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Pilowred®, a Nematode-resistant and Low-vigor Rootstock for *Prunus*

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The demands of the fruit sector in the Mediterranean Basin have shifted in recent years, largely as a result of edaphoclimatic limitations resulting from climate change. These limitations include water scarcity during extreme drought periods, warmer cold seasons, the prevalence of soil diseases such as those related to the presence of root-knot nematodes (RKNs), and iron chlorosis. To address these issues, the *Prunus* spp. rootstock breeding program at the Agrifood Research and Technology Centre of Aragon (CITA) developed red-leaf hybrids from the 'Garfi' × 'Nemared' series (G × N): Garnem®, Felinem®, and Monegro® (Felipe 2009). In addition, CITA selected the plum 'Montizo' as a clonal rootstock, which adapts to wet and heavy soils, reducing tree size compared with other rootstocks (Felipe et al. 1997). In recent years, the adoption of more intensified and technologically advanced cultivation systems has increased the need for low-vigor rootstocks to facilitate sustainable orchard management. In response to these demands, Pilowred® has been released as a valuable rootstock for several stone fruit species, particularly almond. One of its more notable features is its low vigor, which enhances the productivity of the grafted cultivar, leading to greater yields in the orchard. In addition, it exhibits easy vegetative propagation and good graft compatibility. It requires a shorter winter dormancy period, facilitating a rapid entry into production, and it shows greater water use efficiency compared with other G × N hybrids.

Origin

This rootstock is a clone selected from the progeny obtained by crossing the Spanish almond 'Garfi' {*Prunus amygdalus* [syn. *Prunus dulcis* (Mill.) D.A. Webb]} as the female parent with the North American peach 'Nemared' [*Prunus persica* (L.) Batsch] as the pollen donor, which was developed at CITA. The 'Garfi' almond was selected previously for its favorable morphological characteristics, suitability for clonal propagation, and good adaptation to drought stress (Felipe 1989, 2009; Felipe et al. 1995). 'Nemared' was chosen primarily as a source for RKN resistance and for its adaptation to irrigated conditions (Ramming and Tanner 1983).

Description

Unbudded Pilowred® trees exhibit moderate vigor, distinct from other hybrids derived from 'Garfi' and 'Nemared', such as Garnem® (Fig. 1). In the nursery, ungrafted plants display upright growth, with little to no feathering during the first year, and a low propensity to produce root sprouts (Fig. 2). In almond cultivars grafted onto Pilowred®, shorter internodes are observed compared with those grafted onto more vigorous rootstocks such as Garnem® and Rootpac® 40, resulting in fewer sylleptic branches, similar to what is seen with low-vigor rootstocks such as Rootpac® 20 (Fig. 3) (Montesinos et al. 2021, 2022). Regarding the vigor conferred to the grafted cultivar, it has been confirmed that Pilowred® produces a smaller trunk cross-sectional area (TCSA) in the grafted cultivar compared with more vigorous rootstocks such as Garnem® (Fig. 4) and Rootpac® R. These effects of Pilowred® are comparable to those of other low-vigor rootstocks such as Rootpac® 20 (Figs. 5 and 6B). When analyzing TCSA values based on the grafted cultivar, it was observed that, as with Soleta® and 'Lauranne', there were no significant differences among different rootstocks. However, in the case of Isabelona®, Vialfas®, Mardía®, and 'Guara', significant differences were found

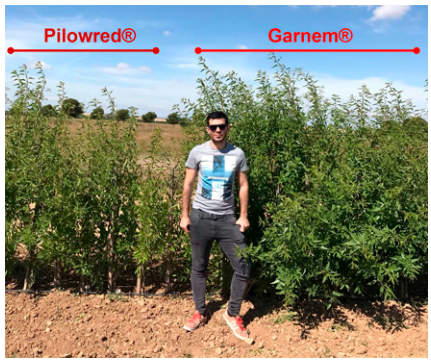

Fig. 1. Vigor differences in 2-year-old Mardía® grafted onto Pilowred® (**left**) and Garnem® (**right**).

