Annals of Silvicultural Research
49(1), 2024: 39-43
https://creajournals.crea.gov.it/asr/

Technical note

Latest results of the breeding activity of hybrid poplars for industrial purposes in Italy

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Received: 1/12/2023 *Accepted: 15/05/2024* *Published online: 17/05/2024*

ABSTRACT In recent years, the Italian breeding activities in the sector of poplar cultivation for industrial purposes have led to the selection of new genotypes with high production potential, resistant to woolly aphid and to the main poplar diseases such as Venturia leaf and shoot blight, *Melampsora* rusts, and *Marssonina* leaf spots. These genotypes, known as "Greater Environmental Sustainability" clones (MSA clones), have shown promising growth and yield outcomes. This note focuses on the performance in growth and yield of a new P. ×*canadensis* genotype, obtained from breeding activity started in the 1990s. The 'Tango' clone shows very high production potential, in many cases higher than older clones used as controls. In particular, it shows faster growth after the 4th or 5th year of cultivation, resulting in higher volumes achieved by the end of the rotation period. In experimental trials, the average annual volume increment of the first 5 stem meters (representing the best wood assortments) ranged between 12.8 and 16.8 m^3 ha^{-1} y^{-1}. These remarkable yields, combined with the resistance to main diseases, make it an excellent candidate for highly sustainable poplar cultivation.

KEYWORDS: 'Tango' clone, growth, yield, wood.

Introduction

In recent years, Italian breeding activity in the sector of poplar cultivation for industrial purposes has selected numerous new genotypes with high production potential, resistant to woolly aphid [*Phloeomyzus passerinii* (Sign.)] and to the main poplar diseases such as Venturia leaf and shoot blight [*Venturia populina* (Vuill.) Fabric.], rusts (*Melampsora* spp.) and Marssonina leaf spots [*Drepanopeziza brunnea* (Ellis & Everh.) Rossman & W.C. Allen] (Gennaro and Giorcelli 2019). Some of the new genotypes, called "cloni a Maggiore Sostenibilità Ambientale" (MSA clones) which means "Greater Environmental Sustainability clones", particularly the *Populus* ×*canadensis* Moench clones 'Tucano', 'Diva' and 'Aleramo', have aroused great interest among nurserymen in Italy and abroad, and are today among the most cultivated poplar genotypes in Europe (Berlin et al. 2023).

Due to their resistance characters and their fast growth, they have been introduced in the recent Italian Rural Development Plans and in forest certification programs like PEFC (Programme for the Endorsement of Forests Certification) and FSC (Forest Stewardship Council) (Vietto et al. 2011, Cadman 2011). The opportunity to cultivate poplar for industrial purposes while minimizing chemical treatments and enhancing yield has drawn significant interest to these clones. They offer increased efficiency in CO_2 absorption, the opportunity for cultivation near populated areas and in environmentally sensitive regions, and suitability for integration into agroforestry systems.

The long-term breeding program of the CREA Research Center for Forestry and Wood is based on the controlled pollination of selected parent trees to generate periodically new hybrid generations (mainly *P.* ×*canadensis*). These generations undergo rigorous selection tests lasting up to 20 years before certain promising clones are proposed for registration in the National Register of Basic Materials (RNMB) and subsequently made available for commercialization. The long duration required for selection is due to two primary factors: *i)* the physiology of poplars and their method of vegetative reproduction, which begins with the original individual (*Ortet*) and gradually produces more and more individuals by cuttings (*Ramet*), and *ii)* the need to assess all the characteristics of new genotypes in mature specimens, typically around 8-10 years of age.

This note presents the growth and yield results of a new *P.* ×*canadensis* genotype derived from breeding activities started in the 1990s. The clone 'Tango' (Fig. 1) originated from a controlled cross conducted in 1994 at the "Mezzi" farm of the CREA Research Centre for Forestry and Wood in Casale Monferrato, Italy. This cross involved the female parent *P. deltoides* Bartr. 'D0-006' from Texas, USA, and the male parent *P. nigra* L. 'N151' from Baiso, Italy. After the initial genotype family evaluation phase, it was experimentally determined that 'Tango' had all the desirable characteristics suitable for commercialization: its growth performances and resistance to diseases were compared with those of poplar clones previously selected and used as controls. Shown here are the results of over 10 years of experimental testing.

