Interim Technical Report on the Vetch Trial

"ESTUDIO DE RESISTENCIA AL FRÍO Y LAS HELADAS DE DOS ESPECIES DE VEZA, VICIA VILLOSA (VARIEDAD ORIUM) Y VICIA SATIVA"

Analysis of Germination, Frost Resistance and Yield per Hectare

Species Evaluated: Hairy Vetch (*Vicia villosa var. Orium*) and Common Vetch (*Vicia sativa*)
Evaluation Period: December 16, 2024 – April 29, 2025

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1. Introduction

Cold and frost resistance is a determining factor for the success of autumn-sown crops in temperate and continental climates, where extreme winter temperatures can negatively impact plant survival and significantly reduce yield. In this context, the species Hairy Vetch (*Vicia villosa* Roth) and Common Vetch (*Vicia sativa L.*), both leguminous plants of recognized agronomic and ecological value, exhibit high genetic variability in their ability to tolerate low temperatures, making them key candidates for integration into sustainable farming systems.

Identifying winter-resistant genotypes would not only allow for the expansion of cultivated areas in frost-prone zones, but also enhance their role as cover crops due to their ability to fix atmospheric nitrogen and improve soil structure and fertility. Moreover, the use of adapted species can increase the protein content of forage, benefiting both livestock production and the sustainability of crop rotations.

Assessing cold tolerance in vetch requires a comprehensive approach that combines field observations under real conditions with the analysis of agronomic parameters such as germination, tolerance to extreme temperatures, post-frost recovery capacity, and forage yield. As demonstrated in legume species like faba bean (*Vicia faba L.*), multi-site and multi-season trials are essential to identify tolerance mechanisms associated with genotype and its interaction with the environment (Arbaoui et al., 2008).

The present trial was conducted at the experimental practice fields of ETSIAAB (Technical University of Madrid), where two vetch species of Spanish origin were evaluated: Hairy Vetch (variety ORIUM) and Common Vetch, through an autumn sowing in December 2024 with an estimated harvest in May 2025. This evaluation period allows for the monitoring of both species' performance during the winter, including frost resistance, recovery capacity, and final yield. The results will help identify varieties with greater adaptability to continental climates, providing valuable insights for genetic selection and the design of agronomic management strategies in winter legumes.

2. Objectives of the Trial

The aim of this study is to evaluate the cold and frost resistance of seeds from two Spanish vetch species: Hairy Vetch (*Vicia villosa* variety ORIUM) and Common Vetch (*Vicia sativa*), through an autumn sowing trial with monitoring until harvest in spring.

The specific objectives are as follows:

- Analyze the germination percentage of both species under field conditions during the winter period.
- Assess tolerance to low temperatures and frost events by quantifying the degree of damage and the plant's recovery capacity.
- Measure forage yield at the end of the cycle, expressed in tons per hectare.
- Determine the crude protein content of the forage as an indicator of nutritional quality.
- Generate technical information that contributes to the selection of varieties better adapted to continental climates and to the genetic improvement of winter legumes, with a positive impact on the sustainability of agricultural and livestock systems.

3. Materials and Methods

Trial Location:
Agronomic Experimentation Fields – ETSIAAB (Figure 1).
Latitude: 40°26'23.16"N | Longitude: 3°44'14.72"W | Altitude: 591 masl.

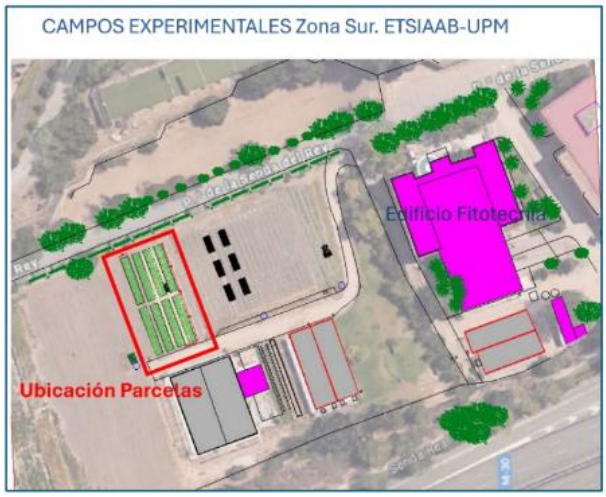

Figure 1 Location of the Vetch Trial Plots.