Fig. 2. Pilowred® plant without grafting in the nursery. Note the upright and sparsely branched growth of this rootstock.

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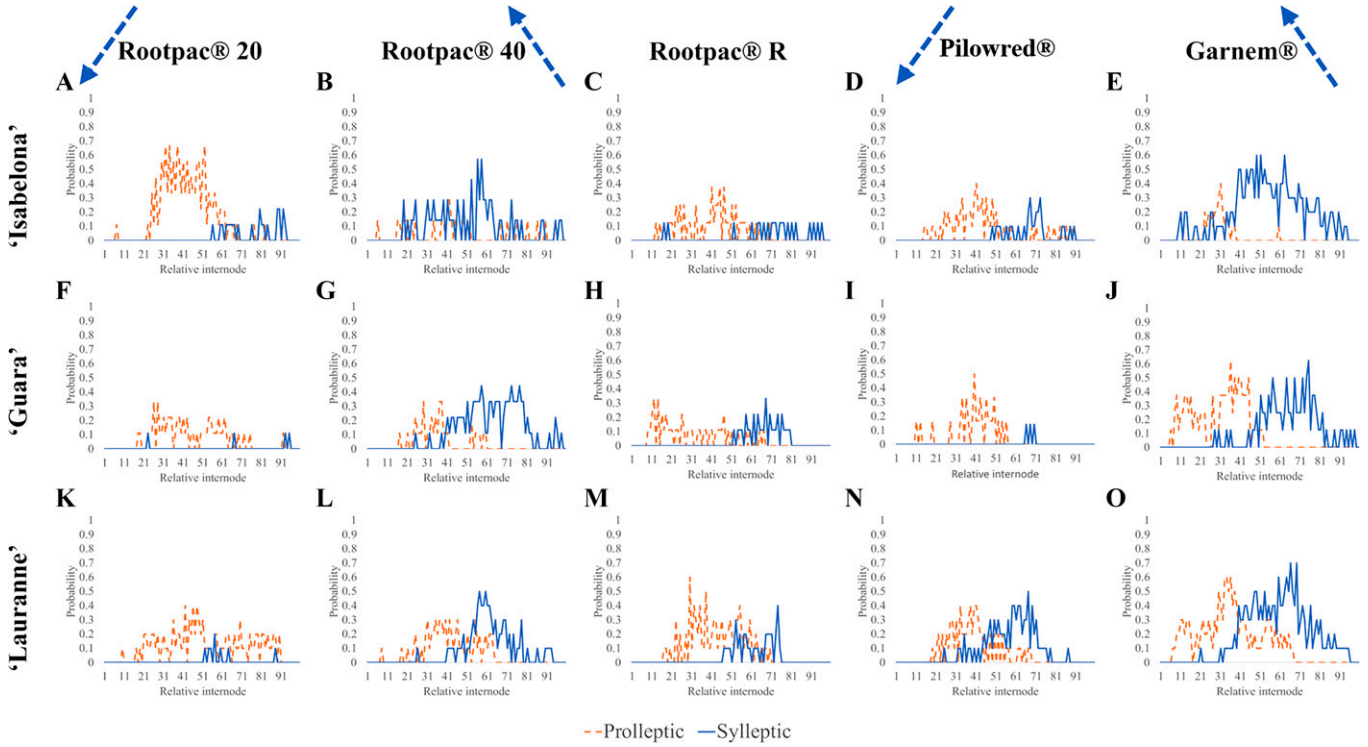

Fig. 3. Probability of proleptic and sylleptic shoot occurrence according to node position along 2-year-old branches of 15 scion–rootstock combinations of almond trees. The probabilities of proleptic and sylleptic shoot formation at each node were converted to their relative position and represented in a branch with 100 internodes. Internode 0 refers to the base of the branch; internode 100 refers to the apex (Montesinos et al. 2022).

between low-vigor rootstocks (Pilowred® and Rootpac® 20) and high-vigor ones (Garnem® and Rootpac® R) (Fig. 6A). A recent study (Bielsa et al. 2022) presented the performance of Pilowred® across different planting systems and cultivars to evaluate its suitability for use in more technologically advanced systems, such as the superefficient system, as well as in more traditional planting frameworks.

