

OLIVE TREES AS OTHER SPECIES ARE AFFECTED BY SOME GRAFT-TRANSMISSIBLE DISEASES THAT CANNOT BE CONTROLLED WITH THE COMMON TECHNIQUES

VIRUSES AND VIRUS-LIKE DISEASES

FUNGUS-VERTICILLIUM WILT

Verticillium dahliae





LIST OF PATHOGENS AFFECTING THE QUALITY OF PROPAGATING MATERIAL

BACTERIA

Olive Knot

Pseudomonas savastanoi
pv *savastanoi*

FUNGI

Verticillium dahliae

NEMATODES

Xiphinema diversicaudatum
Meloidogyne javanica
M. incognita
Pratilenicus vulnus

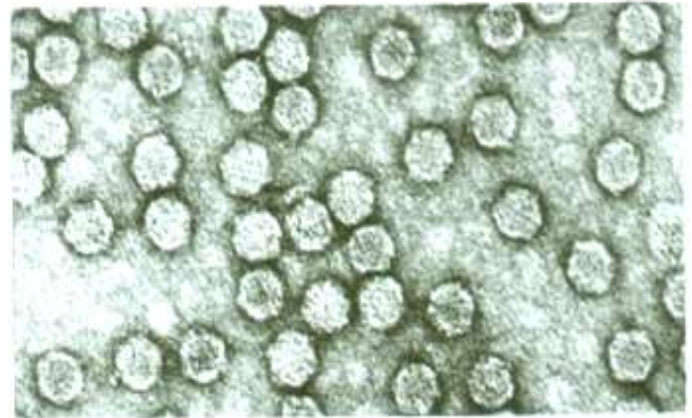
PHYTOPLASMAS

VIRUSES

1. Strawberry latent ringspot virus (SLRV)
2. Arabis mosaic virus (ArMV)
3. Cherry leafroll virus (CLRV)
4. Olive latent ringspot virus (OLRSV)
5. Cucumber mosaic virus (CMV)
6. Olive latent virus 1 (OLV-1)
7. Olive latent virus 2 (OLV-2)
8. Olive vein yellowing associated virus (OVYaV)
9. Olive yellow mottling and decline-associated virus (OMYDaV)
10. Tobacco mosaic virus (TMV)
11. Olive semilatif virus (OSLV)
12. Olive leaf yellowing-associated virus (OLYaV)
13. Tobacco necrosis virus (TNV)

**Bumpy fruits: virus disease
induced by *Strawberry
latent ringspot nepovirus*
(SLRSV)**

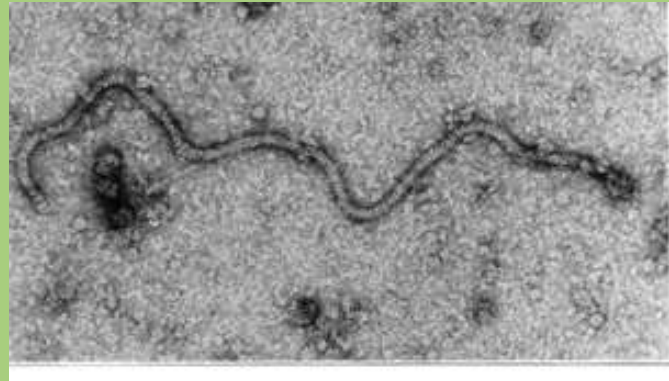
NEPOVIRUS





**Leaf yellowing:
a virus disease
induced by a
closterovirus
(OLYaV)**

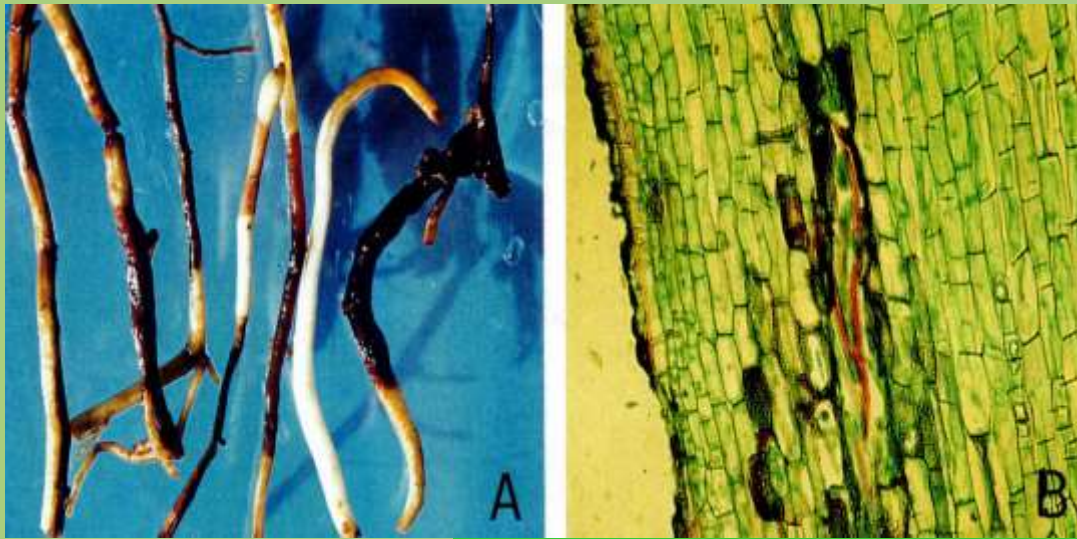
CLOSTEROVIRUS



**Graft transmission
of leaf yellowing**

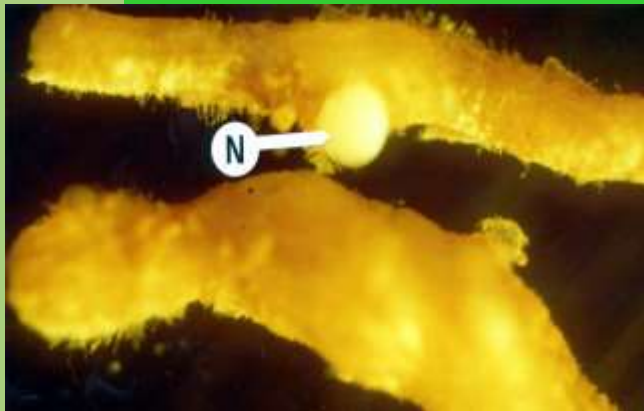
Phytoplasma infections





Endoparasitic nematode
Pratylenchus vulnus

Root-knots nematode
Meloidogyne incognita



Nursery production is subject to quality and phytosanitary regulation which require assessment of true-to-type and sanitary status compulsory

EU Directives (92/34, 93/48)
ITALIAN DECREES
REGULATING

MANDATORY
DM 14/04/1997

VOLUNTARY
DM 24/07/2003
DM 4/05/2006
DM 20/11/2006

CAC
Conformitas Agraria
Comunitatis

VIRUS-FREE
VIRUS-TESTED

WHAT KIND OF MATERIAL CAN A FARMER
USE TO PLANT A NEW OLIVE ORCHARD ?

according italian laws

plants:

C.A.C.

or

CERTIFIED

C.A.C. ***(Conformitas Agraria Communitatis)***

mandatory in all the EU

Minimum status required for the commercialisation of plant propagating material:

- produced and commercialised only by accredited suppliers (nursery man)
- phytosanitary:
 - free from "quality" pathogen organisms
 - biometric: satisfactory vigour and size
 - trueness to type

C.A.C.
(Conformitas Agraria Communitatis)

mandatory in all the EU

Italy adopted the provisions of the Directives
with a Decree of the Ministry of Agriculture
in 1997 (April 14)

WHAT DOES C.A.C. MATERIAL MEAN?

- **Material that derives from founder plants, healthy-looking and vigorous, with true-to-type features and symptomless, selected in commercial orchards**
- **Submitted to laboratory tests for true-to-type and absence of quality-affecting pathogens, representing the minimal phytosanitary requirements established by DM 14/4/1997**

List of harmful and quality-affecting organisms transmitted by propagation material enclosed in EU Directives and national regulation

Genus/Specie

Insects, mites and nematodes at any development stage

Olea europea

- Euzophera pinguis**
- Meloidogyne sp.**
- Saissetia oleae**

Bacteria

Pseudomonas savastanoi pv. savastanoi

Fungi

Verticillium dahliae

Virus and virus-like organisms

ALL

WHAT DOES “CERTIFIED MATERIAL” MEAN?

