LIFE17 / ENV / IT / 000347

Sustainable substrates for agriculture
from dredged remediated marine sediments

From ports to pots
«LIFE SUBSED»

DURATION:
Start: 01/10/2018
End: 30/09/2021

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OBJECTIVES & SCOPE

The main aim of the project is to demonstrate that is possible
convert a waste (the dredged marine sediment)
into a supply (a commercial substrate)

Sediment-based substrates will be applied to:

Nursery production: Cherry Laurel, Olive Tree and Citrus
Ornamental and Flower production: Cherry Laurel, Protea, Calla Lily
Food crops: Blueberry, Strawberry, Citrus and Basil

The performance of the new substrate will be demonstrated by comparison with the typical production cultivated on a peat or cocopeat-based commercial substrates

Treated sediments
(Port of Leghorn – IT)

- Physical and chemical analysis of substrates
- Monitoring rooting and vegetative growth of plants
- Monitoring flowering and fruiting
- Morphological and chemical characterisation of plants and fruit
- Sensorial evaluation of foods
- Life Cycle Analysis

Comparative evaluation of vegetative and productive parameters

- Legal issues on sediments re-use
- Commercial issues for marketing
- Treated sediment preparation and packaging

Pre-commercial trials

Dissemination - Nurseries and Fruitgrowers

TIMETABLE

Action		2018				2019				2020				2021				2022				2023			
Action numbe	Name of the action	I	II	III	IV	I	II	III	IV	I	II	III	IV	I	II	III	IV	I	II	III	IV	I	II	III	IV
A. Preparatory actions (if needed)																									
A.1	Review of the EU and national regulations on the use of sediments for plant nursery and of the analytical protocols				■											■									
B. Implementation actions (obligatory)																									
B.1	Phytoremediated Sediment treated via landfarming process				■	■																			
B.2	Demonstration of the use of remediated sediments as a substrate for nursery production						■	■	■	■	■	■	■	■											
B.3	Demonstration of the use of remediated sediments as substrate for non food crops cultivation (from plantlets to final production: flowers/ornamental)						■	■	■	■	■	■	■	■											
B.4	Demonstration of the use of remediated sediments as a substrate for food crops production						■	■	■	■	■	■	■	■											
B.5	Training courses, workshops and guidelines for project replicability and transferability												■	■	■	■									
B.6	SUBSED Business Plan													■	■	■									
C. Monitoring of the impact of the project actions (obligatory)																									
C.1	Monitoring and validation of treated sediments					■	■																		
C.2	Monitoring and validation of the use of remediated sediments as a substrate for plant nursing and cultivation: non food crops production							■	■	■	■	■	■	■	■										
C.3	Monitoring and validation of the use of remediated sediments as a substrate for nursing and cultivation: food crops production							■	■	■	■	■	■	■	■										
C.4	Monitoring of socio-economic impact of the project and LCA												■	■	■	■									
C.5	Performance indicators monitoring				■					■				■		■									
D. Public awareness and dissemination of results (obligatory)																									
D.1	Project dissemination plan: web-site, material, articles, Layman's report and video				■	■	■	■	■	■	■	■	■	■	■	■									
D.2	Project dissemination plan: events, networking and contacts with Institutions and policy makers				■	■	■	■	■	■	■	■	■	■	■	■									
E. Project management (obligatory)																									
E.1	Project management by FLORA				■	■	■	■	■	■	■	■	■	■	■	■									

ACTION A.1

Review of the EU and national regulations on the use of sediments for plant nursery and of the analytical protocols
All partner

Expected Start Date: IV 2018
Expected End Date: III 2021
Actual Start Date: IV 2018

A1.1 **Preconditioning process for increasing the organic carbon and decreasing the bulk density in order to reach the limit required (expected Bulk density 0.9-0.95, TOC 4-6%) and to produce the clear confirmation that the sediments can be used in agriculture as agronomic substrate.**

A1.2 **A review of the EU and Italian and Spanish regulations/laws regarding the transport and use of dredged sediment based substrates for plant nursery being in force at the date of the project beginning;**

Deliverable Action A1.2

REVIEW OF LEGISLATION
ON DREDGED SEDIMENT MANAGEMENT

Table 1. International Conventions and Main EC Directives

Oslo Convention (1972)	Convention on the Prevention of Marine Pollution by Dumping from Ship and Aircraft
London Convention, Protocol 96 (1972)	Convention on the Prevention of Marine Pollution by Dumping of Wastes and Other Matter
Paris Convention (1974)	Convention on the Prevention of Marine Pollution from land-based sources
OSPAR Convention (1992)	Convention for the Protection of the Marine Environment of the North-East Atlantic
Barcelona Convention, Dumping protocol (1995)	Convention for the Protection of the Marine Environment and the Coastal Region of the Mediterranean
Directives 1999/31/CE (the first EU waste and water directives)	Directive on the landfill of waste; it has a limited impact on the disposal of dredged material along waterways on agricultural land or suitable subaquatic locations are acceptable solutions, but concentrations of contaminants must remain below certain limits.
EU Parliament Decisions 2000/532/CE	Decision on hazardous waste, amended with the Decisions 20001/118/CE, 2001/119/CE and 2001/573/CE, established the European Waste Catalogue, where hazardous waste are defined.