Climatic Conditions:
Throughout the trial period, weather conditions were monitored daily by a meteorological station located at the Agronomic Experimentation Fields – ETSIAAB, which provided accurate data on temperature, precipitation, and reference evapotranspiration (ET_0) (Figure 2, 3).

The minimum temperature recorded at the sowing day (December 16) was -2.98°C, with an extreme low of -9°C on January 15, and an average minimum temperature of -2.14°C. A total of 40 frost days (i.e., days with temperatures below 0°C) were recorded during the trial, imposing significant abiotic stress on the plants. In contrast, the highest temperature recorded was 25.29°C on March 31, with an overall average maximum temperature of 14.60°C across the 127-day trial duration (Figure 2).

Regarding precipitation, the rainiest day was March 11, with a total of 34.6 mm. Total cumulative precipitation for the entire cycle was 294.8 mm, which represented the sole source of water for the crop, as the trial was conducted under rainfed conditions (Figure 3).

With respect to water demand, the accumulated reference evapotranspiration (ET_0) during the trial was 175.1 mm. The highest ET_0 value was recorded on April 8, at 3.8 mm, coinciding with an advanced phenological stage of the crop. These data reflect a progressive increase in water

demand toward the end of the cycle, which is a key factor to consider in future agronomic management strategies aimed at improving water use efficiency (Figure 3).

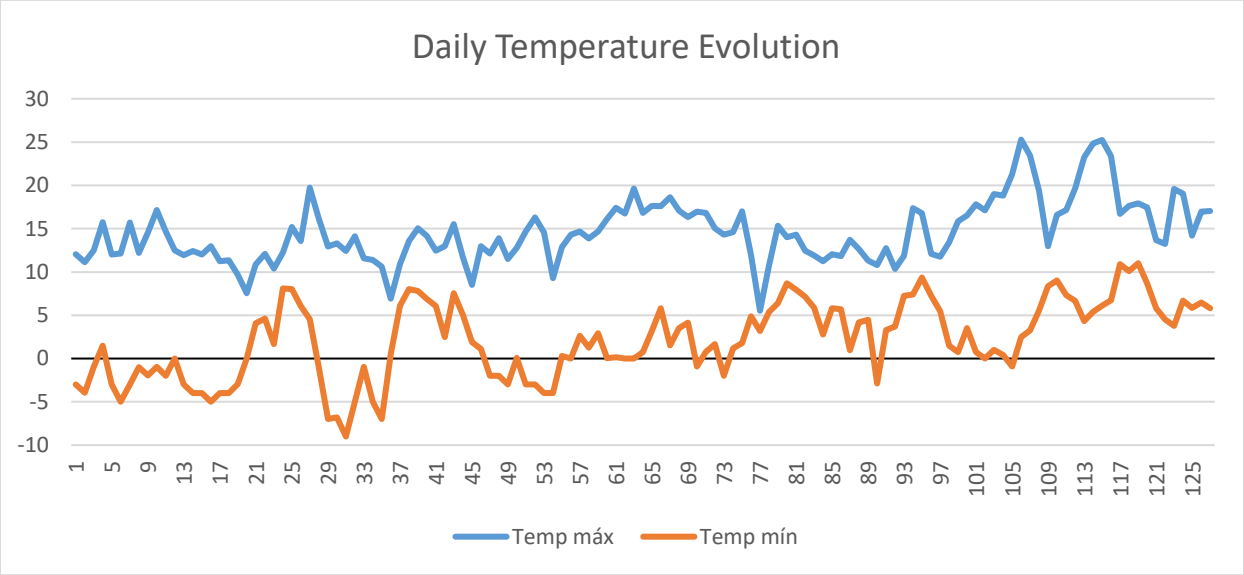

Figure 2 Daily Temperature Evolution over the 127 days of the test. Maximum temperatures are shown in blue, and minimum temperatures are shown in orange.

