



Using Calcium Cyanamide for Control of *Senecio vulgaris* in Leafy Vegetables

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Abstract

Weed management is a critical component of agricultural production, presenting significant challenges to farmers. The efficacy of traditional herbicides has been compromised due to increasingly stringent regulatory measures and the emergence of herbicide-resistant weed populations, complicating weed control efforts in conventional farming systems. Calcium cyanamide, a nitrogen-based fertilizer, has been identified for its phytotoxic properties, which may be leveraged for weed suppression. The necessity for effective weed control is particularly pronounced in some minor crops, where even minimal infestations of harmful weed species can jeopardize the entire yield, e.g. pyrrolizidine alkaloid containing plants in herbs. To assess the viability of calcium cyanamide as a weed management tool in leafy vegetable cultivation, a series of three experimental trials were conducted in Germany. In the initial trial, twenty different weed species were tested for their sensitivity to calcium cyanamide. Notably, *Senecio vulgaris*, an important weed known for its production of pyrrolizidine alkaloids that are toxic to mammals, exhibited a marked sensitivity to the treatment. This finding was corroborated in the subsequent trials, which also investigated the sensitivity of three leafy vegetable crops to calcium cyanamide. Lamb's lettuce and arugula experienced yield reductions due to the application of calcium cyanamide, spinach demonstrated resilience and was unaffected by the treatment. As anticipated, the application of calcium cyanamide resulted in both nutritional and phytotoxic effects, which varied over time. The findings suggest that calcium cyanamide can serve as an effective component of integrated weed management strategies in leafy vegetable production, particularly for controlling *S. vulgaris*. However, it is imperative for growers to consider the degradation timeline of calcium cyanamide to mitigate the risk of phytotoxic damage to crops.

Keywords Weed management · Calcium cyanamide · Phytotoxicity · *Senecio vulgaris* · Pyrrolizidine alkaloids · Leafy vegetables

Introduction

To mitigate the adverse environmental impacts associated with contemporary agricultural practices, the European Union has established objectives aimed at reducing the reliance on synthetic chemical pesticides (European Commission 2019). Concurrently, the intensification of agricultural methods has led to the emergence of herbicide resistance in numerous weed species (Hulme 2023),

thereby diminishing the availability of effective herbicides. This overall reduction in herbicide options presents new challenges for weed management across various crops. In minor crops, where often only a limited number of herbicides is currently registered, a reassessment of traditional weed control strategies is warranted. Calcium cyanamide fertilizers serve a dual purpose by supplying nitrogen and lime to crops. Cyanamide, a natural compound for example found in vetch (*Vicia villosa* ROTH), has been documented to possess weed-suppressive properties (Kamo et al. 2003). However, the European Chemicals Agency (ECHA) has indicated that the application of calcium cyanamide may pose risks to soil organisms in treated fields and to adjacent surface water ecosystems. Specifically, cyanamide can be detrimental to macroorganisms in fertilized soils and, in cases of erosion, may also adversely affect aquatic organisms. Additionally, dicyandiamide, a byproduct formed

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during the soil reaction, can inhibit nitrification and impact soil microbial communities. The potential for cyanamide and dicyandiamide residues to contaminate groundwater has also been noted (European Chemicals Agency 2018). On the other hand, the degradation products of calcium cyanamide exhibit various phytosanitary effects. The application of this fertilizer influences soil microbial populations, initially resulting in a decline in germination rates, followed by a subsequent recovery. A study conducted by Liu et al. (2016) indicated that soil organisms returned to baseline levels approximately 14 days post-application of cyanamide. Calcium cyanamide functions as an herbicide through the generation of the phytotoxic compound cyanamide (Kurth 1960). Recent research has explored the utilization of plant extracts containing cyanamide as allelochemicals for weed control (Kamo et al. 2012; Soltys et al. 2012). Cyanamide undergoes conversion to urea and ammonia in the soil, a process that requires moist conditions and can be expedited by shallow incorporation into the soil matrix (Kurth 1960). When applying calcium cyanamide prior to sowing or planting, it is crucial to allow sufficient time for the fertilizer to convert to urea or ammonium to prevent crop damage from residual cyanamide. According to the application guidelines for the commonly used calcium cyanamide fertilizer Perlka® (Alzchem group 2024), a waiting period of 2 to 3 days per 100 kg of Perlka® per hectare is recommended. Consequently, following a standard application rate of 400 kg/ha, a waiting period of approximately 8 to 12 days is necessary before sowing the crop. Application to established crops is also feasible for certain species, with the timing of application being contingent upon the growth and developmental stages of the crops involved. For instance, Soltys et al. (2012) demonstrated that the phytotoxic effects of cyanamides can significantly reduce the germination and growth of seedlings in both weed and crop species. Numerous studies and recommendations have been published regarding the direct control of weeds using calcium cyanamide fertilizers (Mohr 1951; May and Harvey 1958; Bachthaler 1963), particularly in the latter half of the 20th century. Calcium cyanamide has been recognized for its significant role in managing key weed species in both horticultural and agricultural contexts. Notably, important composite weeds such as *Senecio vulgaris* L. (Orth 1962) and volunteer rice (Ohdaira 2021) have been identified as target organisms for this control method. Recent findings by Krupa and Witkowicz (2023) demonstrated that the application of calcium cyanamide fertilizer resulted in a nearly 39% reduction in the population of dicotyledonous weeds.

The present study aims to investigate the potential of calcium cyanamide in addressing specific weed challenges in specialty crops, particularly lettuce. A growing concern in recent years has been the contamination of harvested crops

by pyrrolizidine alkaloid-containing weeds (Federal Institute for Risk Assessment 2013). Among these, *S. vulgaris* is recognized as the most significant species (Nitzsche et al. 2018). Even a small number of *S. vulgaris* plants can lead to contamination levels that exceed permissible limits for pyrrolizidine alkaloids, rendering the affected crops unsuitable for human consumption or animal feed. Consequently, *S. vulgaris* is classified as a particularly problematic weed due to its high potential for spread in specialty crops, which demand stringent purity standards, especially in herbs and lettuce (Federal Institute for Risk Assessment 2007, 2013, 2016). To evaluate the efficacy of calcium cyanamide in controlling weeds, particularly *S. vulgaris*, a series of three trials was conducted. Given that the effects of cyanamide may also influence crop health, the tolerance of three types of leafy vegetables was assessed in conjunction with weed control efficacy. It was hypothesized that, when applied correctly, calcium cyanamide could serve as a safe and effective method for managing *S. vulgaris* populations in specialty crop production.

