Chapter 3

WP3 3.4.1, 3.4.2, 3.4.3 MONITORING DATA (water, soil, crops) – ARBOREA (Italy)

WP3 Activity 3.4 Comparing baseline value with final data of monitoring water, soil and crops

Enas, in accordance with the current specifications of the Monitoring Plan for the Implementation of the Action Programme of the NVZ of Agricultural Origin and in collaboration with all the public bodies operating in the territory, carried out monitoring of the waters, the soils and crops before and after the construction of the plant in the proximity of 2 piezometers of new realization for testing water impact and give homogeneity to the general monitoring

3.4.1 Water monitoring and 3.4.4 Comparing baseline value with final data

Historical analysis of Arborea NVZ (ARPAS Activity 2018)

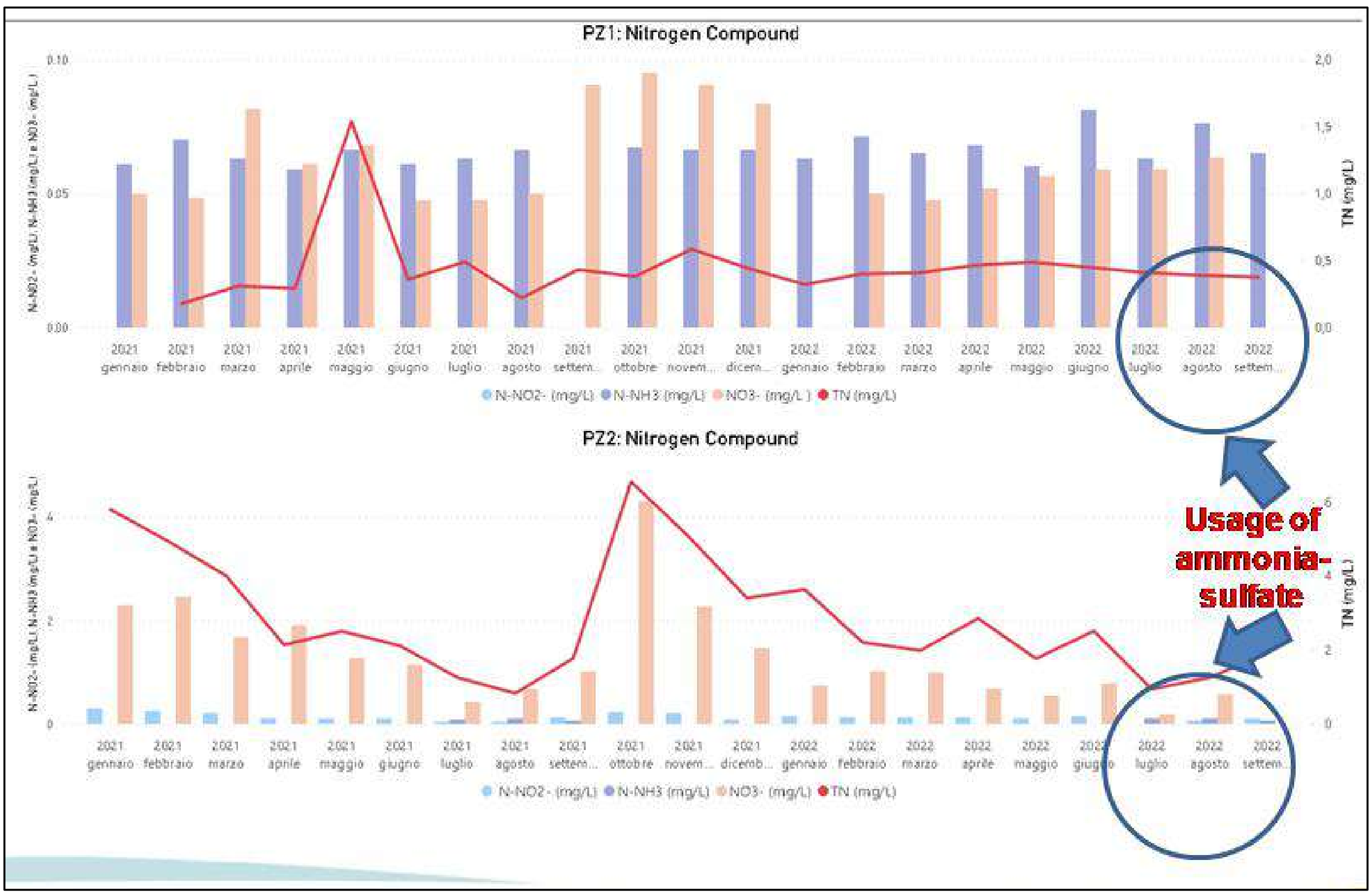
Historical analysis of NVZ site of Arborea is based on 48 monitoring cycles (March 2006-February 2019) focalizing on the nitrate compound content dynamic in the first aquifer, the reason that considers this area is a Nitrate Vulnerable Area (NVZ) under EU Directive 91/676/EEC that indicate the upper limit permitted by EU legislation in 50 mg/L NO3-. The analysis of historical data showed a lack of a seasonal trend, highlighting the punctual pollution of nitrate with marked variability in time and space. Conductivity data (EC), chloride, and sulfate are useful to detect a saline intrusion, one of the main issues in coastal plains like Arborea, till 2015 a stability of values was recorded in both aquifers (1500-2000 µS/cm in the I and 2000-3000 µS/cm in the II). Since 2016 in the Sassu aquifer chloride is often over 1000 mg/L, the same trend was observed for sulfates, over 250 mg/L in some points, showing a trend mainly related to the proximity to the sea and irrigation practices applied for the crops.

Parameter	Method apply	Unit
Temperature	APAT CNR IRSA 2100	°C
pH	APAT/IRSA- CNR 2060	
Conductivity a 25 °C	ISS.BDA.022.rev00	microS/cm
Dissolved Oxigen	HACH LDO METHOD 10360	mg/L
Turbidity	APAT/IRSA- CNR 2110	NTU
Alcalinity	APAT/IRSA- CNR 2010 Metodo B	meq/L
Eh	ISO metodo 11271:2002	mV
Solid resudue 180 °C	APAT/IRSA- CNR 2090 Metodo A	mg/L
Bicarbonate	APAT/IRSA- CNR 2010 Metodo B	mg/L HCO_3^-
Tot Hardness	APAT/IRSA- CNR 2040 Metodo A	°F
Calcium	ISS.CBB.038.rev00	mg/L
Magnesium	ISS.CBB.038.rev00	mg/L
Potassium	ISS.CBB.038.rev00	mg/L
Sodium	ISS.CBB.038.rev00	mg/L
Chlorides	ISS.CBB.037.rev00	mg/L
Nitrates	ISS.CBB.037.rev00	mg/L NO_3^-
Solfates	ISS.CBB.037.rev00	mg/L SO_4^{2-}
Total nitrogen	APAT/IRSA- CNR 4060	mg/L N
Ammonia	APAT/IRSA- CNR 4030 - Metodo A1	mg/L N-NH_3
Nitric Nitrogen	APAT/IRSA- CNR 4050	mg/L N-NO_2^-
Total phosphorus	APAT/IRSA- CNR 4060	mg/L P
Reactive Phosphorus	APAT/IRSA- CNR 4110 Metodo A1	mg/L P-PO_4^{3-}
TOC	APAT/IRSA- CNR 5040	mg/L
Alluminium	EPA 6020b	microg/L
Dissolved alluminium	EPA 6020b	microg/L
Iron	EPA 6020b	microg/L
Dissolved Iron	EPA 6020b	microg/L
Manganese	EPA 6020b	microg/L
Dissolved Manganese	EPA 6020b	microg/L
Arsenic	EPA 6020b	microg/L
Dissolved Arsenic	EPA 6020b	microg/L
Cadmium	EPA 6020b	microg/L
Dissolved Cadmium	EPA 6020b	microg/L
Chromium	EPA 6020b	microg/L
Dissolved Chromium	EPA 6020b	microg/L
Mercury	EPA 6020b	microg/L
Dissolved Mercury	EPA 6020b	microg/L
Nichel	EPA 6020b	microg/L
Dissolved Nichel	EPA 6020b	microg/L
Lead	EPA 6020b	microg/L
Dissolved Lead	EPA 6020b	microg/L
Copper	EPA 6020b	microg/L
Dissolved Copper	EPA 6020b	microg/L
Zinc	EPA 6020b	microg/L
Dissolved Zinc	EPA 6020b	microg/L

The water monitoring was carried out by ENAS labs from January 2021 to September 2022, in 2 piezometers placed in the experimental fields where Ammonium-sulfate produced in the pilot-plant was tested (Photos 1-5), by multiparametric sensors and pumping for purge (Photos 6,7) in field and by laboratory analysis. The water parameters investigated are listed in the table reported below.

The results show small variability in all parameters in particular in Pz1 compared to Pz2. Summarizing the main results of the comparison are E. C. [µS/cm at 25°C] (1020 ± 78.6 in Pz1; 264.5 ± 60.4 in Pz2), Chlorides [mg/L] (168.2 ± 3.61 in Pz1 - 301.8 ± 18.8 in Pz2); Hardness [°F] (15.5 ± 0.5 in Pz1 – 57.8 ± 2.7 in Pz2 ; Nitrates [mg/L NO3-](0.02 ±0.02 in Pz1 – 15.5 ± 0.6 in Pz2); TOC [mg/L] (5.7 ± 0.7 in Pz1; 16.2 ± 6.8 in Pz2).
The monitoring results highlighted a quite stability of water parameters during our investigation in both piezometers compared to historical data recorded in the Arborea plane.

Moreover, the ammonia sulfate produced in the pilot plant was tested in particular on 2 crop fields where were placed the 2 piezometers, with the final goal to evaluate the effect of this fertilizer on groundwaters. Ammonia sulfate was applied during the summer months (July, August, and September 2022), to prevent a dilution effect.
These results, reported in the chart below, are very positive, suggesting that ammonia sulfate unaffected the nitrate concentration in groundwater, reaching one of the main goals of the project.







Photos 1-5 - Piezometers n.1, 2 and the water sampling


Photos 6,7 - Purge pump   Multiparametric case for water quality monitoring

3.4.2,3.4.3 Soil and crops monitoring and 3.4.4 Comparing baseline value with final data

The soil and crops campaign and data elaboration will be carried out by Enas, Laore Regional Agency and Cooperative of Arborea beneficiaries of the results of the project, by its agronomists who followed the vegetation monitoring on field.

The results will be shared and check with the results and the studies being carried out by the regional public-bodies.

About the crops monitoring the Cooperative of Arborea and the Livegreen company both stakeholders of the project, are involved on the activity of experimentation on the crops.

First we will test the fertilizer produced on field crops cultivated with oats (forage for breeding) on an area of about 20 ha as an example to test the reaction and absorption capacity of the soil according to the vegetational cycles. We will test also the ammonium sulphate produced on the cultivation of spirulina algae in greenhouses, an internationally known reality hosted in the Arborea Cooperative and very important for its multiple uses of the algae both in the food sector (also used in third countries such as Africa for the important load energetic), cosmetic, medicinal.

As for the field experimentation, it is carried out on three plots located two in the installation area of the piezometers and in greenhouse: on spirulina alga cultivation.

The monitoring is focused on plant growth level and state of health respect to the previous state before the new fertilizer utilization.

Soil monitoring

The sampling parameters and specifications start from the general indications of ARPAS (Regional Environmental Agency) and the sampling frequency will be carried out annually as indicated in the summary table below.

On September 2022, the soil survey campaign necessary for the recognition of any anomalies after the sulphate of ammonium treatment took place at the Arborea pilot sites.
A survey was carried out on topsoil consistent with that carried out before the installation of the stripping system, the data of which were just implemented in the GIS.
Three representative sites of the study area were identified near the 2 piezometers (n.1 and n.2) built within the MEDISS project and another near the plant.
All were treated with the ammonium sulphate produced in the experiment in the period May-August identified on the basis of the growth phase of the crop (corn).
Three representative sites of the study area were identified in the vicinity of the 2 piezometers (1 and 2) created as part of the MEDISS project and another near the plant.
All were treated with the ammonium sulphate produced in the experiment in the period May-August identified on the basis of the growth phase of the crop (corn) sowing and harvesting.

Matrix	Frequency	Parameters
Soil	Yearly	pH, Conductivity, Organic C. , Tot N,P.Ass Metals (Cu, Zn, As, Cd, Cr, Ni, Pb, Fe, Mn, V)

On the samples relating to 2 piezometer and the one near the plant, given the specificity of the soil, a sampling was carried out on the entire 5 * 5 mesh area (station n. 291_bis) and 4 * 4 (station n.201_bis) and prepared a composite sample. The analyzes were carried out in duplicate between Laore and Enas for a better validation of the data.

The analyzes are being carried out according to the scheme envisaged in the MEDISS monitoring program and the results will soon be verified in relation to the known situation pre-installation of the system.
The sampling was carried out after the mowing and harvesting of the corn grown for animal feed of the numerous heads owned by the Arborea Producers Cooperative.
On each site a descriptive form has been compiled, the data of which will form an integral part of the GIS of the MEDISS project.
The following images show the samplings carried out in the three representative sites (Fig. 1,2,3,4).

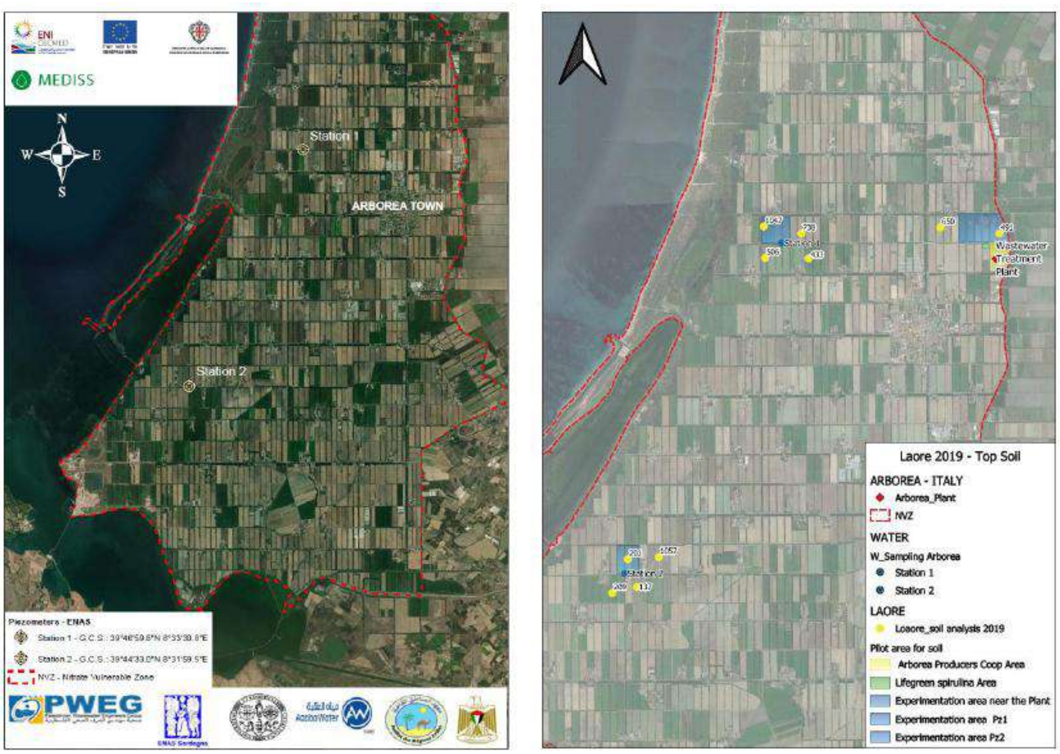


Fig. 1- Piezometer sites on the left and soil sampling points location on the right.