The trunk bark has a rough texture with a bright glaucous green color and round lenticels measuring 0.2 mm in diameter, along with a density of five lenticels per square centimeter. Branches are green with red-purple areas due to anthocyanin pigmentation in the sun-exposed regions. Buds are alternately distributed along the branch, and internode length is medium to long. Young leaves are grayish purple (Fig. 7), whereas mature leaves turn green on the upper surface, but retain the grayish purple coloration on the lower surface. The leaves are large and elongated, with an intermediate morphology between almond and peach. Actively growing shoots display an intense red-purple color (Figs. 2 and 7). In the first year, shoots grow straight, with little to no lateral branching (Fig. 2).

The clone blooms earlier than Garnem®, showing similar low chilling requirements and earlier sprouting after grafting. The flowers are large and rosaceous in type, with pale-pink, rounded–elliptical petals measuring 20.3 mm in length and 16.0 mm in width. Each flower has ~46 stamens and one pistil, with filaments measuring 9.85 mm in length. The fruit are small (diameter, 2.4 cm; length, 2.9 cm), rounded, and covered with a pubescent epidermis. The fruit are green with reddish tones, and the mesocarp is thin and inedible, leaving a free stone. The morphology and performance of this clone can be distinguished from the three red-leaf hybrids by molecular markers.

Resistances and Susceptibilities

The Pilowred® clone was selected to address the need for a low-vigor rootstock with reduced chilling requirements and increased water use efficiency in response to global climate change. It also addresses significant limitations in Mediterranean environments, such as tolerance to iron chlorosis. Its level of tolerance to iron chlorosis is comparable to that of 'GF-677' (Felipe 1989). Pilowred® also performs well under drought conditions, exhibiting higher water use efficiency than the other three red-leaf hybrids Garnem®, Felinem®, and Monegro®, and better than 'GF-677' (Fig. 8) (Bielsa et al. 2018). Like all almond × peach rootstocks, Pilowred® is susceptible to root asphyxia caused by flooding; however, it adapts well if the soils are well drained.

In Spain, the presence of RKNs (*Meloidogyne* spp.) poses a limiting factor in *Prunus* production, particularly in replant situations (Pinochet 1997). Three main species primarily affect stone fruit in Mediterranean regions: *Meloidogyne arenaria*, *Meloidogyne incognita*, and *Meloidogyne javanica* (Esmenjaud et al. 2009). Pilowred® is resistant to *M. arenaria*, *M. incognita* (Fig. 9, Table 1), and *Meloidogyne ethiopica*, but is susceptible to *M. javanica* and *Meloidogyne floridiensis* (Esmenjaud 2021).

Propagation and Compatibility

Pilowred® propagates well by in vitro techniques and also via hardwood and herbaceous cuttings when the trees are well established. Cuttings are easily obtained as a result of the lower level of shoot branching. Nursery operations benefit from the minimal presence of feathers and the distinctive red leaves, coupled with the prolonged vegetative period of the plants. In addition, the early budbreak after dormancy enables quicker production of nursery plants. The bud take percentage is high for all almond cultivars, as well as peach, and nectarine (pers. Comm. with different nurserymen).

This rootstock has been primarily selected for almond and peach, and shows good graft compatibility with numerous almond, peach, and nectarine cultivars, as well as with some diploid plums (Japanese and similar).

Availability

Pilowred® has obtained an American patent (US PP 35468). Pilowred® was sent to examination in 2023 to the European Community Plant Variety Office with application no. 202301807 (Rubio-Cabetas and Felipe 2023).

