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# Effects of integrated grassland renewal strategies on annual and perennial weeds in the sowing year and subsequent production years



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#### ABSTRACT

Appropriate weed control measures during the renewal phase of temporary grasslands are critical to ensure high yields during the whole grassland lifecycle. The aim of this study was to determine which integrated grassland renewal strategy can most effectively control annual weeds in the sowing year and delay perennial weed reestablishment. Four split-plot trials were established at three sites dominated by Rumex spp. along a south-north gradient in Norway. The annual and perennial weed abundance was recorded during the sowing year and two or three production years. Main plots tested seven renewal strategies: 1. Spring plowing, 2. Spring plowing+companion crop (CC), 3. Summer cut+plowing, 4. Summer glyphosate+plowing, 5. Summer glyphosate+harrowing, 6. Late spring glyphosate+plowing, 7. Fall glyphosate+spring plowing+CC. Strategies 1-4 were tested in all four trials, strategy 5 in three trials, strategy 6 in two trials and strategy 7 in one trial. Plowing was performed at 20-25 cm depth, rotary harrowing at 15 cm depth, and glyphosate was applied at 2160 g a.i. ha<sup>-1</sup>. CC was spring barley (Hordeum vulgare). Subplots tested selective herbicide spraying (yes/no) in the sowing year. Results showed that effects of renewal strategies were often site-specific and differed between the sowing year and production years. Spring renewal resulted in higher perennial weed abundance than summer renewal in two out of four trials (by 3 and 12 percentage points, over all production years), and glyphosate followed by harrowing drastically increased Rumex spp. in one out of three trials (by 18 percentage points over all production years). CCs only significantly reduced perennial weed abundance in one trial (by 8 percentage points over all production years). In comparison, the selective herbicides had a strong effect on annual and perennial weeds in the sowing year in all trials. Selective herbicides reduced the weed cover from 32% to 7% cover, and averaged over the production years and sites, the perennial weed biomass fraction was 6 percentage points lower where herbicides had been applied. We conclude that while the tested renewal strategies provided variable and site-specific perennial weed control, selective herbicides were effective at controlling Rumex spp. and other perennial dicot weeds in the first two production years.

## 1. Introduction

Grasslands provide a large proportion of the world's livestock feed (O'Mara, 2012). In the temperate zone, including Norway, much of the grassland area consists of temporary grasslands that are either rotated with annual crops or renewed into a new grassland crop at different intervals (Peeters et al., 2014). Typically, when temporary grasslands are renewed this is due to declining yields and/or altered botanical composition, such as when the sown grassland species (usually perennial grasses, legumes or a mixture of both) are replaced by secondary species, i.e., weeds (e.g., *Rumex longifolius* DC., *Rumex obtusifolius* L, *Ranunculus repens* L., *Taraxacum* spp.). However, renewal is challenging and does not always reduce the proportion of weeds in the sward. In the sowing year, flushes of annual weeds are common (Döring et al., 2017), which can reduce the establishment success of the newly sown species. Inadequate tillage can also lead to proliferation rather than control of perennial weeds present in the sward (Ringselle et al., 2019). As the temporary grassland ages, the proportion of perennial weeds tends to increase due to winter damage and/or non-optimal conditions for the sown species, or weed competition (Håkansson, 2003; Lien et al., 2003; Lunnan et al., 2018). Pesticides are not commonly used in grasslands. For example, in Norway in 2017, pesticides were used on approximately

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6% of the grassland area compared to 90% in other crop areas (Aarstad and Bjørlo, 2019). To prolong temporary grassland longevity, renewal strategies are needed that can minimize the risks of annual weeds impairing the establishment of sown species in the sowing year, and that will delay the buildup of perennial weeds in the subsequent production years.

Weed management in grasslands is complicated by the fact that tillage can only be used during the renewal phase. Moreover, very intensive tillage during the renewal phase may be undesirable as it delays sowing, is time-consuming and resource demanding. Extended periods of bare soil, which are common when multiple tillage operations are performed to control perennial weeds, increase the risk of soil erosion and nutrient leaching (Skøien et al., 2012; Aronsson et al., 2015). The main alternative to tillage is herbicides. However, farmers in the European Union and Norway are obligated to use integrated weed management (IPM) techniques. One of the principles of IPM is that preventive measures should be prioritized over direct control methods, and non-chemical methods over pesticides (Riemens et al., 2022). Consequently, there is a need for efficient IPM strategies for managing weeds during grassland renewal that are not too reliant on either herbicides or tillage, and that are profitable for farmers.

Both plowing and broad-spectrum herbicides (e.g., glyphosate) are effective control methods for perennial weed species, with each affecting the weed flora in different ways (Koning et al., 2019). Combined use of glyphosate and plowing prior to sowing should provide the strongest and most consistent control efficacy as it would target a broad spectrum of weed species and plowing could compensate for a failed or insufficient glyphosate treatment and vice versa. However, using both plowing and glyphosate would also require more time, energy and/or leave the soil bare for longer than only using plowing, and even more so compared to using glyphosate combined with either reduced till (e.g., harrowing without plowing) or no-till. Glyphosate is currently under reevaluation in the European Union. If glyphosate is restricted or phased out in Europe, this will make weed control and grassland renewal more challenging (Silvia et al., 2020).

To control weeds that germinate after sowing, selective herbicides can be used, which have no or minor effect on the sown species (primarily grasses and clovers), but can kill off many dicot weed species such as R. obtusifolius (Donovan et al., 2022), which is a common perennial grassland weed with a global distribution; or its relative R. longifolius, which is wide-spread in the Nordic countries (Ringselle et al., 2019). While established perennial weeds are less vulnerable than newly germinated plants to such selective herbicides (Donovan et al., 2022), the application of selective herbicides may also give a competitive advantage to the sown species over perennial dicot weeds. Intercropping has many positive effects on cropping systems (Tilman, 2020). An annual companion crop (CC) is often sown together with the grassland species to increase the yield in the sowing year (Skjelvag, 1970). In addition to increasing yield, a competitive CC can reduce the weed pressure in the sowing year (Ringselle et al., 2019). Ringselle et al. (2019) showed that the effect of plowing, CC and false seedbeds on Rumex spp. can vary greatly whether the sward is renewed in spring or in summer. Renewing in spring provides more time for the sown species to establish before winter and may allow for 1-2 cuts in areas with a long growing season, while renewing in summer allows for a final cut of the old grassland.

Most studies on weed control in grasslands focus on the short-term effect of a single or a few tools/methods, but IPM strategies necessitate the study of multiple factors over a longer period. In this study, we have investigated how different combinations of renewal time (spring or summer), renewal strategy (plowing, reduced tillage and/or glyphosate use), selective herbicides (yes/no) and use of CCs affected the abundance of annual and perennial weeds in both the sowing year and the following production years. The expectation was that the perennial weed populations would be reduced more and be suppressed for longer when: a) selective herbicides are used in the sowing year, b) the grassland is sown in spring rather than in summer, c) a CC is sown together with the grassland crop, d) glyphosate is applied before plowing, and e) plowing, rather than harrowing, follows the glyphosate application.