Fig. 2 - Soil Profile 201_bis (composite sample) representative areal Piezometer 2 treated with ammonium sulphate fertilizer

Fig. 3 - Soil Profile 1047_bis Areal representative profile 1047_bis Piezometer 1 treated with ammonium sulphate fertilizer

Fig. 4 - Soil profile 491_bis representative of the area close to the stripping plant always treated with fertilizerSoil profile 491_bis representative areal next to the stripping plant always treated with fertilizer

SOIL monitoring - Field experimentation with ammonium sulphate from bovine manure, produced in the pilot plant

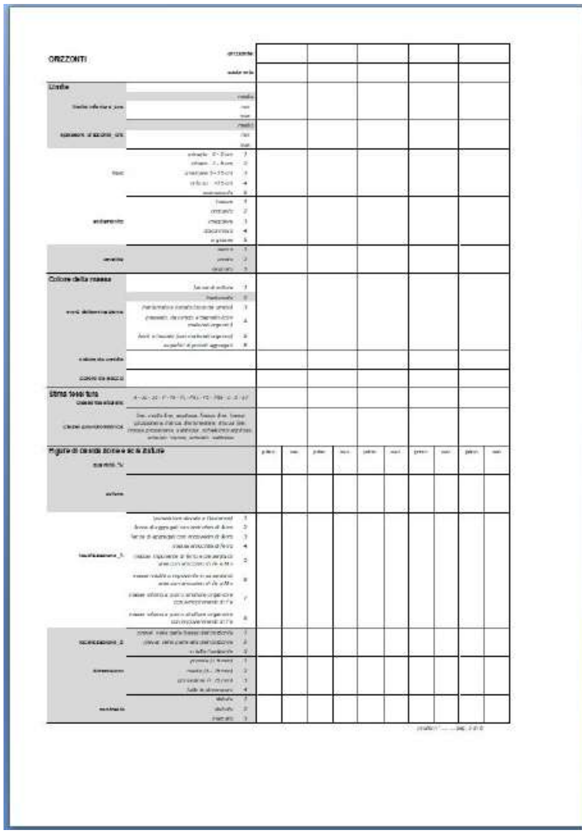
Results of soil campaigns

With the help of the pedological laboratory of Laore, the soil samples taken during the campaign on the pilot sites of Arborea were analyzed after the use of the fertiliser produced by pilot plant (the old campaign before the plant was built are summerized like:

– the total nitrogen shows in all cycles the lack of high values, with normal values for sandy soils and averages in the various cycles between 0.7 and 1.3 g/kg, only some samples of a cycle recorded values up to 20 g/kg;

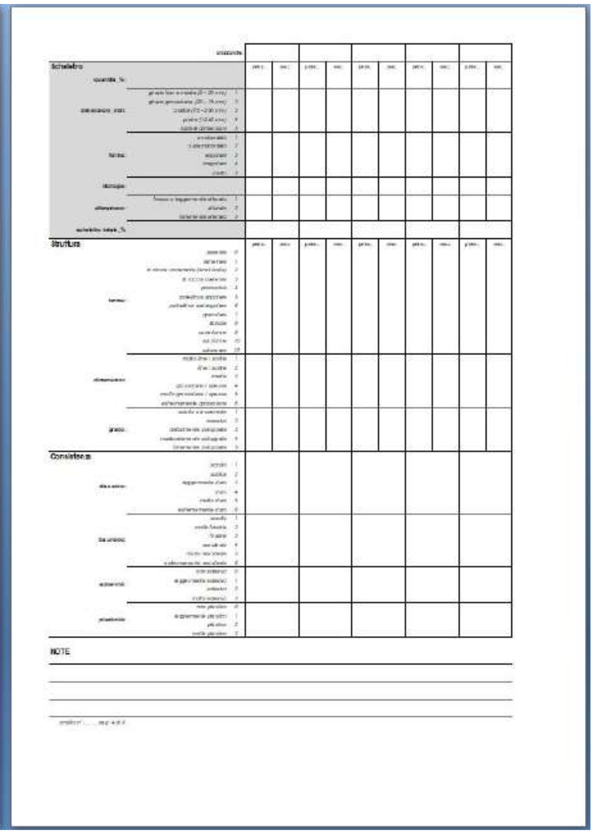
- assimilable phosphorus always shows values higher than the normal values for sandy soils cultivated with similar crops (values between 5-18 mg/kg). Exceedings of the limit value imposed for the NVZ of 200 mg/kg have been recorded, with peaks of even 300-380 mg/kg. In recent years, the phosphorus trend has risen with higher average values than in previous years;
- the organic carbon, linked to the available organic substance, shows oscillations linked to the phases of spreading of animal manure and to the periods of suspension. In general, in coincidence with the spreading periods, the soils show a greater endowment of organic carbon;
- in the periods when manure spreading is prohibited (between November 15th and February 15th), almost half of the samples show an insufficient endowment; metals are contained overall. In fact, the values imposed by Legislative Decree 152/2006 for green areas have never been exceeded, nor have the limits for copper and zinc been exceeded as part of the Action Program for the NVZ;
  soil conductivity is overall quite low, with average values around 0.66-0.51 mS/cm, a completely natural fact if we consider that the samplings concerned only the first 40 cm of surface soil, in most part of the cases consists of porous and very permeable sands, therefore the leaching processes can cause a downward leaching of the salts.

The following form was used for the new soil campaign:


ORIZZONTI

Colore della massa

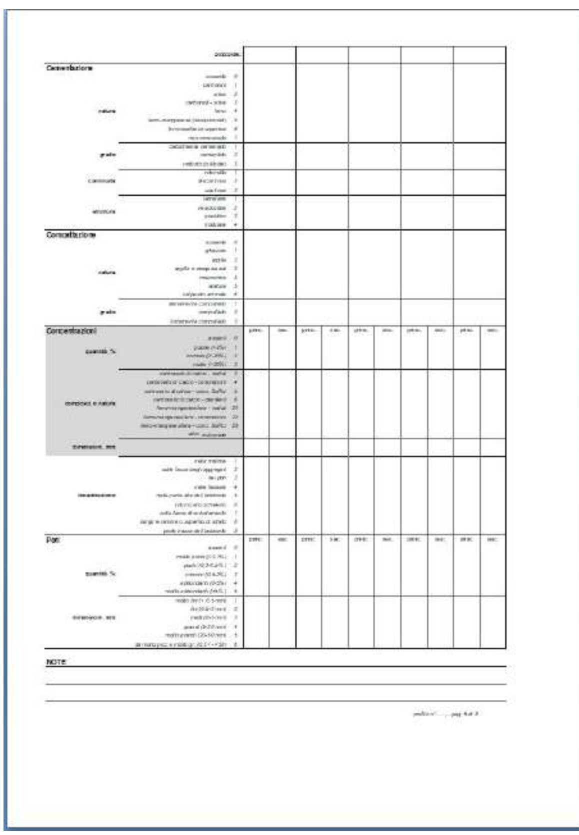
Stima tessitura


Scheletro

Struttura

Consistenza

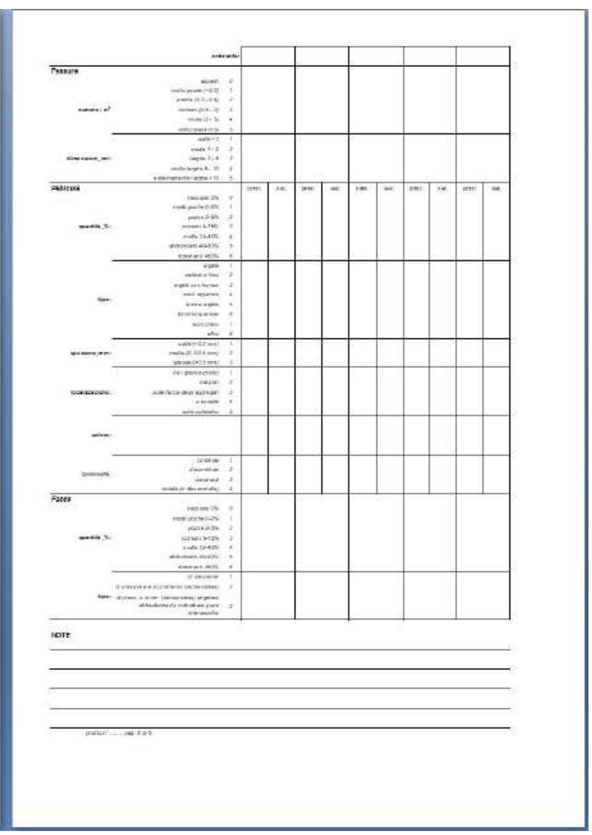
NOTE


Cementazione

Concentrazioni

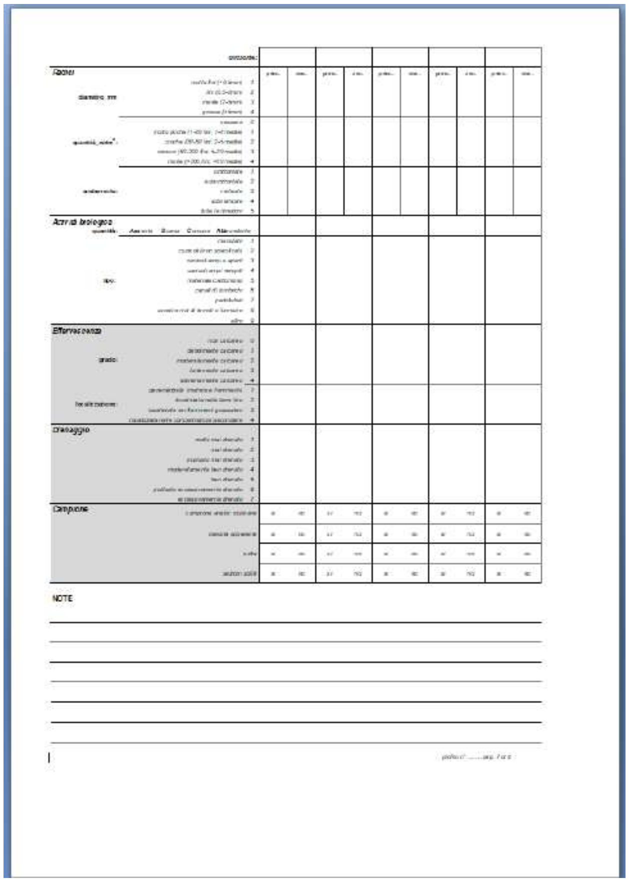
Pori

NOTE


Pellicole

Facce

NOTE



pedonlab

Rapporto di Prova: 2206228
Codice archivio: APJ00038

Latina, 03/10/2022
Codice Cliente: LA8325SA

ANALISI DEL TERRENO

INFORMAZIONI CLIENTE

Azienda/Cliente	LAORE SARDEGNA AG. REGIONALE
Indirizzo	Via Caprera, 8
C.A.P.	09100
Località	CAGLIARI
Provincia	CAGLIARI

IDENTIFICAZIONE CAMPIONE

Identificazione	c1047 bis trattato Arborea Az Pozzato str 21 ovest
Coltura	Non specificata
Area ecologica	Non specificata
Tipo suolo	Non specificato

INFORMAZIONI SUL PRELIEVO

Prelevatore	Cliente
Data Prelievo	20/09/2022

INFORMAZIONE SUL RICEVIMENTO

Data Arrivo	27/09/2022

PROVA ANALITICA

Data inizio Analisi	27/09/2022
Data fine Analisi	30/09/2022

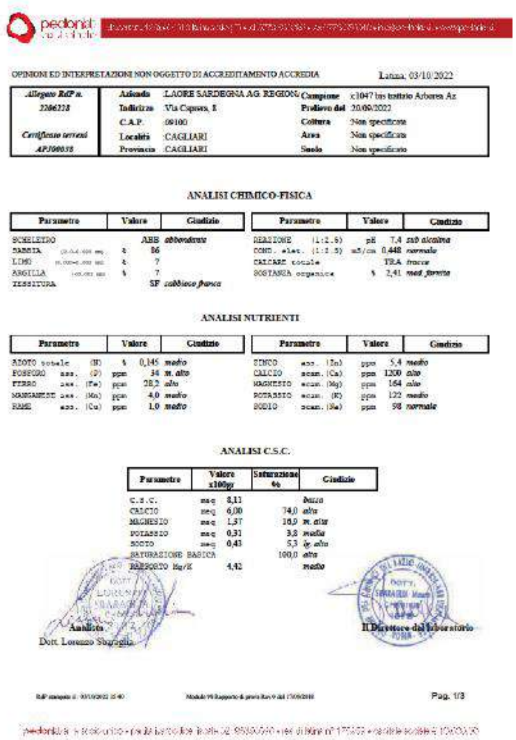
Il Chimico Analista
Dott. Lorenzo Sbaraglia

Il Direttore del Laboratorio
Dott. Mauro Sbaraglia

NOTE

ACCREDIA

5 Analysis certificates and as many agronomic reports have been drawn up where the peculiarities and needs of the soil types analyzed are highlighted, which are an integral part of this work.
In the previous and next table we present an example of 1047 bis simple site:



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OPINIONI ED INTERPRETAZIONI NON OGGETTO DI ACCREDITAMENTO ACCREDIA — Latina, 03/10/2022

Allegato RdP n. 2206228	Azienda	LAORE SARDEGNA AG. REGION.	Campione	c1047 bis trattato Arborea Az
	Indirizzo	Via Caprera, 8	Prelievo del	20/09/2022
	C.A.P.	09100	Coltura	Non specificata
Certificato terreni	Località	CAGLIARI	Area	Non specificata
APJ00038	Provincia	CAGLIARI	Suolo	Non specificato

ANALISI CHIMICO-FISICA

Parametro		Valore	Giudizio	Parametro		Valore	Giudizio
SCHELETRO		ABB	abbondante	REAZIONE (1:2.5)	pH	7,4	sub alcalina
SABBIA	%	86		COND. elet. (1:2.5)	mS/cm	0,448	normale
LIMO	%	7		CALCARE totale		TRA	tracce
ARGILLA	%	7		SOSTANZA organica	%	2,41	mod. fornita
TESSITURA		SF	sabbioso franca				

ANALISI NUTRIENTI

Parametro		Valore	Giudizio	Parametro		Valore	Giudizio
AZOTO totale (N)	%	0,145	medio	ZINCO ass. (Zn)	ppm	5,4	medio
FOSFORO ass. (P)	ppm	34	m. alto	CALCIO scam. (Ca)	ppm	1200	alto
FERRO ass. (Fe)	ppm	28,2	alto	MAGNESIO scam. (Mg)	ppm	164	alto
MANGANESE ass. (Mn)	ppm	4,0	medio	POTASSIO scam. (K)	ppm	122	medio
RAME ass. (Cu)	ppm	1,0	medio	SODIO scam. (Na)	ppm	98	normale

ANALISI C.S.C.

Parametro		Valore x100gr	Saturazione %	Giudizio
C.S.C.	meq	8,11		bassa
CALCIO	meq	6,00	74,0	alta
MAGNESIO	meq	1,37	16,9	m. alta
POTASSIO	meq	0,31	3,8	media
SODIO	meq	0,43	5,3	lg. alta
SATURAZIONE BASICA			100,0	alta
RAPPORTO Mg/K		4,42		medio

Analista
Dott. Lorenzo Sbaraglia

Il Direttore del laboratorio

Pag. 1/3

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OPINIONI ED INTERPRETAZIONI NON OGGETTO DI ACCREDITAMENTO ACCREDIA — Latina, 03/10/2022

ANALISI CHIMICO-FISICA

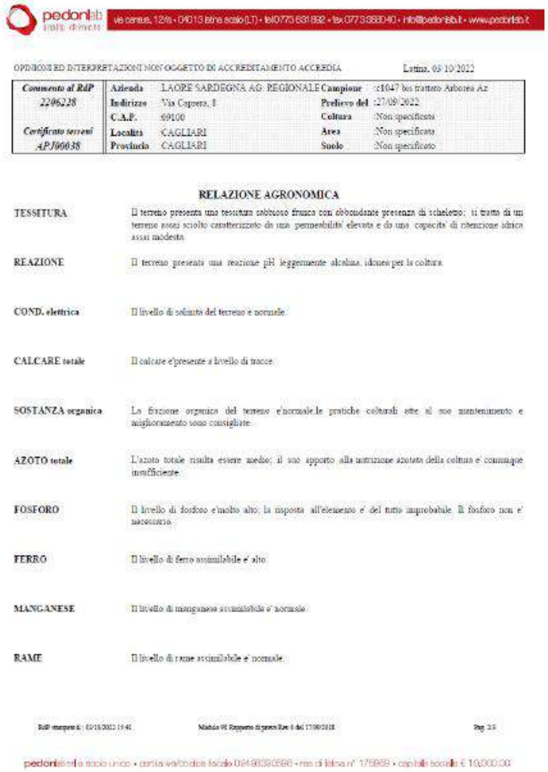
ANALISI NUTRIENTI

ANALISI C.S.C.

Analista
Dott. Lorenzo Sbaraglia

Il Direttore del laboratorio

Pag. 1/3



pedonlab analisi chimiche

OPINIONI ED INTERPRETAZIONI NON OGGETTO DI ACCREDITAMENTO ACCREDIA — Latina, 03/10/2022

Commento al RdP 2206228	**Azienda**	LAORE SARDEGNA AG. REGIONALE	**Campione**	:c1047 bis trattato Arborea Az
	Indirizzo	Via Caprera, 8	**Prelievo del**	:27/09/2022
	C.A.P.	09100	**Coltura**	:Non specificata
Certificato terreni APJ00038	**Località**	CAGLIARI	**Area**	:Non specificata
	Provincia	CAGLIARI	**Suolo**	:Non specificato

RELAZIONE AGRONOMICA

TESSITURA — Il terreno presenta una tessitura sabbioso franca con abbondante presenza di scheletro; si tratta di un terreno assai sciolto caratterizzato da una permeabilita' elevata e da una capacita' di ritenzione idrica assai modesta.

REAZIONE — Il terreno presenta una reazione pH leggermente alcalina, idonea per la coltura.

COND. elettrica — Il livello di soluti del terreno e normale.

CALCARE totale — Il calcare e'presente a livello di tracce.

SOSTANZA organica — La frazione organica del terreno e'normale.le pratiche colturali atte al suo mantenimento e miglioramento sono consigliate.

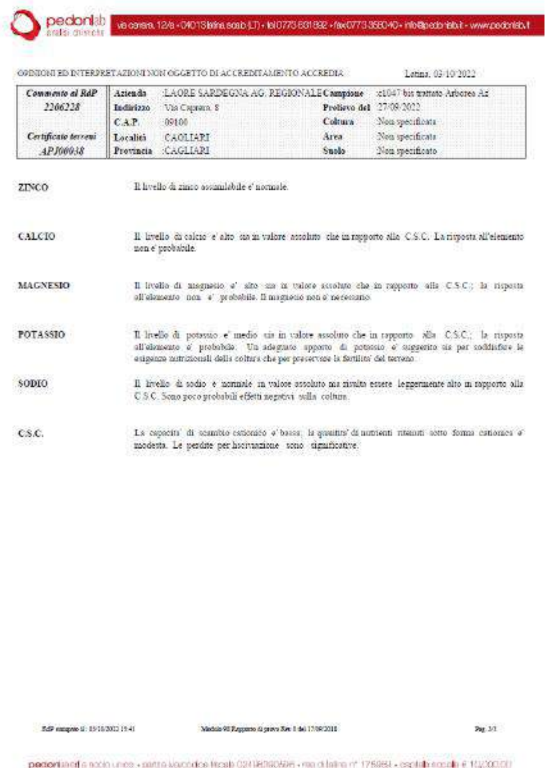
AZOTO totale — L'azoto totale risulta essere medio; il suo apporto alla nutrizione azotata della coltura e' comunque insufficiente.