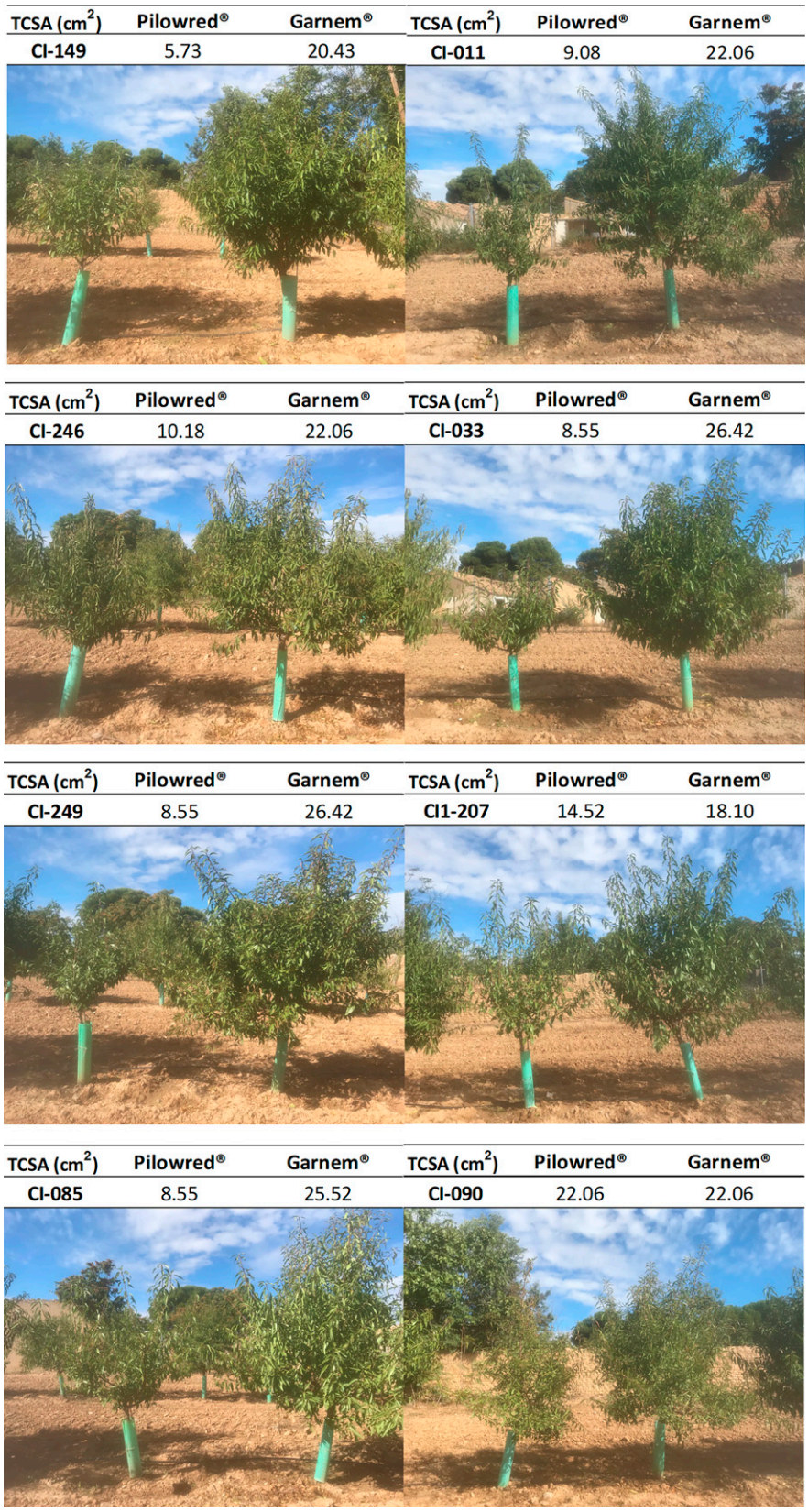

Fig. 4. Comparison of the vigor conferred by Pilowred® (left) and Garnem® (right) to different Agrifood Research and Technology Centre of Aragon (Zaragosa, Spain) almond selections of 3-year-old plants labeled with Cita codes (CI). TCSA = trunk cross-sectional area.