Material and Methods

Three subtrials were conducted at the Julius Kühn-Institute, Federal Research Centre for Cultivated Plants, located in Braunschweig, Germany (52.274563 N, 10.570034 E). In all three experimental approaches Perlka®—a calcium cyanamide fertilizer containing 19.8% nitrogen—was utilized. The first subtrial (7 cm × 7 cm pots) focused on screening potential target weed species for susceptibility to calcium cyanamide. The second subtrial involved a pot experiment (10 L containers) with three types of leafy vegetables in conjunction with *S. vulgaris*. The third subtrial assessed the interaction between *S. vulgaris* and two crops under field conditions (concrete plots measuring 1 m × 2 m). During the execution of all three subtrials the soil treated with calcium cyanamide was maintained in a consistently moist state, rendering the collection of precipitation data unnecessary. The soil in all three trials was steamed before starting and therefore contained no viable weed seeds. The weed seeds were incorporated into the soil to a depth of 0.5–1 cm. Perlka® was applied to the soil surface of the pots.

For subtrial 2 and 3 it is noteworthy that no comparative fertilization treatment, aside from Perlka®, was evaluated. Preliminary trials utilizing an application of urea (80 kg N/ha) indicated no detrimental effects on crop health or growth of *S. vulgaris*. It was thought that urea might be more aggressive than, for example, calcium ammonium nitrate. Consequently, it was determined that nitrogen fertilization would not significantly influence the selectivity of *S. vulgaris* or of the crops. Furthermore, no additional

Table 1 Weed species screened for susceptibility to calcium cyanamide in subtrial 1

Species	Trivial name	EPPO-Code
<i>Alopecurus myosuroides</i>	Blackgrass	ALOMY
<i>Bromus sterilis</i>	Barren brome	BROST
<i>Poa annua</i>	Annual bluegrass	POAAN
<i>Capsella bursa pastoris</i>	Blind weed	CAPBP
<i>Chenopodium album</i>	Common lambsquarters	CHEAL
<i>Datura stramonium</i>	Thorn apple	DATST
<i>Galinsoga parviflora</i>	Small-flowered galinsoga	GASPA
<i>Galium aparine</i>	Cleavers	GALAP
<i>Lamium amplexicaule</i>	Common deadnettle	LAMAM
<i>Matricaria matricarioides</i>	Rayless mayweed	MATMT
<i>Myosotis arvensis</i>	Field forget-me-not	MYOAR
<i>Papaver rhoeas</i>	Common poppy	PAPRH
<i>Senecio vulgaris</i>	Common groundsel	SENVU
<i>Sonchus asper</i>	Rough sow thistle	SONAS
<i>Spargula arvensis</i>	Corn spurrey	SPRAR
<i>Stellaria media</i>	Common chickweed	STEME
<i>Thlaspi arvense</i>	Fanweed	THLAR
<i>Urtica urens</i>	Annual nettle	URTUR
<i>Veronica hederifolia</i>	Ivyleaf speedwell	VERHE
<i>Viola arvensis</i>	Field pansy	VIOAR

foundational fertilization was conducted across any of the experimental variants. Throughout the observation period, all crop plants remained viable, and, while growth differences were noted among the cultivated species, any potential damage was not assessed on an individual plant basis; rather, it was inferred that such damage would be reflected in the overall biomass measurements.

Subtrial 1: Species Screening in Pots

Subtrial 1 evaluated the efficacy of calcium cyanamide on 20 weed species (Table 1) commonly found in agricultural and horticultural settings in Germany (Zwerger 2002). The weed seeds were self-propagated at the JKI experimental site and were not subjected to any pre-sowing treatment or germination testing. Immediately after collection from the seed store, 10 seeds of each weed species were placed into the pots, resulting in a weed density of 2040 seeds/m²;

6 pots were used for each species. An untreated control group (also 6 repetitions) was included in the experiment to facilitate comparative analysis. The pots, measuring 7 cm × 7 cm, were filled with 280 g of 4 mm sieved arable soil. The weed seeds were incorporated into the soil to a depth of 1 cm. Perlka® was applied to the soil surface of the pots immediately following the sowing of the weeds at a rate of 363.4 kg/ha. Initially, a target application rate of 400 kg/ha of Perlka®, containing 19.8% nitrogen, was planned; however, an error in nitrogen content calculation (previously using a calcium cyanamid fertilizer with a 22% nitrogen content) led to an unintended reduction of the application rate, which accounts for the variation in application rates between the first subtrial and the subsequent two subtrials 2 and 3. The number of emergent weeds were recorded weekly. One week after no more weeds emerged (77 days post-sowing), it was concluded that no further weed emergence would occur.

Subtrial 2: 10-litre Containers

In the years 2019 and 2020, a study was conducted to evaluate the efficacy of calcium cyanamide controlling *S. vulgaris* in three crop species: lamb's lettuce (*Valeriana locusta* L.), arugula (*Eruca vesicaria* (L.) Cav.), and spinach (*Spinacia oleracea* L.). The investigation focused on the herbicidal impact of 400 kg/ha of Perlka® on *S. vulgaris*, as well as the tolerance levels of the aforementioned crops to the treatment (Table 2). Harvest was always conducted according to the development of the crop, thus the dates defer between crops and years.

For subtrial 2, 25 seeds of *S. vulgaris* (equivalent to a density of 500 seeds/m²) were sown in 10-litre pots with a radius of 12.62 cm (Table 3). Calcium cyanamide was administered at three distinct intervals: three weeks prior to crop sowing (T3), two weeks prior (T2), and one week prior (T1). Additionally, two control treatments were established without the application of calcium cyanamide: one control included sown *S. vulgaris*, while the other did not (Table 3).