- **Material that derives from plants submitted to sanitary and clonal selection and, if needed, sanitation**
- **individually tested for absence of pathogens transmissible by vegetative propagation**
- **inspected for true-to-type**

DM 20/11/2006 and further modifications

SANITARY STATUS VIRUS-FREE AND VIRUS-TESTED

PATHOGEN	Sanitary status		
	Acronym	Virus-free (VF)	Virus-tested (VT)
VIRUSES :			
Arabis mosaic	ArMV	X	X
Cherry leafroll	CLRV	X	X
Strawberry latent ringspot	SLRV	X	X
Cucumber mosaic	CMV	X	-
Olive latent 1	OLV-1	X	X
Olive latent 2	OLV-2	X	-
Olive leaf yellowing associated	OLYaV	X	X
Tabacco necrosis virus	TNV	X	-
FUNGI:			
<i>Verticillium dahliae</i>		X	X
BACTERIA			
<i>Pseudomonas syringae pv syringae</i>		X	X
PHYTOPLASMA			
NEMATODES			
<i>Meloidogyne incognita</i>		X	X
<i>Meloidogine javanica</i>		X	X
<i>Pratylenchus vulnus</i>		X	X
<i>Xiphineme diversicaudatum</i>		X	X

C.A.C. Category

IDENTIFICATION OF
FOUNDER PLANT WITH
THE MINIMAL
PHYTOSANITARY
REQUIREMENTS

TESTS

SANITARY AND TRUE-
TO-TYPE
ASSESSMENTS

MOTHER PLANT
ORCHARDS (MATERIAL
CAC CATEGORY):
SUPPLYING SOURCE

N
U
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R
Y
M
A
N

Certified Category

CLONAL AND SANITARY SELECTION
IDENTIFICATION OF HEALTHY-LOOKING
VIGOROUS PLANTS, YIELDING AND FREE
FROM VIRUS-LIKE SYMPTOMS

TESTS

SANITARY
ASSESSMENTS

INFECTED
PLANTS

eliminated

SANITATION

“HEALTHY”
PLANTS

SANITARY
ASSESSMENTS

INFECTED
PLANTS

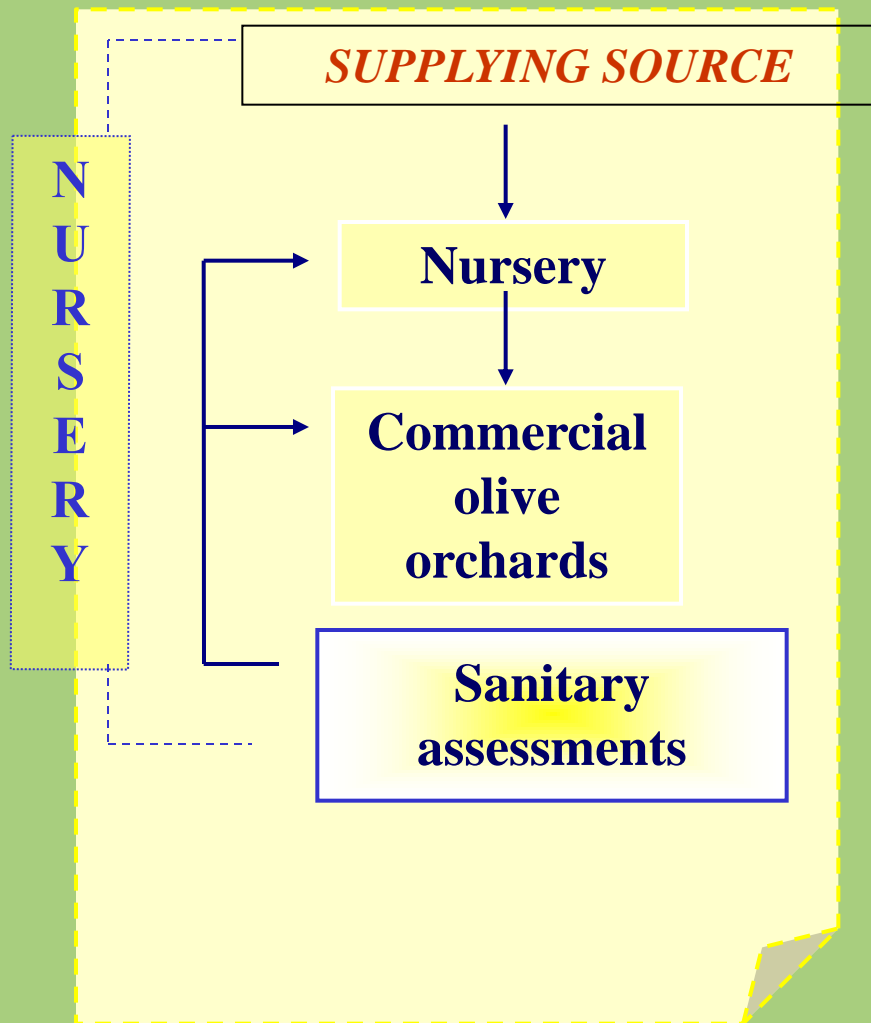
eliminated

Pomological evaluation

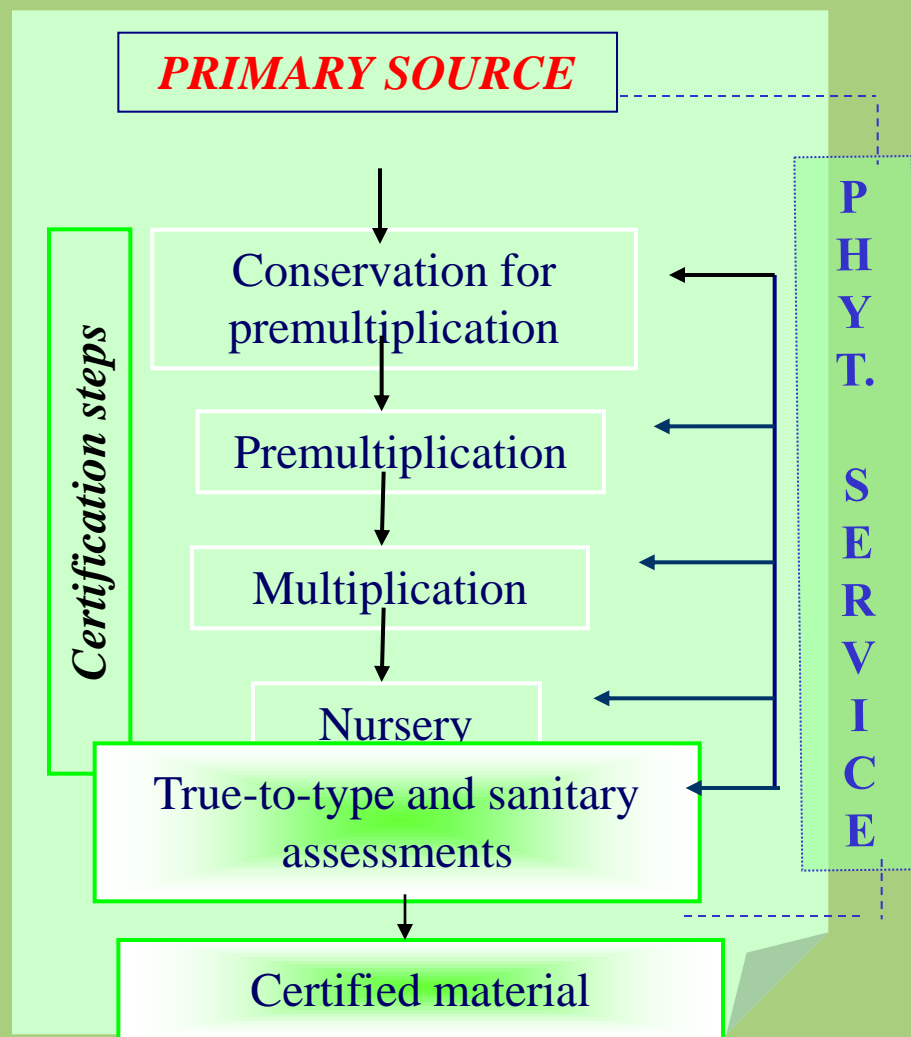
**PRIMARY
SOURCE**

Scientific Institutions

C.A.C. Category



Certified Category



CERTIFICATION PROGRAMMES

A. Production of nuclear stocks

The use of “healthy” plants (accomplishing the phytosanitary requirements) for the new olive groves is a pillar for the quality of crop production

“Healthy” mother plants must be selected through sanitary and clonal selection performed in the framework of certification programme

1. Establishment of the multidisciplinary technical team

**Technological
expertise**

**Pomological
expertise**

**Phytosanitary
expertise**

**Scientific
institutions involved**

SELECTION OF THE PRIMARY SOURCE



Selection of 5-10 plants in each grove according to the variability of the cultivar