FOSFORO — Il livello di fosforo e'molto alto; la risposta all'elemento e' del tutto improbabile. Il fosforo non e' necessario.

FERRO — Il livello di ferro assimilabile e' alto.

MANGANESE — Il livello di manganese assimilabile e' normale.

RAME — Il livello di rame assimilabile e' normale.



pedonlab analisi chimiche

OPINIONI ED INTERPRETAZIONI NON OGGETTO DI ACCREDITAMENTO ACCREDIA — Latina, 03/10/2022

Commento al RdP 2206228	**Azienda**	:LAORE SARDEGNA AG. REGIONALE	**Campione**	:c1047 bis trattato Arborea Az
	Indirizzo	Via Caprera, 8	**Prelievo del**	27/09/2022
	C.A.P.	:09100	**Coltura**	:Non specificata
Certificato terreni APJ00038	**Località**	CAGLIARI	**Area**	:Non specificata
	Provincia	:CAGLIARI	**Suolo**	:Non specificato

ZINCO — Il livello di zinco assimilabile e' normale.

CALCIO — Il livello di calcio e' alto sia in valore assoluto che in rapporto alla C.S.C.. La risposta all'elemento non e' probabile.

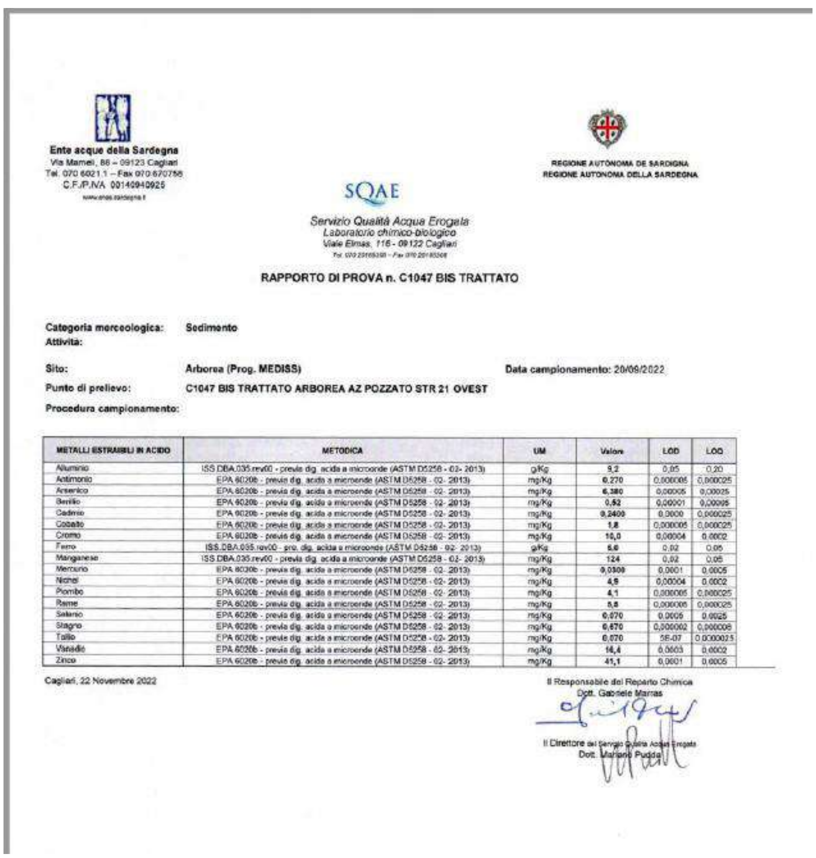
MAGNESIO — Il livello di magnesio e' alto sia in valore assoluto che in rapporto alla C.S.C.; la risposta all'elemento non e' probabile. Il magnesio non e' necessario.

POTASSIO — Il livello di potassio e' medio sia in valore assoluto che in rapporto alla C.S.C.; la risposta all'elemento e' probabile. Un adeguato apporto di potassio e' suggerito sia per soddisfare le esigenze nutrizionali della coltura che per preservare la fertilita' del terreno.

SODIO — Il livello di sodio e normale in valore assoluto ma risulta essere leggermente alto in rapporto alla C.S.C. Sono poco probabili effetti negativi sulla coltura.

C.S.C. — La capacita' di scambio cationico e' bassa; la quantita' di nutrienti ritenuti sotto forma cationica e' modesta. Le perdite per lisciviazione sono significative.

Enas analyzed the metals foreseen in the monitoring plan, but overall the parameters analyzed are more than those foreseen to better characterized the soils, as an example n.1047bis:



Ente acque della Sardegna
Via Mameli, 88 – 09123 Cagliari
Tel. 070 6021.1 – Fax 070 670758
C.F./P.IVA 00140940925

REGIONE AUTÒNOMA DE SARDIGNA
REGIONE AUTONOMA DELLA SARDEGNA

SQAE
Servizio Qualità Acqua Erogata
Laboratorio chimico-biologico
Viale Elmas, 116 - 09122 Cagliari

RAPPORTO DI PROVA n. C1047 BIS TRATTATO

Categoria merceologica: **Sedimento**
Attività:

Sito: **Arborea (Prog. MEDISS)** — **Data campionamento:** 20/09/2022
Punto di prelievo: **C1047 BIS TRATTATO ARBOREA AZ POZZATO STR 21 OVEST**
Procedura campionamento:

METALLI ESTRAIBILI IN ACIDO	METODICA	UM	Valore	LOD	LOQ
Alluminio	ISS DBA.035.rev00 - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	g/Kg	9,2	0,05	0,20
Antimonio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,270	0,000005	0,000025
Arsenico	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	6,380	0,00005	0,00025
Berillio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,52	0,00001	0,00005
Cadmio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,2400	0,0000	0,000025
Cobalto	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	1,8	0,000005	0,000025
Cromo	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	10,0	0,00004	0,0002
Ferro	ISS.DBA.035.rev00 - pro. dig. acida a microonde (ASTM D5258 - 02- 2013)	g/Kg	6,0	0,02	0,05
Manganese	ISS DBA.035.rev00 - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	124	0,02	0,05
Mercurio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,0500	0,0001	0,0005
Nichel	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	4,9	0,00004	0,0002
Piombo	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	4,1	0,000005	0,000025
Rame	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	5,8	0,000005	0,000025
Selenio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,070	0,0005	0,0025
Stagno	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,670	0,000002	0,000008
Tallio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	0,070	5E-07	0,0000025
Vanadio	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	16,4	0,0003	0,0002
Zinco	EPA 6020b - previa dig. acida a microonde (ASTM D5258 - 02- 2013)	mg/Kg	41,1	0,0001	0,0005

Cagliari, 22 Novembre 2022

Il Responsabile del Reparto Chimica
Dott. Gabriele Marras

Il Direttore del Servizio Qualità Acqua Erogata
Dott. Mariano Pudda

Overall we can say that:

*All the soil sampled shows a very drained texture (80/90% sandy)

*The level of N is generally medium and need support by <u>*fertilizer*</u>

*The level of P is high or very high – the crop don't need other P

*The level of Mg and Fe is high or very high – the crop don't need other Mg and Fe

The level of K goes from high to medium.

First year of results shows for this of experimentation that the soils, due to their predominantly sandy textural characteristics and therefore with poor or weak structure on the surface and moderate in depth, show no variations compared to the same samplings carried out in the same sites in 2019 as illustrated in the Monitoring plan in terms of physical and chemical-physical parameters.

***In general the very well drained soil don't assimilate a lot the water and the presence of fertilizer can be an arrangement for the stability of structure of the soil.**

***The difference between the soil health**

respect to the first campaign without the new fertilizer dont change significatively.
In this way is very important the crop plan and fertilization.

<u>CROP monitoring - Field experimentation with ammonium sulphate from bovine manure, produced in the pilot plant</u>

This paragraph presents the results of field trials and monitoring of crops treated with the fertilizer and the related impacts.

Ammonium sulphate in its solid form is already in use in the Arborea area, used specifically for the fertilization of autumn-winter grasslands with excellent technical and production results.

In this sense, it was therefore decided to proceed with testing the same molecule in the liquid form produced by the pilot plant, located at the Centro Ingrasso of the Coop Arborea.

The application of this liquid fertilizer (ammonium sulphate), in the ways and times that will be described later, is aimed at the cultivation of corn.

This crop is of particular importance for the economy of the Arborea area, as it is part of the dairy cattle food chain.

Goals

The study aims to include liquid ammonium sulphate, obtained from the ammonia extraction process (Stripping), present in livestock manure, within the nitrogen fertilization plan for corn.

These effluents are produced in abundance in our area due to the presence of many dairy cattle farms (over 35,000 heads).

At the same time, both the vegetative and reproductive response of the crop under study will be evaluated.

The comparison will concern what will be the cultural responses resulting from the nitrogen input during the crop cycle. On the one hand, we would have the parcel identified as Witness in which agricultural urea will be used as a source of nitrogen (used in part from the transfer to the Cooperative of untreated wastewater), on the other hand the nitrogenous source will be covered by liquid ammonium sulphate (parcel treated).

At the end of the cycle, therefore, a technical-agronomic evaluation will be expressed on the basis of the observations made during the entire cultivation cycle.

Description of the activity

For carrying out the study activity, two precise sites were identified (the first had already been tested on an experimental basis near the pilot plant on oats on 20 Ha), and falling within cultivation areas adjacent to the 2 piezometers installed in precedence (Fig.5,6).

Fig. 5 - Spreading fertilizer on the oat field and meeting with Enas and Cooperative agronomist during the meeting about the fertilizer behaviour on field-crop

The positioning of these piezometers, which are an integral part of the Mediss project for monitoring water quality in the areas subject to treatment with ammonium sulphate, and for descriptive simplification will be identified with Piezometer 1 and Piezometer 2.
Within these cultivation areas, for better applied the fertilize have been delimited micro-plots with a total surface area of 16 square meters each (4*4) were then delimited, clearly identifiable by the presence of wooden stakes and white-red gibbous. In the parcels, at the right time, the product (ammonium sulphate) was distributed, manually applying it along the cultivation inter-row with the aid of graduated jugs designed to ensure correct application of the product.

In summary, therefore, the comparative test envisaged the comparison in the field, and therefore the response of the crop to the parallel administration of agricultural urea on the one hand, and ammonium sulphate of organic derivation on the other, obviously making the same fertilizing units.
It should be remembered that the comparison involved the cultivation of corn, which will be harvested when waxy ripe and intended mainly for feeding dairy cows.

Fig. 6 – graduated jugs used in the experimentation

<u>*Comparative tests description:*</u>

Plot n°1 referred to piezometer n° 1.
Test specifications:
Area under assessment: 16 m^2
Crop: Corn
Variety: Gladius class FAO 700 production cycle 135 days
Sowing date: 15 May 2022
Collection date: August 16, 2022
The company concerned usually administers 300 kg of urea/ha (46% N) divided into 2 interventions to cover crop requirements with mechanical distribution (fertilizer), equivalent to 138 fertilizer units.
In the area under study, a total of 25 liters of ammonium sulphate were administered, during the crop cycle divided into 3 interventions of equal quantities and spaced about 20 days apart.
The distribution was done by hand with a graduated jug in the area between the rows.
The distributed quantity made it possible to make the same nitrogenous fertilizing units made in the witness area with agricultural urea.
First application 06/16/2022
Second application 05/07/2022
Third application 07/24/2022

Plot n°2 referred to piezometer n° 2.
Test specifications:
Evaluation area: 16 m2
Crop: Corn
Variety: P2088 class FAO 700 production cycle 135 days
Sowing date: June 12, 2022
Collection date: September 13, 2022

The company concerned usually administers 350 kg/ha of urea (46% N) divided into 2 interventions to cover crop requirements with mechanical distribution (fertilizer), equivalent to 161 fertilizer units.
In the area under study, a total of 31 liters of ammonium sulphate were administered during the crop cycle divided into 3 interventions of equal quantities and spaced about 20 days apart.

The distribution was done by hand with a graduated jug in the area between the rows.
The distributed quantity made it possible to make the same nitrogenous fertilizing units made in the witness area with agricultural urea.
First application 08/07/2022
Second application 07/29/2022
Third application 08/18/2022

Main observations
As can be seen from the details of the study sites, the two micro-plots share the crop (Maize) and the fertilizer used as a reference (Agricultural Urea); on the other hand they differ in sowing period and quantity of chemical fertilizer distributed.
Despite these differences, the agronomic response, i.e. the cultivation behavior, is the same. Specifically, in the first 45 days of the cycle, the Witness showed more vigorous growth than the Treaty. This answer, in any case, was expected, as the titration of Agricultural Urea and its chemical characteristics always determine a greater vegetative vigor in the initial stages of the crop.
It must also be said that once this phase has been overcome, and on the basis of what has been observed, the vegetative behavior of the crop in the compared theses no longer showed a substantial difference in the subsequent growth phases and identified in sequence with:
• doff
• Emission of male and female inflorescences
• Pollination
• Fruit setting
• Spike enlargement

Technical and Agronomic Evaluations
The product (ammonium sulphate) has satisfactorily shown to support the crop both vegetatively and reproductively.
In fact, for the entire duration of the cultivation cycle, the plants did not show any symptoms attributable to nutritional deficiencies, always showing a typical colour of the crop, accompanied by normal leaf doffing and spreading.

The reproductive part, i.e. the ear of corn, presented itself with normal characteristics. In fact, by evaluating the number of ranks, the size and filling of the grain per ears, it can be stated that pollination took place regularly (Fig.7,8,9).

Conclusions
The work carried out, therefore, made it possible to collect some technical information useful for verifying the potential feasibility of using alternative sources of nitrogen, even more so if derived from virtuous processes of extraction and valorisation of the ammonia part present within the organic waste.
Like all comparative tests, especially in the agricultural field, further evaluations are necessary (this being an experiment linked to the times and activities of the Mediss project).
In fact, it is hoped, for the years to come, the possibility of carrying out a greater number of comparison tests, with larger parcels (e.g. hectare), aimed at guaranteeing an increasingly reliable result.
Obviously such an approach would guarantee the possibility of collecting further data useful to validate the result, such as for example the production on a larger scale (kg/ha).
Summarizing therefore, in principle it has been noted that by adding the same nitrogenous units, the final result, i.e. vegetative-reproductive behavior in the Treated plot, did not show substantial differences compared to the Witness.
On the contrary, going into specifics, we can state that from the point of view of nitrogen efficiency, ammonium sulphate is a product that guarantees a technical response and respect for the environment superior to agricultural urea.

In this sense, in fact, the ammonium sulphate is less exposed to the leaching process (therefore less losses), a very marked phenomenon in sandy soils (80-90% of sandy component) such as those of the Arborea area.

Fig. 7 -The images show the growth sequences of the corn after spreading the fertilizer (Piezometer area 1)

Fig. 8 -The images show the growth sequences of the corn after spreading the fertilizer (Piezometer area 1)

Fig. 9 -The following images show the growth sequences of the corn after spreading the fertilizer (Piezometer area 2)

Spirulina alga test on greenhouse - Field experimentation with ammonium sulphate derived from bovine manure, produced in the pilot plant

According to the results obtained in the laboratory tests using ammonium sulphate produced from the digestate, a preliminary test was performed in two mini-tanks for the cultivation of Spirulina an internationally known reality hosted in the Arborea cooperative, was started and still continues.
The purpose of the experimentation is to evaluate the growth of Spirulina with ammonium sulphate in an environment that simulates the environmental conditions of growth, in order to hypothesize a scaled-up process for feeding the entire Livegreen production plant (Project SpiralG).
The experimentation was carried out in April-May 2022 using two long tanks 2 m and 2 m wide for a total photosynthetic activity area of 18.5 m^2. The culture depth was set at 15 cm.
Ammonium sulphate was provided by a collaboration between MEDISS project and Livegreen company that signed a letter of intent like stakeholders.

The results of the experimentation carried out from April to July (annex 1 of the 3° Interim report) show satisfactory results and suggest in the future important repercussions on the territory given the multiple uses of the algae both in the food sector (also used in third countries such as Africa for the important load energetic), aesthetic, medicinal.
According to the results obtained in the preliminary laboratory tests using ammonium sulphate produced from the digestate by MEDISS project, a first test was performed in two mini-tanks for the cultivation of Spirulina.

A pilot system based on membrane technology has been built and is operational in Arborea to optimize the treatment of the digestate and test the final product through open field tests thanks to the collaboration of local companies.