All crops were sown at a depth of 2 cm across all treatments on a standardized date, which was one week following the final application of calcium cyanamide (Table 3). Each crop was replicated four times. While the *Senecio* seeds were worked into the soil to a depth of 0.5 cm, calcium cyanamide was applied to the surface of the soil. The

Table 2 Sowing and harvesting dates of crops in the subtrial 2

Crop	Variety	Seeds/m ²	Sowing dates		Harvesting dates	
			2019	2020	2019	2020
Lamb's lettuce	Vit	500	10.08	05.02	25.09	29.04
Arugula	Ventia	400	10.08	05.02	25.09	09.04
Spinach	Scorpius	300	10.08	05.02	25.09	28.04

Table 3 Application and sowing dates of calcium cyanamide and *Senecio vulgaris* in subtrial 2

	Treatment	Date of calcium cyanamide application	Sowing dates of <i>S. vulgaris</i> 500 Seeds/m ²	Date of calcium cyanamide application	Sowing dates of <i>S. vulgaris</i> 500 Seeds/m ²
T0	Control (Crops only)	–	–	–	–
T0SENVU	Control (crop species + <i>S. vulgaris</i>)	–	10.08.2019	–	05.02.2020
T1	400 kg/ha Perlka 1 week before sowing the crop species	03.08.2019	03.08.2019	29.01.2020	29.01.2020
T2	400 kg/ha Perlka 2 weeks before sowing the crop species	27.07.2019	27.07.2019	22.01.2020	22.01.2020
T3	400 kg/ha Perlka 3 weeks before sowing the crop species	20.07.2019	20.07.2019	15.01.2020	15.01.2020

experimental setup was designed as a semi-open pot trial, allowing for relocation under protective cover during adverse weather conditions, such as heavy rainfall or hail; however, the pots were primarily positioned outdoors.

Upon harvest, the number of crop and weed plants present, as well as the total fresh biomass for both groups of plants in each replicate, were quantified.

Subtrial 3: Open Field in Small Plots

Subtrial 3 was designed similarly to subtrial 2, but it was conducted in field plots measuring 2 m × 1 m instead of using pots. The primary objective of this setting was to assess the herbicidal efficacy of 400 kg/ha of Perlka® on *S. vulgaris*. As edge effects of pot experiments can be severe, this semi-field approach was assumed to be superior to the pot trial conducted in subtrial 2. As the trial was conducted during the summer, lamb's lettuce was excluded as there was no suitable variety for summer cropping. Therefore, only arugula (*Eruca vesicaria*) and spinach (*Spinacia oleracea*) were included in this trial (Table 4). Perlka® was applied at three different timings (T1–T3) as outlined in Table 5, mirroring the methodology employed in subtrial 2. Control treatments without calcium cyanamide were established with crops only (T0) and crops combined with *S. vulgaris* (T0SENVU). In subtrial 3, *S. vulgaris* was sown at a density of 100 seeds per plot, corresponding to 50 seeds/m²—a more realistic setting than in subtrial 2. Arugula and spinach were sown at densities of 400 seeds/m² and 300 seeds/m², respectively. At the time of harvest, the number of *Senecio* plants in each plot was counted to evaluate the impact of calcium cyanamide as herbicide. Furthermore, the fresh biomass of the crop leaves was measured to assess the crop sensitivity to the treatment. Subtrial 3 was conducted over a single year. However, the results, partic-

ularly regarding the control of *S. vulgaris*, were consistent with those observed in subtrial 2. Therefore, it was not anticipated that repeating the trial in subsequent years would yield further insights.

Statistical Analysis

All statistical analyses were conducted using R software (R Core Team 2024). For subtrial 1, the Mann-Whitney U test was employed with Bonferroni correction, utilizing the coin package (Hothorn et al. 2006). In Subtrials 2 and 3, multiple comparisons were performed using Dunn's test with Bonferroni correction, implemented through the R package dunn.test (Dinno 2024). Figures were generated using the ggplot2 package (Wickham 2016).

Results

Subtrial 1

Alopecurus myosuroides Huds. showed the highest emergence rate in the untreated controls of all weed species, reaching 85% (Fig. 1). Overall however, the dormancy behaviour exhibited by various weed species resulted in uneven or very low average emergence rates (e.g., *Galinsoga parviflora* Cav. at 1% and *Spergula arvensis* L. at 13%), which hindered a comprehensive efficacy assessment for all species tested.

However, most weed species demonstrated a reduction in germination following the application of calcium cyanamide (Fig. 1). A statistically significant reduction in emergence was observed for *A. myosuroides*, *Matricaria chamomilla* L., *Poa annua* L., *Senecio vulgaris* L., *Thlaspi arvense* L., and *Veronica hederifolia* L.. Exceptions were

Table 4 Sowing and harvesting dates of crops in the subtrial 3

Crop	Variety	Seeds/m ²	Sowing dates	Harvesting dates
Arugula	Ventia	400	10.08.2022	05.09.2022
Spinach	Scorpius	300	10.08.2022	15.09.2022

noted for *Bromus sterilis* L., *Chenopodium album* L., and *Datura stramonium* L., which exhibited no change or even an increase in emergence following calcium cyanamide treatment. The germination capacity of these species in the control group ranged from 13 to 38%. It is important to acknowledge that the significance of these results is limited due to the relatively low germination capacity observed. Additionally, species such as *C. album* have been

documented in the literature as being more challenging to control with calcium cyanamide (Kurth 1960).

Subtrial 2

Efficacy

The results from the two testing years 2019 and 2020 have been aggregated for analysis. *S. vulgaris* demonstrated

Fig. 1 Number of germinated individual weed plants from subtrial 1 for plants treated or not treated with calcium cyanamide. The asterisk before the species indicates a significance between treated and untreated