## 2. Materials and methods

# 2.1. Site description

Four trials were established at three locations in Norway, at or close to three experimental stations that belonged to the Norwegian Institute of Bioeconomy Research (NIBIO): Holt, Særheim and Kvithamar. Holt is located in the northern part of Norway, with a subarctic climate and a short growing season (Table S1). Temporary grasslands in the region are generally cut 1–2 times per year and renewed every 6–10 years. Særheim Research Station is located in the southwestern part of Norway, which has a maritime climate with relatively mild winters, high precipitation and a relatively long growing season. Temporary grasslands in the region are generally cut 3–4 times per year and renewed every 2–5 years. Kvithamar is located in the middle part of Norway, with a similar climate to Særheim but with fewer growing days. Temporary grasslands in this region are generally cut 2–3 times per year and renewed every 3–6 years.

At each site, trials were established in spring 2014 (H14, S14 and K14) and at Kvithamar an additional trial was established in the fall of 2014 and sown in 2015 (K15). Each site had established grasslands with large perennial weed populations (Table 1). Each trial included one sowing year and three production years, except K15, which was terminated after two production years. Trial H14 was located on a site with permanent grassland (> 10 years old) close to Holt Research Station (69°42′ N, 18°53′ E, 7 m above sea level (a.s.l.)). Trial S14 was located on Særheim Research Station (58°46′ N, 5°39′ E, 90 m a.s.l.). Trial K14 and K15 were located at Kvithamar Research Station, sub location Værnes (63°27′ N, 10°57′ E, 16 m a.s.l.). Trial H14 was sown with a grass species mixture and the trials S14, K14 and K15 with grass-clover mixture and all trials were fertilized in spring and during summer with mineral fertilizer according to local practice. Details on crop species, fertilization and soil characteristics are given in Table S2.

## 2.2. Treatments and experimental design

The trials were established according to a split-plot design with three replicates. Main plots were  $6 \times 8$  m and the subplots  $3 \times 8$  m at Holt and Særheim (an example of a replicate block given in supplementary material, Fig. S1). At Kvithamar the main plots were  $5.5 \times 7$  m and the subplots  $2.75 \times 7$  m. Between each replicate there was a 10 m bufferzone to allow proper management by the tractor-driven equipment. The buffer zones between the replicates and between adjacent main plots and subplots were mowed or harvested as the surrounding field.

The main plots compared seven different renewal strategies: 1. Spring plowing (Spring plow), 2. Spring plowing and companion crop (Spring plow+CC), 3. Summer plowing 8–17 days after cutting the grassland (Summer cut+plow), 4. Summer plowing 8–17 days after applying glyphosate (Summer glyphosate+plow), 5. Summer harrowing 8–17 days after applying glyphosate (Summer glyphosate+harrow), 6. Late spring plowing 10–26 days after applying glyphosate (Late spring glyphosate+plow), 7. Fall glyphosate application, spring plowing and companion crop (Fall glyphosate+spring plow+CC) (Table 2). H14 and S14 tested strategies 1–6, K14 tested strategies 1–5 and K15 tested strategies 1–4 and 7.

The machines used for management and treatments are given in the supplementary material (Table S3). Glyphosate was applied at 2160 g a. i. ha<sup>-1</sup> (Glyfonova Pluss, 360 g a.i. liter<sup>-1</sup> Cheminova Agro A/S), when perennial dicot weeds had large rosettes/beginning of stem elongation (stem 10–20 cm high), BBCH 31–32 (Hack et al., 1992). Mowing was conducted to 5–8 cm stubble height. Moldboard plowing was conducted to 20–25 cm depth and rotary harrowing to 15 cm depth. The CC was spring barley (*Hordeum vulgare* L.) (Table S2). Seedbed preparation

## Table 1

Weed species present at the experimental sites Holt 2014 (H14), Særheim 2014 (S14), Kvithamar 2014 (K14) and Kvithamar 2015 (K15) in the sowing year pretreatment. The first two sample times were prior to grassland renewal, either by spring plowing or glyphosate, while the third was before selective herbicides were applied. As glyphosate was applied later than spring plowing it meant that more species had time to emerge (indicated by +). The unit displayed is number of weeds per m<sup>2</sup>. Perennial weed species are in bold.

	H14	$\#/m^2$	S14	$\#/m^2$	K14	$\#/m^2$	K15	$\#/m^{2}$
Before spring plowing	Ranunculus repens	38.6	Rumex spp.	0.7	Rumex longifolius	4.2	Rumex longifolius	2.4
	Taraxacum spp.	9.3	Taraxacum spp.	0.1	Taraxacum spp.	1.5		
	Alchemilla spp.	6.5	Symphytum officinale	0.1				
	Rumex spp.	2.7						
	Descampsia cespitosa	1.4						
	Anthriscus sylvestris	0.2						
Before summer glyphosate			+Stellaria media		+Ranunculus repens		+Taraxacum spp.	
							+Ranunculus repens	
Before selective herbicides	Ranunculus repens	77.3	Capsella bursa-pastoris	292.7	Capsella bursa-pastoris	18.5	Gnaphalium uliginosum	64.9
	Stellaria media	11.8	Spergula arvensis	65.9	Taraxacum spp.	12.3	Matricaria matricarioides	25.9
	Poa trivialis	3.2	Stellaria media	64.6	Spergula arvensis	11.9	Capsella bursa-pastoris	19
	Rumex spp.	3	Matricaria matricarioides	13	Rumex longifolius	8.6	Spergula arvensis	13.2
	Alchemilla spp.	0.2	Polygonum aviculare	5.2	Stellaria media	6.3	Stellaria media	12
	Taraxacum spp.	0.3	Persicaria spp.	4.9	Tripleurospermum inodorum	4.9	Lamium purpureum	9.6
	Silene dioica	0.3	Elymus repens	2.2	Gnaphalium uliginosum	4.7	Rumex longifolius	9.5
	Rumex acetoca	0.2	Fumaria officinale	2.1	Chenopodium album	2.7	Elymus repens	4.7
	Descampsia cespitosa	0.1	Rumex obtusifolius	0.2	Viola arvensis	2.3	Ranunculus repens	1.7
							Galeopsis spp.	1.4
							Viola arvensis	1.1
							Taraxacum spp.	0.8

# Table 2

Important dates for field operations in the sowing year. '-' means that the operation was not performed. Please note that at Kvithamar 2015 one of the operations for the Fall gly+spring plow+CC treatment was conducted in the fall prior to the sowing year. Mow=mowing, Gly=glyphosate, Plow=moldboard plowing, RotHar=rotary harrowing.