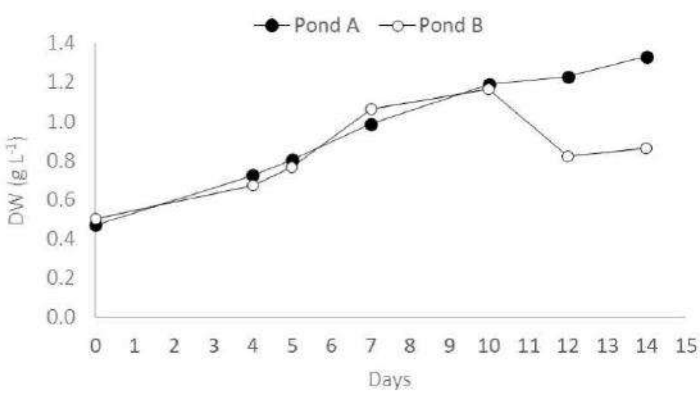


Fig.10 - Graph comparing the cultivation in the two pilot tanks using as nitrogen source, respectively ammonium sulphate (Pond A) and potassium nitrate (Pond B).

The product sampled for our study is ammonium sulfate with a nitrogen concentration of 6 g L^{-1} N-NH4+ and a pH of 2. The inoculum was supplied to have an initial biomass concentration of approximately 0.5 g L^{-1} DW which is the concentration usually set for optimal semi-continuous cultivation in the production facility.
The purpose of this preliminary study was to compare a culture using ammonium sulfate (Pond A) with potassium nitrate (Pond B) which is a common nitrogen source in microalgae cultivation.
Preliminary results indicate that ammonium sulfate can replace potassium nitrate as a source of nitrogen (Fig.10).
As can be seen in Figure 10, both tanks showed the same growth trend during the first 10 days of cultivation. A dry weight trace in Pond B was recorded from day 10, probably due to a contamination that caused a sudden reduction in Spirulina concentration. Both ponds also showed similar average productivity during the experiment being 8.61 g m^{-2} day^{-1} DW and 7.23 g m^{-2} day^{-1} DW for Pond A and Pond B, respectively. These values are in line with the productivity of the SpiralG plant in the same season of the year.
The results of this study demonstrated that ammonium sulphate can be efficiently used as a nitrogen source in the industrial cultivation of Spirulina. Further tests in the production plant are ongoing and planned in order to clearly evaluate the long-term fertilization with ammonium sulphate produced from locally recovered digestate.
From the first experimentation a criticality emerged regarding the evaporation of the ammonium sulphate following the addition of the same in Pond A. The solution at pH 2, in fact, once introduced into the culture at pH 10.5-11 causes the exhalation smell of ammonia.

Another experiment was set up to verify the amount of ammoniacal nitrogen actually available for the crop. The second experiment, still in progress, provides for the administration of ammonium sulphate in both tanks as the only source of nitrogen for the culture: in Pond A nitrogen will be administered through the use of a peristaltic pump (AQUA™ hc 201) for an 8h period each day (8:00-16:00) to maintain a concentration of 50 mg L-1 N-NH4+; in Pond B ammonium sulphate will be administered in a single shift to reach the concentration of 100 mg L-1 N-NH4+.

Both tanks will be sampled every day and the samples will be analyzed to determine the ammoniacal nitrogen present. The protocol used for the determination of ammonia is based on the use of Nessler's reagent and spectrophotometric analysis at À= 410 nm.

The two tanks will also be evaluated for growth. The samples will then be tested for the determination of dry weight and absorbance at À= 750 nm (Fig.11).

Fig. 11 -The use of fertilizer on spirilina alga cultivation in green-house by Live-green company

The purposes of this second experimentation are related to the determination of the ammonia nitrogen actually available for growth, but also to understand which is the optimal concentration of nitrogen and which method of administration is more efficient.

The first data obtained suggest that the continuous administration of ammonium sulphate (Pond A), through the use of the peristaltic pump, guarantees greater growth of the crop compared to the administration in a single shift (Pond B).

Figure 12 shows the different growth trend of the two cultures: Pond A with continuous administration of ammonium sulphate (3.5 l/d) showed a daily productivity of 9.30 g m-2 day-1 DW; Pond B with administration of ammonium sulphate in one shift (5l every two days) showed a lower productivity, equal to 5.59 g m-2 day-1 DW.

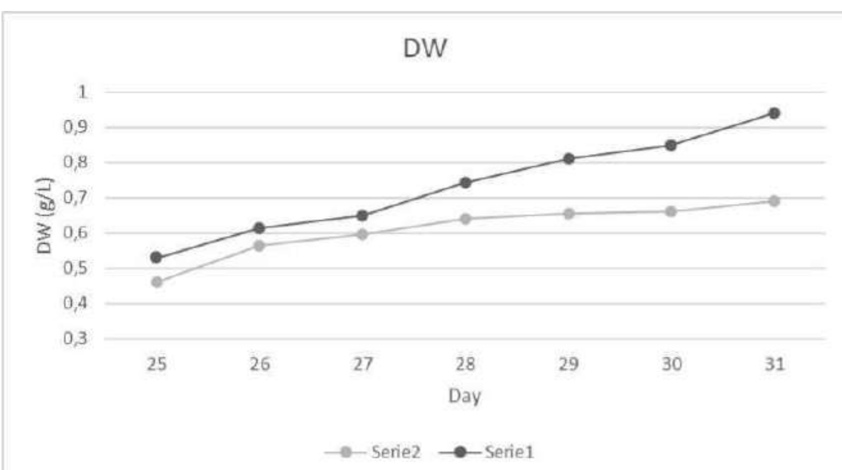


Fig.12 - Graph comparing the growth in terms of dry weight (DW) of the two pilot tanks using ammonium sulphate as the nitrogen source, but with a different method of administration: Pond A continuous administration; Pond B administration in one shift.

This difference in terms of growth could be linked to a lower bioavailability of the nitrogen source due to the method of administration of the nutrient (continuous in Pond A, discontinuous in Pond B). Analyzes on available ammonia nitrogen are still in progress.

Experiments to date clearly indicate that ammonium sulfate can be used as an alternative nitrogen source for Spirulina cultivation, and that the most effective method of administration is continuous administration.

WP3 3.4.1, 3.4.2, 3.4.3 MONITORING DATA (water, soil, crops) – EL HAMMA OF GABES (Tunisie)

WP3 Activity 3.4 Comparing baseline value with final data of monitoring water, soil and crops

3.4.1,3.4.2,3.4.3 Water, soil and crops monitoring and 3.4.4 Comparing baseline value with final data

IRA, in accordance with the current specifications of the Monitoring Plan and in collaboration with all the public bodies operating in the territory, carried out monitoring of the waters, the soils and crops before and after the construction of the plot applications for testing water impact and give homogeneity to the general monitoring

3.4.1, 3.4.2 Water and soils applications and monitoring

Plot applications

For the application on site four small plots were chosen (Fig.1):

P1: Plot not irrigated by wastewater;

P2: Plot irrigated by wastewater for one year;

P3: Plot irrigated by wastewater for 7 years;

P4: Plot irrigated by wastewater for 15 years.

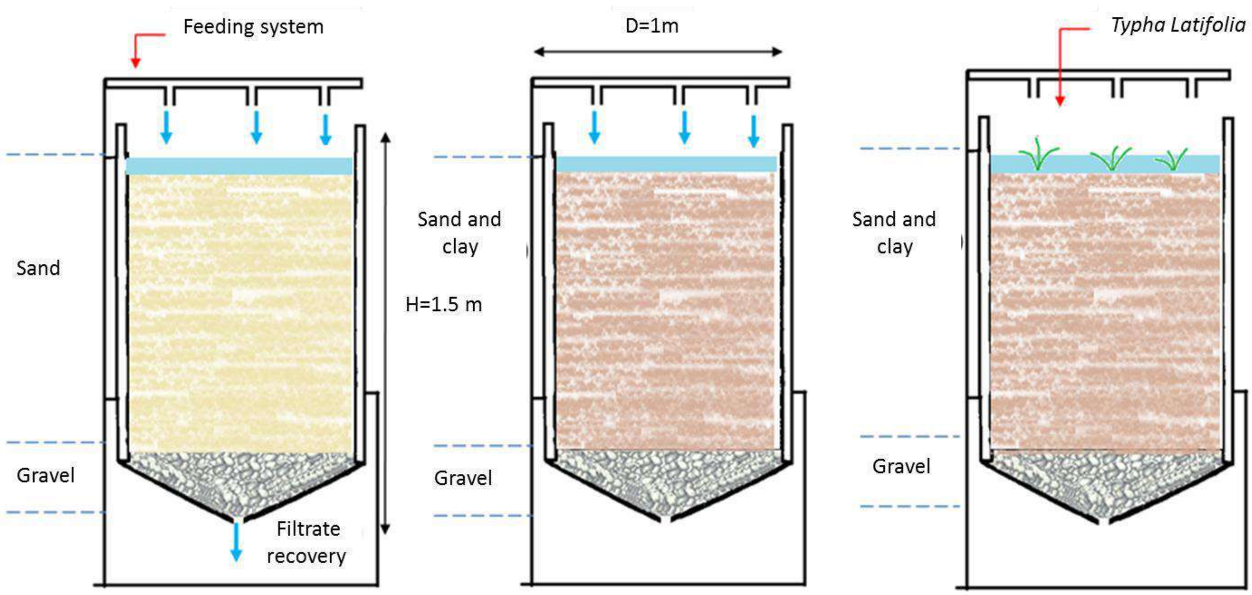


Fig. 1. Schematic presentation of the three silos

In these different plots, IRA sampled at 3 different depths, 0-20 cm, 20-40 cm and 40-60 cm. By this work, IRA will be able to characterize these soils and even predict their future compositions. This study will also allow IRA researchers to study the mobility of pollutants in the soil and their fate after infiltration.

The preparation of the water treatment system was carried out in a succession of steps. From September until December, the monitoring of the water pipe and the preparation of the filter matrix in the system has been ensured. After the preparation step of the filter bed, water sampling and analysis for the wet season have begun in January. The control of the physicochemical and bacteriological water quality, from January to April, has been performed for the wastewater at the input and the outlet of the three treatment reactors. The study of the impact of irrigation with wastewater filtered by the reactors has started in February by a survey of sunflower irrigation.

The filter was fed with secondary effluent from Bechima plant (El Hamma district, Gabes). The operation mode conditions of the wastewater treatment system pilot were performed on a weekly feeding cycle: 1-day flooding period and 2-days drying period. During the feeding operation of the filters (silos), the daily hydraulic load is about 1.5 m3/filter. The feeding is provided by a feeding system to ensure a uniform distribution of the wastewater on the filter surface. Water samples are collected from the input and the outlet of the three silos.

The secondary effluent (see analysis on 2020) and filtrated water were analyzed in laboratory, according to standard methods, for:

*Analyzed parameters for wastewater and filtrated water (**physicochemical and bacteriologic parameters):***

- Temperature, pH, electrical conductivity (EC),
- Total suspended solids (TSS), chemical oxygen demand (COD) and 5-day biochemical demand (BOD_5)
- Total Kjeldahl nitrogen (TKN), ammonium nitrogen (NH_4-N), nitrate nitrogen (NO_3-N)
- Orthophosphate (PO4-P)
- Total and fecal coliforms
- *Escherichia coli*
- Fecal streptococci
- Pseudomonas aeruginosa
- Staphylococcus aureus
- Salmonella

	Ph	T°C	CE(mS/cm)	MES(mg/l)	DCO(mgO2/l)	NO3-N(mg/l)	PO4(mg/l)	NH4(mg/l)	NTK(mg/l)	CT(CFU/100ml)	E,Coli(CFU/100ml)	CF(CFU/100ml)	SF(CFU/100ml)
15/06/2020	7,5	32,9	5,15	620	296	4,1	2,97	78,4	14	610000	20000	230000	160000
24/06/2020	7,58	28,2	5,21	140	730	2	7,15	75,6	14	640000	300000	250000	120000
29/06/2020	7,4	27,6	5,24	320	370	3,8	7,1	53,2	14	70000	-	40000	120000
01/07/2020	7,67	28,4	5,24	170	420	0,6	1,52	36,2	14	310000	-	250000	30000
06/07/2020	7,47	27,2	5,26	330	430	1,3	4,38	42	14	260000	160000		250000
08/07/2020	8,03	30,4	5,18	170	240	1,4	3,76	33,6	11,2	220000	-	70000	40000
13/07/2020	7,63	27,3	5,25	165	1900	1,7	3,77	-	-	310000	-	110000	-
15/07/2020	7,68	29,1	5,22	220	220	1,7	3,87	-	-	280000	-	140000	50000
27/07/2020	7,87	28,2	5,12	520	2190	18,8	18,66	-	-	390000	610000	210000	12000
11/08/2020	7,84	27,9	5,18	210	1890	23	15,9	-	-	1700000	1850000	30000	350000
15/09/2020	8,05	27,4	5,09	160	2500	22,5	10,11	-	-	950000	540000	450000	30000
17/09/2020	7,97	26,4	5,08	90	1260	11,5	13,87	-	-	4100000		-	90000
21/09/2020	8,12	27,6	5,21	240	1430	7,2	5,17	-	-	120000	1000	-	-
23/09/2020	7,82	27	5,26	250	990	11,5	9,33	-	-	210000	-	-	-
28/09/2020	7,85	27	5,7	140	-	16,6	8,77	-	-	21100000	-	10000	-
30/09/2020	7,8	26,1	5,68	180	-	4,5	7,05			25000	-	-	28000
05/10/2020	8,03	27,8	5,32	160	-	4,3	10,86	43,4	53,69	330000	29000	26000	45000
07/10/2020	7,89	24,3	5,32	430	-	4,7	11,17	44	47,8	480000	16000	21000	14000
12/10/2020	8,39	21,9	5,44	110	-	3,2	8,29	35,1	38,5	540000	-	71000	20000
14/10/2020	7,99	22,3	5,45	250	-	3,2	6,08	35	35,8	210000	-	33000	-
20/10/2020	7,8	23,1	5,28	190	-	10,7	8,66	24,6	25,8	470000	20500	30000	11500
22/10/2020	7,65	23,6	5,38	190	-	10,7	11,01	23,38	30,1	1150000	-	-	-
27/10/2020	7,99	22,5	5,4	140	135	3,9	11,06	24,57	26,04	5485000	2950000	-	-
29/10/2020	8,08	23,6	5,39	80	124	2,5	9,11	18,48	22,4	1950000	810000	-	-

Analysis of secondary effluents on 2020

Surveys should also be conducted to accurately determine the influence of changes in the physico-chemical and microbiological composition of the soil, particularly in terms of fertility, crop productivity and human health in the context of sustainable development.

Study area and sampling strategy of soil and wastewater

The experimental plant was near El Hamma of Gabes (Chenchou perimeter), (33◦ 53 ′ 0 ′′ North, 10◦ 7 ′ 0 ′′ East) characterized by an arid climate with an average annual rainfall of 191 mm, an average annual temperature of 24 ◦ C. July is the warmest month of the year, with an average temperature of 32 ◦ C.

The study site was divided into 3 plots that differed in the duration of the WWTP irrigation period 1, 7, and 15 years.

A plot not irrigated by WWTP was taken as a reference.

The plots were cultivated with olive trees and alfalfa, according to the local agricultural practice, and the irrigation was 300 mm per year of water in the control plot and WWTP in the other plots.

Sampling was conducted in July 2020. Therefore, a field experiment was designed, which had a randomized block design with 3 replicates.

There were 12 plots, each with a size of 8 m width × 30 m length (Fig. 1). The field experiment includes four treatments:

Treatment 1: soil non-irrigated with wastewater (control);

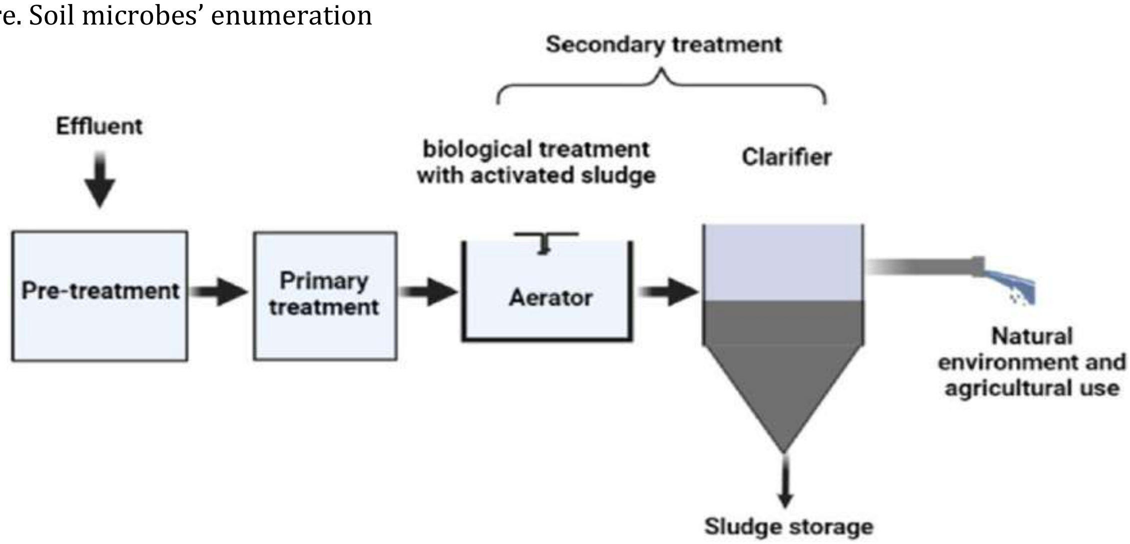
Treatment 2: soils irrigated with wastewater during the last 1 year;

Treatment 3: soils irrigated with wastewater during the last 7 years;
Treatment 4: soils irrigated with wastewater during the last 15 years.