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http://dx.doi.org/10.12899/asr-2532

Figure 1 - The 'Tango' clone in the Cavarzere trial plot.

Methods

The comparison tests of the new genotype with some *P.* ×*canadensis* old genotypes widely used ('BL Costanzo', 'I-214', 'Neva') were carried out in three trials grown in three Po valley sites (Italy) suitable for poplar cultivation but with partially different soil and climate characteristics: Cavarzere, Valenza and Gabiano. Table 1 describes the main characteristics of sites.

Table 1 - Characteristics of the trials for the evaluation of the new hybrid poplar clone.

Site	Geographic coordinates	Plantation year	Age	Soil texture
Cavarzere	45°07'21"N, 12°03'17"E	2007	8	Sandy-loam
Valenza	45°00'20"N, 8°35'37"E	2008	7	Sandy
Gabiano	45°10'12"N, 8°10'42.0"E	2008	12	Silty-clay

The trials were carried out following the conventional Italian model of wood production for industrial purposes in poplar stands (Tedeschi et al. 2005, Berlin et al. 2023). The soil was ploughed to a depth of 30 cm and then harrowed. Chemical weed control was applied during the planting phase. Two-year-old poplar poles without roots and branches were rehydrated in water for three days and manually planted in holes opened with a drill, about 80-100 cm deep, at a spacing of 6 × 6 m, corresponding to a planting density of 278 trees ha^{-1}. Cultural treatments consisted of NPK fertilization at establishment, emergency irrigation and annual winter pruning during the first five years of growth. No treatments were applied against diseases, while insecticide treatments were applied only as needed for xylophagous insects [*Chryptorhynchus lapathi* (L.)] and defoliant insects [*Hyphantria cunea* (Drury)]. All the trials were designed according to randomized complete blocks, with three or more replications of plots including 9 trees (area of 324 m^2).

Stem diameter at breast height (Dbh) and survival were assessed annually on all trees, while stem diameters at 5 m of height were measured during the last rotation year. The volume per hectare of the first five meters of stem (Vol5, which represents the most valuable wood assortment) was estimated for each tree using the equation for the volume of a truncated cone:

$$Vol5 = 5/3\pi (R^2 + rR + r^2) \ [m^3] \quad \text{(eq. 1)}$$

where R is the radius of the major base and r is the radius of the minor base (in m^2).

The ANOVA test was applied on the survival and volume data using R statistical software (R Core Team, 2021. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria). In case of statistical significance, the Duncan test was applied as a *post-hoc* test.

In the presence of expressed symptoms, the incidence of the main biotic adversities [trunk "brown spots", *Marssonina* leaf spots, *Venturia* leaf and shoot blight, *Melampsora* rusts, Poplar Mosaic Virus (PopMV), poplar woolly aphid] was assessed according to the methods reported in the DD 316/2019 (MIPAAFT 2019). Also stem shape characteristics and physical characteristics of the wood were also assessed according to the methods reported in the DD 316/2019.

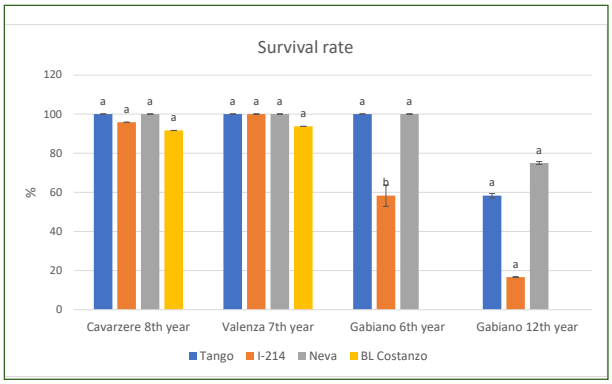
Results

Survival and growth

The last measurements in Cavarzere were carried out at the end of the 8th year of growth. With a 100% survival rate (Fig. 2) and an average stem volume of the first 5 meters of 0.368 m^3, the 'Tango' clone attained an average of 102 m^3 ha^{-1} of assortment suitable for plywood production (Fig. 3). This achievement showed a statistically significant difference compared to the performances of older clones.

