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Employment of radiofrequency technology (RFId) in grapevine nursery traceability

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Abstract: This trial represents the first experience regarding the insertion of microchips (TAG) based on radiofrequency identification (RFId) technology into grapevine plants for nursery purposes. The research was performed considering five economically-important grapevine clones, involving 2500 plants. Microchips were inserted directly into medulla tissue during grafting procedures. Two methods were developed, one based on medulla perforation and the other characterized by a lateral cut on rootstock performed by a specific machine. The presence of electronic tagging (TAG) inside plants did not compromise growth start-up, vegetative growth, sanitary condition, histological status of graft point, or quality of plants, considering the first year of farming. In addition to rapid identification, the TAGs permit the access to a specific online database, in which all plant information is contained, in a particular grapevine certification datasheet with sanitarian status (2005/43/CE). Results obtained suggest RFId technology as a useful tool for replacing traditional labelling and providing an up-to-date traceability system for grapevine nurseries.

1. Introduction

As is well known, in recent years the need for knowledge about the origins and qualitative characteristics of food products commercialized worldwide has increased due to user demands which are more oriented toward safe and wholesome products, supported by stricter regulations for better public health. The chance to acquire information about the "history" of a product, from its origin to the shelf, is known as traceability. In agronomics traceability goes back to the genetic status of products, animal or vegetal. Controls involve different sectors of the production line, using different tools.

With regard to the grapevine nursery, the importance of traceability involves qualitative and sanitary issues. Regulations require that plants produced by nurseries are followed by specific datasheets, reported on coloured labels, regarding type and origins ("base", "certificate" or "standard" materials). In fact some advantages connected with the use of powerful and safe traceability systems in the grapevine sector are linked to legal documentation (68/193/CEE) concerning "certificate" grapevine production which, as is well known, assures a higher genetic-sanitary quality available for marketable products.

The use of traditional labels has represented, till now, the only tool available for commercial grapevine identification. This system, while it is simple and cheap, is not without drawbacks as over time colour and inscriptions fade, are lost or altered and the space for recording information is limited. Moreover, labels are not placed on every single plant, but on groups of grapevines joined by laces. These problems, together with the increasing need for traceability tools, have suggested the development of novel methods of identification, able to supply a wide and complete information package for every plant, with continuous data update. Interesting applications were developed recently for various tree species, using radiofrequency technology (RFId) (Bowman, 2005; Grieco *et al*., 2006), whose potential was described in animal identification systems (Artmann, 1999; Jansen and Eradus, 1999)

Thus, the application of this technology has been studied in the grapevine nursery sector. This paper reports the first results regarding the insertion of RFId TAGs inside this species. Moreover, a specially developed online database was created to match TAGs to information datasheets for users to manage marked plants.

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2. Materials and Methods

The experimental trials reported were conducted from 2007 to 2008 in open-field conditions in a nursery specialized in the production of grafted cuttings.

Plant material

Trials involved grafted cuttings of five grapevine clones used widely in Italy ('Sangiovese I-SS-F9-A5-48', 'Prugnolo gentile I-Bruscello', 'Colorino I-US-FI-PI-10', 'Trebbiano toscano I-S.Lucia 12', 'Vernaccia di S. Gimignano I-VP6'), grafted on one rootstock ('1103P'), supplied by Associazione Toscana Costitutori Viticoli (TOS.CO.VIT., San Piero a Grado, Pisa, Italy, www.toscovit.it).

Electronic material

Transponder glass TAG RFId were used, 2.1 mm diameter and 12 mm length, working at the frequency of 125 KHz (InterMedia Sas, Forlì, Italy, www.rfid360.net) (Fig. 1). TAGs were read electronically by 14-length identification number, and the reading was performed by a Card Flash reader able to identify the microchips from a distance of 20 cm. Data recovery was performed by a palm-PC (Dell Axim X51) on which a database software specifically programmed for storing the data of each plant was installed (Fig. 2).