Sanitary and pomological selection in 50 - 60 year-old grove

Sanitary and Pomological evaluation

Established clone

Sanitary and Pomological assessment

Data processing

PRIMARY SOURCE



Candidate clones

REGISTRATION

Conservation of primary sources

**Primary source
Or
Nuclear stocks**

**in a repository under
conditions ensuring
freedom from re-infection
(insect proof screenhouse)**



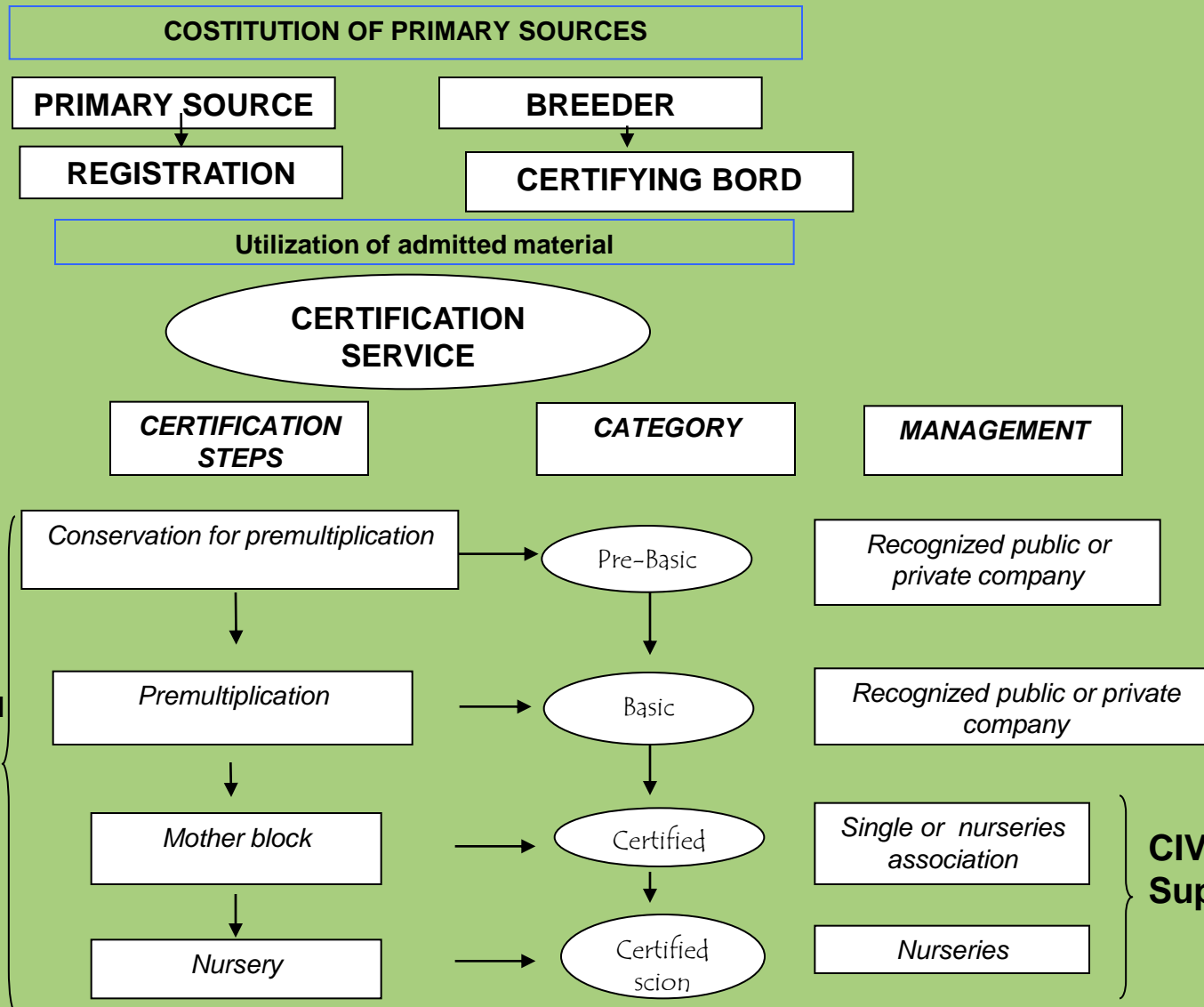
**Responsibility of
the breeder**

OLIVE (*Olea europaea* L.)
PRIMARY SOURCES REGISTERED IN
THE ITALIAN CERTIFICATION
SERVICE

VARIETY	ACCESSION	SELECTION
Aspirina	BN 3/98	Reg. Campania
	CE 1/98	
Biancolilla	12	Reg. Campania
Caiazzana	CE 9/98	Reg. Campania
Cammarotana	23	Reg. Campania
Carboncella	Pianacce B	ARSIAL
Carolea	Oer 0027	IAMUBA
Carpellesse	C	Reg. Campania
Cazzinocchio	Oer0042	IAMUBA
Cellina di Nardò	Oer0056	IAMUBA
Cima di Melfi	Oer0062	IAMUBA
Cipressino	Oer0073	IAMUBA
Coratina	Oer0069	IAMUBA
Corneglia	22	Reg. Campania
Cornia	9	Reg. Campania
Dolce di Cassano	Oer0025	IAMUBA
Femminella	N21	Reg. Campania
	Oer0033	IAMUBA
Frantoio	Francocci 7	ARSIAL
	F46	CNR Scandicci
	13	Reg. Campania
Groia	13	Reg. Campania
	Vallequercia 1/S	ARSIAL
Itrana	Collenero 6	ARSIAL
	Oer0047	IAMUBA
Leccino	Collececco 22	ARSIAL
	L10B	CNR Scandicci
	Pisa 4	Pisa
	Pisa 9	Pisa
	cl ISTE A 30	ISTEA
Marinese	AV 16/98	Reg. Campania
Maurino	M1b	CNR Scandicci
Minutella	Casarè	ARSIAL
Moraiole	Cuccagna 8	ARSIAL
	M11	CNR Scandicci
Nocellara Belice	Oer0010	IAMUBA
Nocellara messinese	Oer0077	IAMUBA
Nociara	Oer0031	IAMUBA
Nolca	Oer0053	IAMUBA
Nostrale	AV 5/98	Reg. Campania
Ogliara	5	Reg. Campania
Ogliarola	Oer0011	IAMUBA
	AV 2/98	Reg. Campania
Oliastro	Oer4000	IAMUBA
Oliva bianca	N16	Reg. Campania
Oliva torsa	AV 12/98	Reg. Campania
Olivella	AV 13/98	Reg. Campania
Olivella appuntita	AV 14/98	Reg. Campania
Olivello	AV 15/98	Reg. Campania
Olivone	AV 17/98	Reg. Campania
Ortice	N22	Reg. Campania
Ortolana	N23	Reg. Campania
Pampagliosa	N24	Reg. Campania
Pasola	Oer0075	IAMUBA
Pasola d'Andria	Oer0081	IAMUBA
	Oer0013	IAMUBA
Pendolino	Oer0013	IAMUBA
	P30	CNR Scandicci

VARIETY	ACCESSION	SELECTION
Picholine	Oer0017	IAMUBA
Pisciottana	A	Reg. Campania
Racioppella	BN 28	Reg. Campania
Ravece	AV 20/98	Reg. Campania
Ritonella	AV 6/98	Reg. Campania
Rizzitella	AV 9/98	Reg. Campania
Rotondella	N6	Reg. Campania
Ruveia	AV 11/98	Reg. Campania
Salella	28	Reg. Campania
Salvia	Montelibretti 6	ARSIAL
Sant'Agostino	Oer0009	IAMUBA
Simone	Oer4010	IAMUBA
Sirole	Soratte 1	ARSIAL
Tenacella	CE 7/98	Reg. Campania
Termine di Bitetto	Oer0097	IAMUBA
Tonda	Ce4/98	Reg. Campania
Toscanella	BN 1/98	Reg. Campania
Toscanina	Oer0035	IAMUBA
Vigna della Corte	AV 4/98	Reg. Campania
Ghiacciolo	cl ISTE A 28/cav51	CAV
Selvatico	cl ISTE A/cav27	CAV
Coreggiolo di Villa Verucchio	cl ISTE A 08 cav	CAV
Nostrana di Brisighella	cl ISTE A 38cav	CAV
Ascolana Tenera	ISPaVe-AN01	ISO-ISPaVE
Ascolana Dura	ISPaVe-AN02	ISO-ISPaVE
Carboncella	ISPaVe-AN05	ISO-ISPaVE
Capolga	ISPaVe-AN06	ISO-ISPaVE
Cornetta	ISPaVe-AN07	ISO-ISPaVE
Coroncina	ISPaVe-AN08	ISO-ISPaVE
Lea	ISPaVe-AN09	ISO-ISPaVE
Mignola	ISPaVe-AN10	ISO-ISPaVE
Nebbia	ISPaVe-AN11	ISO-ISPaVE
Orbetana	ISPaVe-AN12	ISO-ISPaVE
Piantone di Folerone	ISPaVe-AN13	ISO-ISPaVE
Piantone di Magliano	ISPaVe-AN14	ISO-ISPaVE
Roggia	ISPaVe-AN15	ISO-ISPaVE
Rosciola colli Esini	ISPaVe-AN16	ISO-ISPaVE
Sargano di Fermo	ISPaVe-AN17	ISO-ISPaVE
Sargano di S. Benedetto	ISPaVe-AN18	ISO-ISPaVE
Frantoio di villa Verucchio	cl ISTE A 40	ISTEA
Colombina	cl ISTE A 17	ISTEA
Grappuda	cl ISTE A 28	ISTEA
Moraiole	cl ISTE A 26	ISTEA
Carbuncion di Carpineta	cl ISTE A 24	ISTEA
Rossina	cl ISTE A 5	ISTEA
Coreggiolo di Montegrindolfo	cl ISTE A 45	ISTEA