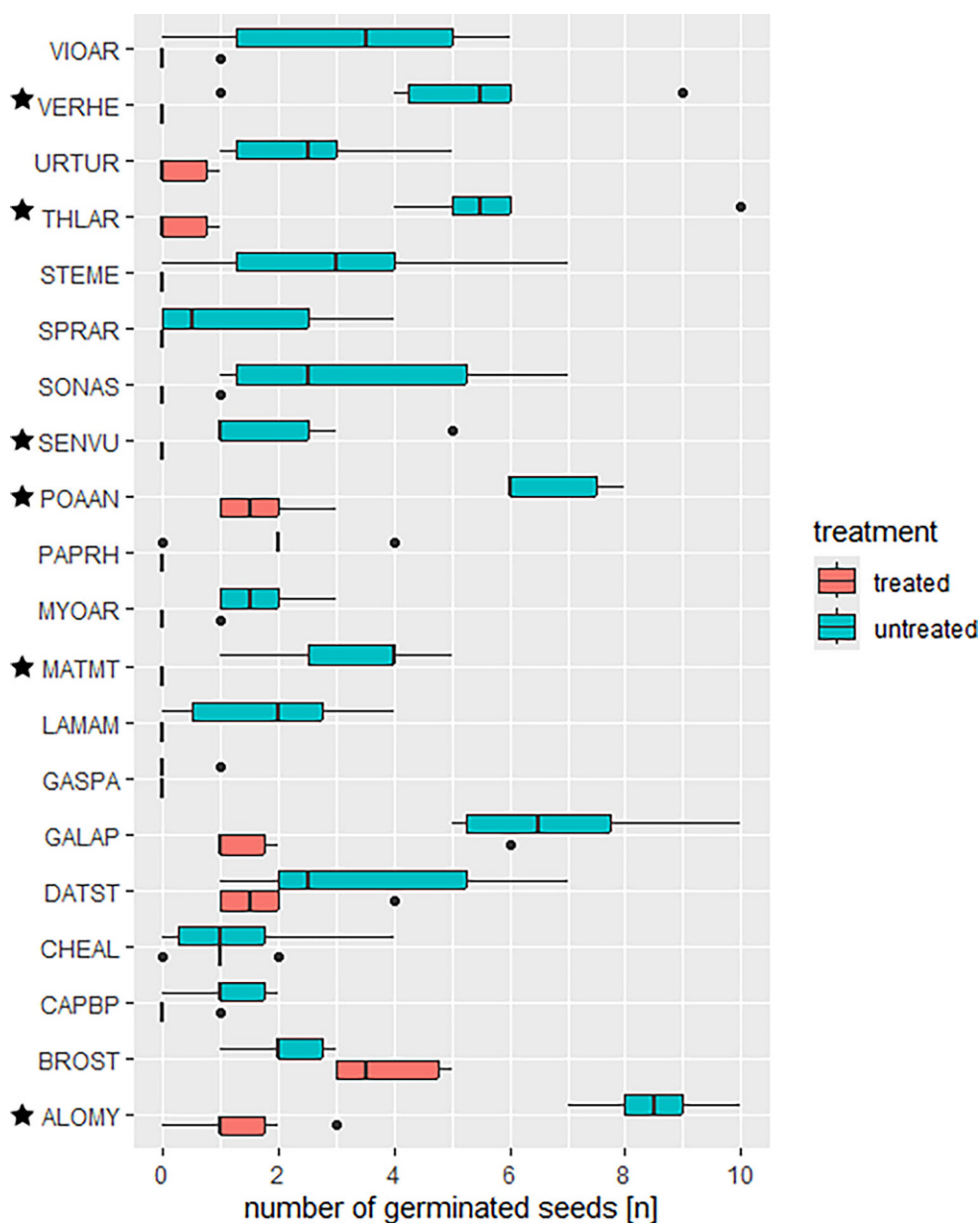


Table 5 Application and sowing dates of calcium cyanamide and *Senecio vulgaris* in subtrial 3

	Treatment	Date of calcium cyanamide application	Sowing dates of <i>S. vulgaris</i> 50 Seeds/m ²
T0	Control (Crops only)	–	–
T0SENVU	Control (crop species + <i>S. vulgaris</i>)	–	10.08.2022
T1	400 kg/ha Perlka 1 week before sowing the crop species	03.08.2022	03.08.2022
T2	400 kg/ha Perlka 2 weeks before sowing the crop species	27.07.2022	27.07.2022
T3	400 kg/ha Perlka 3 weeks before sowing the crop species	20.07.2022	20.07.2022

a high sensitivity to the application of calcium cyanamide. The number of *Senecio* plants at harvest observed across the three crop types was comparable (Fig. 2). However, there was a significant difference in the fresh biomass of *S. vulgaris* among the crops (Fig. 3). Specifically, the more competitive the crop, the lower the fresh mass of the *Senecio* plants. In the case of the low-growing lamb's lettuce, *S. vulgaris* was able to thrive, whereas in arugula, the plants remained extremely small and produced minimal fresh mass. All treatments involving calcium cyanamide exhibited a consistent trend: nearly no weeds were present at the time of harvest (Fig. 2). Correspondingly, the biomass harvested from the calcium cyanamide treatments was almost negligible (Fig. 3). The differences in biomass or plant counts of *S. vulgaris* between the various calcium cyanamide treatments were not statistically significant; however, the differences between these treatments and the control (without calcium cyanamide) were consistently significant.

The efficacy of calcium cyanamide in this trial exceeded 90% across all treatments. The median emergence of *S. vulgaris* in the control group ranged from 12 to 14 plants out of the 25 sown.

Crop Sensitivity

In all three crops, the comparison of the T0 and T0SENVU variants showed that there was no effect on the yield of the crop due to competition from the existing *Senecio* plants. Clear differences in sensitivity were observed among the three crops tested. Figures 4 and 5 illustrate the trends, while Tables 6 and 7 present the significance of the pairwise comparisons of the treatments for all crops.

Lamb's Lettuce: In terms of the number of emerged individuals, lamb's lettuce exhibited a dependence on the time interval between fertilizer application and sowing. Specifically, there was a tendency for lower plant emergence and a decrease in fresh mass the sooner the crop was sown after

fertilizer application, as the interval decreased. However, these differences were not statistically significant.

Arugula: In 2020, all three treatments involving calcium cyanamide resulted in nearly total crop failure for arugula, leading to pronounced differences. In 2019, the plants that were fertilized one week before sowing were primarily affected, showing a reduced number of plants. In contrast, in 2020, only a few plants emerged from these treatments. The variation between the years accounts for the wide range observed in the number of crop plants (Fig. 4) as well as the biomass (Fig. 5).