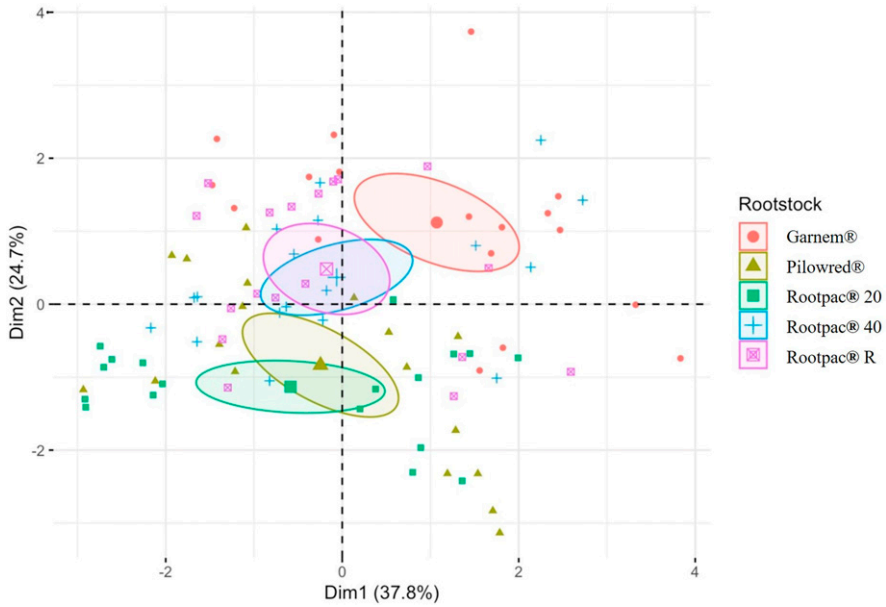

Fig. 5. Principal component analysis of 30 almond scion–rootstock combinations. The combinations were measured using seven descriptors of architecture and vigor: length of the central axis, average length of internodes, number of branches, number of long branches (> 20 cm), number of secondary branches, percentage of branches per internode, and distribution of branches on the trunk (Montesinos et al. 2021). Dim1 = dimension 1; Dim2 = dimension 2.