Table 1. International Conventions and Main EC Directives

Directive 2000/60/CE (Water Framework Directive)	Directive of the European Parliament and of the Council (Water Framework Directive), establishing a framework for Community action in the field of water policy: protection of all waters, protection and enhancement of the status of aquatic ecosystems. (Concerning dredged sediments: member States are required by law to submit proposals for quality standards applicable to the concentration of the main substances in surface water, sediments or biota).
Decision 2455/2001/EC	Decision of the European Parliament and of the Council, establishing the list of priority substances in the field of water policy and amending directive 2000/60/EC.
Directive 2008/32/EC	Directive of the European Parliament and of the Council amending Directive 2000/60/EC establishing a framework for Community action in the field of water policy, as regards the implementing powers conferred to the Commission.
Directive 2008/56/EC	Directive of the European Parliament and of the Council (Marine Strategy Framework Directive), establishing a framework for community action in the field of marine environmental policy, within each Member States shall take the necessary measures to achieve or maintain a good environmental status in the marine environment by the year 2020 at the latest.

Table 1. International Conventions and Main EC Directives

EU 'Waste Directive' (Directive 2008/98/CE)	Directive of the European Parliament and of the Council on waste and repealing certain Directives; it lays down measures to protect the environment and human health by preventing or reducing the adverse impacts of the generation and management of waste and by reducing overall impacts of resource use and improving the efficiency of such use. (Concerning dredged sediments: they are linked to waste because it is not yet clear if they should be considered waste or not waste. Art. 2.3 of the Directive assesses that "Sediments relocated inside surface waters are excluded from the scope of the Waste Directive when they are not hazardous and when they are relocated for the purpose of: managing waters and waterways, preventing floods, mitigating the effects of floods and droughts, land reclamation").
Directive 2008/105/EC	Directive of the European Parliament and of the Council on environmental quality standards in the field of water policy, amending and subsequently repealing Council Directives 82/176/EEC, 83/513/EEC, 84/156/EEC, 84/491/EEC, 86/280/EEC and amending Directive 2000/60/EC of the European Parliament and of the Council; it lays down environmental quality standards (EQS) for priority substances and certain other pollutants as provided for in Art. 16 of Directive 2000/60/EC, with the aim of achieving good surface water chemical status and in accordance with the provisions and objectives of Art. 4 of that Directive.

Table 1. International Conventions and Main EC Directives

Directive 2009/90/EC	Directive of the Commission laying down, pursuant to Directive 2000/60/EC of the European Parliament and of the Council, technical specifications for chemical analysis and monitoring of water status; it lays down technical specifications for chemical analysis and monitoring of water status in accordance with Article 8(3) of Directive 2000/60/EC. It establishes minimum performance criteria for methods of analysis to be applied by Member States when monitoring water status, sediment and biota, as well as rules for demonstrating the quality of analytical results.
Directive 2013/39/EU	Directive of the European Parliament and of the Council amending Directives 2000/60/EC and 2008/105/EC as regards priority substances in the field of water policy.
Directive 2018/851/EU	Directive of the European Parliament and of the Council of May 30th, 2018 amending Directive 2008/98/EC concerning wastes

Table 2.

Resume in chronological order of laws[1] that have directly addressed the topic of sediment management and SIN areas

LG.D. n. 99/1992	it implements Dir. 86/278/CEE applying the land-spreading regime of purified sludge in agriculture, in particular: conditions for use, bans, jurisdiction of the state and regions, authorizations and discipline, sludge and land analysis, penalis
LG.D. n. 22/1997:	it is the first legislation affecting environmental issues. Annex I reproduces the European Waste Catalogue (E.W.C.) identifying with the code 17 05 00 the dredged soil and materials and with the code 17 05 02 the dredged soil. This Decree has been abrogated and replaced by LG.D. n. 152/2006.
M.D. 05.02.1998:	it identifies non-hazardous waste that, according to LG.D. n. 22/1997 (today 152/2006), may be subject to simplified procedures for recovery (the recovery may be started 90 days after a simple communication to the competent Province).
Article 35 LG.D. n. 152/1999:	it regards water protection against pollution. It implements Directive 91/271/EEC concerning urban waste-water treatment and Directive 91/676/CEE concerning the protection of waters against pollution caused by nitrates from agricultural sources. Article 35 allows the immersion at sea, under authorization, of *«excavated materials from marine or salty bottoms or emerged coastal soils»*, provided that it is proved that there is the *«technical or economical impossibility to use them for the purposes of beach nourishment or recovery or alternative disposal»*. This LG.D. was also abrogated and replaced by LG.D. n. 152/2006, in particular art. 109 (see below).
M.D.n.468/2001	National remediation and environmental restoration program: it identifies, along with others, the SIN area of Leghorn
M.D.n.161/2002	Regulation implementing art. 31 and 32 of LG.D.n.22/1997, concerning the identification of hazardous waste that can be admitted to simplified procedures
M.D.24.02.2003	It perimeters the SIN area of Leghorn
M.D.n.269/2005	Regulation implementing art. 31 and 32 of LG.D.n.22/1997, concerning the identification of hazardous waste from ships, that can be admitted to simplified procedures

Table 2. Resume in chronological order of laws[1] that have directly addressed the topic of sediment management and SIN areas