Methods of implantation

TAGs were inserted inside medulla of rootstocks just after grafting, following two different specifically designed procedures, submitted for patent registration.

In the first procedure, TAGs are inserted following removal of a portion of medulla by drilling (Figs. 3, 4).

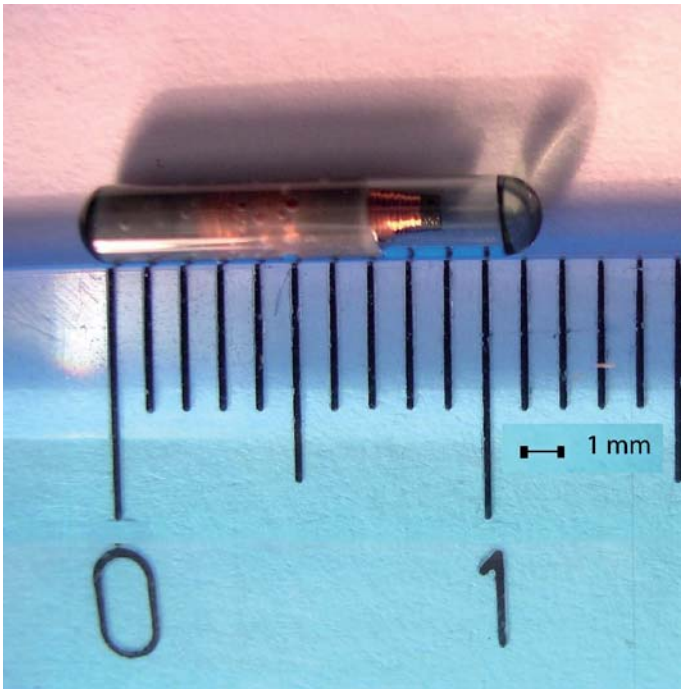

Fig. 1 - TAG inserted within plant.

Fig. 2 - Palm-PC with reader for TAG identification.

The second procedure consists of inserting the TAG laterally in the rootstock by a "door" created by a specially designed machine (Figs. 5, 6).

For each procedure, the average time (s) for TAG insertion was registered.

Fig. 3 - Drilling of medulla within rootstock.

Fig. 4 - Insertion of TAG within rootstock.

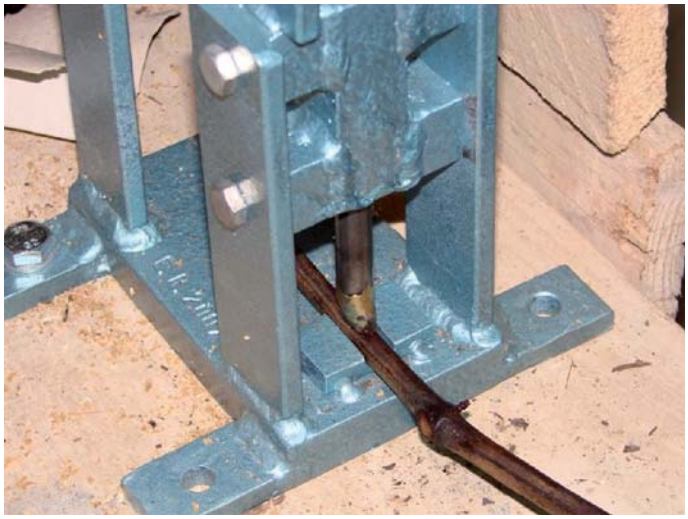

Fig. 5 - Lateral cutting by specially designed machine.