The soil was sampled at three depths: 0–20, 20–40, and 40–60 cm. Soil samples were sieved (2 mm), placed inside vented plastic bags, and stored at 5 ◦ C for 15 days for biochemical and microbiological analysis.
Municipal wastewater, named secondary treated wastewater (WWTP), used in this study was treated in the treatment plant of El Hamma, part of the administrative division of the governorate of Gabes and the delegation of El Hamma. The El Hamma wastewater treatment plant is in the north-west and about 4 km from the city of El Hamma. The municipal wastewater treatment plant covers an area of 8.2 ha (ONAS, 2016). The wastewater plant in the Hammet Gabes region of the south of Tunisia treats a wastewater flow of 32,407 m3/day (ONAS, 2019). It is an activated sludge extended aeration plant with a mechanical screen, grit removal tanks, primary sedimentation, extended aeration, and final sedimentation tanks (Fig. 2). *2.2.*

Wastewater and soil physical, chemical, and microbial characterization
Suspended solid, pH, electrical conductivity (EC), COD, BOD5, total Kjeldahl nitrogen (N), phosphates, ammonium, and nitrates of WWTP samples were performed according to the standard methods for the examination of wastewater and soil (APHA, 1998). All wastewater samples were examined within 24 h for screening and numbering pathogenic bacteria (total coliform, fecal coliforms, *E. coli*, and fecal streptococci) by using the MPN method and following the 3 replications 5 dilutions (Hidri et al., 2021). Soil samples before characterization were sieved (2 mm) and dried at room temperature. The total and active CaCO3, soil granulometric fractions were assessed according to Soil Sampling and Analysis Method 2 (Carter and Gregorich, 2007). Organic carbon was determined by the Walkley and Black dichromate oxidation method (Jackson and Messick, 1958). Cation exchange capacity (CEC) was determined at the actual soil pH by the cobalt hexamine method (Orsini and Remy, 1976). Soil granulometric fractions were determined by laser granulomas (type Fritsch Analysette 22). All these characteristics were observed in the dry seasons.

Microbial enumeration in soil
Bacteria and fungi
Bacteria and fungi were extracted by vortexing soil samples with 9 % sterile NaCl solution and the homogenous soil suspension was serially diluted tenfold with sterile saline solution. Mesophilic Aerobic Bacteria (MAB) count was carried out by spreading 100 μL of appropriate dilutions on Plate Count Agar as recommended by Al-Lahham et al. (2003), and incubated for 48 h at 28 ◦ C. Only plates showing between 10 and 100 colonies per plate of 90 mm were counted. For fungi enumeration, the appropriate soil dilution was spread on potato dextrose agar (PDA). The number of developed colonies was recorded after 7 days of incubation at ambient room temperature. Soil microbes' enumeration



was expressed as the number of colony-forming units (CFU) per gram of dry weight soil (Hidri et al., 2021).

Statistical analysis
All results are the average of three determinations. Analysis of variance (ANOVA) was carried out using the SPSS 21.0 statistical program (SPSS for Windows; SPSS, Inc., Chicago, IL, USA), and means were separated according to the Tukey post-hoc test ($p < 0.05$). Before running, PCA data were log10 transformed. Principal component analysis (PCA) was performed to evaluate differences among all variables in the control, 1, 7, and 15 years WWTP irrigated soil. The PCA results were shown as a bi-plot to highlight the interaction between the samples and variables. The PCA was performed by the SPSS program package (IBM Corp. Released 2012. IBM SPSS Statistics for Windows, version 21.0).

Result and discussion

Treated wastewater quality
The results of the chemicals and microbiological of the WWTP sample are presented in Table 1. The WWTP showed a sub-alkaline pH of 7.72 and a high EC of 5.31 mS cm^{-1} (Table 1). As expected, a greater value of suspended solid was found during the dry season 228.3 mg L.
The level of biochemical oxygen demand (BOD) and chemical oxygen demand (COD) were 126 and 575.65 mg L^{-1}, respectively (Tab. 1). An important value of phosphate and nitrate content of 53.15 and 2.06 mg L was observed, respectively. The microbial characteristics of the WWTP used in this study are also listed in Table 1. The pathogen bacteria analysis included total coliforms, fecal coliforms, *Escherichia coli*, and fecal streptococci. In the effluent from secondary treatment, the number of total coliforms, fecal coliforms, *E. coli*, and fecal streptococci were $36.57 \cdot 10^5$, $23.56 \cdot 10^5$, $11.43 \cdot 10^5$, and $10.23 \cdot 10^5$ MPN/100 mL^{-1}, respectively. In addition, the number of the pathogenic bacteria was bigger than 1000 MPN mL^{-1}, threshold fixed as standard by the World Health Organization (WHO) for "irrigation of crops likely to be eaten uncooked" (WHO, 1989).

Table 1
Chemical and microbiological characteristics of secondary municipal wastewater used for soil irrigation (Mean ± standard deviation).

Parameters	Unit	Values
BOD_5	(mg L^{-1})	126.0 ± 0.1
COD	(mg L^{-1})	575.65 ± 55.8
Ammonium	(mg L^{-1})	13.43 ± 1.14
Nitrates	(mg L^{-1})	2.06 ± 1.23
N	(mg L^{-1})	53.15 ± 19.67
Phosphates	(mg L^{-1})	4.62 ± 1.69
Suspended solid	(mg L^{-1})	228.3 ± 64.13
pH	–	7.72 ± 0.19
EC	mS cm^{-1}	5.31 ± 0.03
Total coliforms	(MPN/100 mL)	$36.57 \cdot 10^5 \pm 7.76$
Fecal coliforms	(MPN/100 mL)	$23.56 \cdot 10^5 \pm 4.04$
Escherichia coli	(MPN/100 mL)	$11.43 \cdot 10^5 \pm 2.51$
Fecal Streptococci	(MPN/100 mL)	$10.23 \cdot 10^5 \pm 1.15$

EC: Electrical conductivity; OC: Organic Carbon; COD: Chemical oxygen demand; N: Nitrogen; BOD_5: Biochemical oxygen demand; $n = 3$; (In brackets): Standard deviation with $n = 3$.

Besides, it is proven that using saline water for irrigation allowed an enrichment and increase in the soil salt content (Hidri et al., 2021). As a result, it is essential and important to monitor the soil salinity during WWTP irrigation practice (Shakir et al., 2017). Soil irrigation with treated wastewater of good quality is one of the best practical and management rules of wastewater since it introduced some vital nutrients into soil and plants like nitrogen, phosphates, potassium, and ultima trace elements such as copper, zinc, etc. All these residual compounds usually provided by treated wastewater are made available and valuable to plants, microbes, etc. (Melloul et al., 2002). The bacteriological quality of the water worsened slightly when reaching the plot. This result appeared to be related to residual and stagnant organic compounds in the water distribution network (Lonigro et al., 2016). Based on these results, we can conclude that the load of fecal bacteria considered as opportunistic pathogens and carried by the treated wastewater from the El-Hammet Gabes WWTP is conform to the limits set by WHO standards. The Tunisian Standard (NT 106.03) does not specify the bacteriological quality of wastewater that can be used in agriculture. According to the World Health Organization's 1975 recommendation, only biologically treated wastewater should irrigate vegetables intended for raw consumption, and the samples should have no >100 coliforms per 100 ml of contamination. Pathogens in fresh vegetables and fruits are those originating from the gut; like Salmonella, Shigella, Escherichia, and Klebsiella. The results of the microbiological

WWTP characterization followed those published by Coppola et al. (2004), Rosan (n.d), and Klay et al. (2010) in other studies of treated urban wastewater conducted in Sardinia, Tunisia, and Jordan (2010).

Wastewater irrigation's effects on the physical and chemical characteristics of the soil

All determined physical and chemical soil characteristics were shown in Tables 2 and 3. The results showed globally that the varied period of wastewater soil irrigation and soil depths has not influenced largely the variation of pH since the pH showed on average a slight maximum variation of around 0.5 pH-unit. Thus, in the control plot, the pH showed a slight increase of 0.5 pH-unit with the increase of depth, 8.30 pH-unit for horizon 0–20 cm, and 8.73 pH-unit for horizon 40–60 cm. For the plot irrigated for 15 years with wastewater, the soil pH showed no significant variation with the soil depths and an average pH of about 7.8 pH-unit. The same result was observed after 1 or 7 years of WWTP irrigation ($P < 0.05$). Whereas, the soil electric conductivity (EC), reflecting the general soil salinity, showed a significant value of 3.59 mS cm^{-1} in the soil control, at 40–60 cm and after 1 and 7 years of WWTP irrigation.

Table 2

Main soil properties of the different plots investigated for 0, 1, 7 and 15 years and at different soil horizons of 0–20, 20–40 and 40–60 cm.

Sites	Depth (cm)	pH	EC (mS cm^{-1})	C	N	C/N	OM (%)	CEC (meq + 100 g^{-1})
Site 1 (control as annual rainfall)	0–20	8.30 ± 0.02 b	2.52 ± 0.02 d	0.104 ± 0.007 f	0.321 ± 0.017 e	0.32 ± 0.36 c	0.18 ± 0.01 f	3.09 ± 0.04 d
	20–40	8.43 ± 0.03 b	2.71 ± 0.05 d	0.098 ± 0.004 f	0.126 ± 0.015 g	0.77 ± 0.80 b	0.17 ± 0.03 f	2.61 ± 0.47 d
	40–60	8.73 ± 0.03 a	3.59 ± 0.05 b	0.168 ± 0.007 e	0.133 ± 0.011 g	1.26 ± 1.50 a	0.30 ± 0.04 e	2.97 ± 0.18 d
Site 2 (1 years)	0–20	8.14 ± 0.02 c	2.58 ± 0.05 d	0.238 ± 0.008 d	0.472 ± 0.042 c	0.51 ± 0.38 c	0.41 ± 0.01 d	3.23 ± 0.10 c
	20–40	8.09 ± 0.07 c	2.50 ± 0.02 d	0.203 ± 0.002 d	0.218 ± 0.003 f	0.93 ± 0.36 a	0.35 ± 0.01 d	3.54 ± 0.19 b
	40–60	8.06 ± 0.08 c	2.15 ± 0.01 e	0.174 ± 0.005 e	0.216 ± 0.003 f	0.81 ± 0.26 b	0.30 ± 0.01 e	3.19 ± 0.06 c
Site 3 (7 years)	0–20	8.01 ± 0.05 d	2.21 ± 0.01 e	0.308 ± 0.016 c	0.688 ± 0.039 b	0.45 ± 0.36 c	0.53 ± 0.01 c	3.11 ± 0.13 c
	20–40	8.03 ± 0.06 d	2.52 ± 0.02 d	0.203 ± 0.03 d	0.297 ± 0.012 e	0.68 ± 0.71 b	0.35 ± 0.09 d	3.29 ± 0.03 b
	40–60	8.16 ± 0.10 c	3.01 ± 0.07 c	0.377 ± 0.006 b	0.321 ± 0.017 e	1.17 ± 0.36 a	0.65 ± 0.01 b	3.31 ± 0.33 b
Site 4 (15 years)	0–20	7.81 ± 0.05 d	2.30 ± 0.04 e	0.680 ± 0.005 a	0.808 ± 0.024 a	0.84 ± 0.15 b	1.17 ± 0.01 a	5.41 ± 0.29 a
	20–40	7.83 ± 0.09 d	4.14 ± 0.01 a	0.401 ± 0.004 b	0.375 ± 0.044 d	1.20 ± 1.97 a	0.69 ± 0.05 b	5.34 ± 0.28 a
	40–60	7.94 ± 0.02 d	2.64 ± 0.06 d	0.290 ± 0.007 c	0.486 ± 0.020 c	0.60 ± 0.24 b	0.50 ± 0.01 c	2.56 ± 0.25 d

EC: Electrical conductivity; C: Carbon; N: Nitrogen; OM: Organic matter; CEC: Cation exchange capacity; n = 3; (In brackets): Standard deviation.
Different lower-case letters showed significant differences among sampling sites and depths in according to the Tukey post-hoc test ($p < 0.05$).

Table 3

Main mineral constitution of the soil of the different plots investigated for 0, 1, 7 and 15 years and at different soil horizons of 0–20, 20–40 and 40–60 cm.

Sites	Depth (cm)	P_2O_5 (%)	K (meq + 100 g^{-1})	Ca (meq + 100 g^{-1})	Mg (meq + 100 g^{-1})	Na (meq + 100 g^{-1})	Total $CaCO_3$ (%)	Active $CaCO_3$ (%)
Site 1 (control as annual rainfall)	0–20	0.08 ± 0.05 d	412.3 ± 2.9 d	5.3 ± 0.1 d	0.36 ± 0.01 b	0.04 ± 0.01 f	9.1 ± 0.2 b	7.7 ± 0.7 c
	20–40	0.07 ± 0.05 d	439.2 ± 41.2 d	5.8 ± 0.1 c	0.37 ± 0.01 b	0.04 ± 0.01 f	7.2 ± 0.4 d	6.5 ± 0.4 d
	40–60	0.06 ± 0.01 d	488.4 ± 2.9 c	4.5 ± 0.2 e	0.44 ± 0.02 a	0.06 ± 0.01 f	7.3 ± 0.3 d	6.3 ± 0.6 d
Site 2 (1 years)	0–20	0.12 ± 0.02 c	439.4 ± 41.0 d	6.7 ± 0.1 b	0.37 ± 0.02 b	0.24 ± 0.01 d	9.5 ± 0.3 a	8.7 ± 0.3 b
	20–40	0.11 ± 0.01 c	413.9 ± 2.9 d	4.7 ± 0.6 d	0.35 ± 0.03 b	0.20 ± 0.01 d	9.7 ± 0.1 a	9.4 ± 0.3 a
	40–60	0.06 ± 0.01 d	506.9 ± 26.9 b	4.2 ± 0.1 e	0.28 ± 0.02 c	0.17 ± 0.01 e	8.7 ± 0.2 b	8.1 ± 0.4 b
Site 3 (7 years)	0–20	0.16 ± 0.02 b	488.4 ± 2.9 c	2.7 ± 0.2 f	0.31 ± 0.03 c	0.31 ± 0.01 c	8.7 ± 0.2 b	7.7 ± 0.9 c
	20–40	0.15 ± 0.02 b	380.1 ± 33.2 e	3.3 ± 0.3 f	0.38 ± 0.03 b	0.13 ± 0.01 e	8.8 ± 0.1 b	8.3 ± 0.6 b
	40–60	0.11 ± 0.01 c	358.2 ± 23.2 e	6.3 ± 0.7 b	0.45 ± 0.02 a	0.29 ± 0.01 c	8.4 ± 0.3 c	7.6 ± 0.7 c
Site 4 (15 years)	0–20	0.22 ± 0.02 a	558.6 ± 57.9 b	4.8 ± 0.4 d	0.47 ± 0.02 a	0.68 ± 0.01 a	9.9 ± 0.5 a	8.4 ± 0.7 b
	20–40	0.19 ± 0.01 a	573.8 ± 31.5 b	7.3 ± 0.4 a	0.42 ± 0.02 a	0.40 ± 0.01 b	8.5 ± 0.1 c	8.2 ± 0.5 b
	40–60	0.17 ± 0.01 a	688.9 ± 83.9 a	7.4 ± 0.7 a	0.37 ± 0.01 b	0.38 ± 0.01 b	10.5 ± 0.7 a	9.9 ± 0.5 a

P2O5: Phosphorus; K_2O: Potassium; Ca: Calcium; Mg: Magnesium; Na: Sodium; $CaCO_3$: Calcium carbonate.
Different lower-case letters showed significant differences among sampling sites and depths in according to the Tukey post-hoc test ($p < 0.05$).

the year of wastewater application, by excluding the first year of investigation and where no significant differences were observed as compared to the ones registered for the control plot (Fig. 3A). The average number of MAB in the topsoil (0–20 cm) ranged from 2.07·105 in the control plot to 3.05 and 3.46·105 CFU g^{-1} 1 dry soil in the 7 and 15 years treated wastewater irrigated plots. On the soil horizons of 20–40 cm and 40–60 cm, the same result observed for horizon 0–20 cm was registered, but with a lesser intensity (Fig. 3A). This result seemed related to the sandy texture and the high availability of organic matter and oxygen in the soil profile. A similar result was observed for the soil fungal number (Fig. 3B), and no significant differences were found between the number of fungi in the soil control plot and the area irrigated by the WWTP for 1 year (Fig. 3B). By increasing the years of WWTP irrigation (7 and 15 years), a significant increase in the fungi number was observed (Fig. 3B). This value decreased by increasing the soil profile depth (0–20 cm > 20–40 cm > 40–60 cm). However, the number of fungal showed an increase after 15 years of WWTP irrigation at 0–20 cm soil depth and with around 2.97·104 CFU g^{-1} soil dry weight. Altogether, these data revealed that long-term irrigation with treated wastewater produced and caused a significant microbial abundance and activity in soil. This microbial growth and reactivity could be explained by the large availability of nutrients related to abundant and continuous condensed soil fertilization conditioned by wastewater irrigation and varied compound provision (Hidri et al., 2021). Microorganisms are predominantly heterotrophic and soil carbon is limited, and the observed differences in the microbial distribution in different soil depths must be related to the availability and quality of carbon sources provided following wastewater applications (Adrover et al., 2012; Truu et al., 2017). But these changes are not always useful and the stimulation of the soil microbial abundance can also additionally have poor effects on other soil properties (Chen et al., 2008). For instance, Becerra-Castro et al. (2015) determined that the bacterial development after irrigation with wastewater helped to shape the biofilm, with the concomitant clogging of the pore area among particles, resulting in a negative implication on the soil hydraulic conductivity. The practice of wastewater for irrigation may be sometimes sources of deleterious or beneficial soil bacteria. For example, introducing bacteria that monitor the nitrogen cycle or other bacteria with the ability to repair and remediate contaminated soil, with main recalcitrant residual compounds like HAP, pesticides, heavy metals, or antibiotics, processes that may help for improving the general soil conditions and quality (Hanjra et al., 2012; Becerra-Castro et al., 2015).