Figure 4 shows the growth curves of stem circumference at breast heigh. In this trial, the clones 'Tango' and 'Neva' showed similar growth patterns; however, 'Tango' surpassed 'Neva' during the fourth year.

Figure 2 - Survival rate (expressed as a percentage of the number of trees planted) of the clones compared (for Gabiano test, the data are taken before and after the storm and flood events).

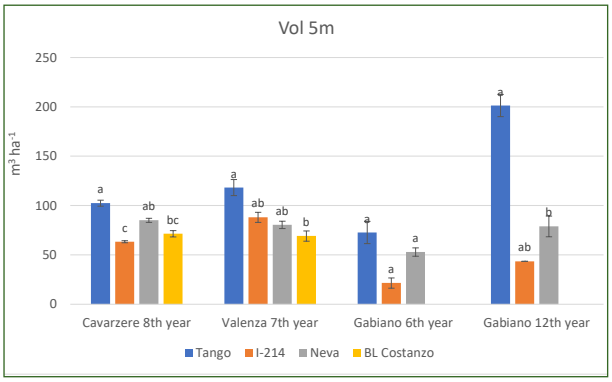


The last measurements in Valenza were carried out at the end of the 7th year of growth. With 100% trees survival rate (Fig. 2) and an average stem volume of first 5

meters of 0.424 m³, the clone 'Tango' achieved an average of 118 $m^3\ ha^{-1}$ of assortment suitable for plywood production (Fig. 3).

Figure 5 shows the growth curves of stem circumference at breast height. In this trial, the 'Tango' clone showed a growth capacity superior to other clones from the first years, and in particular after the fifth year.

Figure 3 - Wood yield of first 5 m of stem of the clones compared (for Gabiano test, the data are taken before and after the storm and flood events).



The last measurements in Gabiano were carried out at the end of the 12th year of growth. During this trial, a violent storm in 2011 and a flooding event in 2016 destroyed many trees; for several years it was impossible to reach the trial site for measurements. Figure 2 shows the average survival rate of clones before (6th year) and after (12th year) the flooding and storm events. Several factors may have influenced the survival of the trees, including clone resistance and robustness, as well as the position of the experimental plots in relation to the flood wave. In the Figure 3 it is possible to see that the clone 'Tango' had a higher average stem volume even before the catastrophic events. The events destroyed about 40% of 'Tango' trees but stem growth was very high. The final production highlighted a very high difference between 'Tango' and the other two clones.

Figure 4 - Growth curves of stem girth at breast height of the clones compared in the Cavarzere trial plot.

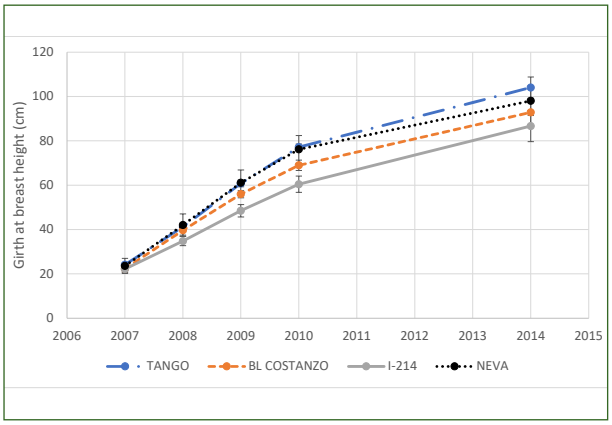
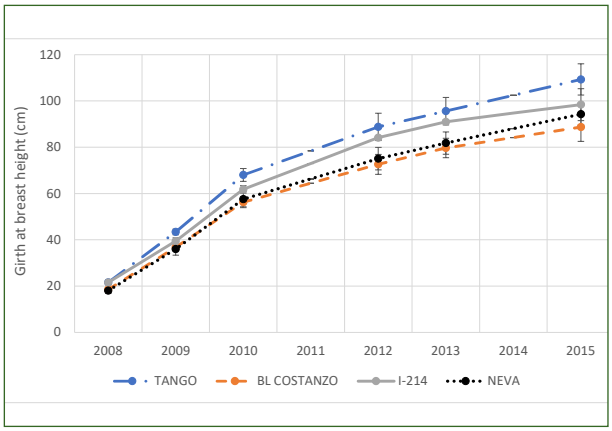