Spinach: Spinach was the least sensitive of the crops tested, demonstrating a high tolerance to calcium cyanamide. No significant effects on the number of emerged plants were detected (Fig. 4). However, there was a noticeable increase in biomass variance as the distance between sowing and the application of calcium cyanamide increased (Fig. 5). In all three crops, comparisons between the T0 and T0SENVU variants indicated that there was no impact on crop yield due to competition from the existing *Senecio* plants.

Subtrial 3

For subtrial 3 the same treatments as in subtrial 2 were used, but only arugula and spinach as crop were tested. In addition, the trial was conducted as a semi-field trial. Semi-field trial means that the trial took place in the field, but in concrete boxes, each measuring 2 m × 1 m. The concrete plots are much less affected by external factors than pots and therefore almost comparable to the open field.

Efficacy

The efficacy of calcium cyanamide was determined by counting *S. vulgaris* plants in the plots at harvest. In the calcium cyanamide-free treatment (T0SENVU), an average of 70 to 80 plants were counted at harvest (Fig. 6), emerged from 100 seeds per plot. In contrast, in the calcium cyanamide-treated treatments, a maximum of 13 plants

were counted at harvest. Most of the differences of the treatments were proven to be significant by the Dunnett Test (Table 8). Overall, efficiencies of well over 90% were achieved in the calcium cyanamide-treated treatments. As most of the plots had very few *S. vulgaris* plants, no fresh mass determination was carried out.

Crop Sensitivity

Overall, crop biomass was significantly higher in all treatments with calcium cyanamide than in those without (Fig. 7; Table 9). No damage was seen in either crop, despite the fact that the calcium cyanamide application was only one week earlier. Furthermore, there was no significant difference in yield between the treatments with calcium cyanamide for the arugula. However, there was significant higher yield for spinach with a one week interval (T1) between calcium cyanamide application and sowing of the crop.

Discussion

The herbicidal effect of calcium cyanamide is certainly a secondary purpose, as it is mainly used to supply mineral nutrients. Nevertheless, the beneficial effects of its use in agriculture and horticulture in terms of weed control and reduction of plant diseases are readily exploited (Heitefuss et al. 1977). Calcium cyanamide has either a corrosive effect on the surface of the leaf tissue of young plants or a toxic effect in soil on the roots of some monocotyledonous grass weeds and dicotyledonous seed weeds at the germination stage (Koch 1970; Soltys et al. 2011). During degradation cyanamide is broken down to urea, a process accelerated by increasing temperature (Vilsmeier and Amberger 1980).

In the current study, the application of calcium cyanamide was intended to provide weed control against *S. vulgaris* in three types of leafy vegetable. The fertiliser was therefore applied at the same time as the *S. vulgaris* was sown and the soil was kept sufficiently moist. This is because the degradation of calcium cyanamide is primarily dependent on soil moisture and temperature (Amann 1956).

Control of *S. vulgaris* by Calcium Cyanamide

The results of the series of trials presented here demonstrates the basic suitability of calcium cyanamide for the control of some weeds. This is consistent with the results of other studies which also found good activity against a number of weeds (Koch 1970; Ohdaira 2021; Krupa and Witkowicz 2023). In the screening of 20 weed species (subtrial 1, Fig. 1), only *C. album*, *D. stramonium* and *B. sterilis* were found to be relatively insensitive to the application of calcium cyanamide, indicating a wide range of possible target weeds for calcium cyanamide. However, these three insensitive species are important and wide spread weeds in various agricultural and horticultural crops. Thus, calcium cyanamide alone might not be sufficient to achieve satisfactory weed control in many cases and additional measures of weed control will be necessary. As the emergence of some weeds was low, below 20% (e.g. *Galinsoga parviflora* Cav. at 1% and *Spergula arvensis* L. at 13%), the significance of this trial is naturally limited. A higher number of weed seeds might result in a clearer picture. However, the control of *S. vulgaris* was very promising, which was further investigated in subtrials 2 and 3.

Subtrials 2 and 3 showed very similar results in three different years and two different setups (pot and field), although subtrial 3 was only conducted in one year. On the one hand, calcium cyanamide showed very good control of

Fig. 2 Number of *S. vulgaris* plants at harvest in three crops in four treatments. *S. vulgaris* was always treated with 400 kg/ha calcium cyanamide when sown. Only in the treatment T0SENVU there was no calcium cyanamide applied

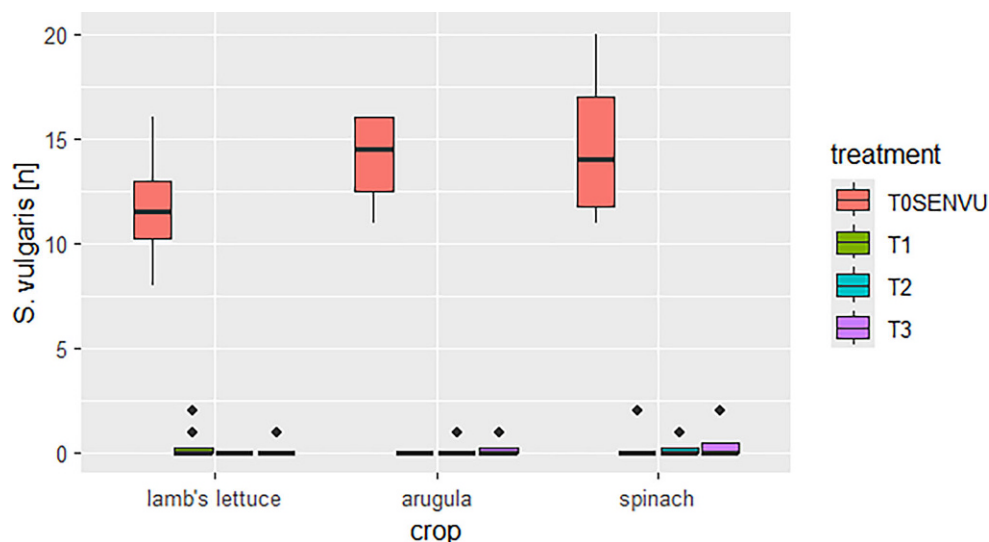


Fig. 3 Biomass of *S. vulgaris* plants at harvest in three crops in four treatments. *S. vulgaris* was always treated with 400 kg/ha calcium cyanamide when sown. Only in the treatment T0SENVU there was no Calcium Cyanamid applied