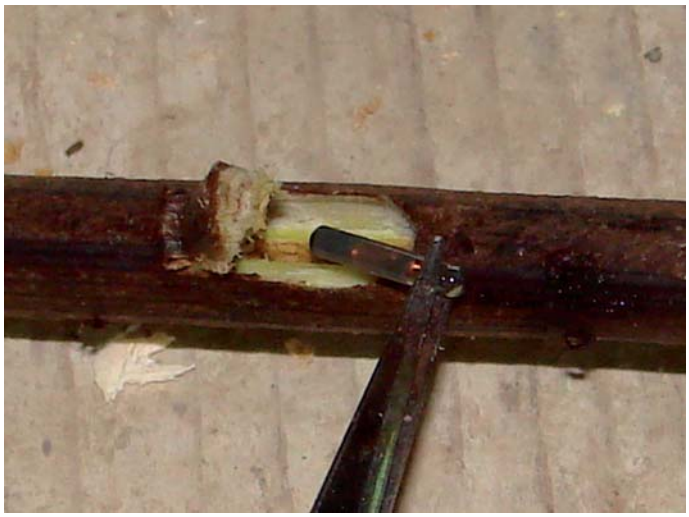

Fig. 6 - TAG insertion throughout "door" in rootstock.

Plants assay

Mortality, plant growth, sanitary status, histological status of graft point and quality of plants were evaluated after one year of farming.

Mortality was evaluated counting dead plants after planting in nursery (May 2007), and after planting in vineyard (June 2008).

For plant growth, shoots length (cm) were measured after 45 days from planting. After last winter polling (January 2008), shoots were measured as fresh weight.

Plants were constantly monitored for onset of pathological symptoms. Histological observations were performed on the trunk section cut at the graft point at the end of the growing period.

Plant quality was evaluated as the number of "first choice" plants produced after removal from nursery. Classification of plants into the "first choice" category was performed by a nursery specialist, as commonly done.

Experimental design

Trials involved 2500 grafted cuttings, with 500 plants for each clone. Growth data were analyzed by analysis of variance in a randomized design, and differences were assessed by Duncan's Multiple Range test at the 5% level.

Online database

The system aimed to permit access to a datasheet by users involved in grapevine production - from grapevine constitutor to farmer - using the identification codes linked to TAGs. Datasheets and their access had to be designed considering privacy policy, with regards to each type of user. The online database for managing marked plants was developed by InterMedia Sas.

3. Results and Discussion

Tag implantation

The principal objective of this first part of the research was to evaluate the possibility of inserting TAGs based on RFId technology within grapevine plants without damaging tissues or compromising growth. To achieve these targets, two different procedures were tested, with the following results.

Considering operational issues, the first procedure based on drilling seems simpler than the second procedure, using common, inexpensive tools. Even the working time seems faster: 16±1 s versus 18±1 s, respectively.

Moreover, considering that TAG insertion was performed just after grafting, drilling regards only the medulla, without damage to other tissues as tissue reforms over that grafting point. In contrast in the second procedure, additional tissue areas are interested by cicatrisation, due to the lateral cut performed by drift. Moreover, an extensive wound area is generated, increasing exposure to external agents, even if the surface of wounded tissues is covered by mastic immediately after grafting.

Mortality, vegetative activity, sanitary condition, histological observations and quality of plants

With regard to mortality, most differences between marking and unmarking were noted after nursery planting in 2007, with losses comparable to level of death of grafted cuttings generally reported in nursery farming (mean data of whole production were 13.0% for medulla drilling, 19.2% for lateral cutting, 13.2% for control). No significant losses occurred after vineyard planting in 2008 (mean data of whole production were 6.7% for medulla drilling, 10.34% for lateral cutting, 7.0% for control). Only in several lateral cutting groups (i.e. Colorino and Vernaccia 2007, Vernaccia 2008) was the mortality slightly higher in modified plants compared to control.

Considering all clones, growth of shoots did not show significant differences in marked plants compared to control (Table 1). Generally, the two investigated procedures, did not have significant impact on plant growth after nursery planting or after polling: moreover, marked plants did not differ from control plants with regard to fresh weight of vine-branches collected after last winter polling (data not shown).