Soil pathogenic bacteria determination

The whole results of pathogenic bacteria registered in the soil are reported in Table 4 and Fig. 4. These results allowed primary to realize that the number of pathogenic bacteria in the WWTP irrigated soil was greater than the one registered in the control plot, where water is only provided by usual rain. Thus, the number total coliform in the soil showed 62.2 MPN g^{-1} soil dry weight at the top horizon of 0–20 cm of the control plot and 877.8 MPN g^{-1} soil dry weight at the same horizon of 0–20

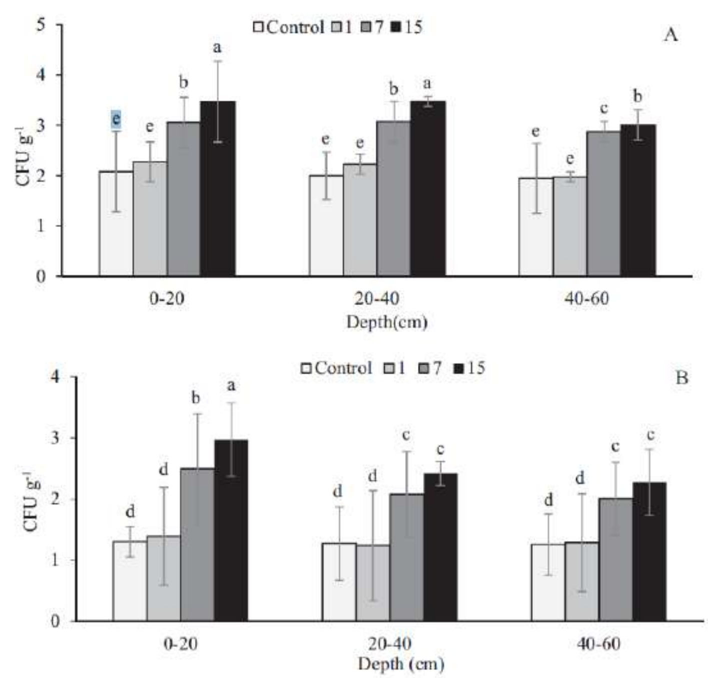


Fig. 3. A) Mesophilic Aerobic Bacteria (MAB) species and B) Fungi biomass affected by wastewater application (0, 1, 7 and 15 years) at different soil depths 0–20, 20–40 and 40–60 cm. Different lower-case letters indicate significant differences among sampling sites and depths in according to Tukey post-hoc test ($p < 0.05$).

cm of the plot irrigated with treated wastewater for 15 years. Besides, the average number of these opportunistic pathogenic bacteria such as TC, FC, *E. coli*, ST, and FS showed a significant increase with the wastewater irrigation of 15 years at the horizon 20–40 cm, and with 952.4, 903.4, 649.8, 160.2, 130.1 MPN g^{-1} 1 soil dry weight, respectively (Table 4). This result reflected the high numbers of fecal bacteria colonizing and residing in the wastewater used for irrigation. The *Pseudomonas* sp., *Staphylococcus*, and *Salmonella*/*Shigella* determination achieved by the count Agar technique showed that *Salmonella*/*Shigella* were not detected in the soil control plot, and after 1 year in the soil horizon of 20–40 cm. But, they were prevailing with an important number in the WWTP irrigated soil for 7 and 15 years at 0–20 cm depth, 3.75 and 3.76 CFU g^{-1} 1 soil dry weight, respectively. For the two horizons, 20–40 cm and 40–60 cm, a significant decrease in the number of *Salmonella*/*Shigella* was observed; the value decreased up to 1.50 CFU g^{-1} 1 soil dry weight soil at 40–60 cm of soil depth (Fig. 4). The average number of *Staphylococci* strains at 0–20 cm showed a significant increase over the years of the WWTP application, this number showed the average of 2.59 and 4.10 CFU g^{-1} soil dry weight for the control plot soil and the 15-year WWTP irrigation soil, respectively (Fig. 4). The same tendency with the similar average value was observed for the two soil depths of 20–40 cm and 40–60 cm (Fig. 4). The *Pseudomonas* sp. strain was always detected in the control plot soil and wastewater irrigated soil at the three soil horizons of 0–20 cm, 20–40 cm and 40–60 cm. On the horizon of 0–20 cm, the average number of *Pseudomonas* sp. was around 2.4 CFU g^{-1} soil dry weight in the control plot and showed up to 4.50 CFU g soil dry weight in the WWTP irrigated soil. No significant differences in the number of *Pseudomonas* sp. were observed during the different periods of treated wastewater application of 1, 7 and 15 years (Fig. 4). At 20–40 cm and 40–60 cm of soil depths and only after 7 and 15 years of wastewater soil irrigation, a significant increase in the number of *Pseudomonas* sp. was observed showing on average 3.45 and 2.98 CFU g^{-1} soil dry weight, respectively (Fig. 4). Thus, world water scarcity has largely influenced the fact and practice of treated wastewater using, exploitation and recycling in heavy industries and/or in agriculture. But this last practice may carry some important opposed and known negative effects on the soils and crops,

Table 4

Soil fecal bacteria determined following secondary wastewater application for 0, 1, 7 and 15 years at different soil profiles of 0–20, 20–40 and 40–60 cm.

Sites	Depth (cm)	TC MPN g^{-1} soil dry weight	FC MPN g^{-1} soil dry weight	*E. coli* MPN g^{-1} soil dry weight	TS MPN g^{-1} soil dry weight	FS MPN g^{-1} soil dry weight
Site 1 (control as annual rainfall)	0–20	62.2 ± 17.9 e	51.8 ± 17.9 e	<3 f	34.5 ± 5.6 d	<3 e
	20–40	49.5 ± 22.1 e	35.4 ± 6.1 e	<3 f	49.5 ± 22.1 d	<3 e
	40–60	<3 f	36.4 ± 6.9 e	<3 f	39.6 ± 11.3 d	<3 e
Site 2 (1 years)	0–20	95.0 ± 26.3 d	87.4 ± 13.2 e	76.0 ± 28.7 e	110.2 ± 13.2 c	38.0 ± 6.6 d
	20–40	86.2 ± 13.0 d	74.9 ± 28.3 e	74.9 ± 28.3 e	56.2 ± 19.5 d	41.2 ± 6.5 d
	40–60	66.6 ± 19.2 e	66.6 ± 19.2 e	40.7 ± 6.4 e	37.0 ± 6.4 d	<3 e
Site 3 (7 years)	0–20	179.3 ± 35.3 c	156.5 ± 6.2 d	145.9 ± 24.7 d	88.9 ± 12.3 c	88.9 ± 12.3 c
	20–40	164.7 ± 50.3 c	160.9 ± 6.3 d	160.9 ± 6.3 d	113.4 ± 12.7 c	113.4 ± 12.7 ab
	40–60	157.9 ± 21.0 c	157.9 ± 21.0 d	125.6 ± 14.0 d	109.4 ± 24.3 c	44.6 ± 7.0 d
Site 4 (15 years)	0–20	877.8 ± 166.2 a	768.1 ± 72.1 b	472.9 ± 26.2 b	117.3 ± 13.1 c	109.7 ± 13.1 b
	20–40	952.4 ± 84.8 a	903.4 ± 84.8 a	649.8 ± 179.3 a	160.2 ± 23.1 b	130.1 ± 17.2 a
	40–60	417.7 ± 75.1 b	374.3 ± 75.1 c	252.2 ± 18.1 c	244.3 ± 6.8 a	59.1 ± 20.5 d

TC: Total Coliforms; FC: Fecal Coliforms; *E. coli*: *Escherichia coli*; FS: Fecal Streptococci.
Different lower-case letters showed significant differences among sampling sites and depths in according to the Tukey post-hoc test ($p < 0.05$).

specifically following the contamination by potential toxic elements, residual organic by-products and pathogenic microbes (Bayomi et al., 2019). Palese et al. (2009) evidenced that the non-treated and treated wastewater irrigated soils always showed different repartition and distribution for TC, FC, *E. coli*, and FS with the soil depth, registering the highest values in the upper layer and a significant decrease by increasing soil depth. This result is linked to the soil filtration effectiveness and availability of nutrients in the soil profile. The results of Belaid et al. (2019) agreed with ours since the soil microbial communities are largely affected by the physical and chemical soil properties and macro- and microbiological soil components. In addition, this effect is supposed to depend on the direct input of exogenous microbiota, which, in an improbable worst-case scenario, would lead to eliminating autochthonous microorganisms of competition (Becerra-Castro et al., 2015). It is thought and reported by Al-Lahham et al. (2003) that using a less contaminant irrigation method or better-quality effluents might further reduce the risk of transmission of fecal pathogenic bacteria.

Changes in soil microbial biomasses C and N

Microbial components released by fumigation and directly extracted can determine the size of the soil biomass, in particular microbial biomasses C and N (MBC and MBN) as earlier reported by Brookes et al. (1985) and Vance et al. (1987).

Microbial biomass carbon (MBC).

The results obtained in this study about the changes in MBC are shown in Fig. 5a. The average soil MBC showed that MBC is being simultaneously affected by the duration of wastewater soil irrigation, the number of years of WWTP application, and the soil depth. The MBC registered for the horizon 0–20 cm showed a significant increase from 10.16 mg kg in the soil plot control to 15.99 mg kg in the WWTP irrigated soil for 15 years (Fig. 5a). In addition, after just only 1 year of WWTP irrigation of 0–20 cm soil depth, the average MBC showed a significant increase to 12.37 mg kg. At 20–40 cm of soil depth and only after 7 or 15 years of WWTP irrigation, the average MBC showed a significant increase as compared to the control soil plot with respective values of 19.24 and 25.47 mg kg (Fig. 5a). But at 40–60 cm, the WWTP application showed a significant raise in the MBC, with respective values of 23.36, 27.33, and 30.24 mg kg, through the 1, 7, and 15 years of WWTP irrigation (Fig. 5a). These results confirmed the ones found in the common literature that main bacterial populations and communities are usually concentrated and lived in soil horizons rich in organic matter and nourishing compounds (Palese et al., 2009). The common organic matter existing into soil matrices showed an easy deeply migration from the top to the bottom. This operation is especially registered for sandy soils. The duration of wastewater irrigation showed a crucial proportional effect on the soil microbial biomass C evolution. The increase in soil microbial biomass C registered in the present following WWTP irrigation looked comparable to the one observed in soil amended with organic compost, as reported by Jedidi et al. (2004).

Microbial biomass nitrogen (MBN).

The average MBN registered into soils showed a general significant change according to the number of years of WWTP irrigation (Fig. 5b). Thus, MBN recorded was very important in the deeper soil horizons of 20–40 cm and 40–60 cm, especially after 15 years of WWTP application (Fig. 5b). Also, important amounts of MBN of 33.20 mg kg and 47.35 mg kg were registered at 40–60 cm soil depth following 1 and 7 years of WWTP application1, respectively (Fig. 5b). After 15 years of WWTP application, a significant increase was also observed in 20–40 cm and 40–60 cm with 36.25 and 49.41 mg kg, respectively; (Fig. 5b). These results could be explained as changes in soil nitrogen stock depending essentially on the main microbial and plant activities. According to soil physical and chemical conditions, the microbial pool has and performed different effects on the mineral and organic nitrogen soil stock. According to Jedidi et al. (2004), soil microbes can release (mineralize) or trap nitrogen (tissue) confirmed by the abundance of total coliforms, fecal coliforms, *E. coli*, and fecal Streptococci at 20–40 cm and 60–40 cm of soil depths. Adrover

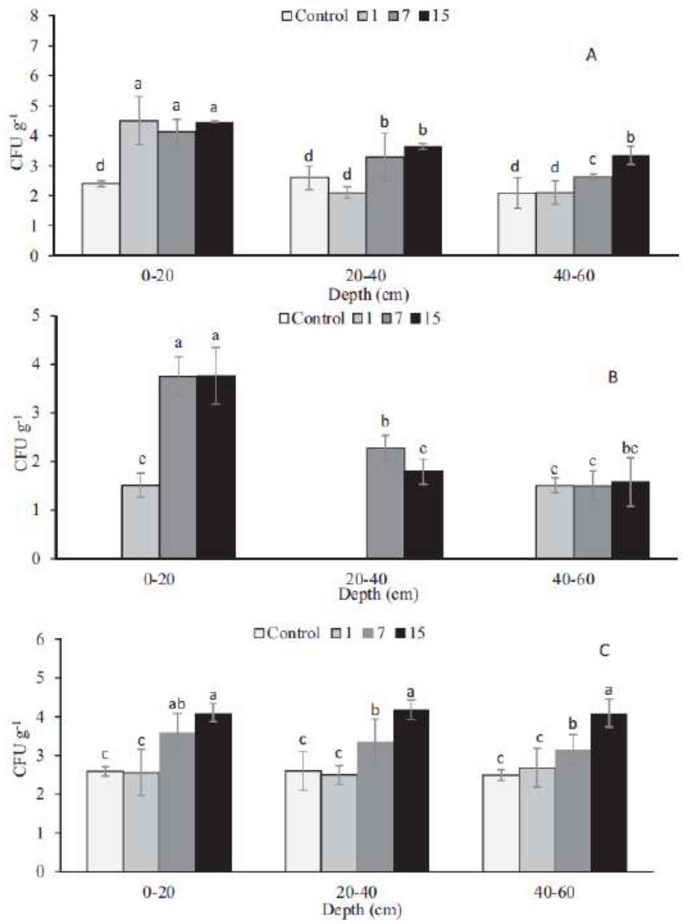


Fig. 4. Soil pathogen microbial biomass (*Salmonella/Shigella*: A, *Staphylococcus*: B and *Pseudomonas* sp.: C) affected by the duration of wastewater application (0, 1, 7 and 15 years) at different soil depths 0–20, 20–40 and 40–60 cm. Different lower-case letters indicate significant differences among sampling sites and depths in according to Tukey post-hoc test ($p < 0.05$).

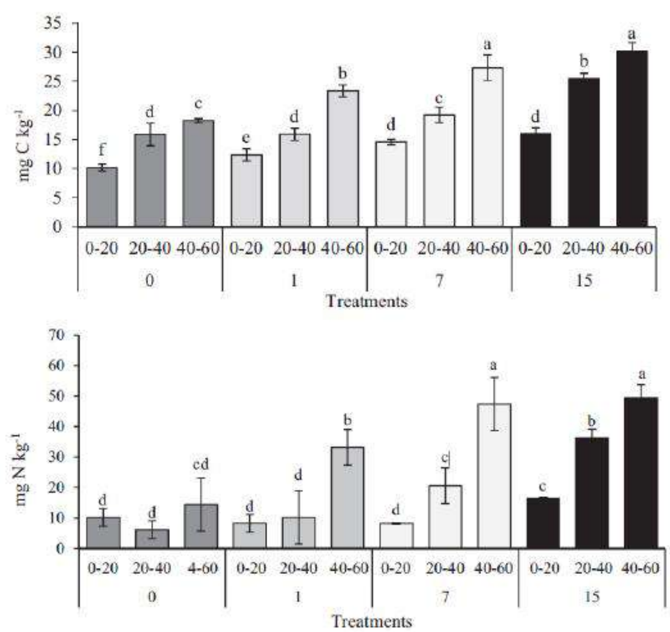


Fig. 5. Soil microbial biomass carbon MBC (a) and microbial biomass nitrogen MBN (b) affected by the duration of wastewater application (0, 1, 7 and 15 years) at different soil depths 0–20, 20–40 and 40–60 cm. Different lower-case letters indicate significant differences among sampling sites and depths in according to Tukey post-hoc test ($p < 0.05$).