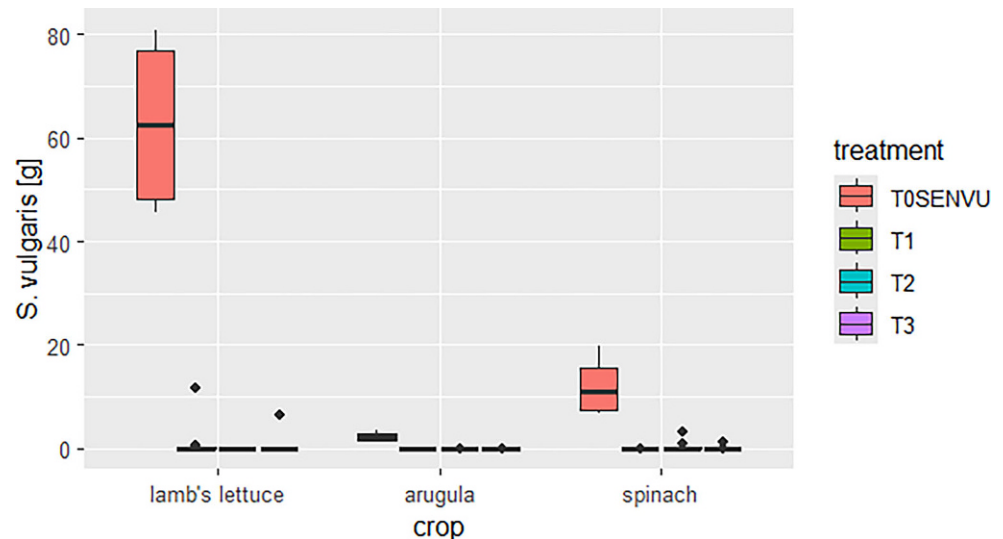
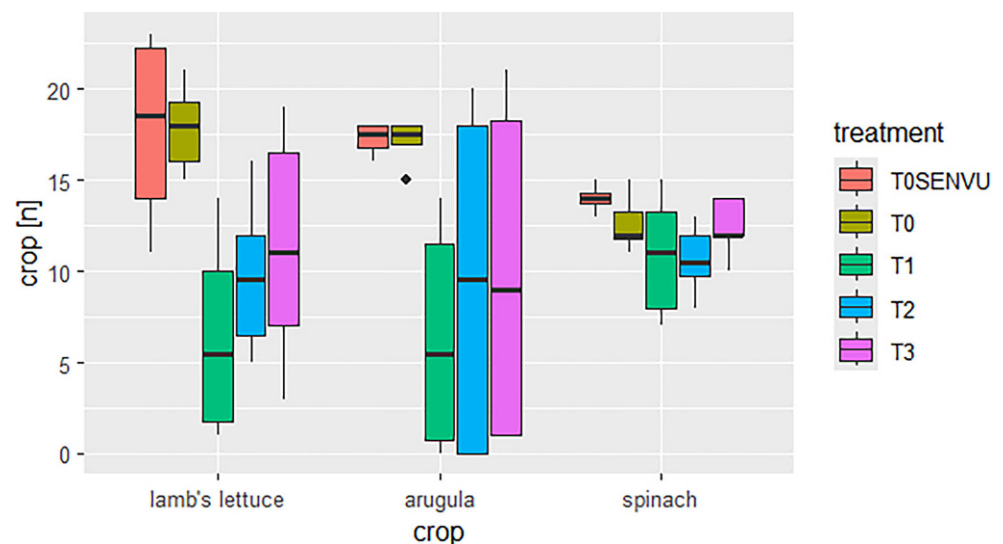


Fig. 4 Number of harvested crop plants in three treatments with calcium cyanamide (T1, T2, T3) and two treatments without calcium cyanamide (T0SENVU, T0) for subtrial 2. T1, T2, and T3 were treated with calcium cyanamide 1, 2, and 3 weeks before sowing of the crops, respectively. Table 6 shows the significances of the multiple comparisons



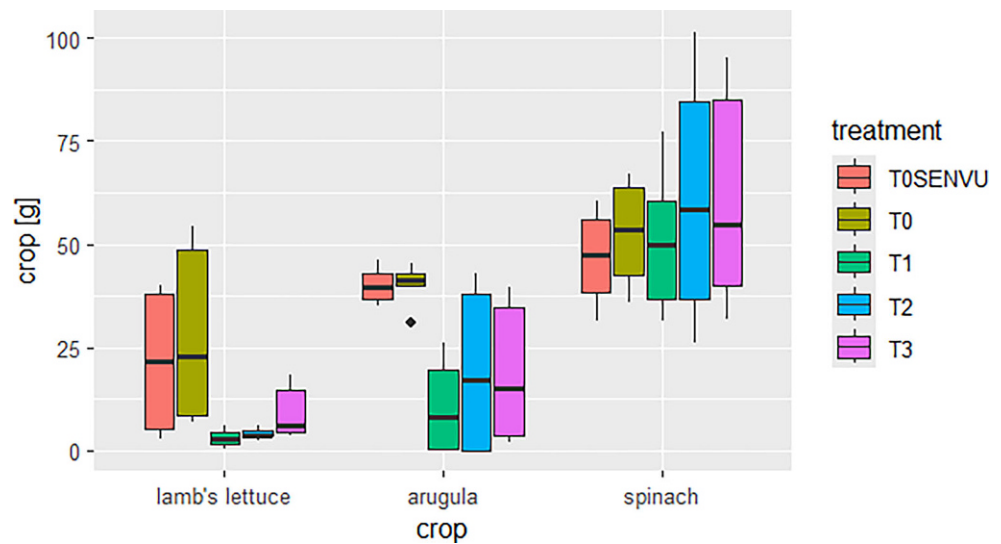
S. vulgaris, certainly matching the desired control (>90%) of other herbicides used in these crops (Figs. 2 and 6). The germination rate of *S. vulgaris* was very low when calcium cyanamide was applied in the subtrials 2 and 3, confirming the results of subtrial 1. The biomass of *S. vulgaris* measured in subtrial 2 was close to zero. Nevertheless, few *S. vulgaris* plants were able to establish in the different calcium cyanamide treatments in both subtrials, so that complete control was not achieved. The surviving plants remained small and did not compete with the crop (Figs. 5 and 7). However, the question remains how many parts of *S. vulgaris* plants could end up in the harvested crop under field conditions, causing a contamination of the harvest with the toxic pyrrolizidine alkaloids.