Sanitary differences were also considered during the monitoring, with no symptomatic conditions noted considering all plants. Moreover, histological observations of the graft point in plants subjected to drilling of medulla did not show any alteration of tissues, with damage of medulla only. In fact, even primary xylem was always untouched and the few xylem vessels damaged did not outnumber the control. Lateral cutting caused, as predictable, a necrotic area in proximity to cuts (the "door" area), but this damage was always covered by formation of new tissues produced by the plant during the first year of growth, recovering the normal vascular activity and, presumably, limiting damage in terms of growth. An indicative comparison of graft point section can be observed in figure 7.

Finally, the quality of plants produced did not differ in modified plants compared to control, with a "first choice" production of more than 70% compared whole plants production, similar to control production.

Considering these results, the insertion of TAGs does not cause significant damage to plants during the first year, in terms of growth, mortality and health status.

Online database

The database was set-up following the scheme reported (Fig. 8). As shown, TAGs can be useful for identifying plants (genetic and certification type), and users involved in production and farming can add information to the datasheet regarding specific areas of action (i.e. certification type and date, sell date, farming details).

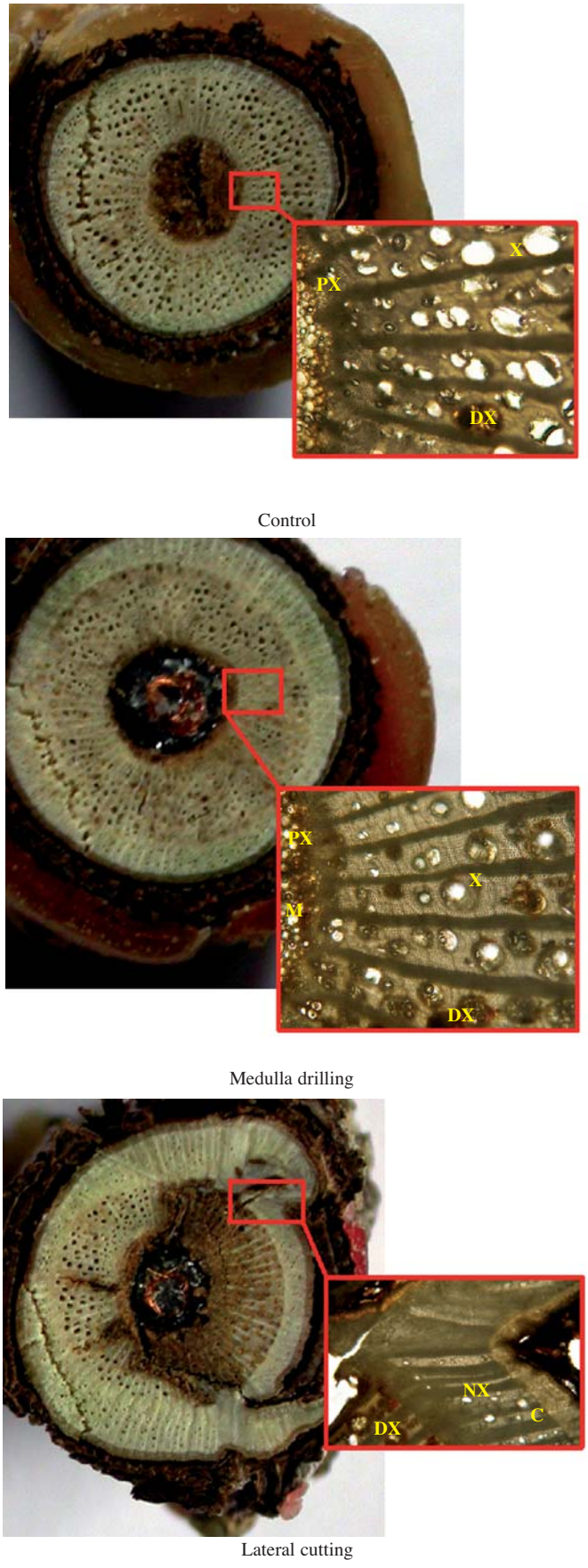

Fig. 7 - Graft point fresh sections, mounted in water; image enlargement 1X and 100X (20 µm thickness). X: xylem vessels; PX: primary xylem; NX: novel xylem post-trauma; DX: damaged xylem; M: medulla; C: cambium; T: TAG.