Steps and classification of plant propagating material in the Certification scheme



Sanitary and genetic control by Regional Phytosanitary Service

CIVI-Italia Support



CERTIFICATION SET UP

CONSERVATION FOR PREMULTIPLICATION



c/o Dipartimento di protezione delle Piante e Microbiologia Applicata Azienda Martucci, Valenzano (BA)

PREMULTIPLICATION CENTRE



c/o Centro di Ricerca e Sperimentazione in Agricoltura "Basile Caramia" Locorotondo (BA)

MULTIPLICATION CENTRE



c/o Consorzio Vivaistico Pugliese, Valenzano (BA)
c/o Coop. Rinascita Agricola, Melendugno (BA)

NATIONAL AND REGIONAL PHYTOSANITARY SERVICE

CONTROLS

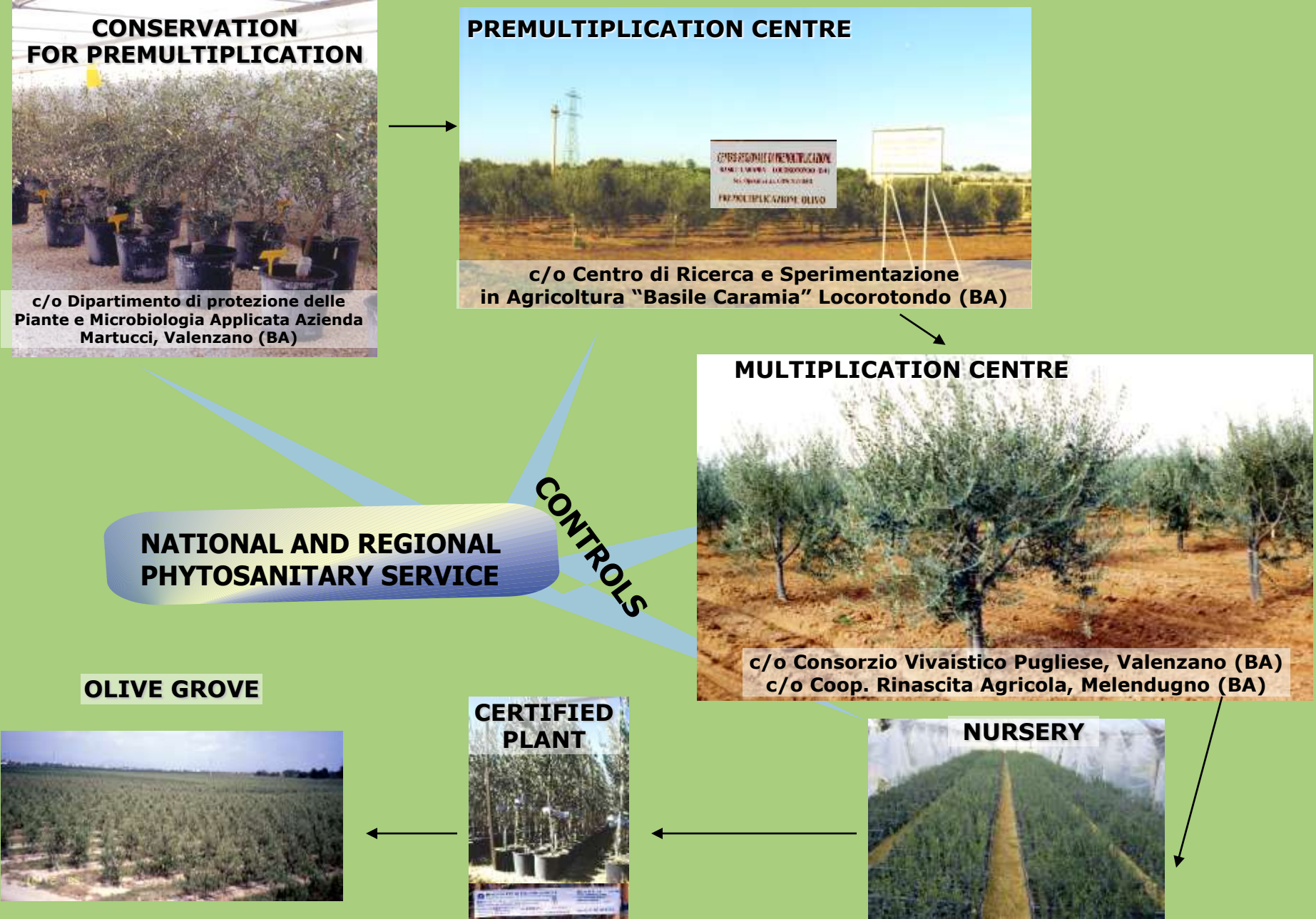
OLIVE GROVE



CERTIFIED PLANT



NURSERY



Conservation for premultiplication

**primary
source**

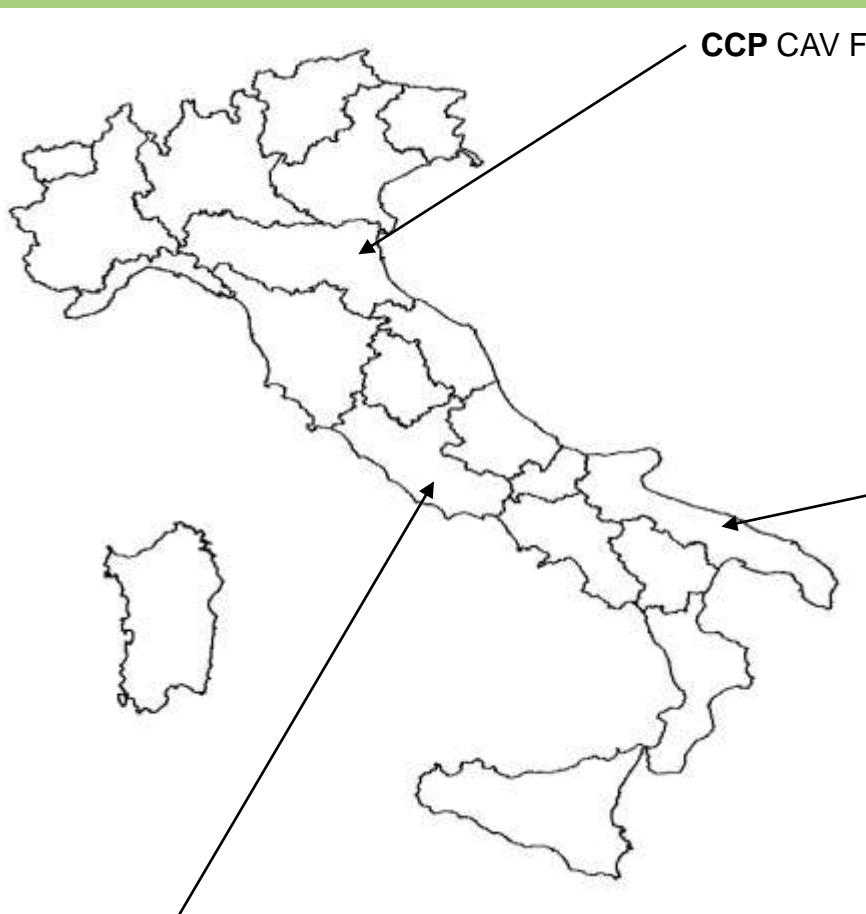
pre-basic



in a repository

**Responsability of Public or
private institutions officially
recognized from MiPAAF**





CCP CAV Faenza - Reg. E. Romagna



CCP DPPMA UBA - Reg. Puglia



CCP C.R.A. IsPaVe Roma - Reg. Lazio



**Conservation centre for
Premultiplication working in
the Italian certification scheme**

Premultiplication

pre-basic

Basic ●

Rootstocks and varieties grown in open field, in soil free from virus-vector nematodes and fungi