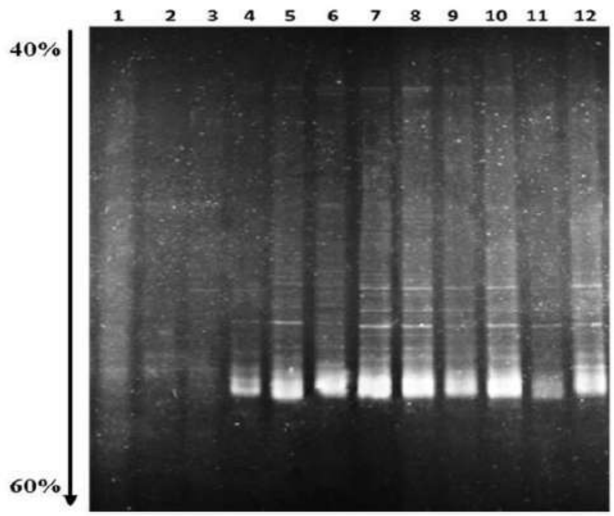


Fig. 6. DGGE patterns show the bacterial community structure of various soils samples based on the variability of the V3–V5 region of 16S rRNA by using urea and formamide gradient ranged from 40 % to 60 %. 1, 2 and 3: control 0–20, 20–40 and 40–60 cm, respectively; 4, 5 and 6: 1 years 0–20, 20–40 and 40–60 cm, respectively; 7, 8 and 9: 7 years 0–20, 20–40 and 40–60 cm, respectively; 10, 11 and 12: 15 years 0–20, 20–40 and 40–60 cm, respectively.

et al. (2012) observed comparable effects with better microbial biomass after 20 years of wastewater irrigation. Friedel et al. (2000) observed a similar increase in microbial biomass and dehydrogenase activity in long-term irrigated soil with raw wastewater (Friedel et al., 2000; Brzezinska, 2006). The stratification of microbial biomass observed according to the soil depth corresponded to the decrease in C organic and N organic contents, as classically observed (Lejon et al., 2007). Altogether, that information revealed that the long-time period of irrigation with wastewater caused a giant “boom” in soil microbial abundance. This “boom” of microorganisms was probably related to the availability of easily degradable compounds or substrates brought following wastewater irrigation (Ramirez-Fuentes et al., 2002). Indeed, microorganisms are in particular heterotrophic and carbon restrained in soil, and the microbial variation was related to better availability of the carbon supply from wastewater irrigation. This might cause a transitory increase in soil microorganisms with a considerable resiliency conditioned by the stock of clean organic compounds (Lejon et al., 2007).

Bacterial community structure in the different WWTP-irrigated soils The different bacterial communities present in WWTP-irrigated soils over the short and long term are analyzed by denaturing gradient gel electrophoresis (DGGE) techniques, and the results are summarized in Fig. 6. The DGGE profiles showed significant variability in the structure of the bacterial communities in the different samples. The DNA bands increase in both number and intensity by increasing the year of WWTP irrigation. Thus, more DNA bands were detected in irrigated soil samples over 15 years (Fig. 6). These results of the molecular analyzes follow the results found by the agar enumeration culturing technique; so treated wastewater soil irrigation mainly leads to an raise, an enrichment and diversification of the soil microbiota. A positive correlation was found

Conclusion

This study reported the special effects of treated wastewater on agricultural soil irrigated for different duration of 1, 7, and 15 years and depths of 0–20, 20–40, and 40–60 cm. The reuse of treated wastewater in agricultural soil was essentially operated to balance water demand and supply in the world. A long duration of WWTP soil application allowed for an improvement of the organic and mineral stock into soil, the stabilization of soil pH, and the increase of soil cation exchange capacity. In addition, the practice of wastewater soil application largely affects the soil microbiota with an increase of microbial biomasses C and N and developing microbial activity with distinct changes in microbial community structure. Therefore, we can conclude that using treated wastewater in agriculture, in arid areas such as Tunisia, seems promising. However, WWTP Gabes used for irrigation looked contaminated with some pathogenic bacteria, and for improvement of the wastewater microbiological quality. Thus, a tertiary disinfection treatment is necessary for safely applying the treated wastewater and the harvesting and collection of safe crops.

3.4.3 Crops applications and monitoring

About the agricultural aspect this work represents the impact of alfalfa irrigation with treated wastewater on the microbiological characteristics of the soil

Using the counting method, the evolution of the number of cultivable mesophilic bacteria (PCA medium) and fungi (PDA medium) of alfalfa soil irrigated by the different wastewater treatments was determined in order to conclude the impact of the Alfalfa irrigation with treated wastewater on the microbiological characteristics of the soil. Figure 7 clearly represents the number of aerobic bacterial colonies counted and the number of fungi on the

PCA and PDA agar media from the different treatments of the alfalfa soil irrigated with the raw and treated EUS. The use of this combined Phytoremediation/Bio-augmentation process in the treatment of wastewater contaminated by phosphates aims to accelerate the phenomenon of elimination of this pollutant and to ensure its transformation into assimilable phosphorus by alfalfa and therefore the bio-fertilization of the soil by a purely biological refining.

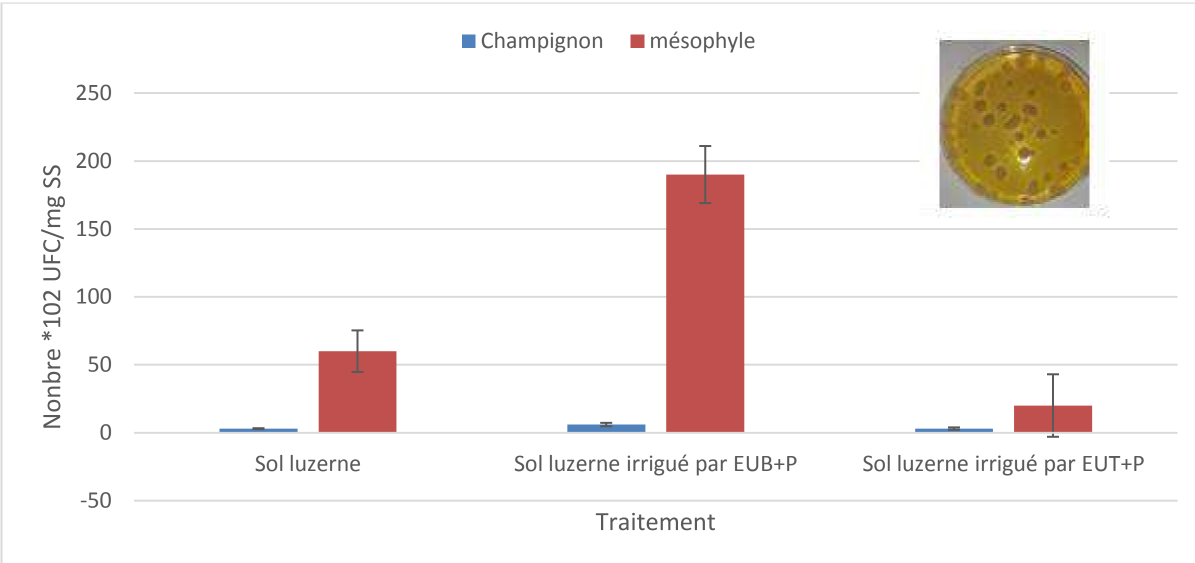


Fig.7: Result of enumeration of soil mesophiles Number of fungi (102 CFU/ml) in the different soil treatments

Introduction

In recent years, new alternatives have been mise en place, which consists in the use of EUTs to irrigate fodder crops such as alfalfa, mainly in Gabes, planted for livestock feed and dietetics. Alfalfa is widely cultivated in all climatic zones of the world, except the tropical climate. The main production regions are North and South America and Europe. Only 3% of the world's land area is cultivated in Africa but production is significant in North African countries, where it is grown under irrigation. More than 106,000 ha of alfalfa are cultivated in Morocco (i.e. 22.8% of the total fodder area of the country) 36,000 ha in Egypt. In Algeria, alfalfa covers around 10,000 ha. In Tunisia, alfalfa is the main source of food where it is grown on about 13,000 ha (77% in the oases).

This work was developed within the framework of the MEDISS project "Mediterranean integrated system for water supply". This project will test innovative solutions in the use of EUTs and the desalination of brackish water as well as specific awareness initiatives aimed at end users challenging their reluctance towards unconventional resources and formulating best agricultural practices will help reduce the pressure on fresh water as well as water supply costs. This work represents a comparative study between alfalfa irrigated by treated wastewater and that irrigated by borehole water in the oasis.

Study areas

- The oasis of Chenini which is a coastal oasis located on the coastal plain of Djefara which extends from Sfax to the Libyan border, it runs along the northern edge of the city of Gabès and extends for about 10 km towards West (Fig.8).
- Bechima which is located in the North-West and about 4 km from the city of el Hamma. It covers an area of 50 ha completely irrigated by treated wastewater.

These two areas have similar soil and hydrological characteristics while they differ in terms of climatic characteristics.

Preparation of pots and description of the method of harvesting alfalfa

The experimental part of this work was carried out in March and May at the Institute of Arid Regions of Gabès (IRA Gabès). Twenty jars were prepared and cleaned for the experiment. In each pot a pebble has been placed on the drainage hole to prevent it from clogging. Then, a bed of gravel was placed, equivalent to 20% of the pot volume to ensure good drainage and prevent root rot. The oasis soil mixed with a little compost (loam) is placed on the gravel and used as support. Twenty small alfalfa plants of the same age and same size were selected and repotted from the plots studied. (Photo 1,2).

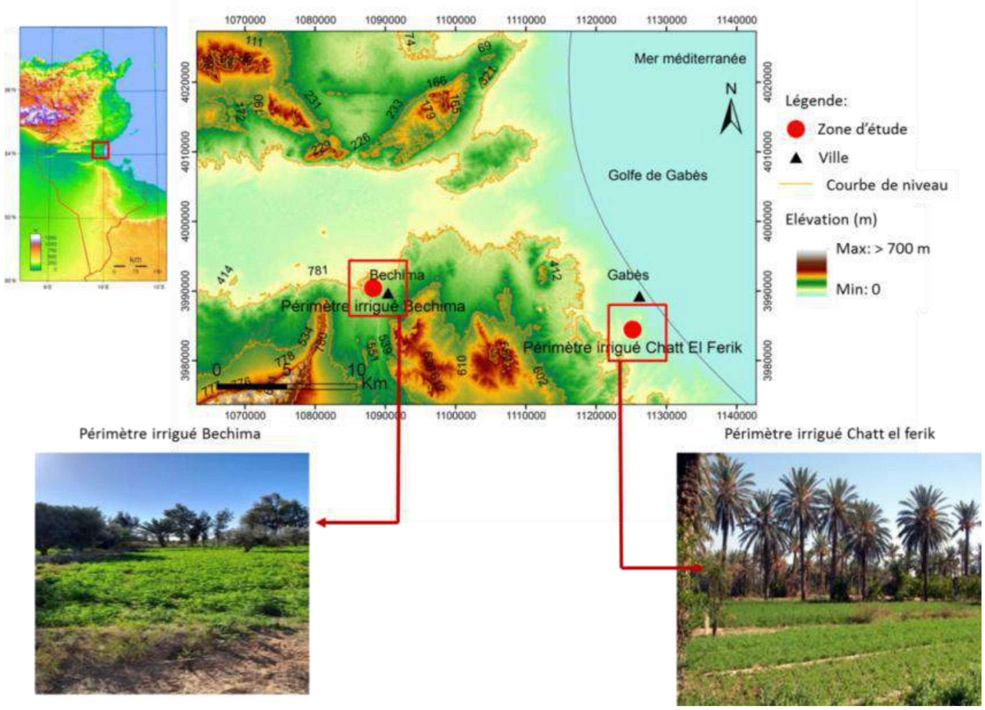


Fig.8 - Présentation de deux zones d'études.

Vegetative parameters of alfalfa

Effect of treated wastewater on production. Figure 9 represents the variation in production as a function of the two water qualities (EF and EUT). The comparison between the values of the MF of alfalfa shows that the EUT has a significant effect on the production in MF. This improvement can be expressed by the development of leaf area and stem length and is attributed to the enrichment of wastewater in nitrogenous elements and phosphorus. Our results are consistent with those found in the study by El Moussaoui et al. 2019 in Morocco.Dry matter is plant matter derived from organic matter that remains after elimination of all the water it contains. The results illustrated in figure 3 show the weight of the DM in g/plant. It should be noted that the DM of alfalfa irrigated by EUT exceeds that of the control with values of the order of 1.38 and 1.014 g respectively. These values are consistent with the results observed in other research (Fars et al, 2003; Rusan et al, 2007), which found an increase in dry biomass in forage plants irrigated by EUTs such as wheat and barley.

Photos 1,2: Transplantation de la luzerne dans les pots.

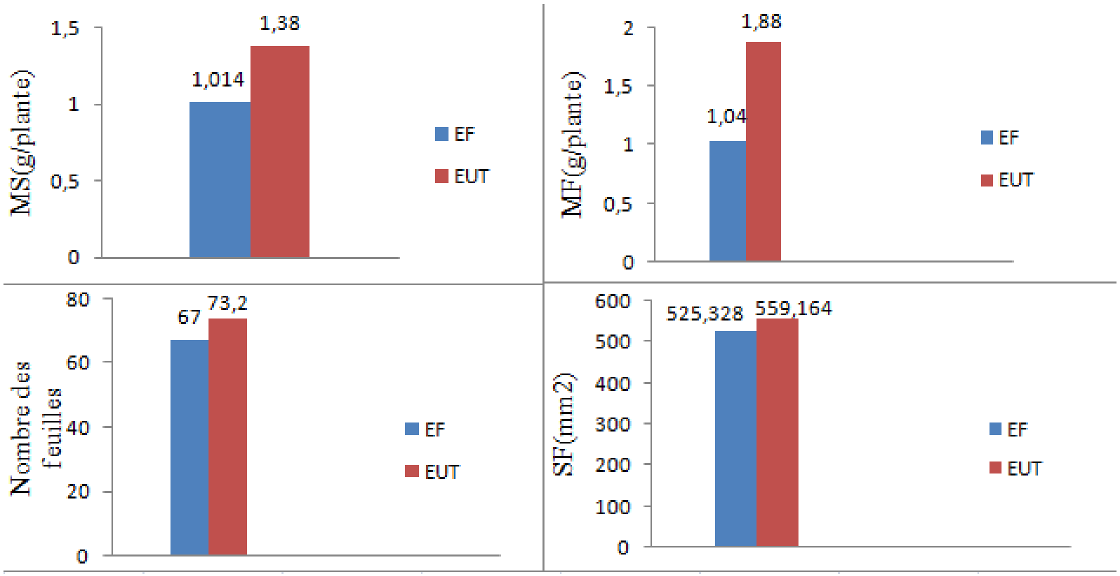


Fig. 9: Effect of treated wastewater on vegetative parameters in alfalfa.

The relative water content (TRE)

The relative water content is one of the preliminary parameters characterizing the water status of plants and therefore its decrease reflects dehydration of plant tissues. Figure 10 represents the variation of the ERR of the leaves according to two qualities of water carried out on the alfalfa. Reading the results reveals that the difference between the two means of ERR is not significant with values of 73.9% and 70.2% respectively for plants irrigated by EUT and EF. Knowing that the plant's ERR can vary between 68% and 84% under good water conditions, and from 45% to 63% in the presence of a water deficit (Radhouane, 2008). does not affect leaf turgidity in alfalfa and does not cause water stress.

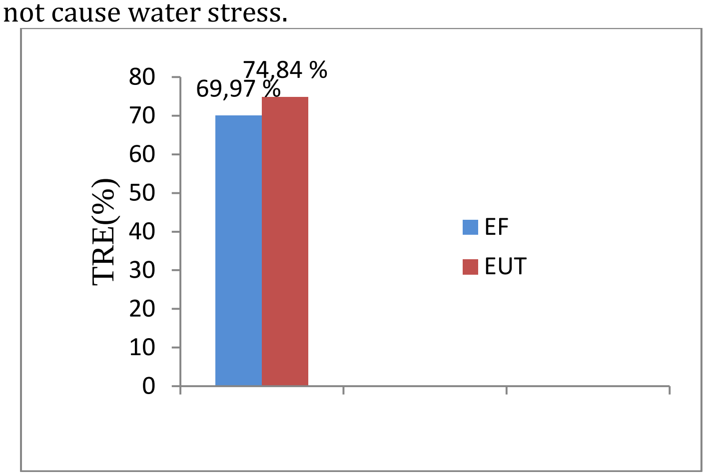


Fig. 10: Relative water content in alfalfa.

Variation des nodosités racinaires des plantes dans les deux parcelles

The effect of EUT irrigation on the number of plant nodules in the experimental plots (perimeter irrigated by borehole water and Bechima irrigated by treated wastewater) is shown in Figure 11. The results reveal a significant difference in the number of nodules in plants irrigated by EUT (3nodule/plant) compared to control plants (4 nodules/plant). The reduction in the number of nodules can be explained by the richness of EUT in nitrogen which generates the reduction of nodules in alfalfa. In fact, an excess of nitrogen in the soil inhibits the symbiosis since the plant no longer has any interest in investing energy in the nodules if it can simply absorb the nitrates from the soil . In this sense, Gassama-Dia, 1996 indicates that the quantity of nitrogen fixed by the rhizobial symbiosis is very variable according to the environmental conditions.

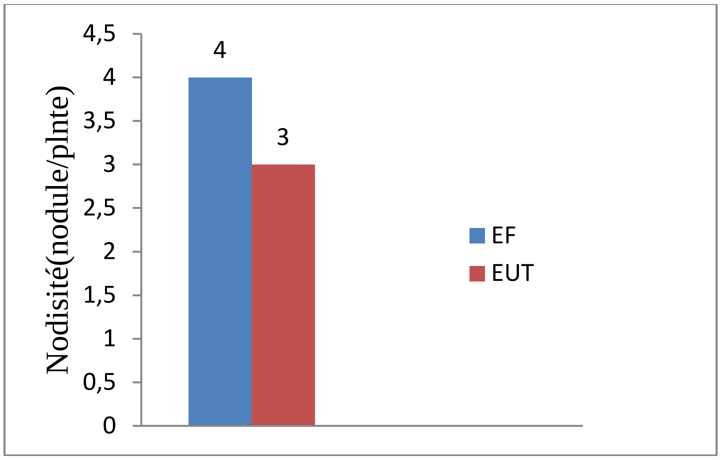


Fig. 11 - The effect of irrigation with treated wastewater on the number of alfalfa nodules in the experimental plots (Chatt Ferik and Bechima).