According to Nitzsche et al. (2018), *S. vulgaris* is the main source of contamination with pyrrolizidine alkaloids of food in Germany. The co-harvesting of *S. vulgaris*,

or other pyrrolizidine containing weeds, with the crops is a way that harvested crops can be contaminated with pyrrolizidine-containing plant material. It is therefore essential to keep crops free of *S. vulgaris* plants—in case of doubt, even by hand weeding. However, it is currently not possible to completely prevent the contamination of plant raw materials with pyrrolizidine alkaloids (Lebensmittelverband Deutschland e.V. 2019). New processes utilising robotics and AI can bring further improvements here.

The different development of *S. vulgaris* in the cultivated crops was striking in the subtrials 2 and 3. Although the number of *S. vulgaris* plants was comparable in all crops (subtrial 2), the biomass of *S. vulgaris* determined in the control was about 10 times higher in the lamb's lettuce crop (Fig. 3) than in the spinach crop and about 20 times higher than in the arugula crop. Arugula significantly inhibited the

Fig. 5 Biomass of three harvested crop species in three treatments with calcium cyanamid (T1, T2, T3) and two treatments without calcium cyanamid (T0SENVU, T0) for subtrial 2. T1, T2, and T3 were treated with calcium cyanamid 1, 2, and 3 weeks before sowing of the crops, respectively for subtrial 2. Table 7 shows the significances of the multiple comparisons



growth of *S. vulgaris* in both years, spinach less, and lamb's lettuce proved to be a very weak competitor, probably due to its slower development.

Tolerance of the Crop to Calcium Cyanamid

It is more difficult to interpret crop tolerance respective sensitivity to an application of calcium cyanamid than to assess the efficacy of the treatment. Because calcium cyanamid firstly causes toxic effects to plants and only then it serves as fertilizer. The crop needs to be sufficiently

Table 6 *P*-value showing the significance of differences in crop plants at harvest between the treatments for lamb's lettuce, arugula, and spinach. The Dunnett test with Bonferroni correction for multiple comparison was used. Figure 4 shows the corresponding counts of crop plants

Comparisons	Lamb's lettuce	Arugula	Spinach
T0—T0SENVU	1	1	1
T0—T1	0.017	0.051	0.541
T0SENVU—T1	0.001	0.008	0.003
T0—T2	0.12	0.94	0.228
T0SENVU—T2	0.021	1	0
T1—T2	1	0.631	1
T0—T3	0.643	1	1
T0SENVU—T3	0.387	1	0.059
T1—T3	0.427	0.404	1
T2—T3	1	1	1

Table 7 *P*-value showing the significance of differences in crop biomass between the treatments for lamb's lettuce, arugula, and spinach. The Dunnett test with Bonferroni correction for multiple comparison was used. Figure 5 shows the corresponding biomass of crop plants

Comparisons	Lamb's lettuce	Arugula	Spinach
T0—T0SENVU	0.399	1	1
T0—T1	0.448	0.071	1
T0SENVU—T1	0	0.001	1
T0—T2	0.786	0.388	1
T0SENVU—T2	0	0.024	0.623
T1—T2	1	1	1
T0—T3	1	0.411	1
T0SENVU—T3	0.012	0.027	0.575
T1—T3	0.667	1	1
T2—T3	1	1	1

Fig. 6 Number of weeds for two crops in three treatments with calcium cyanamide. T1, T2, and T3 were treated with calcium cyanamide 1, 2, and 3 weeks before sowing of the crops, respectively. The treatment T0SENVU was not treated with calcium cyanamide. Table 8 shows the significances of the multiple comparisons

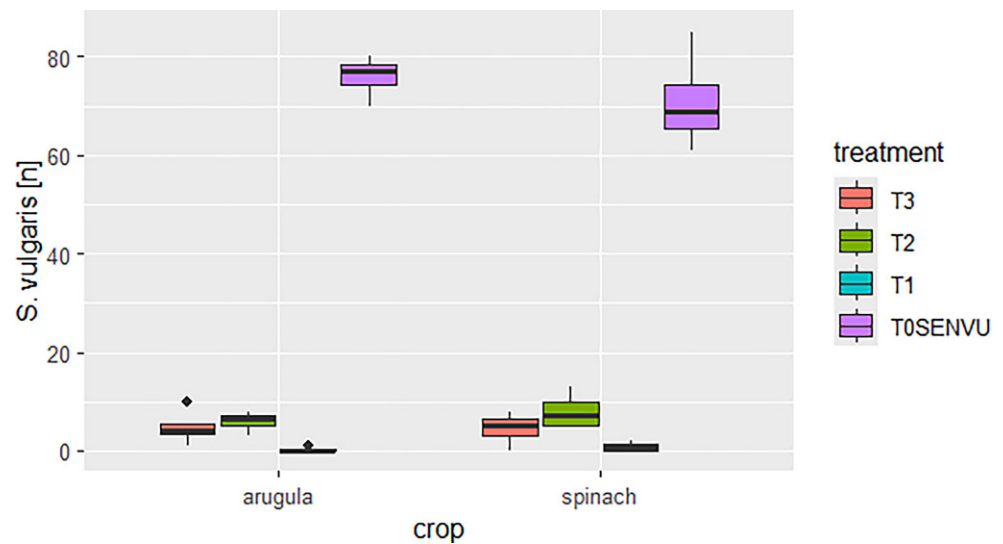
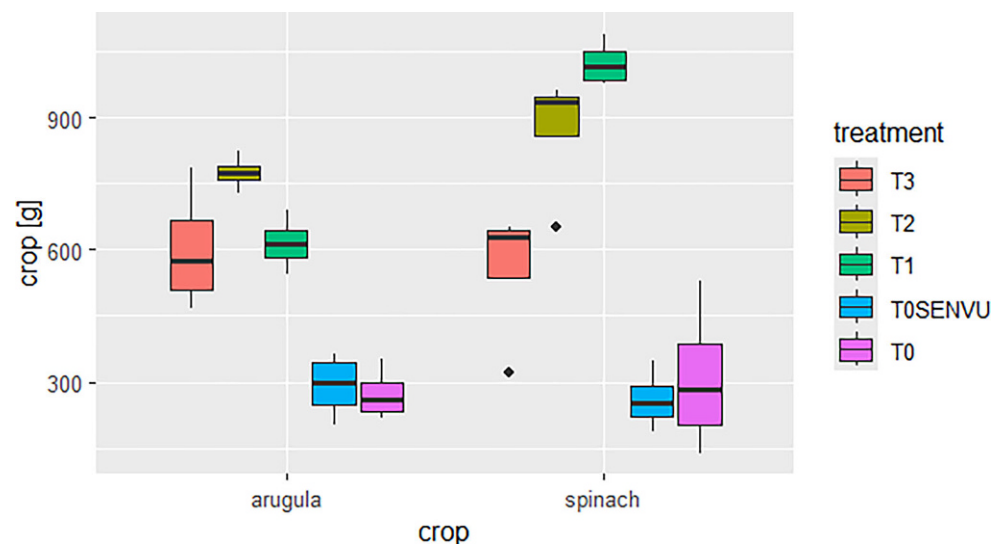