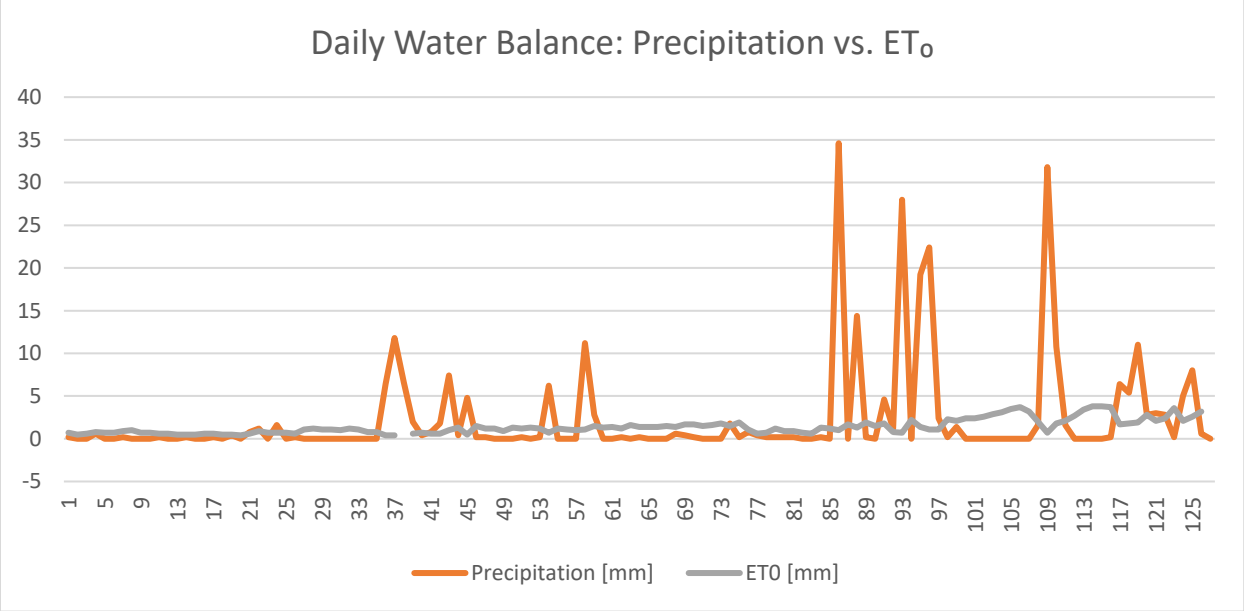

Figure 3 Daily Water Balance over the 127 days of the test. Precipitation mm in orange and ET_0 mm in grey.

Experimental Design:

- Randomized complete block design (RCBD) with 4 replications per variety.
- Plot size: 36 m² (15 x 2.4 m)
- Sowing density: 10 g/m²

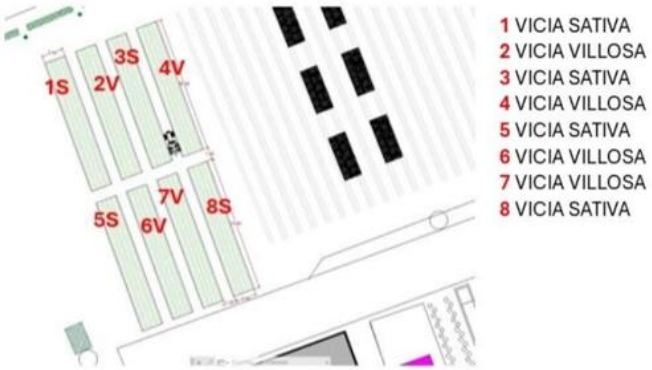

Figure 4 Experimental Design Plot Distribution.

Key Dates:

- **Sowing:** Day 1 - December 16,2024
- **Germination assessment:** Day 40 - January 24, 2025
- **Frost damage evaluations:** Day 53 - February 6, 2025
- **Plant development assessment:** Day 65 - February 18, Day 101 - March 26, 2025
- **Harvest:** Day 135 - April 29, 2025

Figure 5 Vetch trial sowing December 16, 2024.