Figure 5 - Growth curves of stem girth at breast height of the clones compared in the Valenza trial plot.



Resistance to biotic adversities

In the last ten years, despite several observations in experimental plantations located in the Po Valley, the hydrophysiological disorder in the trunk known as "brown spots" has never exhibited its typical symptoms of bark necrotic lesions in 'Tango'. Therefore, 'Tango' shows greater resistance compared to 'I-214', which serves as the control clone for this disease (DD 316/2019, Appendix B, Annex IV).

As regards Marssonina leaf spots, surveys conducted over the past decade on leaf disc samples from both nursery and adult trees, along with observations of defoliation levels in experimental plantations located in Po Valley, consistently indicate that 'Tango' exhibits resistance to the disease when compared with 'I-214' (listed among control clones). Foliar brown spots or phylloptosis have never been observed. In the following years, 'Tango' has always demonstrated this behavior. However, in a context of increasingly rare Marssonina attacks, the particular drought conditions experienced in 2022 further limited their occurrence, confirming the resistance of the clone.

Surveys conducted over the last twenty years in experimental plantations in Casale Monferrato allowed the annual observation of potential symptoms of Venturia leaf and shoot blight. Disease symptoms were only detected in 2001, with 'Tango' showing a minimal 0.83% of affected shoots, compared to the highly susceptible 'Boccalari' clone (32.9%), designated as the control clone, and 'Neva' clone (40.7%). This behavior is not significantly different from that of 'I-214', the clone control for resistance, which showed 0% affected shoots. Subsequently, the attacks were detected annually, but with zero or almost zero results concerning *Venturia*. It is important to note that due to the gradual disappearance of plantations historically composed of more susceptible clones and the unfavorable climate conditions for such disease in recent decades, Venturia leaf and shoot blight has significantly declined. Currently, its general incidence in poplar cultivation areas is almost absent.

As regards Melampsora rusts, the surveys on R1S1 trees (one year old stem with one year old root) carried out in

late summer mainly concerned *M. larici-populina* Kleb., predominant in northern Italy and in the rest of Europe, both from nursery infections and from laboratory inoculations. The intensity of the attack, measured according to the protocol developed by INRA-FCBA and INBO, ranged between 0.13% (degree 6 from DD 316/2019, table in Appendix B, Annex IV) in 1999 and 20.7% (degree 5 from the cited table) in 2011. These percentages were consistently much lower than those observed on clone 'I-214', which serves as the control. Following laboratory investigations, 'Tango' did not present uredinia of *M. larici-populina* E3 race and presented 0.80% of foliar affected area (degree 6 from the cited table) by E4 race. Therefore, we can say that 'Tango' is more resistant than control clone 'I-214', which is relatively resistant to *Melampsora* spp.; these data have been consistently confirmed over the last 5 years through repeated observations in the nursery.