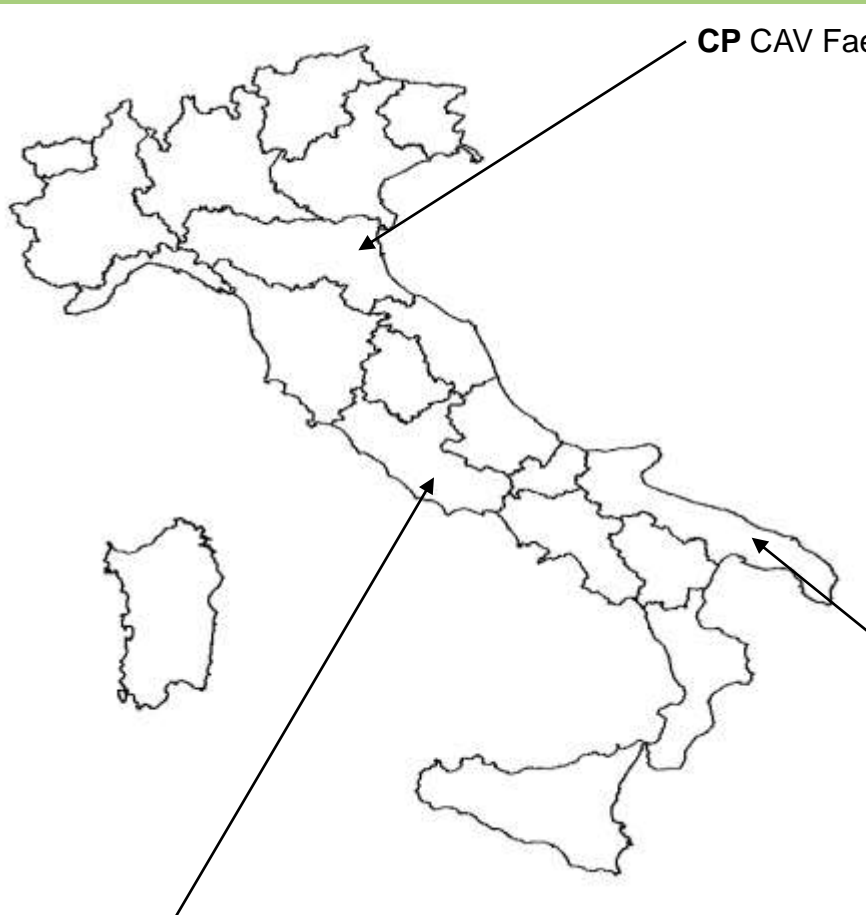
Nursery activity

To provide mother plants for Multiplication Centre.



Responsability of Public or private institutions officially recognized from MiPAAF





CP CAV Faenza - Reg. E. Romagna



CP CRSA "B. Caramia" Locorotondo - Reg. Puglia



CP C.R.A. IsPaVe Roma - Reg. Lazio



**Premultiplication centre
working in the Italian
certification scheme**

MULTIPLICATION → **In open field** → **Non-cultivated border (10m)**

"Certified mother plants"

Phytosanitary controls

PLANTS

1) VISUAL INSPECTIONS

2) LABORATORY TESTS:

Viruses	dsRNA for VF plants and RT-PCR for VT plants: on 5% of plants each year, starting of the year fifth Tissue: scraping from 1-2 years old cutting collected in spring or autumn
<i>V. Dahliae</i>	Only suspicious plants must be tested by isolation from 1-2 years old cuttings
Phytoplasmas	Only suspicious plants must be tested by PCR

SOIL

LABORATORY TESTS:

V. dahliae
X. diversicaudatum

MULTIPLICATION "Nurseries" → **In open field** → **Non-cultivated border**
Grafted or self-rooted plants or in pots

Plots and box used for seed-bed, for growing grafted or self-rooted plants must include a non-cultivated border, isolated from water surface, if pots are used they must not be put directly on the soil but on inert material ensuring drainage and isolation



Phytosanitary controls

2) VISUAL INSPECTIONS for the plants

1) LABORATORY TESTS: for the soil and substrate

V. dahliae, *X. diversicaudatum*,
P. vulnus, *Meloidogyne* spp.



1



Plants propagated by cutting

2



3



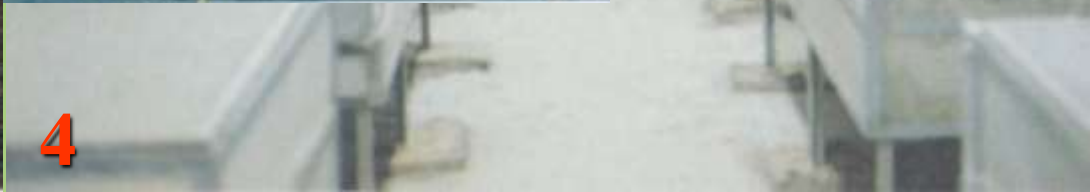
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5



4



Choice of the suitable propagating material

A plant propagated by cutting is characterised by:

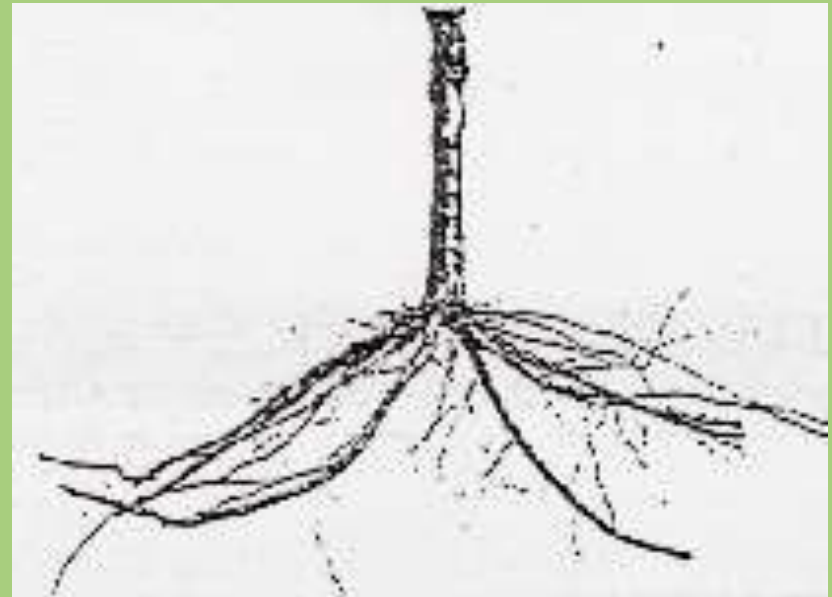
A shallow fasciculate root system

Poor anchorage to the soil

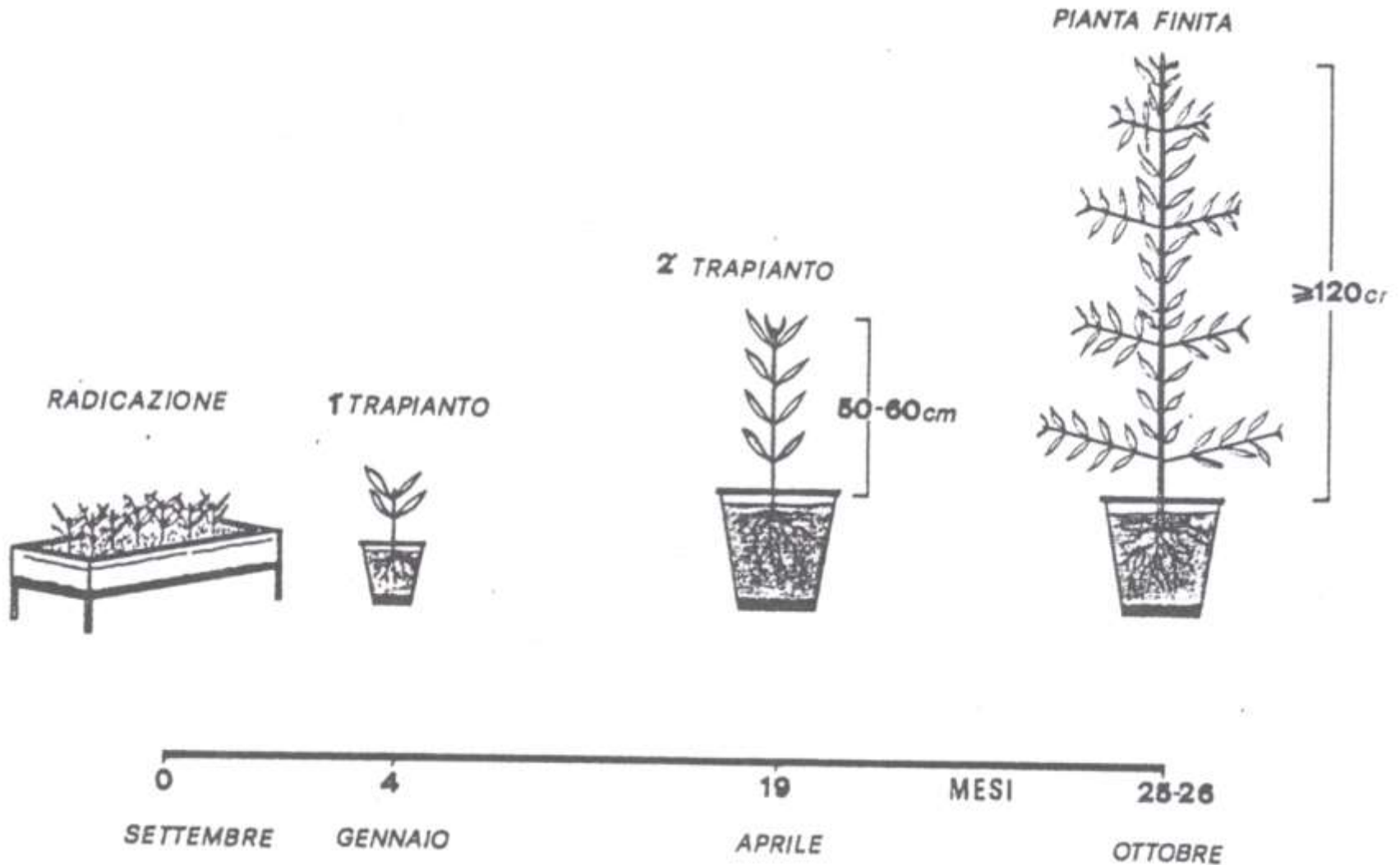
Higher susceptibility to water stress

Difficulty to explore hard soils

Low resistance to mechanical stresses (mechanical harvest).



Production cycle of olive trees by cutting



PROTOCOLS FOR THE PRODUCTION OF OLIVE SELF ROOTED PLANTS IN APULIA REGION

Provincia	Foggia	Bari	Brindisi	Taranto	Lecce
Vivaisti (n)	1	2	3	1	6
Preparazione talee (mese)	Ago	Mar-Ago	Set-Ott	Ott	Ago-Ott
Uso fitoregolatori (si/no)	si	si	si	si	si
da preparare	100,0%	50,0%	66,7%	-	100,0%
già preparato	-	50,0%	33,3%	100,0%	-
Principio attivo prevalente	IBA	IBA	IBA	IBA	IBA
Formulato					
Liquido (L)	-	50,0%	100,0%	-	-
Polvere (P)	100,0%	50,0%	-	100,0%	100,0%
Concentrazione (ppm)	2.000	2-4.000	4-5.000	2-4.000	2-4.000
Durata trattamento	5"	3-15"	6-10"		2-3"
Luogo	serra	serra	serra	serra	serra
radicazione:					
su bancale	100,0%	50,0%		100,0%	66,7%
in cassette	-	50,0%	66,7%		33,7%
in vaso	-		33,3%		
substrato	perlite	perlite	perlite	perlite	perlite
Metodo controllo umidità:					
nebulizzazione	100,0%	50,0%	66,7%		100,0%
umidificazione	-	50,0%	33,3%	100,0%	
Sistema controllo umidità:					
computerizzato	100,0%	50,0%	66,7%	-	66,7%
orologeria	-	50,0%	33,3%	100,0%	33,3%
Riscaldamento basale:	si	si	si	no	si
vivai (%)	100,0%	50,0%	100,0%	-	66,7%
ad acqua		-	33,3%	-	100,0%
ad aria	100,0%	100,0%	66,7%	-	-
Spaziatura talee (cm x cm)	3x3	2x2-6x6	1x2-4x4	1x2	2x2-5x5
Raccorciamento foglie (% vivai)	100,0%	50,0%	66,7%	no	66,7%
Tempo medio radicazione (settimane)	8	3-12	8-12	8	8-10
Radicazione media	45%	75%	60%	70%	45%
Durata indurimento (mesi)	6	5-13	3-10	8	2-6
Trapianto: mese	Mar	Feb-Mar	variabile	Sett	Apr-Giu
in vaso terracotta	-	33,3%	-	-	-
in vaso plastica	100,0%	66,7%	100,0%	100,0%	100,0%