Fig. 7 Biomass of harvested crop species arugula and spinach in five treatments. T1, T2, and T3 were treated with calcium cyanamide 1, 2, and 3 weeks before sowing of the crops, respectively. The treatments T0 and T0SENVU were not treated with calcium cyanamide. *S. vulgaris* was sown in all treatments but not in T0. Table 9 shows the significances of the multiple comparisons



tolerant to the first phase to benefit from the later. For the tested crops, a different sensitivity could certainly be determined. In literature a severe difference in sensitivity was observed for other crops as well (Soltys et al. 2012; Sun et al. 2024).

In subtrial 2, lamb's lettuce was found to be very sensitive to calcium cyanamide. The application reduced the number of plants harvested (Fig. 4) and yield (Fig. 5) decreased significantly. A similar, but less pronounced, effect was seen with arugula. Here the variance in both the number of plants harvested and the biomass harvested was significantly increased when calcium cyanamide was applied compared to the control.

Unlike the other two crops, spinach proved to be almost insensitive to the toxic cyanamide phase of calcium cyanamide. Neither the biomass nor the number of plants decreased significantly. But it should be noted that the vari-

ance increased when the crop was sown with a distance of two to three weeks to the treatment. This indicates the possibility of a certain level of phytotoxic effect for spinach as well. It was also striking that the unfertilised treatment in subtrial 2 produced the same yield in all crops as the fertilised treatment without weeds. Fertilisation did not increase yield of spinach, which is usually the aim of nitrogen fertilisation and thus at least one aim of calcium cyanamid application. In contrast, subtrial 3 showed a clear fertiliser effect on yield of spinach. All fertilised treatments showed a higher yield than the unfertilised control. The fertilising effect of nitrogen completely superimposed other phytotoxic symptoms on the crop and the calcium cyanamide fertilised treatments achieved significantly higher yields than the unfertilised treatments for both crops. This discrepancy between the subtrials may be explained by differences in the experimental setting. While subtrial 2 was planted in pots

Table 8 *P*-value showing the significance of differences in number of weeds of the treatments for arugula and spinach as well as both crops combined. The Dunnet test with Bonferroni correction for multiple comparison was used. Figure 6 shows the corresponding number of weeds at harvest

Comparisons	Arugula	Spinach	All crops
T0 SENVU—T11	0.001	0.002	0
T0 SENVU—T2	0.171	0.187	0.02
T1—T2	0.144	0.063	0.005
T0 SENVU—T3	1	1	1
T1—T3	0.001	0.002	0
T2—T3	0.302	0.408	0.079

Table 9 *P*-value showing the significance of differences in biomass between the treatments for arugula and spinach as well as both crops combined. The Dunnet test with Bonferroni correction for multiple comparison was used. Figure 7 shows the corresponding biomass of crop plants

Comparisons	Arugula	Spinach	All crops
T0—T0SENVU	1	1	1
T0—T1	0.243	0.005	0.001
T0SENVU—T1	0.182	0.006	0.001
T0—T2	0.009	0.116	0
T0SENVU—T2	0.006	0.135	0.001
T1—T2	1	1	1
T0—T3	0.279	1	0.124
T0SENVU—T3	0.211	1	0.154
T1—T3	1	0.211	0.618
T2—T3	1	1	0.456

with soil without a previous crop, subtrial 3 was planted in plots with a previous crop. It can be assumed that this resulted in different nutrient levels in the soil. While the plants in subtrial 2 were well supplied without additional fertilisation even in the treatments without calcium cyanamide, the plants in subtrial 3 were undersupplied with nitrogen without fertilisation and were therefore dependent on fertilisation for yield. Thus, an additional treatment with an alternative N-supply would allow a deeper interpretation of this effect.

A reduction in yield would have been expected due to the good establishment of *S. vulgaris* in subtrial 2 and 3. But even in treatment with the most successful establishment and strong growth of *S. vulgaris* lamb's lettuce (Fig. 3), the yield of the crop was not reduced by the weed (Fig. 5, T0SENVU—T0). *S. vulgaris* showed no measurable competitive effect on the crops. Thus, the pyrrolizidine alkaloid content is the main challenge this weed poses to crop production.

Conclusion

Calcium cyanamide is certainly an option for adding a module to integrated crop protection. However, the conversion behaviour of calcium cyanamide, which depends on weather and soil conditions, can make application difficult and requires very close attention to the application instructions. Calcium cyanamide showed excellent activity against *S. vulgaris* in the current study, but as the degradation and thus the establishment of the toxic phase of calcium cyanamide is weather dependent (Vilsmeier and Amberger 1980) the actual timing of application and sowing of the crop poses practical difficulties, especially in sensitive crops such as lambs lettuce. Variability in efficacy must be taken into account, especially if weeds such as pyrrolizidine-containing weed species are to be removed from the crop as completely as possible. Monitoring of the crop and removal of individual *S. vulgaris* plants is therefore necessary, especially when used in special crops.

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Conflict of interest H.-P. Söchting and C. von Redwitz declare that they have no competing interests.

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