Article 109 LG.D. n. 152/2006:	it repeals the mentioned Article 35. This provision has recently been modified by D.L. n. 5/2012 (it removed the condition concerning the *«technical or economical impossibility to use them for the purposes of beach nourishment or recovery or alternative disposal»*).
Article 1, par. 996 L. n. 296/2006:	it modifies Law n. 84/1994 (*Italian Law on ports*), introducing specific provisions for dredging operations to be conducted within *remediation Sites of National Interest* (in Italian S.I.N.). As reported in Article 5, dredging operations and remediation activities may be conducted concurrently on the base of a project approved by the competent authority not being detrimental to the site remediation; dredged materials may be, under authorization i) immersed at sea if their characteristics are similar to the background level of their original site, they are suitable for the destination site and they are not positive to eco-toxicity tests, otherwise ii) filled in coastal retaining structures if they are non-hazardous.
M.D. 7 November 2008:	it deals with technical provisions for dredging operations within *remediation Sites of National Interest* (S.I.N.). This decree have been partially modified by M.D. 4 August 2010 introducing in Annex A the new table A2 (chemical analyses to be conducted on port sediments about to be dredged and related thresholds).
D.L. 172/ 2008	(Converted in law by L. n.210/2008): extraordinary measures for the emergence of waste disposal in Campania and urgent measures for environmental protection.
Article 13 LG.D. n. 205/2010:	it has entirely modified Article 185 of LG.D. n. 152/2006 dedicated to exclusions from the waste legislation; paragraph 3 of Article 185 textually reproduces what provided by Directive 2008/98/EC: *«without prejudice to obligations under other relevant Community legislation, sediments relocated inside surface waters for the purpose of managing waters and waterways or of preventing floods or mitigating the effects of floods and droughts or land reclamation shall be excluded from the scope of Part Four of this Decree if it is proved that the sediments are non-hazardous pursuing Commission Decision 2000/532/EC»*.
Article 39, par. 13 of LG.D. n. 205/2010:	it specifies that the notion of *by-product* also applies *«to the material removed, exclusively for hydraulic security reasons, from the bed of rivers, lakes and creeks»*.

Table 2. Resume in chronological order of laws[1] that have directly addressed the topic of sediment management and SIN areas

L. n. 27/2012:	it has abrogated par. 11-bis to 11-sexies of Article 5 L. n. 84/94 replacing them with new Article 5-bis. According to par. 1-7, dredging operations of ports or marine-coastal areas within *remediation Sites of National Interest* (S.I.N.) and remediation activities may be conducted concurrently on the base of a project approved by the competent authority not being detrimental to the site remediation; dredged materials may be, under authorization, i) immersed in the same body of water if their characteristics are similar to the background level of their original site, they are suitable for the destination site and they are not positive to eco-toxicity tests, otherwise ii) reused on land pursuing the conditions of a dedicated ministerial decree if pollutants contained in them do not exceed certain thresholds, otherwise iii) filled in coastal retaining tanks if they are non-hazardous. According to par. 8 materials dredged from the bottom of ports outside a S.I.N. may be immersed at sea pursuing Article 109 of LG.D. n. 152/2006, otherwise they may be used for beach nourishment, even with spill in the part of the active submerged beach, or for the construction of coastal retaining structures in ports.
Art. 24 LG.D n. 5/2012 as converted into law by L. 35/2012:	it modifies Art. 109 of the LG.D. n.152/2006: it established that the competent body to authorize all possible management options for sediments dredged in a port not located inside a Contaminated Sites of National Relevance is the Region (with the exception of sea dumping for sea dumping inside the Italian marine protected areas, whose authorization is still released by the Ministry of the Environment).
M.D. n. 161/2012:	it regulates terms and conditions at which *excavated materials* may be reused as *by-products* and thus managed as *non-waste*.
M.D. n. 22/2013	Regulation governing the cessation of the status of waste of certain types of secondary solid fuels.
L. 98/2013	Contains provisions for further simplification for excavated earth and rocks deriving from small sites
M.D. 22.05.2014	New definition of the perimeter of the SIN area of Leghorn. Are excluded from the SIN area: the ground areas included the tanks of fill in the sea, the industrial channels, **the port areas within the breakwaters** and the areas between breakwaters and 3 kilometers from the coast which have got values <VI.

Table 2. Resume in chronological order of laws[1] that have directly addressed the topic of sediment management and SIN areas

LG.D n. 91/2014 as converted into law by L. 116/2014:	it introduces important changes in the regulation of dredging materials: new art. 184-quater of LG. D. n. 152/2006, which is specifically dedicated to the use of dredging materials.
LG.D n. 133/2014, as converted into law by L. 164/2014	Art. 7, Comma 8-bis: art. 185, comma 3, del D.lg. 152/2006, Exclusion from the regulations on waste of sediments moved within the hydraulic appurtenances.
LG.D. n.221/2015	Art.78, if the pollutants are below specific thresholds the sites are excluded from the SIN areas.
M.D. 264/16	regulation which provides indication to prove that a production residue is a by-product and not a waste
M.D. 351/16	procedure for deriving the reference values in marine and brackish areas inside the SIN.
M.D. 172/16	regulation on the rules of the methods and technical standards for dredging operations in SIN. It must be approved by 'Conferenza di servizi'.
M.D 173/16	regulation on methods and technical criteria for the immersion in sea of seabed excavation materials.
LG.D DPR 120/2017	the sediments were excluded from the list of "excavation materials" because included in other regulations.

