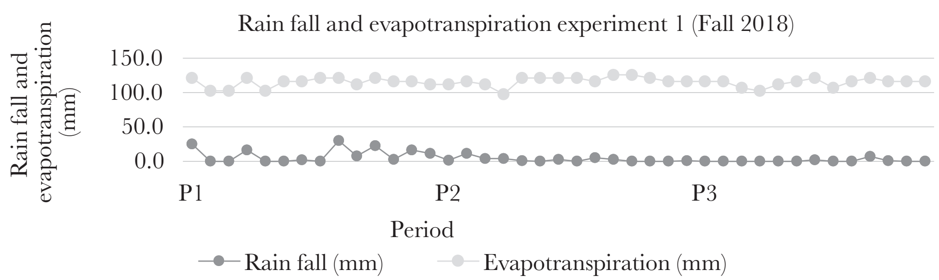

Figure 4. Evapotranspiration and rainfall recorded during experiment 1.

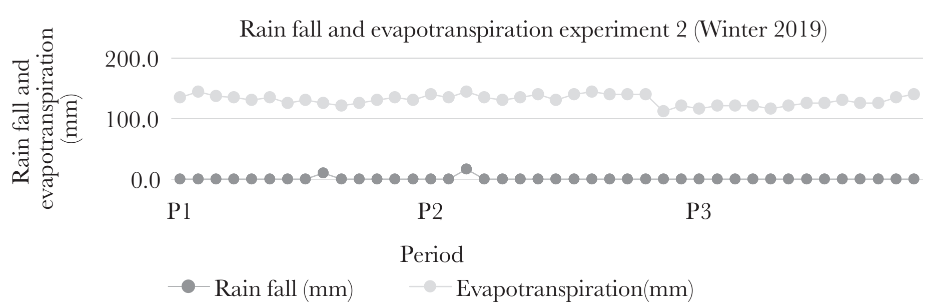

Figure 5. Evapotranspiration and rainfall recorded during experiment 2.

area, and the water available to producers for irrigation, derived from a water catchment-dam. Such water is distributed locally during the dry season but is limited (López-González *et al.*, 2020).

Botanical composition between grasslands

Significant differences ($p<0.05$) were recorded between grasslands for the botanical composition of kikuyu, tall fescue, white clover, and the proportion of dead tissue, as shown in Table 2. Both treatments presented a similar production of living tissue depending on their contribution to botanical composition without significant differences ($p>0.05$). Significant differences ($p<0.05$) were recorded in terms of the proportion of grass in each of the grasslands. Kikuyu performed best in both grasslands, because it is a very competitive species with easy growth. Despite the high stocking density and limited irrigation which are characteristics of SMPS in the study area.

On the other hand, white clover also recorded significant differences ($p<0.05$) and a greater presence in the KY+TB grassland, which could have influenced a lower proportion of dead tissue, probably due to the complementarity between species because of the grass-legume combination (Muciño-Álvarez *et al.*, 2021; Plata-Reyes *et al.*, 2021). In contrast with the values in the CJ+KY+TB grassland, with a higher mean ($p<0.05$) statistically significant, as a function of the proportion of dead tissue. Finally, wild plants neither showed significant differences ($p>0.05$) in terms of their contribution on each grassland.

Botanical composition by season

By season, all components showed significant differences ($p<0.05$), except white clover ($p>0.05$), as shown in Table 3. It is observed that kikuyu grass had a significant greater contribution ($p<0.05$) during the winter compared to tall fescue. Regardless thermal tolerance ranges between 7-38 °C (Fraser *et al.*, 2017), within the 42 d of duration of experiment 2, 33 d were measured with temperatures below 7 °C (Figure 2). Greater resilience and resilience were observed in kikuyu grass that contributed directly to adaptability through its latency capacity (Marais, 2001; Bell *et al.*, 2013; García *et al.*, 2014).

Table 1. Weather conditions recorded during experiments.

Experiment	Period	Precipitation (mm)	Maximum temperature (°C)	Temperature Minimun (°C)	Temperature Average (°C)	Evapotranspiration (mm)
Exp1	P1	10.7	23.7	10.0	16.9	115.1
	P2	3.1	24.1	11.6	17.9	117.1
	P3	0.8	23.6	10.1	16.8	114.4
Exp2	P1	0.7	27.1	4.6	15.9	131.0
	P2	1.1	28.1	6.4	17.3	135.8
	P3	0.0	25.8	4.3	15.0	124.8

Exp1=*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (KY+TB)
Exp2=*Lolium arundinaceum* cv. Cajun II+*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (CJ+KY+TB)

Table 2. Botanical composition results by grassland (g per 100g DM).