Methodology for each parameter:

- ***Germination:*** Counting emerged seedlings within a delimited area 1 x 1 m.
- ***Frost Damage:*** Visual assessment of damage levels (Zero, Partial, Severe) within a delimited area 1 x 1 m.
- ***Yield:*** Harvest of plots, adjust for moisture content and extrapolation to kg/ha.

4. Results

4.1 Germination percentage assessed on 24 January 2025

Species	Germination (%)	Observations
Vicia Villosa	70 %	Dull color, partial emergence
Vicia Sativa	77%	Vibrant color, full emergence

During the evaluation period, notable differences were observed in the germinative behavior of the two species analyzed (Figure 6). The *V. sativa* exhibited a higher percentage of germination, indicating greater seedling vigor compared to the *V. villosa*, which showed a lower rate of emerged seedlings.

In addition to the germination percentage, early morphological differences were identified between both species. The *V. sativa* showed a more intense and vibrant coloration in the emerged seedlings, which may be related to a higher chlorophyll content and improved initial photosynthetic capacity (Figure 6). In contrast, seedlings from the *V. villosa* species presented a duller color, which could reflect lower physiological vigor.

Figure 6 Germination percentage assessed on 24 January 2025 (Day 40). *Vicia villosa* (left) *Vicia sativa* (right).

In terms of emergence, the *V. sativa* also demonstrated faster and more uniform seedling emergence, with a higher number of seedlings emerging within the first few days after sowing.

4.2 Frost Resistance assessed on February 6, 2025

Frost Date	Min Temp (°C)	Species	Zero Damage	Partial Damage	Severe Damage	Observations
06/02/2025	-4 °C	***Vicia Villosa***	52.5%	38.8%	8.8%	Mild necrosis on upper leaves, stem elongation, thinner leaves.
06/02/2025	-4°C	***Vicia Sativa***	94%	5.5%	0.5%	Zero damage, no visible signs of frost injury.

Regarding the frost damage evaluation, significant differences were observed between the species analyzed (Figure 7). The *V. sativa* demonstrated notable tolerance to low-temperature stress, with 94% of the plants showing no visible symptoms of damage (zero damage) (Figure 8). In *V. sativa*, no lesions were found on leaves or stems, indicating good physiological adaptation to cold conditions, possibly associated with morphological and anatomical traits that provide greater resistance to low temperatures.

In contrast, the *V. villosa* exhibited a less favorable response, with only 52.8% of individuals showing no visible damage, while the remainder displayed typical symptoms of cold stress, such as thin leaves, foliar necrosis, and abnormal stem elongation (See Fig. 8). These symptoms suggest a lower acclimation capacity to frost, which could negatively impact vegetative development and ultimately reduce yield under more continental conditions where low temperatures are frequent during crop establishment.