From Waste to By-Product

LG.D. 152/2006 and subsequent amendments

•It has been subjected to a recovery operation including recycling and preparation for reuse;

•The substance is commonly used for specific purposed;

•the substance or object can be used directly without any further treatment other than normal industrial practice;

•There is a market or a demand for this substance;

•The substance or object meets the technical requirements for the specific purposes and complies with the existing legislation and standards applicable to the products;

•The use of the substance will not lead to overall negative impacts on the environment or on human health will be used, during the course of the same or a subsequent production or use process, by the manufacturer or third parties

Table 3 – Spanish regulation on dredging management

Law 22/1988, 28 July	of the Coastal law. The present law aims the determination, protection, use and police of maritime-terrestrial and especially seashore public domain. Article 56 establishes that discharges at sea from ships and aircraft shall be regulated by specific legislation, while Article 63 indicates that, in order to obtain the authorization for aggregates extraction and dredging projects, a previous study showing the potential impact on land and maritime public domain is required, regarding the place of extraction or dredging and discharged area as well. This law was amended by Law 2/2013, 29th of May.
Ministerial Order MAM 304/2002, 8 February	through which the dispositions for the recovery operations, dregs disposal and the European waste list are published.
Law 41/2010, 29 December	for the protection of the marine environment; its main objective was to achieve or maintain good environmental status of the marine environment at the latest in 2020, for which planning tools of marine environment are were created. The assessment of the environmental status of the marine sites should be carried out through 11 descriptors included in its Annex I, three of them related to dredging operations and product management of dredging at sea.
Royal Decree-law 60/2011, 21 January	regards the environmental quality standards in the context of water policy. This Royal Decree aims to: 1. Establish environmental quality standards (EQS) for priority substances and other contaminants listed in annex I in order to achieve good chemical status of surface water. 2. Establish EQS for priority substances listed in annex II and fix the procedure to calculate the EQS not contained in annexes I and II of contaminants of annex III in order to achieve good ecological status of surface waters or a good ecological potential of these waters, when proceed.

Table 3. Spanish regulation on dredging management

Law 22/2011, 28 July	is related to dregs and contaminated soil. This law aims at regulating the dregs management by promoting measures that prevent its generation and mitigate adverse impacts on human health and the environment associated with its generation and management, improving resources use efficiency. It also aims to regulate the legal status of contaminated soils.
Royal Legislative Decree-law 2/2011, 5 September	approves the revised text of the state ports law and merchant marine. The objectives of this law are: a) Determine and classify ports falling within the jurisdiction of the State General Administration. b) Regulate the planning, construction, organization, management, economic and financial regime and police them. c) Regulate the provision of services in these ports as well as their utilization. d) Determine the state port organization, providing ports of general interest of a regime of functional autonomy and management for the exercise the competences attributed by this law, and regulate the autonomous communities of Port authorities government. e) Establish the regulatory framework of the merchant marine. f) Regulate own administration of merchant marine. g) Establish the regime of infractions and sanctions applicable in the merchant marine area and in the state competency port. The Article 64, Paragraph 3 of this royal legislative decree-law, indicates that dredging projects will include a study of the management of products dredging, and in particular the dumping area location and its treatment, as well as, in general terms, the necessary studies from the environmental point of view.
Law 2/2013, 29 May	Protection and sustainable use of the coastal.

Table 3. Spanish regulation on dredging management

Royal Decree-law 817/2015, 11 September	established the criteria for monitoring and evaluating the surface water status and environmental quality standards. In order to protect water, this royal decree states: I. The basic and homogeneous criteria for the design and implementation of surface water masses state monitoring programs and for additional control of protected areas. II. The environmental quality standards (EQS) for priority substances and other contaminants in order to achieve good chemical status of surface waters. Establish EQS for priority substances and fix a procedure to calculate specific contaminants EQS in order to achieve good ecological status of surface waters or good ecological potential of these waters, when proceed. III. Reference conditions and limits class status indicators of biological, physicochemical and hydromorphological quality elements to classify the ecological or potential status of surface water masses. IV. Minimum standards for the information exchange concerning water status and quality between the state general administration and administrations with responsibility for water, aiming at compliance of laws that regulated the rights of access to information and public participation.
	Guidelines for the characterization of dredged material and its relocation in waters of the maritime-terrestrial public domain. 2015. Interministerial Commission on Maritime Strategies. Spain
	Guidelines for the characterization of dredged material and its relocation in waters of the maritime-terrestrial public domain. 2017. Interministerial Commission on Maritime Strategies. Spain

A1.3 An update (if advisable, and in relation to the findings of the previous task) of the list of the analysis to be performed on substrates and products (plants and fruits).

A1.4 Defining a common protocol for the analysis (sampling, extraction, quali- quantification, etc.).

A1.3 e A1.4
Procedures for **Olive** (grafted plantlets)
Cultivar: FRANTOIO
Experimental unit: 5 plants
Replicates: 3 **Treatments:** 5 (TS0 – TS25 - TS50 – TS75 - TS100) x 3 water regimes

Total plants = 225
Number of samplings: 20

1. Plants and leaves
Data collection (non-destructive analysis) on 20 grafted plantlets/treatment (or as differently indicated)
Recurrence: every 20 days from planting time (included) or as differently indicate in each parameter

Cultural practices: irrigation, pest control and fertilization to be performed as typically done in the area of production

1.1. Mortality (number; percentage)
1.2. Stem diameter
1.3. Stem height (tallest part in absolute)

1.4. Leaf surface (on 20 leaves from each cv-substrate plot) (early September)
1.5. Leaf blade colour (L, a, b coordinates) (on 20 leaves from cv-substrate plot) (early September)
1.6. Chlorophyll content (on 20 leaves from each plot in early September)

1.7. Nutritional foliar analysis (on one sample per cv-substrate) once soon after fruit harvesting: N, P, K, Ca, Mg, Na, B, Mn, Fe, Zn, Cu, Mo

1.8. Appearance of leaves

Pictures for each cv-substrate every 40? days

Data collection (destructive analysis) - Recurrence: only at the end of the trial
1.9. Fresh weight of the whole plant
1.10. Dry weight of the whole plant
1.11. Fresh weight of stem (aerial part)
1.12. Dry weight of stem
1.13. Fresh weight of root system
1.14. Dry weight of root system
1.15. Root system structure: pictures.