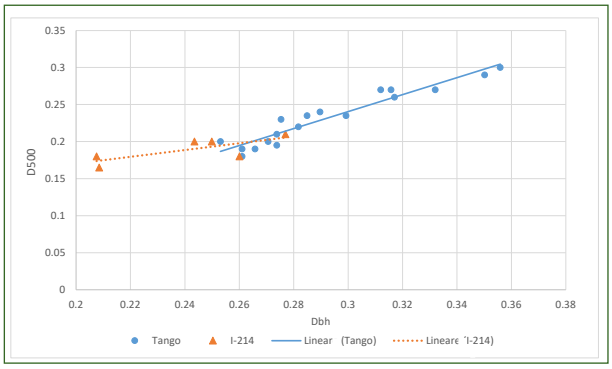
Based on observations spanning two decades in the laboratory according to the method outlined by Arru (1974), 'Tango' has demonstrated high resistance to woolly aphid infestations. In terms of average colonization of the cuttings, compared to those of 'I-214' (designed as the control clone declared in DD 316/2019, Appendix B, Annex IV, highly susceptible and considered as equal to 1, as reference, after 21 days), 'Tango' exhibited values ranging from 0.13 in the year 2000, during the winter on quiescent cutting, to values even lower than 0.07 in the year 2021 on active cuttings during the summer, further substantiating its resistance.

Considering susceptibility to PopMV, the 'Tango' clone, being a hybrid genotype of *P. ×canadensis*, showed no symptoms (class 0, according to the scale declared in DD 316/2019, Appendix B, Annex IV) during the observations carried out on R2S1 (trees with two-years-old roots and one-year-old stem) and R3S1 (trees with three-years-old roots and one-year-old stem). However, it should be noted that PopMV primarily affects North American native genotypes of *P. deltoides*, in particular those of "Carolinian" morphology, such as 'Lux', which is designated as the control clone.

Physical characteristics of wood

The 'Tango' clone has a rather regular shape of the stem without basal buttresses or extreme curvatures; compared to the 'I-214' it has a reduced curvature index (1.305 for the 'Tango' versus 2.022 of the 'I-214'). Compared to older clones such as 'I-214' and 'Neva', it maintains a greater cylindricity of the stem in height, allowing to obtain a greater volume of workable wood from the first 5 m of stem. In Figure 6 the ratio between diameter at breast height (Dbh) and diameter at 5 m height of the stems measured in the Casale Monferrato trial is shown; even if it was not possible to compare many plants of the same size for the two clones, observing the interpolations of the data it is possible to see that with the same Dbh values the 'Tango' clone is able to maintain a greater diameter measured at 5 m of height, compared to 'I -214', increasing the production of peeling logs.

Figure 6 - Relationship between stem diameter at breast height (Dbh, in m) and stem diameter at 5 m height (D500, in m) of clone 'Tango' compared to clone 'I-214'.



The new clone has a rather high basal density (395 kg m^3) and volumetric density (680 kg m^3).

Discussion

The new *P. ×canadensis* clone 'Tango' has been selected for its fast growth, disease resistance and high-quality wood. The experimental tests were conducted in three sites suitable for poplar cultivation, each with distinct pedoclimatic characteristics. The 'Tango' clone showed remarkable production potential, exceeding that of the control clones: even if starting at plantation with similar stem size as the other clones, it subsequently tends to show faster growth, particularly after the 4th or 5th year, resulting in larger stem diameters at the end of the rotation. Furthermore, in the trial conducted in Gabiano, 'Tango' showed greater tolerance to catastrophic events such as flood and storm.

In terms of biotic adversities, the 'Tango' clone has been found to have a higher resistance compared to the control clones against all tested threats. Apparently immune to the attacks of "brown spots", PopMV and *Marssonina* leaf spots, it has shown to possess the same resistance to *Venturia* as the 'I-214' clone, and even higher resistance to woolly aphids (almost immune) compared to the same clone. Limited attacks by *Melampsora* rusts were also observed, which should not raise concerns since they do not impact wood production and may even prevent the emergence of new pathotypes, which is more likely in the presence of fully resistant hosts. Thus, 'Tango' is well-suited for highly sustainable poplar cultivation.

Conclusion

The new *P. ×canadensis* clone 'Tango' (Fig. 1) achieved registration with the RNMB in March 2024, making it commercially available for planting in pure or polyclonal commercial poplar stands to sustainably produce high quality wood.

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