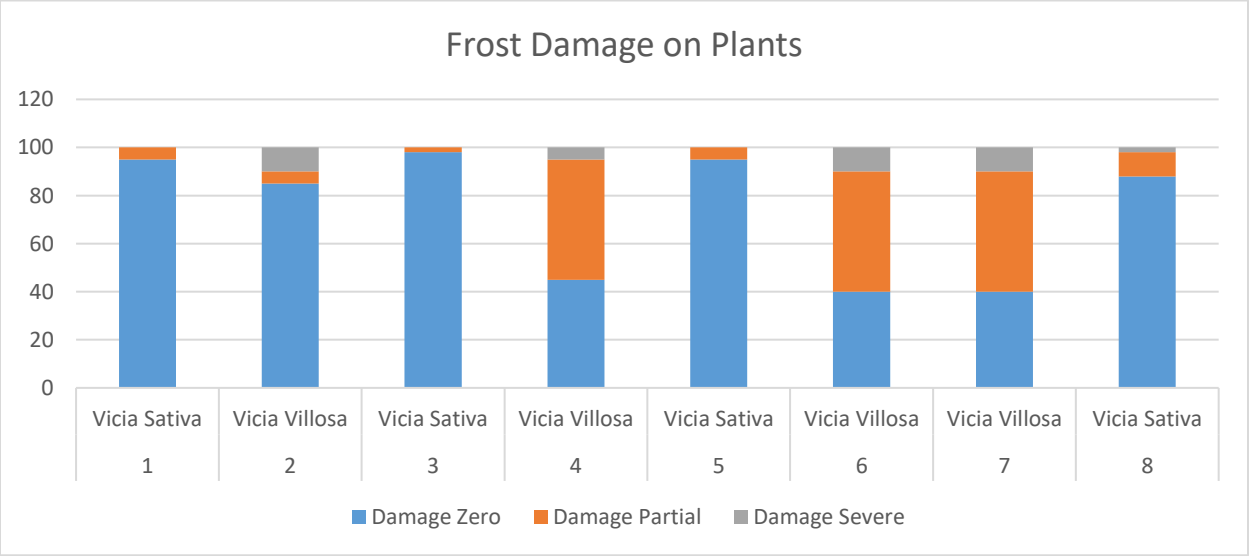

Figure 7 Frost Damage in the Eight Test Plots assessed on February 6, 2025 (Day 53). Zero damage in blue, Partial damage in orange, and Severe damage in gray.

Figure 8 Frost Damage on some *V. villosa* plants assessed on February 6, 2025 (Day 53).

4.3 Yield per Hectare at harvesting in April 29, 2025

Species	Gross Yield (kg fresh weight/ha)	Moisture (%)	Net Yield (kg dry matter/ha)
Vicia Villosa	49,640	87.04%	6,428
Vicia Sativa	42,000	86.18%	5,818

The yield results demonstrate clearly favorable performance of *V. villosa*, which achieved a net yield of 6,428 kg dry matter ha^{-1} (Figure 9, 10), surpassing the *V. sativa*, which recorded 5,818 kg dry matter ha^{-1} (Figure 11, 12). This outcome is particularly noteworthy, given that during the early stages of the trial, especially during germination and initial establishment, the *V. villosa* exhibited greater vigor, more uniform emergence, and apparent resistance to early stress, traits that initially suggested superior productivity (Figure 13).

However, as the growing period progressed and precipitation events increased, the *V. villosa* species showed a strong adaptive response, experiencing steady growth and demonstrating a greater capacity for water use under rainfed conditions. This behavior suggests that the *V. villosa* species possesses good phenotypic plasticity and water use efficiency, which were key to achieving these high yields under limited water availability. These characteristics position it as a promising option for extensive dryland production systems.

Figure 9 *Vicia villosa* growth at day 105 of the project 26 of March 2025 (Day 101).

Figure 10 *Vicia villosa* at harvest on April 29 (Day 135).

Figure 11 *Vicia sativa* growth at day 105 of the project 26 of March 2025 (Day 101).

Figure 12 *Vicia sativa* harvest on April 29 (Day 135).

Figure 13 Harvest of *Vicia sativa* on the right and *Vicia villosa* on the left on April 29 (Day 135).

5. Conclusion

During the winter evaluation period, both species exhibited marked differences in terms of germination, frost resistance, and yield. The main findings were as follows:

- **Germination:** *V. sativa* showed a higher germination percentage (77%) than *V. villosa* (70%), suggesting better initial adaptability to late sowing in December under cold conditions.
- **Frost Resistance:** *V. sativa* exhibited greater tolerance, with an average partial damage rate of 5.5%, in contrast to *V. villosa*, which suffered more severe frost damage with 38.75% partial damage.
- **Yield:** In terms of production, *V. villosa* outperformed in net yield in dry matter per hectare, reaching 6,248 kg dry matter ha^{-1}, whereas *V. sativa* yielded 5,818 kg dry matter ha^{-1}.

Recommendation: Although *Vicia villosa* showed lower germination and greater susceptibility to frost damage compared to *Vicia sativa*, its final yield (6,248 248 kg dry matter ha^{-1}) was the highest among the two species evaluated. These results point to a potentially high productive capacity under rainfed conditions, despite its sensitivity to early cold stress. Further trials could help identify management strategies—such as early sowing—that enhance establishment and consistency across seasons, thereby maximizing their high yield potential.

In Madrid, 27 May 2025

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