Field	Treatment	Treatment before sowing		Sowing time		Herbicide application	Harvests		
	no.	Before soil tillage	Soil tillage	Cereal cover crop	Forage mixture	(subplots)	Before sowing	After sowing	
Holt 2014	1	-	Plow 27/5	-	2/6	1/7	-	11/9	
	2	-	Plow 27/5	2/6	2/6	1/7	-	6/8 <sup>a</sup>	
	3	Mow 18/7	Plow 4/8	-	5/8	22/8	1/7	-	
	4	Gly 18/7	Plow 4/8	-	5/8	22/8	1/7	-	
	5	Gly 18/7	RotHar 4/8	-	5/8	22/8	1/7	-	
	6	Gly 6/6	Plow 16/6	-	16/6	10/7	-	11/9	
Særheim 2014	1	-	Plow 28/4	-	14/5	17/6	-	19/7, 14/8Mow, 29/9	
	2	-	Plow 28/4	14/5	14/5	17/6		19/7, 14/8Mow, 29/9 <sup>a</sup>	
	3	Mow 8/7	Plow 18/7	-	23/7	27/8	13/6	14/8Mow, 29/ 9Mow	
	4	Gly 8/7	Plow 18/7	-	23/7	27/8	13/6	14/8Mow, 29/ 9Mow	
	5	Gly 8/7	RotHar 18/ 7	-	23/7	27/8	13/6	14/8Mow, 29/ 9Mow	
	6	Gly 9/5	Plow 4/6	-	6/6	8/7	-	29/9Mow	
Kvithamar 2014	1	-	Plow 22/4	-	6/5	6/6	-	5/8, 19/9	
	2	-	Plow 22/4	6/5	6/5	6/6	-	7/8 <sup>b</sup>	
	3	Mow 2/7	Plow 10/7	-	11/7	7/8	10/6	19/9Mow	
	4	Gly 2/7	Plow 10/7	-	11/7	7/8	10/6	19/9Mow	
	5	Gly 2/7	RotHar 10/ 7	-	11/7	7/8	10/6	19/9Mow	
Kvithamar 2015	1	-	Plow 4/5	-	5/5	2/6	-	10/8, 12/9	
(+2014)	2	-	Plow 4/5	4/5	5/5	2/6	-	3/9 <sup>b</sup>	
	3	Mow 6/7	Plow 14/7	-	15/7	11/8	14/6	12/9	
	4	Gly 6/7	Plow 14/7	-	15/7	11/8	14/6	12/9	
	7	Gly 15/9–14	Plow 4/5	4/5	5/5	2/6	-	3/9 <sup>b</sup>	

<sup>a</sup> Barley harvested as green forage.

<sup>b</sup> Barley harvested as grain yield

followed normal practice for the area with levelling and tine harrowing in plowed plots. Sowing was done with a combi machine (cereals), grass seed sower or experimental plot seeder. Fertilization was done with a combi machine together with sowing cereals, with a plot fertilizer or manually. After sowing the fields were rolled. The spring strategies (1, 2 and 7) were conducted as soon as possible in spring when soil had dried after winter/snow had melted and the soil was not too dry or too wet for tillage. The summer strategies (3-5) were conducted on perennial weed regrowth after the first cut (mowing or glyphosate at least 2–3 weeks after first cut, soil tillage (plowing or rotary harrowing) at least 8–10 days after mowing or glyphosate), while the late spring strategy (6) started when perennial dicot weeds had reached the recommended growth stage for glyphosate application (large rosettes/beginning of stem elongation, stem 10–20 cm high) and plowing 10 d or more after glyphosate application. The actual dates of the various operations are given in Table 2.

The subplots tested selective herbicides (sprayed or not sprayed), applied when grass seedlings had 2–3 leaves (BBCH 12–13) and red clover seedlings had one trifoliate leaf (the second leaf developed, BBCH 12) (Table 2). The herbicide mixture tribenuron-methyl+MCPA (3.75 +375 g a.i. ha<sup>-1</sup>, as Express, 500 g a.i. kg<sup>-1</sup>, Du Pont de Nemours + MCPA 750 Flytende, 750 g a.i. litre<sup>-1</sup>, Nufarm Deutschland Gmbh) was used in fields seeded with a grass+red clover mixture (S14, K14, K15), while the herbicide mixture fluroxypyr+clopyralid+MCPA (80 + 40 + 400 g a.i. ha<sup>-1</sup>, as Ariane S, 40 + 20 + 200 g a.i. litre<sup>-1</sup>, Dow AgroSciences), was used in the field at H14, which was seeded with a grass-only mixture. The herbicides were applied with a backpack sprayer driven by compressed air (NOR sprayer (modified Oxford sprayer), produced by Olav T. Langmyr, Kristiansand, Norway), 110° flat fan nozzles (XR TeeJet 11002), a nozzle pressure of 1.5–2 bar and water volume of 250 L ha<sup>-1</sup> (see weather conditions in the sowing year in supplementary Fig. S1).

In the production years, forage yield was harvested three times per year at Særheim and Kvithamar and two times at Holt. The fields were fertilized in spring and after each cut (except after the last cut), with NPK mineral fertilizer 18-3-15 (but NPK 22-3-10 at K14 in spring and after 1st cut in 1st production year 2015) corresponding to 240 kg N ha<sup>-1</sup> at Særheim (distributed 45:30:25 in spring, after 1st and 2nd cut, respectively), 230 kg N ha<sup>-1</sup> at Kvithamar (distributed 47:27:27 in spring, after 1st and 2nd cut, respectively) and 174 kg N ha<sup>-1</sup> at Holt (distributed 60:40 in spring and after 1st cut, respectively). At Holt, an additional 16.5 kg P and 31.5 kg K per ha were applied to replicate 1, since soil samples collected in the first production year (2015) showed a lack of these nutrients in replicate 1. In the production years, spring fertilizations were conducted in late April at Særheim (21-24/4) and Kvithamar (28-29/4). The three cuts were conducted at these sites in mid-June (S14: 1-15/6, K14/K15: 14-23/6), July/August (S14: 26/7-11/8, K14/K15: 20/7-1/8) and September/October (S14: 23/9-2/10, K14+K15: 14-15/9). At Holt, where the growing season is shorter, spring fertilization was done between 18/5-12/6, depending on start of spring. The 1st cut was made between 12 and 13/7, and the 2nd cut between 1 and 19/9.