Component	Experiment	Praire	Mean	SEM	Value of P
Kikuyo	Exp1	KY+TB	43.9[b]	8.3***	0.000
	Exp2	CJ+KY+TB	22.4[a]		
Tall Fescue cv. Cajun II	Exp1	KY+TB	0.0[a]	17.0***	0.000
	Exp2	CJ+KY+TB	44.1[b]		
White clover cv. Ladino	Exp1	KY+TB	22.8[b]	6.3***	0.000
	Exp2	CJ+KY+TB	6.4[a]		
Dead	Exp1	KY+TB	25.5[a]	4.8**	0.002
	Exp2	CJ+KY+TB	37.7[b]		
Live	Exp1	KY+TB	63.4[a]	1.6[NS]	0.208
	Exp2	CJ+KY+TB	59.1[a]		
Other	Exp1	KY+TB	7.9[a]	0.8[NS]	0.592
	Exp2	CJ+KY+TB	9.0[a]		

Exp1=*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (KY+TB); Exp2=*Lolium arundinaceum* cv. Cajun II+*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (CJ+KY+TB); SEM=standard error of the mean; NS=p>0.05; *=p<0.05; **=p<0.01; ***=p<0.001; [abc]=different letters indicate significant differences between grasslands.

This capacity of kikuyu grass shown during this study is worth noticing; despite it is a tropical species, its growth does not stop in conditions of low temperatures. Meanwhile, tall fescue also registered a significantly higher proportion ($p<0.05$) during the autumn of 2018, where an average 8.21 mm rainfall was reported, compared to the 2.91 mm average rainfall in the winter 2019 with average temperatures between 16.8-17.9 °C. This agrees

Table 3. Botanical composition (g per 100g DM) by season (2018-2019).

Component	Season of the year	Experiment	Mean	SEM	Value of P
Kikuyo	Autumn 2018	Exp1	23.2[a]	7.7***	0.000
	Winter 2019	Exp2	43.1[b]		
Tall Fescue cv. Cajun II	Autumn 2018	Exp1	28.1[b]	4.7*	0.023
	Winter 2019	Exp2	15.9[a]		
White clover cv. Ladino	Autumn 2018	Exp1	13.9[a]	0.5[NS]	0.585
	Winter 2019	Exp2	15.3[a]		
Dead	Autumn 2018	Exp1	53.8[b]	12.2***	0.000
	Winter 2019	Exp2	9.4[a]		
Live	Autumn 2018	Exp1	31.8[a]	22.7***	0.000
	Winter 2019	Exp2	90.7[b]		
Other	Autumn 2018	Exp1	0.6[a]	6.1***	0.000
	Winter 2019	Exp2	16.3[b]		

Exp1 (Otoño 2018)=*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (KY+TB); Exp2 (Invierno 2019)=*Lolium arundinaceum* cv. Cajun II+*Cenchrus clandestinus*+*Trifolium repens* cv. Ladino (CJ+KY+TB); SEM=standard error of the mean; NS=p>0.05; *=p<0.05; **=p<0.01; ***=p<0.001; [abc]=different letters indicate significant differences between grasslands.

with Muciño-Álvarez *et al.* (2021), who mentioned that tall fescue improved its growth during the fall. Regardless, the contribution of tall fescue was like that of kikuyu during the autumn probably because the tropical grass tolerates better the water deficit and adverse conditions of the study region (Figure 2).

Botanical composition by experimental period

Table 4 presents the botanical composition recorded during the experiments. Kikuyu grass was the component with the highest presence (33%) in each of the experimental periods, tall fescue accounted for 22% and white clover 15%, reflecting the higher proportion of grasses and legumes over wild plants and dead tissue. These are favorable aspects that have an impact on the nutritional quality of forage in grasslands under continuous grazing (Edwards *et al.*, 2011; Claffey *et al.*, 2020).

The proportion recorded of living tissue showed significant differences ($p<0.05$); it can be observed how it decreased during the experimental periods, the proportion of dead tissue per period was significant ($p<0.05$) (Table 4). According to what was reported by Muciño-Álvarez *et al.* (2021), the combination of grasses and legumes allows complementarity between species, resulting in constant forage production. In this regard, Lowe *et al.* (2009) mentioned that the number of species established in a mixed grassland allows each of the species to be properly established.

Table 4. Botanical composition (g 100 g^{-1} DM) of both grasslands by experimental period.

Variable	Period	Mean	SEM	Value of P
Kikuyo	P1	28.9[a]	2.15[NS]	0.476
	P2	34.6[a]		
	P3	35.9[a]		
Tall Fescue cv. Cajun II	P1	23.3[a]	3.16[NS]	0.085
	P2	14.2[a]		
	P3	28.6[a]		
White clover cv. Ladino	P1	15.8[a]	1.34[NS]	0.752
	P2	13.5[a]		
	P3	14.5[a]		
Dead	P1	29.4[a]	6.03**	0.003
	P2	24.8[a]		
	P3	40.6[b]		
Live	P1	61.9[a]	2.29**	0.003
	P2	68.6[a]		
	P3	53.2[b]		
Other	P1	7.7[a]	2.97[NS]	0.174
	P2	11.3[a]		
	P3	6.3[a]		

SEM=standard error of the mean; NS=$p>0.05$; *=$p<0.05$; **=$p<0.01$; ***=$p<0.001$; [abc]=different letters indicate significant differences between grasslands.

CONCLUSIONS

The performance of kikuyu grass in the experiments during the two seasons of the year is highlighted, which is according to its reported agronomic characteristics. Thus, it has a better survival potential compared to the agronomic management carried out by the producers of the area. This management is characterized by a high stocking density and little availability of irrigation water. The proportion of white clover may have influenced forage growth in the kikuyu grassland due to the complementarity between species, which is a result of the combination between grasses and legumes.

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