Table 1 - Average growth of vine-branches (cm) after 45 days from planting

Thesis		Clones				
Methods of TAG implantation	Presence +/- of TAG	Sangiovese	Prugnolo gentile	Colorino	Trebbiano toscano	Vernaccia San Gimignano
Medulla drilling	+	32.4 b*	24.4 a	19.1 a	21.3 b	22.9 a
Medulla drilling	-	43.7 a	22.0 a	17.7 a	31.5 a	25.2 a
Lateral cutting	+	32.9 b	24.0 a	16.7 a	17.9 b	20.0 a
Lateral cutting	-	24.4 b	25.0 a	9.4 b	24.0 b	20.9 a
Control	-	31.2 b	30.3 a	18.9 a	25.1 ab	16.9 a

* Values in the same column followed by the same letter do not significantly differ according to Duncan's Multiple Range test (P=0.05).

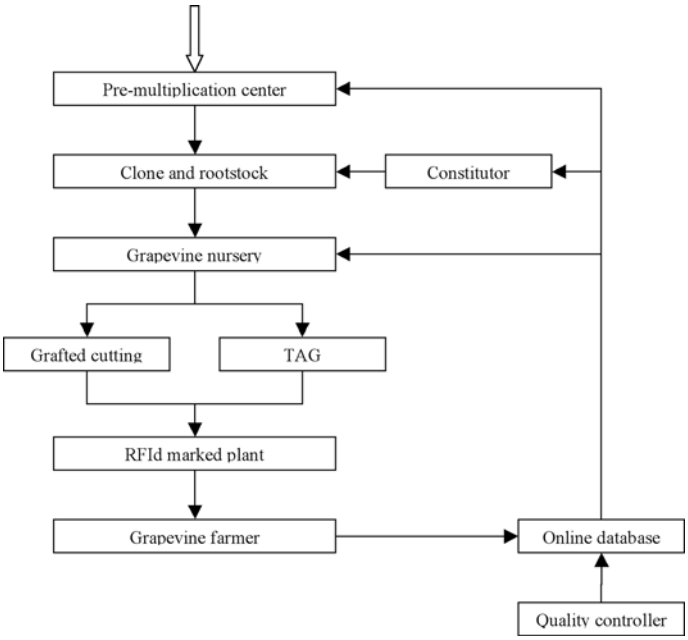

Fig. 8 - Scheme of online database for traceability purpose.

Fig. 9 - Grafted cuttings with microchips planted in nursery. Cicatrisation callus is visible for plants subjected to lateral cuttings.

4. Conclusions

This paper presents an innovation regarding identification tools for grapevine produced in nursery. For the first time radiofrequency technology is detailed for inserting TAGs inside grapevine plant considering biological and sanitarian issues. Our findings suggest that TAGs inside plants did not compromise growth start-up, vegetative growth, sanitary condition, histological status of graft point, or quality of plants, considering the first year of farming, with slightly better performance obtained with the medulla drilling procedure (faster and safer in terms of mortality).

The current research, which considered a large population of grapevine, focused on the substitution of traditional labels with radiofrequency chips inserted directly - and permanently - within the plants (Fig. 9), making it possible to identify every single grafted cutting produced by a nursery and to track after planting the whole plant history and certification data. Moreover, in addition to allowing rapid identification, the microchip is linked to a specific database which stores a datasheet of useful information about the plant.

In light of the results obtained during the first year of trials, the RFId technology can be considered a useful and up-to-date tool for satisfying the need for high quality, safety and traceability along the grapevine production line

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