# 2.3. Crop and weed assessments

Four assessments techniques were used: 1. Visual estimates of the ground cover (weeds + crop + soil = 100%), 2. visual estimates of the botanical composition (biomass of weeds + crop = 100%), and 3. weed counts. Visual estimates were conducted over the whole plots. Weed counts were either conducted per plot or per m<sup>2</sup> (quadrants of 0.25 m x 0.25 m put randomly 4 times in each plot). Weed cover, botanical composition and weed numbers were divided into weed species. Included in the crop were both sown and unsown species of grass (timothy (*Phleum pratense* L), meadow fescue (*Festuca pratensis* L.), perennial ryegrass (*Lolium perenne* L.), smooth meadow-grass (*Poa pratensis* L.), common bent (*Agrostis capillaris* L.)) and clover (red clover (*Trifolium pratense* L), white clover (*Trifolium repens* L.)).

The ground cover was assessed at: a) Spring during the sowing year (Y0) (i.e., pre-treatment except treatment Fall glyphosate+Spring plow+CC), b) 3–4 weeks after spraying selective herbicides Y0, c) Fall Y0, and d) at spring fertilization in the production years (Y1-Y3). The botanical composition was assessed prior to all cuts in Y1-Y3. The weed density was assessed for all weeds in spring Y0, and prior to and at 3–4 weeks after selective herbicide spraying. In addition, the density of *Rumex* spp. plants was also determined during Y1-Y3 in spring and at all cuts.

# 2.4. Statistical analyses

The results were analyzed in RStudio 2021.09.0 build 351 (RStudio, PBC) using R 4.1.1 (R Foundation) with the tidyverse, emmeans, car,

nlme and readxl packages. Each trial was analyzed separately as each site had a different weed flora and not all treatments were included at all sites. The response variables, crop and weed cover, perennial weed biomass fraction (perennial weed biomass/total grassland biomass) and weed density, were analyzed in mixed linear models using the lme function (Tables S4-S6). Prior to analyzes, weed numbers were converted to density per m<sup>2</sup> and square root transformed, and perennial weed biomass fractions were log+1 transformed, both to achieve approximate homoscedasticity. Except for the selective herbicide effect 3-4 weeks after spraying, analyzes were conducted as repeated measures using the corCAR1 correlation structure. Main factors (Renewal strategy, Herbicides, Time) and their interactions were analyzed as fixed, and Replicate, Main plots, and Subplots as random. The weed cover prior to grassland renewal was used as a covariate for the perennial weed biomass fraction and the Rumex spp. density prior to grassland renewal as the covariate for the Rumex spp. density analyzes. No covariate was used for the crop/weed cover analyzes since the pretreatment measurement was included in the analysis. At K15 no covariate was used since there was no pre-treatment measurement for the Fall glyphosate+spring plow+CC treatment. Tukey's HSD tests at  $\alpha = 0.05$ were used for post-hoc analysis.

# 3. Results

# 3.1. Weed flora in the sowing year (Y0)

There were one to four perennial weed species at K15 (*R. longifolius*), K14, (*R. longifolius* and *Taraxacum spp.*) and S14 (*R. longifolius, R. obtusifolius, Taraxcum* spp. and *Symphytum officinale* L.) in the grass sward in spring Y0 before renewal strategy treatments started, while H14 had six prevalent perennial weed species (Table 1). Before the glyphosate treatments, *Rumex* spp. and *Taraxacum* spp. were present in all trials and *Ranunculus repens* in all trials except S14. After grassland swards were sown, annual weeds (particularly *Capsella bursa-pastoris* (L.) Medik.) dominated the weed flora (57–84% of the total weed cover at three-four weeks after spraying in the plots without selective herbicide spraying; Table 3) until fall in all trials except at H14, where perennial weeds dominated both during the sowing and production years.

# 3.2. Weed flora during the production years (Y1-3)

During the production years, 82% of the weed biomass consisted of R. longifolius at K14 and K15. The remaining 18% of the weed biomass consisted of Taraxacum spp., R. repens, other perennial dicots, and at K14, Elymus repens (L.) Gould appeared in the last production year (Fig. 1). At H14, most of the weed biomass consisted of Rumex spp. (40%) and R. repens (33%). H14 was also the trial that had the most monocotyledonous weeds, with the weed biomass consisting of 11% E. repens and 9% other grass species (e.g., Fig. 2). At S14, most of the weed biomass consisted of Rumex spp. (35%), Taraxacum spp. (13%) and other perennial dicot weeds (17%). In the last production year at S14, R. repens and E. repens appeared (Figs. 1 and 2). S14 was the only trial with S. officinale (13%; but only in spring-plowed plots) and the only trial with a significant amount of annual weeds during the production years; the annual weeds consisted primarily of Poa annua L., which occurred in all production years, with annual peaks in spring (data not shown).

## 3.3. Effect of selective herbicides

Overall, selective herbicides provided effective control of many, but not all (e.g., not *S. officiale* or *Gnaphalium uliginosum* L.), annual and perennial dicot weeds in both the sowing and production years. Threefour weeks after spraying, selective herbicides had a strong negative effect on the weed cover (reducing it from 32.2% to 6.5% cover, on

#### Table 3

Means of weed and crop cover (%) or weed number per  $m^2$  (#) three-four weeks after spraying selective herbicides at the experimental sites Særheim 2014 (S14), Holt 2014 (H14), Kvithamar 2014 (K14) and Kvithamar 2015 (K15) in the sowing year. Not all treatments were sampled at all sites for all weeds and in some cases differed between weed cover and weed density. Data on different renewal strategies are not show. Symbols show significance levels (no symbol = p > 0.1, ' =  $p \le 0.1$ , \* =  $p \le 0.05$ , \*\* =  $p \le 0.01$ , \*\*\* =  $p \le 0.001$ ).

		No herbicides	Herbicides		No herbicides	Herbicides		
		%	%	Sign. level	#	#	Sign. level	
H14	Ranunculus repens	5	1	¢	59	43	د	
	Elymus repens	2	4					
	Rumex spp.	2	0	*	3	1	***	
	Stellaria media				12	1	*	
	Sum weeds	9	5					
	Grass crop	32	32					
S14	Capsella bursa-pastoris	46	2	***				
	Persicaria spp.	4	0	•				
	Stellaria media	3	0	***				
	Rumex spp.	1	1					
	Spergula arvensis	0	2					
	Polygonum aviculare	1	2					
	Sum weeds	64	10	***				
	Grass crop <sup>a</sup>	27	74	***				
K14	Capsella bursa-pastoris	18	2	***	19	1	***	
	Chenopodium album	9	3					
	Rumex longifolius	8	0	**	7	1	**	
	Spergula arvensis				18	3	***	
	Taraxacum spp.				4	1	***	
	Tripleurospermum inodorum				8	1	***	
	Lamium purpureum				2	1		
	Stellaria media				7	0	***	
	Sum weeds	35	9	***				
	Clover crop	5	4	*				
	Grass crop	32	43	**				
K15	Capsella bursa-pastoris	11	0	***	37	5	***	
	Rumex longifolius	8	0	**	5	3		
	Stellaria media	1	0		20	5	*	
	Gnaphalium uliginosum	0	1	ć	84	94		
	Ranunculus repens				2	1		
	Viola arvensis				1	3	*	
	Spergula arvensis				28	7	***	
	Lamium purpureum				8	4	4	
	Matricaria discoidea				34	7	•	
	Sum weeds	21	2	***				
	Clover crop	5	5					
	Grass crop	74	91	***				

<sup>a</sup> Cover of clover crop not assessed at this time at S14.