Analysis of contaminants (end of trial)
1.16. Heavy metals on roots
1.17. Heavy metals on stems
1.18. Heavy metals on leaves
1.19. Other contaminants (to be defined in relation to analytical results of sediments)

A1.3 e A1.4 BLUEBERRY

Cultivars: Duke and Bluecrop
Experimental unit: 4 plants
Replicates: 3
Treatments: 3 (TS0 - TS50 - TS100) x 3 water regimes
Total plants = 108 Duke + 108 Bluecrop

1. Plants and leaves

Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 30 days from planting time (included) or more

Cultural practices: irrigation, pest control and fertilization to be performed as typically done in the area of production

1.1. Plant mortality (number; percentage)
1.2. Plant width (widest part)
1.3. Plant height (tallest part))
1.4. Number of shoots (longer than 15 cm)
1.5. Leaf surface (on 10 leaves from each plot; 30 leaves per cv-substrate) soon after fruit harvesting
1.6. Leaf blade colour (L, a, b coordinates) (on 10 leaves from each plot; 30 leaves per cv-substrate) soon after fruit harvesting
1.7. Chlorophyll content (SPAD) (on 10 leaves from each plot; 30 leaves per cv-substrate) soon after fruit harvesting
1.8. Nutrient content (N, P, K, Ca and Mg) in the blueberry leaves (on one sample per cv-substrate) once soon after fruit harvesting.

Pictures for each cv-substrate at every monitoring

Data collection (destructive analysis) - Recurrence: only at the end of the trial

1.9. Fresh weight of the whole plant
1.10. Dry weight of the whole plant
1.11. Fresh weight of stem (aerial part)
1.12. Dry weight of stem
1.13. Length of root system
1.14. Fresh weight of root system
1.15. Dry weight of root system
1.16. Root system structure: pictures.

2. Phenology

2.1. Re-growth time (second year)
2.2. Flowering time (first flower - first two completely open flowers in one plant; peak of flowering - 50 % of completely open flowers; last flower)
2.3. Ripening time (first fruit, peak, last fruit)

A1.3 e A1.4 BLUEBERRY

3. Fruits

3.1. Total fruit production per plant (calculated by blocks)

Morphological and physical parameters.

Data on 10 fruits for each block (30 fruits per cv-substrate) during the period of maximum productive peak

3.2. Fruit fresh weight
3.3. Fruit dry weight
3.4. Maximum diameter
3.5. Shape in longitudinal section (UPOV - 21)
3.6. Depth of calyx basin (UPOV - 25)
3.7. Skin colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$
3.8. Flesh firmness (6 mm diameter plunger hand penetrometer)

Chemical parameters

(on juice of 10 fruits collected for each block at peak ripening time; 2 lectures)

3.9. pH
3.10. Titratable acidity (pH 8 with a 0.1 N solution of NaOH)
3.11. Total Solid Soluble (TSS)
3.12. Total polyphenols content
3.13. Total anthocyanin content
3.14. Antioxidant activity: FRAP, ABTS and DPPH
3.15. Organic acids (malic, citric, ascorbic, tartaric)
3.16. Sugars (glucose, fructose, sucrose)

3.17. Analysis of organic and inorganic contaminants

Pictures comparing fruits from treatments with ruler, entire and in longitudinal section

4. Sensorial analysis

4.1. Official panel test to be performed only after the analysis of contaminants and after discussion. Methods: ISO 3972:2011 and ISO 5496:2006.
4.2. Visual and olfactory analysis for each fruit sample.

A1.3 e A1.4
Procedures for Woodland Strawberry
(plant and fruit evaluation)

Cultivar: REGINA DELLE VALLI
Experimental unit: 5 plants
Replicates: 3
Treatments: 3 (TS0 - TS50 - TS100) x 3 water regimes
Total plants = 135

1. Plants and leaves
Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 30 days from planting time (included) or more

Cultural practices: irrigation, pest control and fertilization to be performed as typically done in the area of production (Murcia and Tuscany); stolons to be removed as soon as they appear

1.1. Plant mortality (number; percentage)
1.2. Crown diameter
1.3. Plant height (tallest part in absolute)
1.4. Number of fully expanded leaves

1.5. Leaf surface (on 10 leaves from each plot; 30 leaves per cv-substrate) soon after fruit harvesting
1.6. Leaf blade colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (on 10 leaves from each plot; 30 leaves per cv-substrate) soon after fruit harvesting
1.7. Chlorophyll content (on 10 leaves from each plot; 30 leaves per cv-substrate) soon after fruit harvesting
1.8. Nutritional foliar analysis (on one sample per cv-substrate) once soon after fruit harvesting (N, P, K, Ca, Mg).

Pictures for each cv-substrate at every monitoring

Data collection (destructive analysis) - Recurrence: only at the end of the trial
1.10. Fresh weight of the whole plant
1.11. Dry weight of the whole plant
1.12. Fresh weight of stem (aerial part)
1.13. Dry weight of stem
1.14. Length of root system
1.15. Fresh weight of root system
1.16. Dry weight of root system
1.17. Root system structure: pictures.