Plants propagated by cutting



**AGROMILLORA
CATALANA
System**



6



Plants propagated by grafting

8



7

12



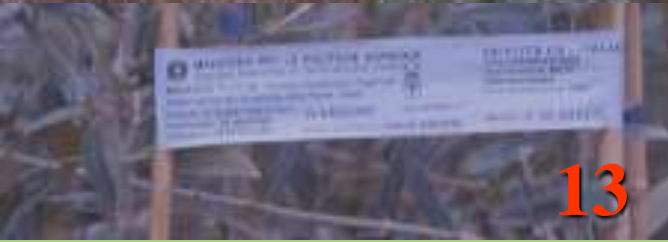
11



9



13



10

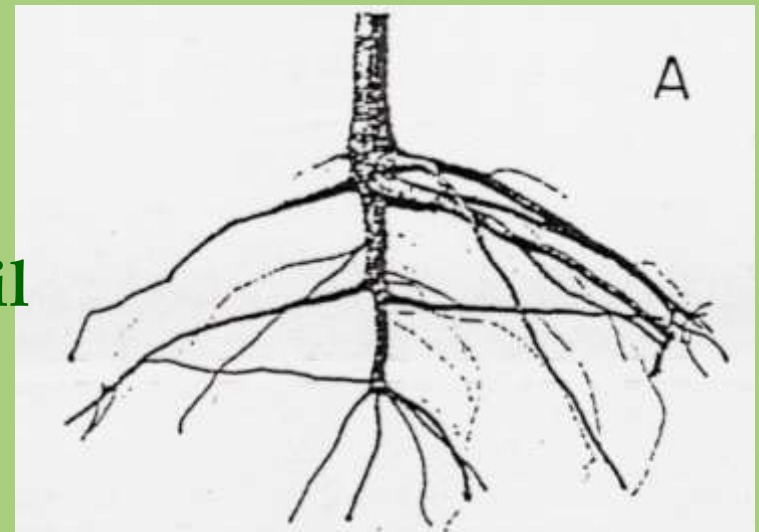


Choice of the suitable propagating material

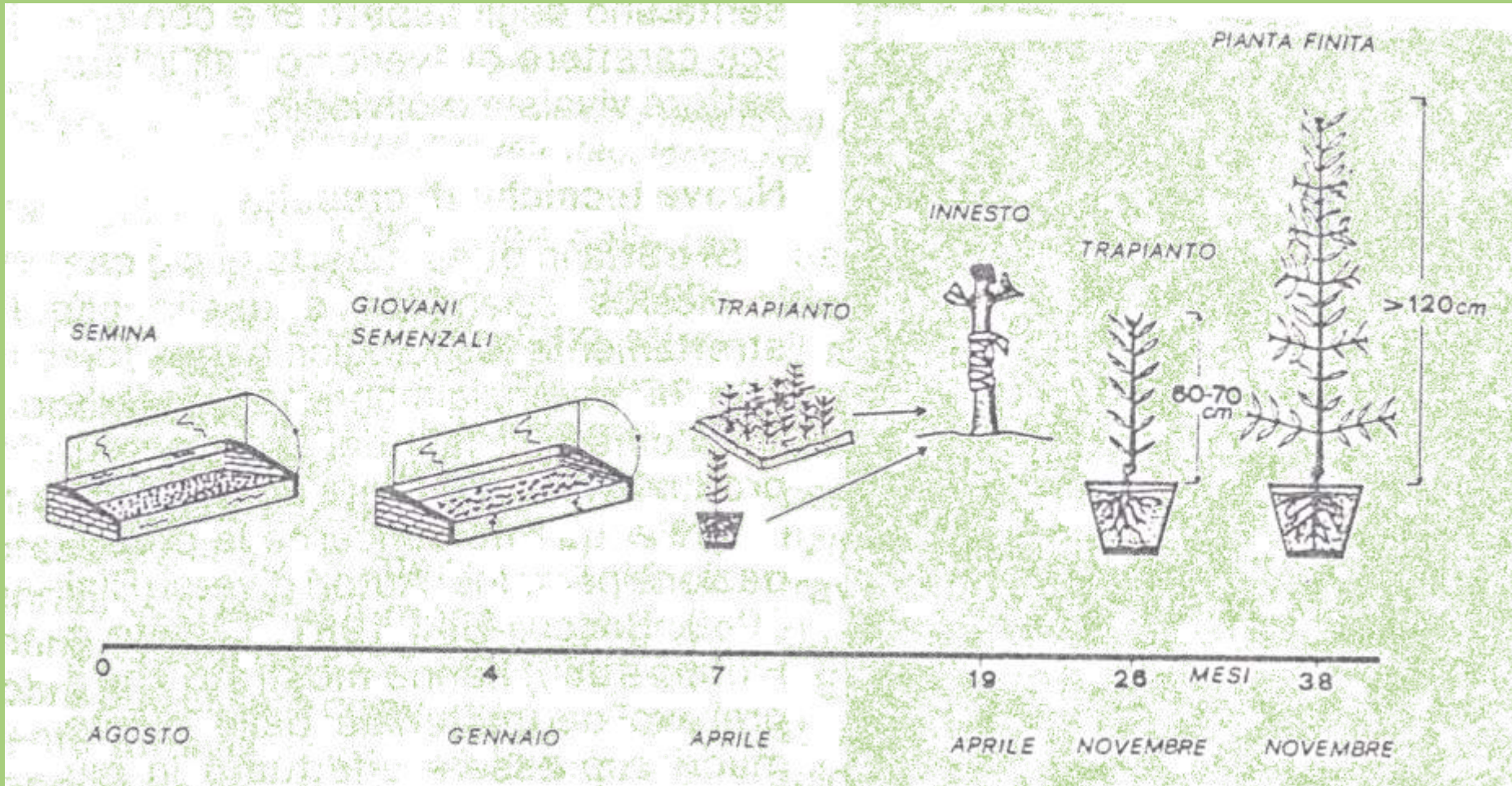
A plant propagated by grafting is characterised by:

A tap root system which deepens in the soil

- Better anchorage to the soil
- In the first years of planting, it better explores and exploits the soil water resource
- It is less sensitive to water stress
- It forms ovular masses at the plant base sooner.



Production cycle of olive trees by grafting



Plants propagated by grafting



**Seedlings
production**

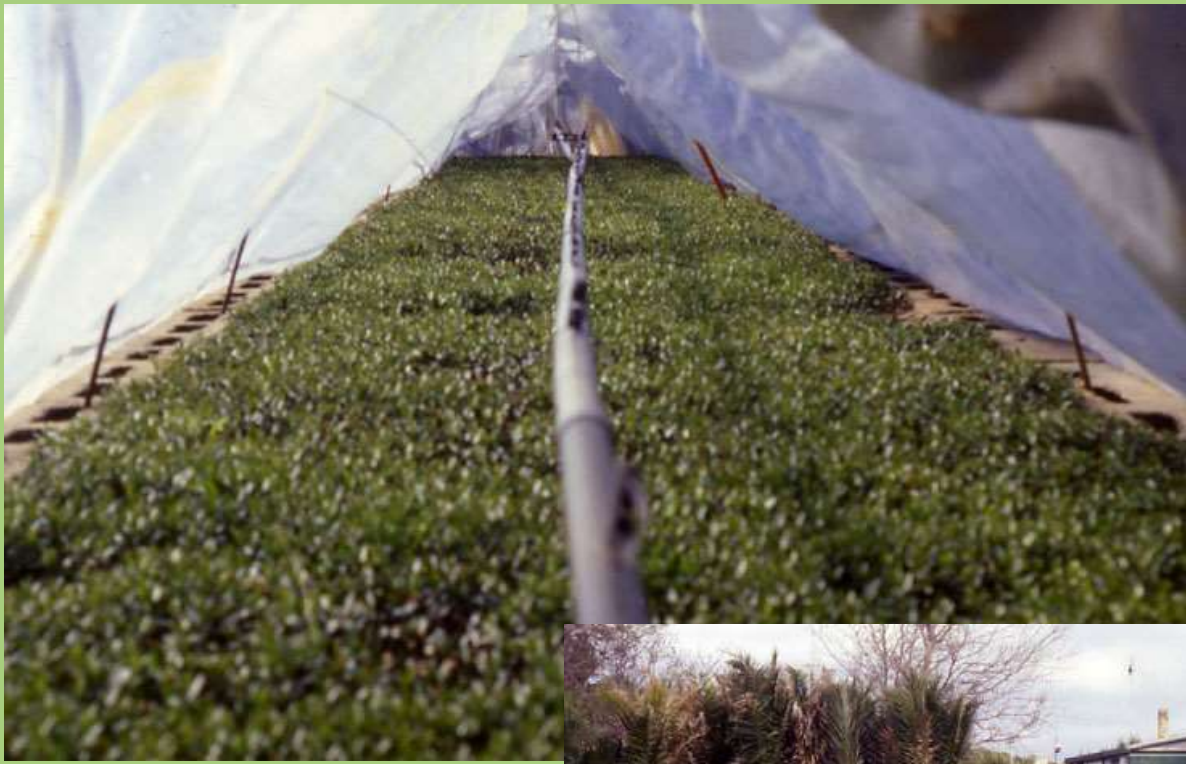




















Plants propagated by grafting



**Mother
block**



**Budsticks
collection**



































Insetti nocivi alla crescita e formazione delle piante

MARGARONIA



Insetti nocivi alla crescita e formazione delle piante

RODILEGNO GIALLO



Insetti nocivi alla crescita e formazione delle piante

OZIORRINCO



Insetti nocivi alla crescita e formazione delle piante

CICALE



Funghi nocivi alla crescita e formazione delle piante

OCCHIO DI PAVONE







 **MINISTERO PER LE POLITICHE AGRI**
Servizio Nazionale di Certificazione Vegetale

REGIONE PUGLIA - Servizio Fitosanitario Regionale
Osservatorio per le Malattie delle Piante - BARI

PIANTA DI OLIVO INNESTATA
Portinnesto: **OLIVASTRO** cv **TOSCANINO**
Categoria: **CERTIFICATO** Stato sanitario: **V**



MINISTERO DELLE POLITICHE AGRICOLE E FORESTALI

Servizio Nazionale di Certificazione Volontaria

REGIONE PUGLIA - Servizio Fitosanitario Regionale

Osservatorio per le Malattie delle Piante - BARI

Piantone di olivo innestato Portinnesto: **OLIVASTRO**



Cv LECCINO (selez. IAM-UBA/Cer-47)

Categoria: **CERTIFICATO** Stato sanitario **VIRUS ESENTE**

QUALITÀ CE - ITALIA

Cod. Prod.: 02506990726

Cod. Fornitore: BA 0002 / FRU

Olea europaea

Serie **VE/ 05** N° **098089**







Certified grafted plants



Plant's delivery



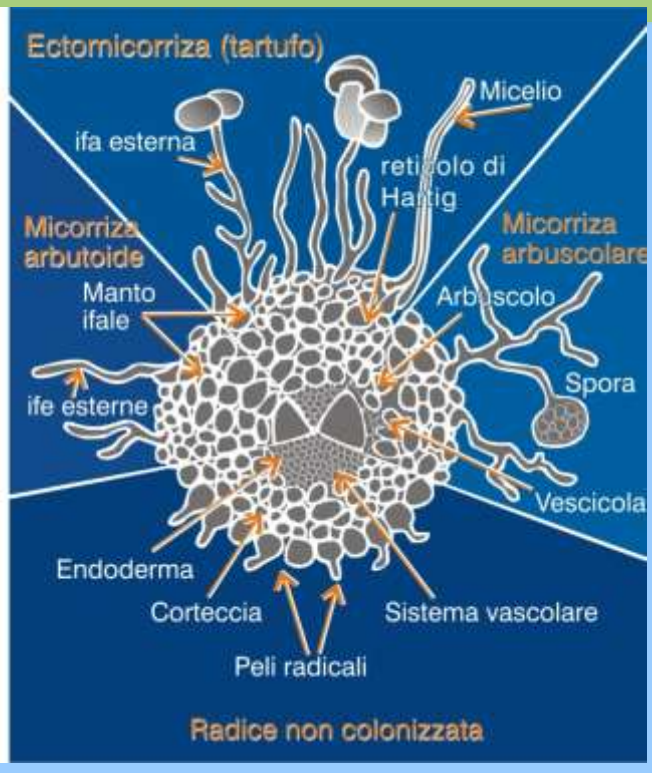
Bare roots certified plants processed to export to Australia