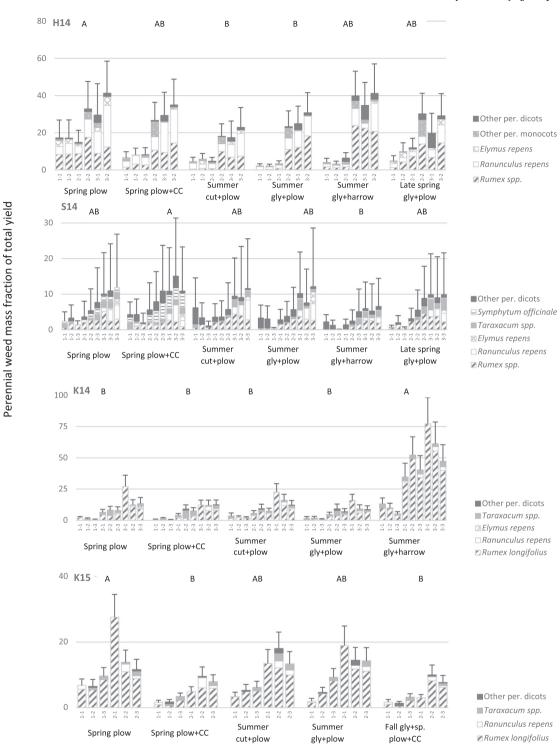
average) and weed density (on average by 75%, excluding R. repens, Viola arvensis Murray and G. uliginosum that were not effectively controlled) in all trials, except at H14, where most of the remaining weed cover (3.9% out of 4.8%) consisted of the grass weed E. repens (Table 3). The selective herbicides' effect on the weed cover remained in the fall of the sowing year (reduced from 19% to 6% cover, on average) and even into the last production year (reduced by 6 and 4 percentage points at K14 and K15, respectively, and tended to be lower at H14 (P = 0.1)) (Table 4). At S14, the weed cover was only significantly reduced by selective herbicides in the first production year (by 6 percentage points; Table 4). The selective herbicides showed a similar reduction in perennial weed biomass fraction (by 6 percentage points, on average over all sites) and Rumex spp. density (from, on average, 1.7 to 0.8 plants  $m^{-2}$ ) over the production years at all sites (Fig. 3; Table 3). There were significant interactions between selective herbicides and time at all sites except H14 (Table S5), which indicated that the perennial weed biomass was not only lower, but also increased slower over time in the herbicide treated plots (Fig. 2).

The selective herbicides had a strong immediate effect on the grass crop cover (increasing it by an average of 25 percentage points; CC and clover cover excluded) in all trials, except at H14 (Table 3). However, in the fall of the sowing year and in the production years, the crop cover was only higher in the herbicide treated plots at K14 (by 7.5 percentage points, on average, over the production years; Table 4).

# 3.4. Effect of renewal strategies

The interaction between renewal strategy and time was significant at all sites for crop and weed cover (Table S4), perennial weed biomass fraction (Table S5) and for *Rumex* spp. density (Table S6), with only a couple of exceptions where the main effects were significant but not the interaction (perennial weed biomass fraction at K14 and *Rumex* spp. density for S14). Additionally, at K14, there was a 3-way interaction for weed cover between renewal strategy, time and selective herbicides (Table S4).

Renewal strategy did affect weed and crop cover, but the effects were generally only significant in the sowing and/or first production year, except at K14 (Table 4). At K14, the Summer glyphosate+harrow treatment had a much higher weed cover (27% vs. 9% cover, average over all years) and lower crop cover (43% vs. 65% cover, averaged over all years) than the other treatments throughout the trial (Table 4). The 3way interaction for weed cover at K14 was because the difference between unsprayed and sprayed plots was only 3 percentage points in spring-renewed plots compared to 37 percentage points in summerrenewed plots. At H14, the Spring plow+CC treatment had the highest weed cover in the sowing year (primarily due to a 30% cover of *R. repens*), but in the first production year it had the highest crop cover (Table 4). At S14, the summer-renewed treatments had higher weed covers (20% vs. 11% cover) and lower crop covers (51% vs 64% cover) than the spring-renewed treatments, but only in the first production year (Table 4). At K15, the Fall glyphosate+spring plow+CC treatment had

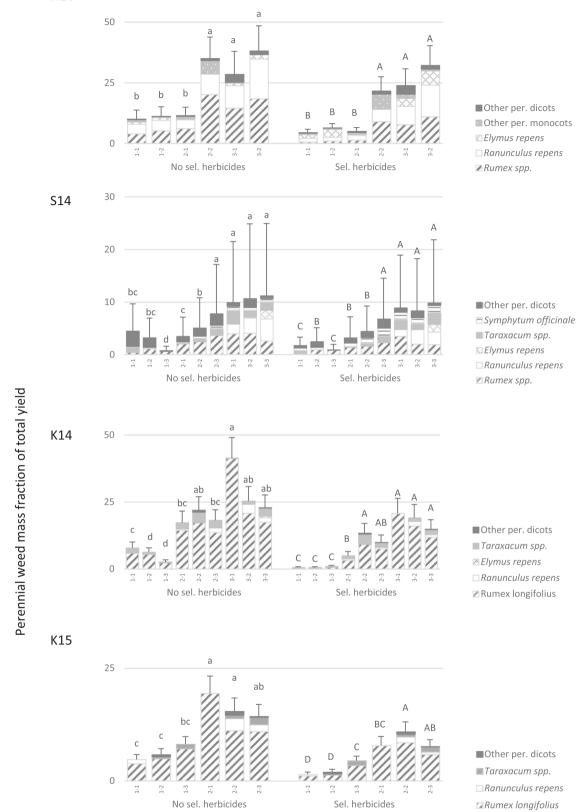


**Fig. 1.** Means of the perennial weed biomass fraction for the interaction between Time (cuts during the grassland years) and Renewal Strategy, at the four trials (H14 =Holt 2014, S14 =Særheim 2014, K14 = Kvithamar 2014, K15 =Kvithamar 2015). On the x-axis, the two values indicate the year and the cut (e.g., 1–1: year 1 and cut 1). Letters show the results of Tukey test between the main effects of Renewal Strategies, not the interaction between Renewal Strategies and Time. Error bars are standard errors back-transformed using the delta method. Please note the difference in y-axis maximum between the trials.

almost no spring weed cover in the sowing year (due to the fall application of glyphosate), and the Spring plow treatment had a lower crop cover in the first production year compared to the other treatments (68% vs. 83% cover).