2. Phenology
2.1. Re-growth time (second year)
2.2. Flowering time (first flower - first two completely open flowers in one plant; peak of flowering - 50 % of completely open flowers; last flower)
2.3. Ripening time (first fruit, peak, last fruit)

A1.3 e A1.4

3. Fruits

3.1. Total fruit production per plant (calculated by blocks: total weight of one block/5)

Morphological and physical parameters.

Data on 10 fruits for each block (30 fruits per cv-substrate) during the period of maximum productive peak. Select primary or secondary fruits.

3.2. Fruit fresh weight
3.3. Fruit dry weight
3.4. Maximum diameter
3.5. Fruit - Shape (UPOV - 30)
3.6. Fruit - Evenness of skin colour (UPOV - 33)
3.7. Fruit - Evenness of surface (UPOV - 35)
3.8. Fruit cavity (UPOV - 45)
3.9. Skin colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$
3.10. Flesh firmness (durometer or 6 mm diameter plunger hand penetrometer)

Chemical parameters (on juice of 10 fruits collected for each block at peak ripening time; 2 lectures)

3.11. pH
3.12. Titratable acidity (pH 8 with a 0.1 N solution of NaOH)
3.13. Total Solid Soluble (TSS)
3.14. Total polyphenols content
3.15. Antioxidant activity: FRAP, ABTS and DPPH
3.16. Organic acids (malic, citric, ascorbic)
3.17. Sugars (glucose, fructose, sucrose)
3.18. Analysis of organic and inorganic contaminants

Pictures comparing fruits from treatments with ruler, entire and in longitudinal section

4. Sensorial analysis

4.1. Official panel test to be performed only after the analysis of contaminants and after discussion. Methods: ISO 3972:2011 and ISO 5496:2006.
4.2. Visual and olfactory analysis for each fruit sample.

Alloro, Laurel

Laurus nobilis (Fam. Lauraceae)

Lauro, Cherry-laurel

Prunus laurocerasus (Fam. Rosaceae)

Action A1.3 e A1.4

Procedures for *Prunus laurocerasus* (rooted cuttings)

Common name: Cherry-Laurel

Cultivar: Novita

Experimental unit: 20 cuttings=10 pots (2 rooted cuttings per 6 L pot)

Replicates: 3

Treatments: 7 (PB100 – PB75/TS25 – PB50/TS50 – CF75/TS25 – CF50/TS50 – WF75/TS25 – WF50/TS50) x 3 water regimes

TS - Treated Sediment **PB** - Peat Based Commercial Substrate **CF** - Coconut Fiber **WF** - Wood Fiber

Total plants: 1260 cuttings = 630 pots

Planting period: end of winter

Cultivation period: 1 year

Cultivation environment: open air

Cultural practices: pest control and fertilization to be performed as typically done in the area of production (Tuscany), except for irrigation, to be differentiated in 3 different regimes

Expected results: 1260 laurel rooted cuttings growing and developing saplings

1. Plants and leaves

Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 30 days from planting time (included) or more

1.1. Plant mortality (number; percentage)
1.2. Base stem diameter (every 3 months)
1.3. Maximum plant height (every 3 months)
1.4. Number of vegetative sprouts (every 3 months)
1.5. Length of vegetative sprouts (every 3 months)
1.6. Number of fully expanded leaves on vegetative sprouts
1.7. Leaf blade colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 leaves per each treatment replicate)
1.8. Photosynthetic activity by CIRAS-2 (on 10 leaves per each treatment replicate)

Data collection (destructive analysis) - Recurrence: only at the end of the trial on 10 samples per each treatment replicate

1.9. Leaf area (on 5 leaves per plant)
1.10. Chlorophyll content
1.11. Fresh weight of the whole plant
1.12. Dry weight of the whole plant
1.13. Fresh weight of stem (aerial part)
1.14. Dry weight of stem
1.15. Length of root system
1.16. Fresh weight of root system
1.17. Dry weight of root system

Pictures for each cv-substrate at every monitoring

Expected results:

- Typization of the dose-effect relationship, aimed at assessing the range of sediment % which can be added to a growing substrates in horticultural nursing.
- Validation of the use of remediated sediments for horticultural nursing.
- Waste management: amount (%) of reduction of the use of peat and its substitution with treated sediments (10-20 % of substitution are expected).
- Reduction of CO2 emission (kg) due to the substitution of peat by treated sediments (expected to be of about 80-90% less).

A1.3 e A1.4 non food crops cultivation (from plantlets to final production: flowers/ornamental)

1) Procedures for *Zantedeschia aethiopica* (cut flower production)

Common name: Calla lily
Experimental unit: 30 rhizomes per bench, at a distance of 50 cm (21 m^2 surface, 20 cm depth)
Replicates: 3
Treatments: 3 (PB100 – PB75/TS25 – PB50/TS50)
Total plants: 270
Planting period: end of summer
Cultivation period: 2 vegetative cycles (2 years)

Cultivation environment: greenhouse
Cultural practices: pest control and fertilization to be performed as typically done in the area of production (Tuscany), except for irrigation, to be differentiated in 3 different regimes

Expected results: 270 calla lily plants developing and flowering

1. Procedures for *Zantedeschia aethiopica*

A1.3 e A1.4 non food crops cultivation (from plantlets to final production: flowers/ornamental)

1. Plants and leaves

Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 30 days from planting time (included) or more

1.1. Plant mortality (number; percentage)
1.2. Number of stems
1.3. Maximum stem height
1.4. Number of flowers
1.5. Length of spathe
1.6. Spathe colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 flowers per each treatment replicate)
1.7. Leaf blade colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 leaves per each treatment replicate)
1.8. Photosynthetic activity by CIRAS-2 (on 10 leaves per each treatment replicate)