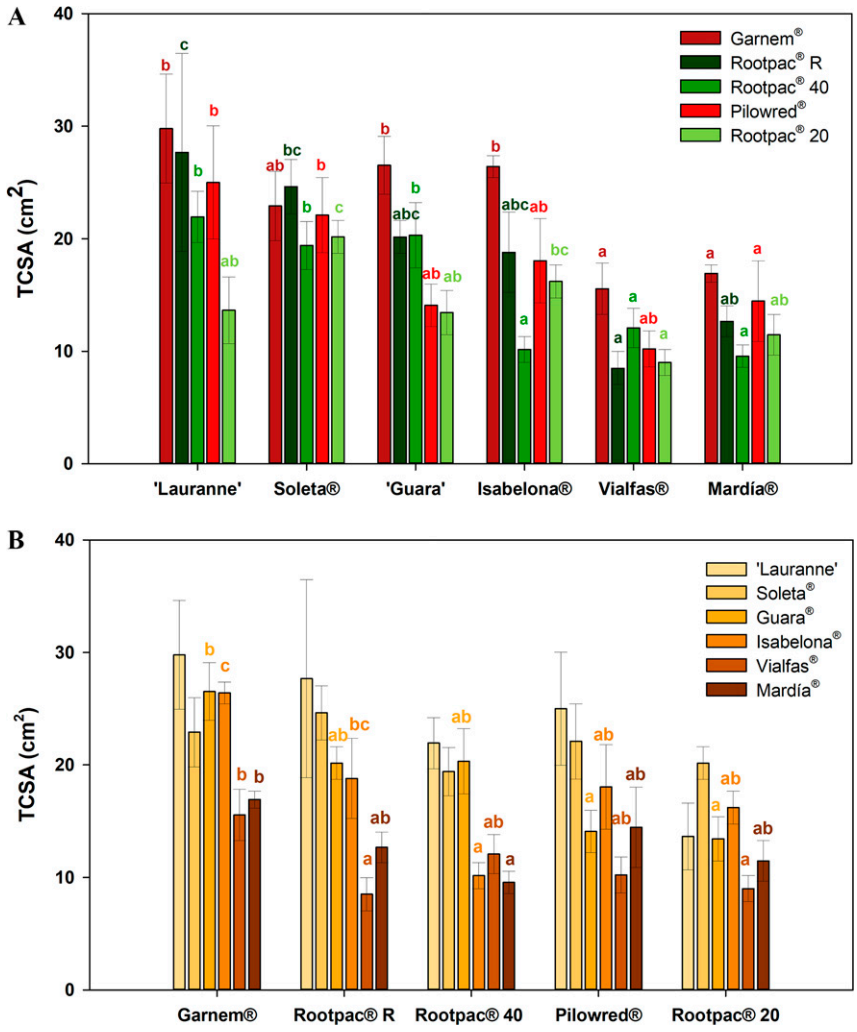

Fig. 6. Trunk cross-sectional area (TCSA) values in six commercial almond cultivars grafted onto five rootstocks with varying vigor in 2-year-old plants (Bielsa et al. 2023). (**A**) Differences among rootstocks within each cultivar. (**B**) Differences among cultivars for each rootstock. Bars with lowercase letters indicate significant differences ($P \leq 0.05$) according to Tukey's multiple range test after a one-way analysis of variance. Error bars represent the standard error of the mean.

Fig. 7. Pilowred® in the nursery. Detail of its characteristic red leaf is typical of the 'Garfi' × 'Nemared' series.

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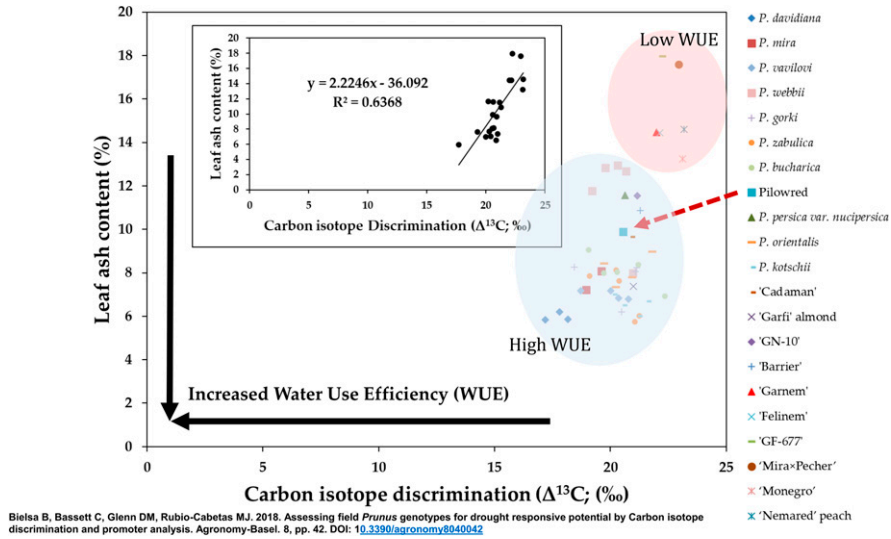

Fig. 8. Water Use Efficiency estimation through the analysis of carbon isotope discrimination ($\Delta^{13}C$) in several *Prunus* spp., including almond wild-relative species and commercial rootstocks (Bielsa et al. 2018).

Fig. 9. Root-knot nematode inoculation in roots. Resistant phenotypes of Pilowred® (**right**) and Garnem® (**left**) to *Meloidogyne incognita*.

Table 1. Evaluation of resistance using a gall index rating from 0 to 5 points for the accessions Pilowred® and Garnem® toward the root-knot nematode species *Meloidogyne arenaria, Meloidogyne incognita, Meloidogyne javanica*, and *Meloidogyne floridensis*.

	Gall index rating (pt)			
Rootstock	*M. arenaria*	*M. incognita*	*M. javanica*	*M. floridensis*
Pilowred®	0	0	2.5	4.6
Garnem®	0	0	2.2	4.5

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