Regardless of renewal strategy, the perennial weed biomass fraction generally started to increase in the second production year (Fig. 1). However, averaged over all production years, the highest proportion of perennial weeds were found in the Spring plow treatment at H14 and K15, Spring plow+CC at S14 and Summer glyphosate+harrow at K14 (Fig. 1). At S14 and K15, similar to K14, the interaction between renewal strategy and selective herbicides (Table S5) showed that the selective herbicides had a greater effect in the summer treatments than the spring treatments (Fig. 4).

The *Rumex* spp. density mostly followed the same pattern as the perennial weed biomass fraction, being highest in Spring plow treatment at H14 (though not significantly), Spring plow+CC at S14 and Summer



**Fig. 2.** Means of the perennial weed biomass fraction for the interaction between Time (cuts during the grassland years) and Selective Herbicides, at the four trials (H14 =Holt 2014, S14 =Særheim 2014, K14 = Kvithamar 2014, K15 =Kvithamar 2015). On the x-axis, the two values indicate the year and the cut (e.g., 1–1: year 1 and cut 1). Tukey test shows significant difference within herbicide treatments, using upper and lower case letters to indicate that Tukey test comparisons are only made within herbicide treatments. Error bars are standard errors back-transformed using the delta method. Please note the difference in y-axis maximum between the trials.

#### Table 4

Means of crop cover (C (%)) and weed cover (W (%)) at the experimental sites Særheim 2014 (S14), Holt 2014 (H14), Kvithamar 2014 (K14) and Kvithamar 2015 (K15) in the sowing year (Y0) and subsequent grassland production years (Y1-Y3). Bold indicate significant differences and the letters show where significant differences existed, within rows, between renewal strategies or herbicide treatments as a result of Tukey groupings at  $\alpha$ =0.05. Sp. = spring, Su. = Summer, gly = glyphosate, CC = companion crop, CI = 95% Confidence interval.

Site	Time	Cover	Sp.	Sp. plow+CC	Su.	Su. gly+plow	Su. gly+harrow	Late sp.	Fall	CI renewal	Herbi	cides	CI herbicides
			plow		cut+plow			gly+plow	gly+sp. plow+CC	strategies	No	Yes	
H14	Y0 Spring	C (%)	62	77	60	67	70	70		$\pm 23$	68	67	$\pm 17$
H14	Y0 Fall	C (%)	38a	39a	40a	39a	18b	38a		$\pm 23$	35	35	$\pm 17$
H14	Y1 Spring	C (%)	67a	82a	40b	40Ь	21b	76a		$\pm 23$	52	56	$\pm 17$
H14	Y2 Spring	C (%)	89	92	95	94	93	90		$\pm 23$	91	93	$\pm 17$
H14	Y3 Spring	C (%)	47	55	52	46	36	40		$\pm 23$	45	47	$\pm 17$
H14	Y0 Spring	W (%)	38	23	40	33	30	30		$\pm 25$	32	33	$\pm 17$
H14	Y0 Fall	W (%)	3c	37a	18b	10bc	28ab	2c		$\pm 25$	24a	9b	$\pm 17$
H14	Y1 Spring	W (%)	23	8	8	4	22	8		$\pm 25$	15	9	$\pm 17$
H14	Y2 Spring	W (%)	11	9	5	6	7	9		$\pm 25$	9	6	$\pm 17$
H14	Y3 Spring	W (%)	38	25	20	26	45	40		$\pm 25$	36	29	$\pm 17$
S14	Y0 Spring	C (%)	50	32	50	51	51	44		$\pm 27$	46	47	$\pm 16$
S14	Y1 Spring	C (%)	69ab	58ab	45b	49b	58ab	83a		$\pm 27$	55	66	$\pm 16$
S14	Y2 Spring	C (%)	89	81	85	83	76	85		$\pm 27$	81	85	$\pm 16$
S14	Y3 Spring	C (%)	68	58	63	69	77	63		$\pm 27$	67	66	$\pm 16$
S14	Y0 Spring	W (%)	5b	22a	6b	5b	14ab	3b		$\pm 12$	10	9	±7
S14	Y1 Spring	W (%)	9bc	13abc	24a	20ab	15abc	3c		$\pm 12$	17a	11b	±7
S14	Y2 Spring	W (%)	5	10	6	6	7	4		$\pm 12$	7	5	±7
S14	Y3 Spring	W (%)	11	18	12	11	8	11		$\pm 12$	12	11	±7
K14	Y0 Spring	C (%)	40	41	38	39	36			$\pm 18$	39	39	$\pm 12$
K14	Y0 Fall	C (%)	96a	87a	61b	61b	42c			$\pm 18$	64b	74a	$\pm 12$
K14	Y1 Spring	C (%)	88a	88a	75ab	73ab	57b			$\pm 18$	72b	80a	$\pm 12$
K14	Y2 Spring	C (%)	80a	80a	81a	85a	56b			$\pm 18$	74	79	$\pm 12$
K14	Y3 Spring	C (%)	51a	48a	43a	43a	23b			$\pm 18$	40b	47a	$\pm 12$
K14	Y0 Spring	W (%)	12	10	12	12	13			$\pm 13$	11	12	±7
K14	Y0 Fall	W (%)	2c	4c	22b	22b	38a			$\pm 13$	29a	6b	±7
K14	Y1 Spring	W (%)	3b	2b	8b	7b	23a			$\pm 13$	12a	5b	±7
K14	Y2 Spring	W (%)	9b	7b	7b	6b	36a			$\pm 13$	16a	10b	±7
K14	Y3 Spring	W (%)	7b	9b	13b	7b	27a			$\pm 13$	15a	9b	±7
K15	Y0 Spring	C (%)	73a	73a	75a	73a			0Ь	$\pm 12$	58	59	$\pm 8$
K15	Y0 Fall	C (%)	95	97	95	93			99	$\pm 12$	94	97	$\pm 8$
K15	Y1 Spring	C (%)	68b	83a	83a	81a			87a	$\pm 12$	80	81	±8
K15	Y2 Spring	C (%)	55	56	56	53			54	$\pm 12$	53	57	$\pm 8$
K15	Y0 Spring	W (%)	13a	13a	12a	13a			0b	$\pm 6$	11	9	±4
K15	Y0 Fall	W (%)	4	3	4	6			2	$\pm 6$	5	3	±4
K15	Y1 Spring	W (%)	8	6	8	9			4	$\pm 6$	9a	5b	±4
K15	Y2 Spring	W (%)	10	5	7	8			5	$\pm 6$	9a	5b	±4

glyphosate+harrow at K14. However, at K15 the Summer cut+plow treatment had the highest *Rumex* spp. density, average over all years, due to a large increase in *Rumex* spp. plants in the first cut of the second production year (Fig. 3).