Data collection (destructive analysis) - Recurrence: only at the end of the trial on 10 samples per each treatment replicate

1.9. Leaf area (on 5 leaves per plant)
1.10. Chlorophyll content
1.11. Fresh weight of the whole plant
1.12. Dry weight of the whole plant
1.13. Fresh weight of stem (aerial part)
1.14. Dry weight of stem
1.15. Length of root system
1.16. Fresh weight of root system
1.17. Dry weight of root system
1.18. Malondialdehyde (MDA) analysis for oxidative stress
1.19. Heavy metal analysis (2 replicates per each treatment)

2. Phenology

2.1. Re-growth time (second year)
2.2. Flowering time (first flower - first two completely open flowers in one plant; peak of flowering - 50% of completely open flowers; last flower)

Pictures for each treatment at every monitoring

A1.3 e A1.4 non food crops cultivation (from plantlets to final production: flowers/ornamental)

2) Procedures for *Protea cynaroides* (flower pot plant)

Common name: King protea
Cultivar: Little Prince
Experimental unit: 10 plants (1 rooted cuttings in 2,8 L pot)
Replicates: 3
Treatments: 7 (PB100 – PB75/TS25 – PB50/TS50 – CF75/TS25 – CF50/TS50 – WF75/TS25 – WF50/TS50) x 3 water regimes
TS - Treated Sediment **PB** - Peat Based Commercial Substrate **CF** - Coconut Fiber **WF** - Wood Fiber

Total plants: 630
Planting period: spring
Cultivation period: 2 vegetative cycles (2 years)

Cultivation environment: greenhouse
Cultural practices: pest control and fertilization to be performed as typically done in the area of production (Tuscany), except for irrigation, to be differentiated in 3 different regimes

Expected results: 630 protea plants growing and flowering

A1.3 e A1.4 non food crops cultivation (from plantlets to final production: flowers/ornamental)

2. Procedures for *Protea cynaroides*

1. Plants and leaves

Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 30 days from planting time (included) or more

1.1. Plant mortality (number; percentage)
1.2. Number of stems with flowers
1.3. Base diameter of stems
1.4. Maximum stem height
1.5. Number of flowers
1.6. Shape of inflorescence
1.7. Size of inflorescence
1.8. Colour of flower cluster (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 flowers per each treatment replicate)
1.9. Colour of surrounding bracts (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 bracts per each treatment replicate)
1.10. Colour of paddle-shaped leaves (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 leaves per each treatment replicate)
1.11. Photosynthetic activity by CIRAS-2 (on 10 leaves per each treatment replicate)

Data collection (destructive analysis) - Recurrence: only at the end of the trial on 10 samples per each treatment replicate

1.12. Leaf area (on 5 leaves per plant)
1.13. Chlorophyll content
1.14. Fresh weight of the whole plant
1.15. Dry weight of the whole plant
1.16. Fresh weight of stem (aerial part)
1.17. Dry weight of stem
1.18. Length of root system
1.19. Fresh weight of root system
1.20. Dry weight of root system
1.21. Malondialdehyde (MDA) analysis for oxidative stress
1.22. Heavy metal analysis (2 replicates per each treatment)

2. Phenology

2.1. Re-growth time (second year) ???
2.2. Flowering time (first flower - first two completely open flowers in one plant; peak of flowering - 50 % of completely open flowers; last flower)

Pictures for each cv-substrate at every monitoring

A1.3 e A1.4 non food crops cultivation (from plantlets to final production: flowers/ornamental)

3 Procedures for *Prunus laurocerasus* (ornamental)

Cultivar: Novita
Experimental unit: 10 plants (one-year-old rooted cuttings in 10 L pot)
Replicates: 3
Treatments: 7 (PB100 – PB75/TS25 – PB50/TS50 – CF75/TS25 – CF50/TS50 – WF75/TS25 – WF50/TS50) x 3 water regimes
TS - Treated Sediment **PB** - Peat Based Commercial Substrate **CF** - Coconut Fiber **WF** - Wood Fiber

Total plants: 630
Planting period: spring
Cultivation period: 2 growing cycles ??

Cultivation environment: open air
Cultural practices: pest control and fertilization to be performed as typically done in the area of production (Tuscany), except for irrigation, to be differentiated in 3 different regimes

Expected results: 630 laurel saplings growing and developing

A1.3 e A1.4 non food crops cultivation (from plantlets to final production: flowers/ornamental)

3. Procedures for *Prunus laurocerasus*

1. Plants and leaves

Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 30 days from planting time (included) or more

1.1. Plant mortality (number; percentage)
1.2. Base stem diameter (every 3 months)
1.3. Maximum plant height (every 3 months)
1.4. Number of vegetative sprouts (every 3 months)
1.5. Length of vegetative sprouts (every 3 months)
1.6. Number of fully expanded leaves on vegetative sprouts
1.7. Leaf blade colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 leaves per each treatment replicate)
1.8. Photosynthetic activity by CIRAS-2 (on 10 leaves per each treatment replicate)

Data collection (destructive analysis) - Recurrence: only at the end of the trial on 10 samples per each treatment replicate

1.9. Leaf area (on 5 leaves per plant)
1.10. Chlorophyll content
1.11. Fresh weight of the whole plant
1.12. Dry weight of the whole plant
1.13. Fresh weight of stem (aerial part)
1.14. Dry weight of stem
1.15. Length of root system
1.16. Fresh weight of root system
1.17. Dry weight of root system
1.18. Malondialdehyde (MDA) analysis for oxidative stress
1.19. Heavy metal analysis (2 replicates per each treatment)

2. Phenology

2.1. Re-growth time
2.2. Sprouting time

Pictures for each cv-substrate at every monitoring

Expected results:

- Typization of the dose-effect relationship, aimed at assessing the range of sediment % which can be added to a growing substrates in horticultural nursing
- Validation of the use of remediated sediments for horticultural nursing
- Waste management: amount (%) of reduction of the use of peat and its substitution with treated sediments (10-20 % of substitution are expected)
- Reduction of CO2 emission (kg) due to the substitution of peat by treated sediments (expected to be of about 80-90% less).