**New olive
grooves
constituted
with certified
plant**

Innovative nursery techniques



MICORRIZA

AZIONI

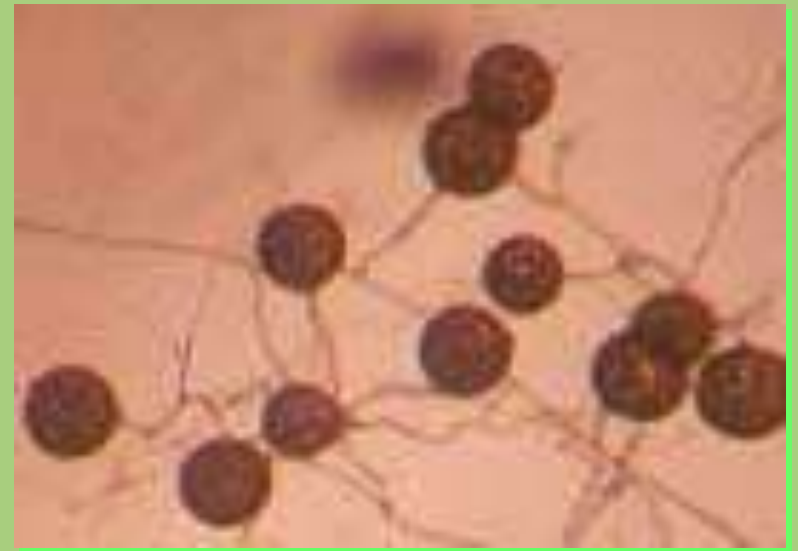
1. Aumento dell'apparato radicale
2. Migliora l'assorbimento degli elementi minerali del suolo
3. Migliora la resistenza in post-trapianto
4. Aumenta la disponibilità d'acqua (resistenza alla siccità)
5. Conferisce tolleranza alla salinità
6. Aumenta la tolleranza a determinati patogeni
7. Azione antistress sulla pianta
8. Azione miglioratrice sulla struttura del terreno

COS'E' LA MICORRIZA

E' chiamata così l'associazione simbiotica (benefica per entrambi) fra le radici delle piante e alcuni funghi

Il fungo colonizza le radici della pianta fornendole elementi minerali ed acqua, che estrae dal suolo attraverso la sua rete esterna di ife, mentre la pianta fornisce al fungo essudati organici radicali

Il fungo non danneggia la pianta!!







CORATINA
18 mesi innesto
TESTIMONE NON
TRATTATO
10 mesi dal trapianto

CORATINA
18 mesi d'innesto
AEGIS
1 applicazione 8 gr.
10 mesi dal trapianto

CORATINA
18 mesi innesto
AEGIS
1 applicazione 16 gr.
10 mesi dal trapianto



“In Vitro” Propagation of Olive



Micropropagation

is the key to:

- rapid propagation on a mass scale in limited spaces
- production of healthy and genetically uniform plants
- propagation of genotypes difficult to multiply by cutting
- production independent from the seasonal events



EASY adaptation to vitro



Hojiblanca



Barnea



Carolea



Coratina COVIP

EASY adaptation to vitro

Correggiolo



Difficult adaptation to vitro

Picholine



Difficult adaptation to vitro

Leccino



Cultivars of easy adaptation

It is possible to produce nursery material on a mass scale within 12 months (about 100,000 plants)

Cultivars of medium adaptation

It is possible to produce plantlets on a mass scale but optimization is needed

Cultivars of difficult adaptation

Material is “in vitro” and sterile, but mass scale production is not yet possible; further studies are needed

Advantages of olive micropropagation

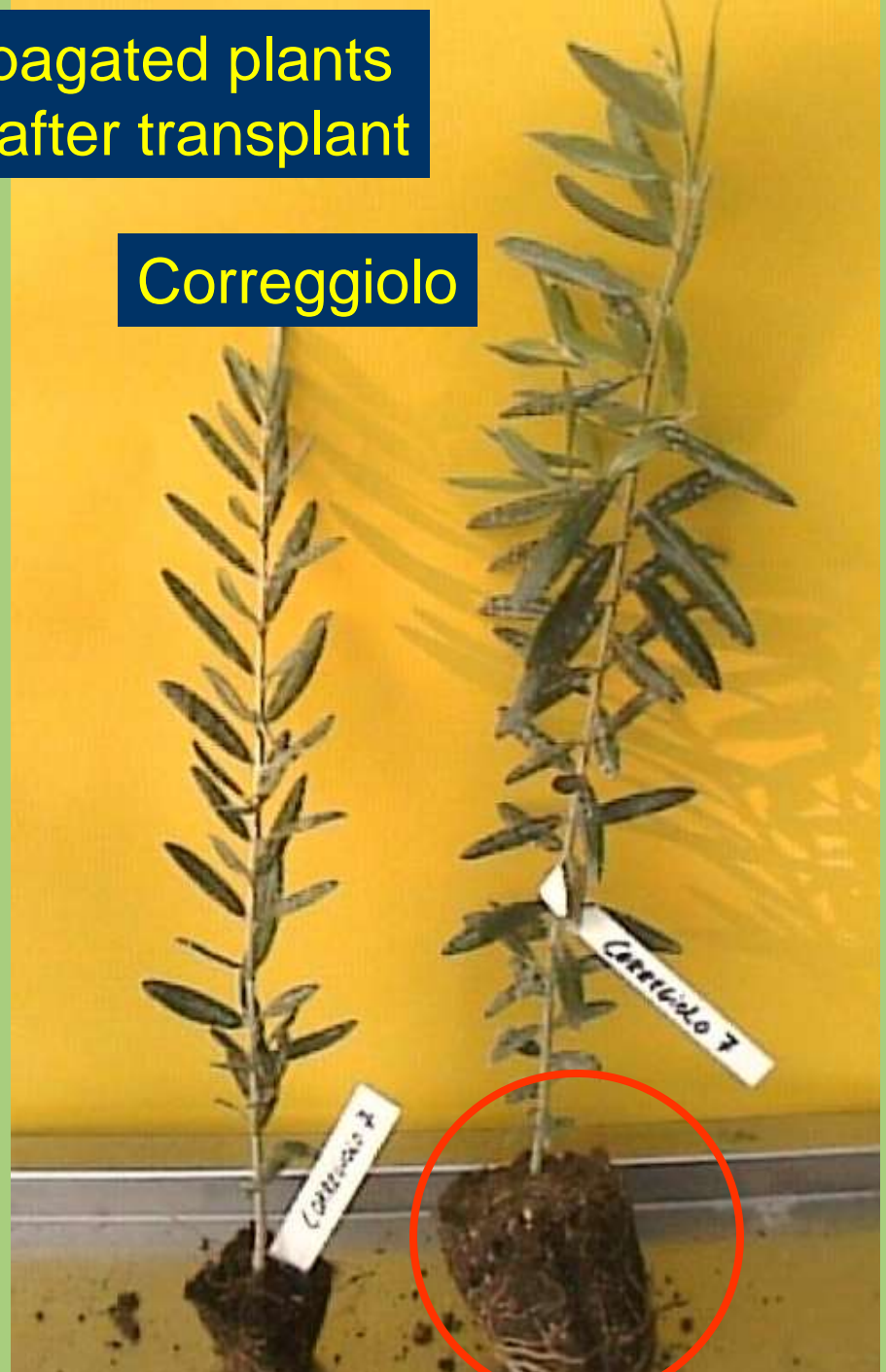


- Rooting phase in sterile rockwool can be used to export to countries with severe phytosanitary laws

Micropropagated plants
120 days after transplant

Hojiblanca

Correggiolo



Final Conclusions - Olive micropropagation

- It is extremely important to use efficient protocols, suitable for commercial labs' requirements (media, fragmentation of material).
- First stages of “in vitro” adaptation are the most critical ones (necessary to balance auxine and cytokinins levels); rooting stage is not so critical.
- Olive is a “slow-growing” culture both “in vitro” and “in vivo”; excessive fragmentation of microcuttings causes too long subcultures, reducing the main tissue culture advantage (rapidity).

Final Conclusions - Olive micropropagation

- Can contribute to the evolution and progress of olive cultivation in the world, by multiplying the cultivars suitable for high density systems, for mechanical harvest and for high quality production.
- For cultivars with easy or medium adaptation to “vitro” conditions, it may represent an alternative to the traditional nursery techniques. For the difficult ones further investigations are needed.