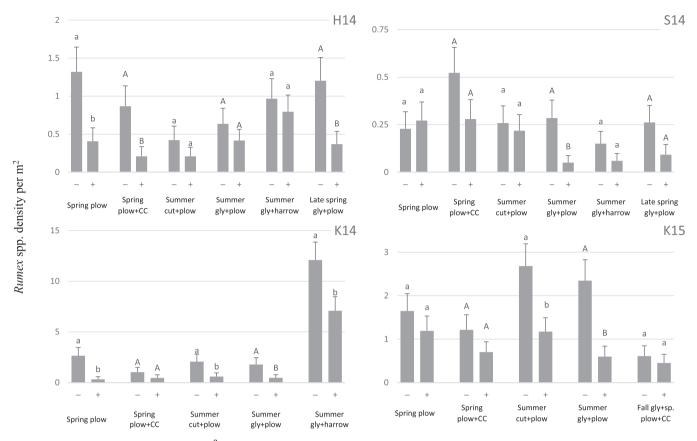
## 4. Discussion

Our main hypothesis was that the grassland renewal strategy and/or selective herbicide use would greatly affect the weed abundance in sowing year and the subsequent production years. The hypothesis was supported for selective herbicides, but not for renewal strategy. Selective herbicides reduced both the annual and perennial weed abundance in the sowing year and the effect on perennial weeds lasted well into the production years in all trials except at S14 where this was only the case in the summer treatments. This is in line with other studies such as Donovan et al. (2022) who found that selective herbicides could suppress *R. obtusifolius* populations until the fourth year after application, with *R. obtusifolius* seedlings being far more vulnerable to herbicides than older plants. As expected, the selective herbicides had little effect on *E. repens* and other perennial grass weeds, and the efficacy differed between dicot species (cf. labels of the herbicides).

We expected spring renewal to suppress perennial weeds more than summer renewal. However, at H14 and S14, spring renewal resulted in a higher perennial weed biomass fraction than summer renewal. At H14, there was a large proportion of *E. repens* in the Spring plow treatment, which could be explain by the fact that *E. repens* is a tillage-tolerant species that usually requires more intensive tillage than plowing alone to be controlled (Ringselle et al., 2016; Brandsæter et al., 2017). At S14, the early spring plowing seemed to have been ineffective, resulting in the survival of many *S. officinale* and *Rumex* spp. plants. In contrast to our results, Ringselle et al. (2019) found that spring renewal resulted in far less *Rumex* spp. than summer renewal, though the effects generally did not carry over from the sowing year. Harrington et al. (2013) found no difference in efficacy on *R. obtusifolius* or *R. repens* populations when grassland renewal time directly affect *Rumex* spp. abundance. However, renewal time is certainly important for establishing the most competitive crop and it determines which control measures can be used.

We expected the addition of a CC to suppress perennial weeds. However, sowing a CC together with the grassland species did not have a consistently strong effect on the perennial weed flora; significantly reducing it at K15, tending to reduce it at H14 and K14 and tending to increase it at S14. This is likely due to the particularly large starting populations of *Rumex* spp. and *S. officinale* in the CC plots, which had survived spring plowing in this trial. Ringselle et al. (2019) found that a CC reduced the *Rumex* spp. plants density, but the effect was much greater after summer renewal than during spring renewal.

We expected that using glyphosate prior to plowing would reduce the perennial weeds more than using cutting prior to plowing. However, in no trial did glyphosate before plowing in summer reduce the perennial weed abundance more than cutting the grassland prior to plowing. The relatively low efficacy of glyphosate could be caused by cold (S14,



**Fig. 3.** Means of the *Rumex* spp. density per  $m^2$  for the interaction between Renewal Strategy and Selective Herbicides, at the four trials (H14 =Holt 2014, S14 =Særheim 2014, K14 = Kvithamar 2014, K15 =Kvithamar 2015). + and – denote the use or no use of selective herbicides in the seeding year, respectively. Tukey test shows significant difference between herbicide use within renewal strategies, using alternating upper and lower case letters to indicate that Tukey test comparisons are only made between herbicide treatments, within renewal strategies. Error bars are standard errors that were back-transformed using the delta method. Please note the difference in y-axis maximum between the trials.

K14, K15) or dry conditions (H14) prior to spraying, which can result in slowly growing weed plants and thus less translocation of glyphosate to roots and other plant parts (Adkins et al., 1998). This can be contrasted with the high short-term efficacy of the Fall glyphosate+spring plow+CC at K15, though it was not higher than Spring plow+CC in the production years.

We expected that it would be less effective to follow the glyphosate treatment with harrowing instead of plowing. At S14 there was little difference in total perennial weed biomass fraction between harrowing or plowing after glyphosate application, but at H14 there was a larger amount of Rumex spp. after harrowing. More strikingly, at K14, harrowing after glyphosate resulted in a huge increase in *Rumex* spp., most likely due to dry conditions both at spraying and sowing with poor germination of the newly seeded crop giving a competitive advantage to the weeds (Supplemental information, Fig. S1). At S14, Summer glyphosate+harrow treatment was more successful probably due to more optimal soil moisture conditions after sowing. The plowing depth was larger at K14 than at H14 and S14 and larger plowing depth may give better effect on perennial weeds than more shallow depth (Brandsæter et al., 2011; Ringselle et al., 2019) and can thus explain larger difference between harrowed plots and plowed plots at K14 than at S14 and H14. In cereal cropping systems, it has been shown that harrowing (shallower than in the current study) can result in more perennial weeds than plowed plots even if glyphosate is used (Tørresen and Skuterud, 2002). Thus, harrowing can be a risky strategy against large populations of Rumex spp., since, unlike plowing, it cannot compensate if the glyphosate is not sufficient to control the Rumex spp. plants. In general, optimal conditions for crop germination are important for successful establishment of the new forage crop and can interact with the renewal strategies.