A1.3 e A1.4 Food crop production

Procedures for *Ocimum basilicum* (seeds)

Common name: Basil
Cultivars: 2
Experimental unit: 40 pots ~ 800 putative plantlets (20 seeds in 0.75 L pot)
Replicates: 3
Treatments: 3 (PB100 – PB50/TS50 – TS100)
Total seeds: 14.400
Planting period: end of summer
Cultivation period: 2 months

Cultivation environment: open air
Cultural practices: pest control and fertilization to be performed as typically done in the area of production (Tuscany), except for irrigation, to be differentiated in 3 different regimes

Expected results: 14.400 basil seeds sowed

A1.3 e A1.4 Food crop production

Procedures for *Ocimum basilicum*

1. Plants and leaves

Data collection (non-destructive analysis) on all the plants (or as differently indicated)
Recurrence: every 7 days from planting time (included) or more

1.1. Seed germination (number; percentage)
1.2. Plant mortality (number; percentage)
1.3. Number of leaves

Data collection at the growth stage of 2-4 couple of true leaves (end of demonstration) on 10 samples per each cv-substrate replicate

1.4. Number of leaves
1.5. Maximum plant height
1.6. Leaf blade colour (L, a, b coordinates) and chroma index $(a^2 + b^2)^{1/2}$ (10 leaves per each cv-substrate replicate)
1.7. Leaf area (on 5 leaves per plant)
1.8. Chlorophyll content
1.9. Fresh weight of the whole seedling
1.10. Dry weight of the whole seedling

Chemical parameters

3.11. Total polyphenols content
3.12. Antioxidant activity: FRAP, ABTS and DPPH
3.13. Organic acids (malic, citric, ascorbic)
3.14. Sugars (glucose, fructose, sucrose)
3.15. Essential oil content
3.16. Analysis of organic and inorganic contaminants

Pictures for each cv-substrate at every monitoring

Expected results:

- Typization of the dose-effect relationship, to assess the range of sediment % which can be added to typical substrate for food crops propagation/cultivation
- Good chemivcal and organoleptic quality of products
- Full marketability of food crops products
- Waste management: amount (%) of peat use reduction and its substitution with treated sediments (10-20 % of substitution is expected)
- Reduction of CO_2 emission (kg) of the whole production cycle due to the substitution of peat with treated sediments (expected to be 80-90% less)

ACTION B.1

Phytoremediated Sediment treated via landfarming process
ISECNR

Expected Start Date: IV 2018
Expected End Date: I 2019
Actual Start Date: IV 2018
Actual End Date: I 2019

ACTION C.1

Monitoring and validation of treated sediments
ISECNR

Expected Start Date: I 2019
Expected End Date: II 2019
Actual Start Date: IV 2018

ACTION C.5

Performance indicators monitoring FLORA TOSCANA and all the partners

This monitoring activities consists of measuring the efficiency of the performance indicators defined in the attached LIFE SUBSED project specific indicators excel document, in order to fulfil the environmental and social viability of the LIFE SUBSED process.

The group of indicators will be revised in each of the projects coordination meetings in order to check any irregularities.

Subaction C.5.1 KPI Webtool

All partners will monitor and measure during all the project duration the LIFE SUBSED performance indicators in order to support the coordinating beneficiary FLORA TOSCANA in updating and reporting the KPI Webtool upon throughout the project implementation.

Expected Start Date: IV 2018

Expected End Date: III 2021

FUTURE ACTIONS

B.2

Demonstration of the use of remediated sediments as a substrate for nursery production

B.3

Demonstration of the use of remediated sediments as substrate for non food crops cultivation (from plantlets to final production: Flowers - ornamental)

B.4

Demonstration of the use of remediated sediments as a substrate for food crops production

FLORA TOSCANA and all the partners

Expected Start Date: II 2019
Expected End Date: I 2021

C.2
Monitoring and validation of the use of remediated sediments as a substrate for plant nursing and cultivation: non food crops production

C.3
Monitoring and validation of the use of remediated sediments as a substrate for nursing and cultivation: food crops production

CarbonSink, CREA, UMH

Expected Start Date: III 2019
Expected End Date: II 2021

C.4

Monitoring of socio-economic impact of the project and LCA
CarbonSink, FLORA TOSCANA and all the partners

Expected results:

- socio-economic impact of the project
- LCA document with the elaboration and analysis of project data in terms of technical and environmental impact of the project process

Expected Start Date: IV 2020
Expected End Date: III 2021

B.5

Training courses, workshops and guidelines for project replicability and transferability

Expected Start Date: IV 2020
Expected End Date: III 2021

B.6

SUBSED Business Plan

Expected Start Date: I 2021
Expected End Date: III 2021

FLORA TOSCANA and all the partners