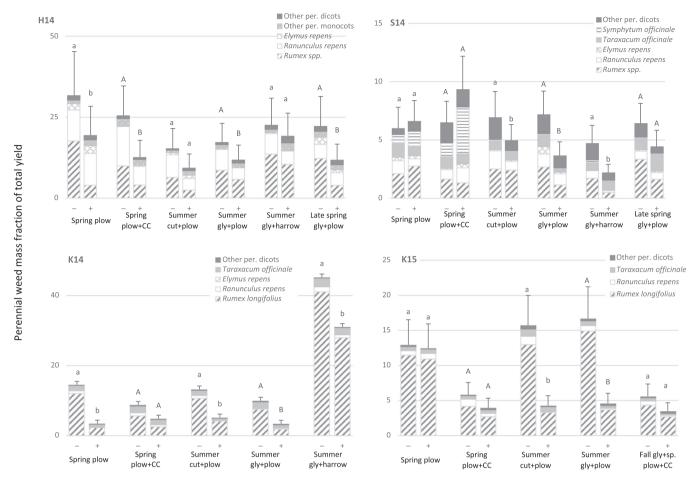
#### 4.1. Implications for management

Since *R. longifolius* and *R. obtusifolius* were the most dominant species in the trials, the conclusions drawn will be most relevant for these species. However, at H14 there was a similar effect on *R. repens* as on *Rumex* spp. At S14 *Taraxacum* spp. became more prominent in production year two and three. However, since *Taraxacum* spp. seeds can easily spread from neighboring areas by wind (Stewart-Wade et al., 2002) it is difficult to determine if the increase at S14 was due to failure to control existing *Taraxacum* spp. plants or the establishment of new seed plants.

The need for managing weeds is not equal for all production systems and varies depending on the weed flora. For example, Lewis and Hopkins (2000) have pointed out that loss of production due to weeds is likely to be of greater significance, and control measures more profitable, under intensive than extensive grassland management. Moreover, while a high proportion of species like Taraxacum spp. does not necessarily affect yield and yield quality negatively (Bakken et al., 2009), weeds are more detrimental for the yield if the temporary grasslands are infected by Rumex spp. (Zaller, 2004), Anthriscus sylvestris (L.) Hoffm. (Miller and D'Auria, 2011), or the low-yielding grasses P. annua and Alopecurus geniculatus L. Thus, in grasslands where Rumex spp. are a major problem, the present study offers strong support for the inclusion of selective herbicides as part of an integrated weed management strategy. However, selective herbicides are unlikely to be sufficient on their own as the perennial weed biomass was high in most trials after 2-3 years, though this depends on how established the weeds are when sprayed (Donovan et al., 2022).

Even though there were no large or consistent differences between the renewal strategies in the present study, one cannot conclude that renewal strategy does not matter regarding perennial weed control. The

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**Fig. 4.** Means of the perennial weed biomass fraction perennial weed biomass fraction for the interaction between Renewal Strategy and Selective Herbicides, at the four trials (H14 =Holt 2014, S14 =Særheim 2014, K14 = Kvithamar 2014, K15 =Kvithamar 2015). + and – denote the use or no use of selective herbicides in the seeding year, respectively. Tukey test shows significant difference between herbicide treatments on perennial weed biomass fraction of all perennial weed species combined, using alternating upper and lower case letters to indicate that Tukey test comparisons are only made between herbicide treatments, within renewal strategies. Error bars are standard errors that were back-transformed using the delta method. Please note the difference in y-axis maximum between the trials.

large variation between trials in both weed abundance and environmental conditions mean that there may be differences in treatment effects that were not captured. However, some conclusions can still be drawn.

Firstly, the drastic increase in perennial weed abundance after some treatments in some trials underscores the importance of using the weed control tools at optimal environmental conditions and at the optimal weed growth stage. This is exemplified by the difference of the Summer glyphosate+harrow treatments at S14 and K14. In general, correct adjustment, depth and use of tillage implements/plowing are important for the result. Deep plowing can for instance give better control of perennial weeds than shallow plowing (Brandsæter et al., 2011). If glyphosate or plowing fails, it increases the need for more frequent renewal/renovation or back-up plans.

Secondly, even though a CC did not consistently provide a significant reduction in weed abundance, the addition of a CC was generally not detrimental either. As a CC usually increases the overall harvestable yield in the sowing year (Skjelvag, 1970; Fitjar, 2020), a CC can therefore most likely be recommended as long as it can be harvested for grain or fodder, even if the effect on the perennial weed abundance is likely to be variable and unpredictable. However, the impact of CC on forage yield in the first production year need to be taken into consideration as in some areas (e.g., those with a short growing season) and years (especially if the CC is cut late) as the inclusion of a CC can also cause a yield reduction in the first production year (Fitjar, 2020).

Thirdly, the lack of clear preference for a specific renewal time as the best for controlling *Rumex* spp., means that other factors must decide. For example, the Late spring gly+plow treatment may only be recommended in exceptional cases as it delayed the growth of the grassland compared to Spring plow, and it did not provide the extra harvest of the old grassland crop as the summer treatments did.

Fourthly, the lack of clear connection between effects on perennial weeds in the sowing year and the effect in the production years, clearly show the weakness of studies that only look at effects in the sowing year.

# 4.2. Conclusions

The effect of renewal strategies was generally site-specific and often there was little correlation between the effects in the sowing year (when both annual and perennial weeds were present) and the effect in the production year (when primarily perennial weeds were present). The effect of grassland renewal timing (spring or summer) and the addition of a CC on perennial weed abundance was site-specific, and thus their use must be adapted to local conditions. When there was an effect, it was sometimes quite significant. Glyphosate combined with plowing did not reduce *Rumex* spp. abundance more than cutting followed by plowing, but glyphosate followed by harrowing drastically increased *Rumex* spp. in one out of three trials. Glyphosate in fall followed by plowing in spring and a companion crop (only tested at one site) gave a better overall efficacy than glyphosate followed by plowing in summer, but not better than Spring plow+CC. Compared to the renewal strategies, the selective herbicides had a more consistent and stronger reductive effect on the annual and perennial weed abundance (especially *Rumex* spp.) in the sowing year (75% on average excluding species that were unaffected) and the effect on the perennial weeds lasted for at least two production years (on average 6 percentage points). Perennial weed abundance generally increased drastically in the second production year regardless of site and treatment.

Future research should investigate the importance of glyphosate for grassland renewal. If we can manage without glyphosate this is of interest if glyphosate use is limited or banned in the future. Moreover, it is important to study the factors affecting glyphosate efficacy when renewing grasslands – especially if followed by rotary harrowing. Of special interest is to further investigate the effect on short- and long-term weed control on renewal in spring with plowing combined with a companion crop with or without glyphosate in the preceding fall. Further it is also of interest to investigate more cutting prior to plowing. Considering the site-specificity of the results of the different renewal strategies it would be a great value to further study how grassland renewal strategies should be adopted to local conditions.

# CRediT authorship contribution statement

**Björn Ringselle:** Formal analysis, Writing – original draft, Visualization Writing – review & editing. **Anne Kjersti Bakken:** Conceptualization, Methodology, Investigation, Resources, Writing – review & editing. **Mats Höglind:** Conceptualization, Methodology, Investigation, Resources, Writing – review & editing. **Marit Jørgensen:** Conceptualization, Methodology, Investigation, Resources, Writing – review & editing. **Kirsten S. Tørresen:** Funding acquisition, Resources, Writing – original draft, Visualization, Writing – review & editing.

## **Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

# Data Availability

Data will be made available on request.

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# Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.eja.2023.126799.

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