Trace minerals

The essential elements studied are calcium (Ca2+), potassium (K), magnesium (Mg2+) and sodium (Na+). The results show that plants irrigated by EUT are richer in potassium, magnesium and sodium with values respectively equal to 20.58ppm; 6.61µg/ml and 17.41µg/ml (table 5). This richness is due to the large quantity of nutrients in the EUTs. They are rich in nutrients such as nitrogen, phosphorus and potassium. For example, in France, the rate of wastewater discharge into rivers is estimated at 38% for nitrogen, 18% for phosphorus and 96% for potassium (Esculier et al, 2018). This proves that irrigated alfalfa by EUT can be good quality fodder for livestock feed in terms of quantity and quality. Except that we must respect international standards. From Table 2 we observe that Mg2+ is significantly more abundant in alfalfa leaves irrigated with EUT. This is a good indicator since Mg2+ plays an important role in photosynthesis and protein synthesis and any deficiency influences the quality of the forage (Chen et al. 2017). On the other hand, we note that the Ca2+ contents are much lower at the level of plants irrigated by EUT than those irrigated by EF. This is due to the lack of calcium in EUT or in the soil of the Bechima region. Also, this decrease in Ca2+ can be caused by the increase in K+ in the aerial parts of plants irrigated by EUTs (20.58 m/l) following the phenomenon of K/Ca antagonism (Dilmaghani et al, 2007).

Like the concentrations of macronutrients (N, P, K, …), the contents of micronutrients (Zn, Fe and Cu) in the plant can be affected by wastewater irrigation (El Moussaoui et al, 2019).

The results of the analyzes of these microelements in the aerial part of alfalfa are shown in Table 1 and show the abundance of Fe followed by Zn. It is observed that the iron (Fe), zinc (Zn) and copper (Cu, classified with heavy metals) contents are high in plants irrigated by EUT compared to those irrigated by EF. By irrigating with EUT, the Zinc content almost doubled from 0.33 to 0.76 mg/l compared to irrigation with groundwater. These results justify concerns about the bioaccumulation of microelements and heavy metals present in EUTs and used for plant irrigation. The risk of this accumulation comes down to their transfer to plants and their eventual entry into the food chain (El Moussaoui et al, 2019). These results obtained on the increase in these trace elements are in agreement with other research studying the accumulation of microelements caused by irrigation with treated and untreated wastewater (Rattan et al. 2005; Adrover et al. 2012).A long-term effects study conducted on forage plants in the United States irrigated with EUTs found a 68% increase in Zn concentration, without exceeding the allowable limits, while Cu levels decreased by 8%. (WHO, 2013).

Tabl.1: Trace mineral contents for alfalfa (irrigated by EUT and EF).

Elément	EF	EUT
Ca(mg/l)	17,54	12,71
K(ppm)	19,39	20,58
Mg (µg/ml)	4,79	6,61
Na(µg/ml)	16,64	17,41
Fe(mg/l)	1,25	0,79
Zn(µg/ml)	0,33	0,76
Cu(mg/l)	0,26	0,27

Conclusions

The results obtained during this study present values that are largely in line with national and international standards that regulate irrigation water. Indeed, these waters can convey concentrations of chemical elements positively or negatively affecting the physiological evolution of these crops, therefore we must remain vigilant regarding the use of treated wastewater in agriculture.

Our results also show that the irrigation of the alfalfa variety (Medicago sativa L) responds favorably to the use of EUTs in the Bechimma El Hamma wastewater treatment plant compared to EF (Control). This may be due to its better assimilation thanks to the nutritive contributions of these waters. This gave positive results on the percentage of germination, the leaf area, the dry matter and the fresh matter, the relative water content, the number of leaves and the number of nodules.

So we can conclude that the efficiency of the reuse of this water in agriculture which requires taking into account the initial composition of the soil, the nutrient load, moreover the recovery of this water preserves natural resources.

Following the promising results of this work, it seems necessary to consider a study on the optimization of the conditions of reuse of treated wastewater in the agricultural field. It is also essential to minimize the health risks generated by these waters and to expand the range of fodder crops irrigated by these waters.

Test site for soil monitoring - Tunisia

WP3 3.4.1, 3.4.2, 3.4.3 MONITORING DATA (water, soil, crops) – AL RISHA (Jordan)

WP3 Activity 3.4 Comparing baseline value with final data of monitoring water, soil and crops

3.4.1 Water monitoring and 3.4.4 Comparing baseline value with final data

The Wadi Araba basin is divided into a northern sub- basin and a southern sub-basin. The drainage divide between the two coincides with the groundwater divide and lies to the north of Qa' es Sai'diyeen on the western side of Jabal Al Risha, about 75 km to the north- northeast of Aqaba. The southern sub-basin drains into the Gulf of Aqaba while the northern one drains into the Dead Sea (National Water Master Plan (NWMP), 2004).

The monitoring of the water quality was carried out first on the reference well for the Al Risha Well No. 9 plant, and following the construction of the desalination plant, other measurements were carried out both on the well and on the water leaving the plant, intended for both irrigation and drinking use after tertiary treatment.

After the recent failures that occurred in the drinking water treatment and distribution plant managed by AWC, the MEDISS plant is guaranteeing drinking water to around 3000 people, as an alternative to the regional one. It is certainly the best result that MEDISS could achieve.

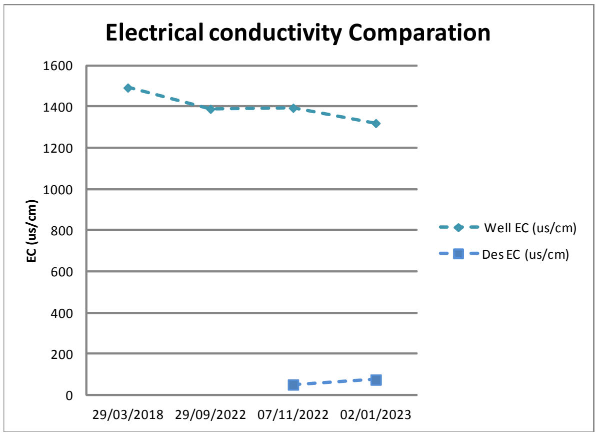
As for the irrigation used by Qaa Al-Saadain Association, it is from their own wells for their own farms as well.

The table show the results of the comparison of the tests between the company's well and the association's wells (29/9/2022) water properties:

	associations		*company*
Water well	Qa'a Al-Saadeen	Qa'a Al-Saadeen	Al-Risha well n.9
EC us/cm	2550	2480	1389
TDS mg/l	1632	1588	880
Turb. NTU	1.4	1	0,5
pH Unit	8,1	7,9	7,9

All the comparison after Al-Risha well n.9 and water produced from desalination plant was presented in the table and in the graphic below:

	Al-Risha well No.9				Desalination Plant				Desalination Plant (Brine Water)			
Data Sampl	Well EC (us/cm)	Well TDS (mg/l)	Well TUR (NTU)	Well pH	Des EC (us/cm)	Des TDS (mg/l)	Des TUR (NTU)	Des pH	Brine W EC (us/cm)	Brine W TDS (mg/l)	Brine W TUR (NTU)	Brine W pH
29/03/2018	1492											
29/09/2022	1389	880	0,5	7,9								
07/11/2022	1394	892	0,08	7,5	52	28	0,01	7,3				
02/01/2023	1320	845	0,2	8	76	41	0,2	6,7	5600	3024	3	8,1



In the table is showed also the brine water able to control the level of salt in the reject after water production in the plant. You can observe the difference between the EC values present in the well (hight) and those following the desalination of the water (very low) and how much salt has accumulated in the rejects leaving the plant.

3.4.2,3.4.3 Soil and crops monitoring and 3.4.4 Comparing baseline value with final data

The objective of this short-term experiment is to assess the feasibility of irrigating commercial vegetable crops with a suitable blend of fresh water with high-salinity water, the latter being the reject of the activity of one desalination plant, recently set up in the village of Al-Reesh (Araba Valley), through the MEDISS project. The plant, which started its operations in December 2022, treats the brackish underground water and makes it drinkable for the local community. The so called "brine water" is the reject of the plant, namely a very salty water fraction which contains the excess of salts, and it is typically considered as waste. The experiment has also the goal of identifying the best combination (in terms of relative proportion) between the underground water and the brine water, with the purpose of recycling the whole amount of the latter in agricultural activities instead of throught it away)(Fig.1).

Table 1 shows electrical conductivity and other chemical and physical properties of the 3 kinds of waters mentioned above: the underground water (low salinity) which is abstracted by a well and treated by the desalination plant; the desalinated water, which is supplied to the local community, and the brine water which currently represents a reject (waste, not reused water).

Fig.1 One component of the new desalination plant set up in Al-Risha by the AWC

Table 1 Electrical conductivity, salt concentration, turbidity and pH of the 3 kinds of water

Results for water wells (Al-Rishah well No. 9 and Al-Rishah Desalination Plant and Reject)

Date: 2/1/2023

Analysis	Al-Rishah well No.9	Al-Rishah Desalination Plant	Al-Rishah Desalination Plant (Brine Water)
EC us/cm	1320	76	5600
TDS mg/l	845	41	3024
Turb. NTU	0.2	0.2	3.0
pH Unit	8.0	6.7	8.1

The experiment

The farming environment and actors involved in the experiment

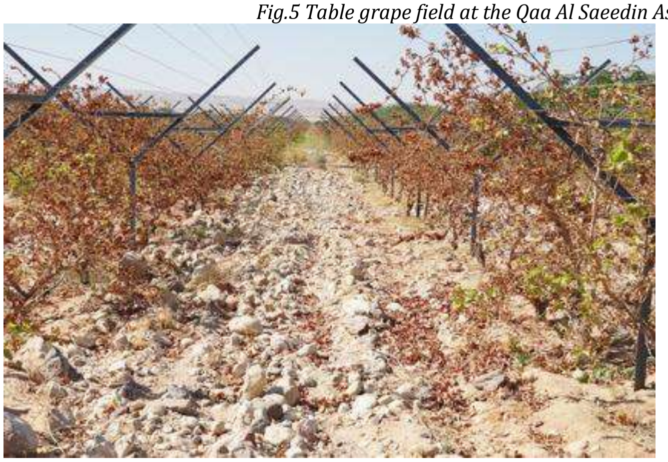
The farming environment is characterised by low rainfall and high plant evapotranspiration. High temperature and dry conditions make irrigation (through underground water) strictly necessary for obtaining agricultural production (Figure 2). Smooth winter temperature of the area, however, allows more crop harvests over the year. Produce is sold in the Aqaba market.

Fig. 2 The dry agricultural landscape of the Araba Valley

The Qaa Al Saeedin Association (Fig.3,4,5) is the agricultural partner of the Aqaba Water Company in this experiment. It is an agricultural association which owns 80 hectares of cropland. Thirty hectares are leased to an agricultural investor and the 50 remaining hectares are managed by the Association. There are two deep wells for irrigating the whole area; the abstracted water is then pumped in a water pool, and from there pumped to the fields. Main grown crops are tomato, water melon and potato. Table grape is grown too.

Fig.3 On the right, mr Abu Mshari, president of the Qaa Al Saeedin Association

Fig.5 Table grape field at the Qaa Al Saeedin Association (L) and the water pool used for irrigation (R)



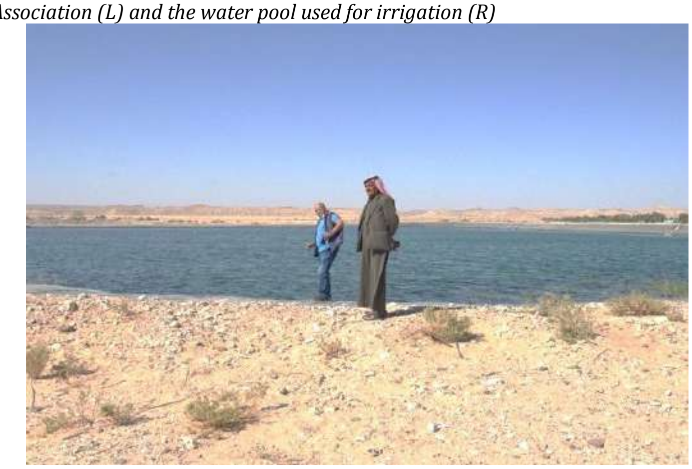


Fig. 4 Farmers of the Qaa Al Saeedin Association and tomato crop

Methods

In order to achieve the foreseen objectives of the experiment, the following methodology has been discussed and agreed with the person in charge of the AWC and with mr Abu Mshari, president of the Qaa Al Saeedin Association, who is the executor of the trial.

a) One plot of around 1,000 m2 shall be planted with water melon plants, according to the local standard technique.

b) Half of this plot shall be irrigated with underground water (treatment 1) and the other half with the "MEDISS water" (treatment 2), which is composed in part by the brine water coming from the desalination plant and in part by underground water. Table 31 shows chemical-physical characteristics of both the waters; groundwater is abstracted from the “Al-Reesh well no.9”.

c) Considering the EC of respectively brine water and water from "Al-Risha well no. 9", the calculated proportion is:

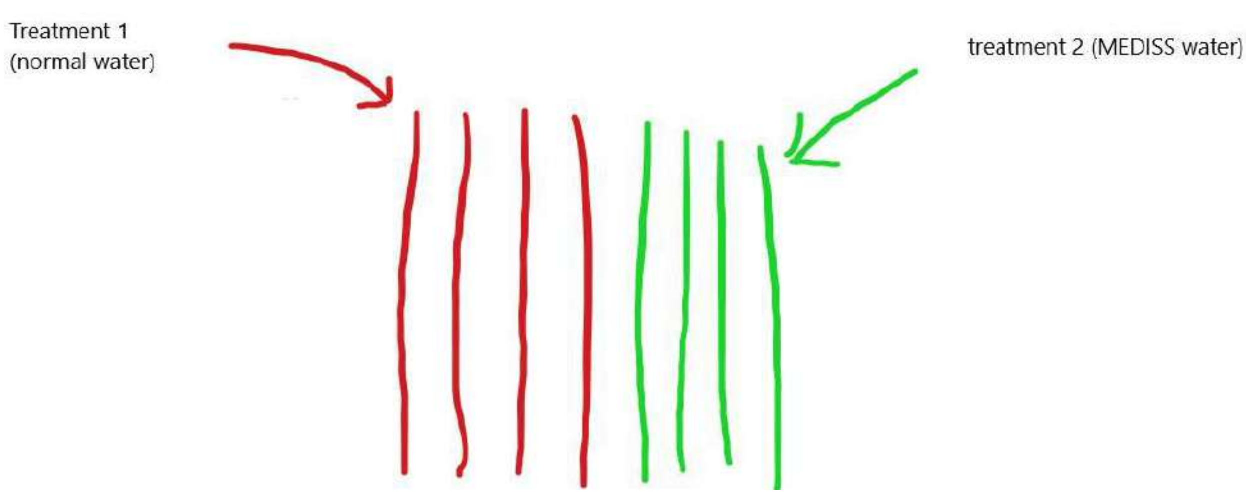
80%, water from "Al-Risha well no. 9" + 20%, brine water

E.g., if the irrigation dose is 1,000 liters, 200 liters must be of brine water and 800 liters of "Al-Risha well no. 9". **These two quantities must be mixed in a pond and then the resulting solution supplied to the plants, under treatment 2**. By doing so, the resulting EC will be 2.17 dS/m, which is deemed as an acceptable trade-off.

d) Summarising, the above-mentioned mix of the 2 waters will be used in treatment 2; while for treatment 1 only water from the "Al-Risha well no. 9" shall be used for irrigating the water melon plants (Fig. 6,7).

e) At the end of the period, differences in plant growth between the 2 treatments, and, possibly, in crop’s yield and quality, will be assessed by the consultant.

Fig. 6 The sketched layout of the trial



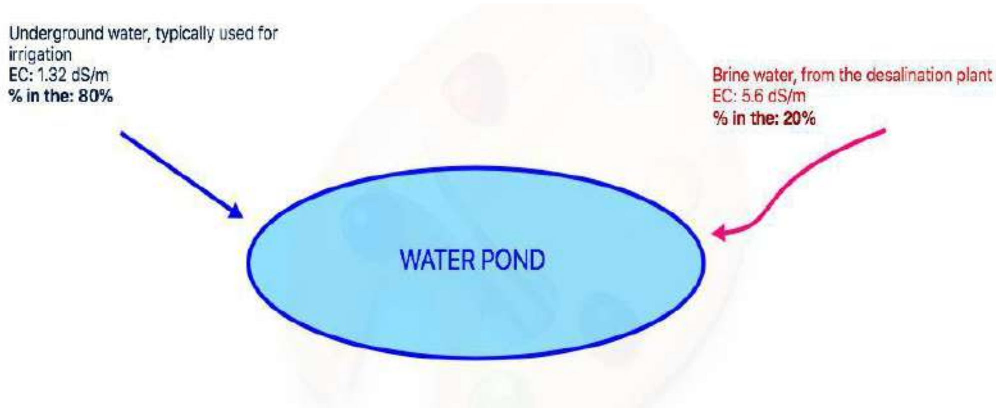


Fig.7 Mixing the 2 kinds of water in a pond

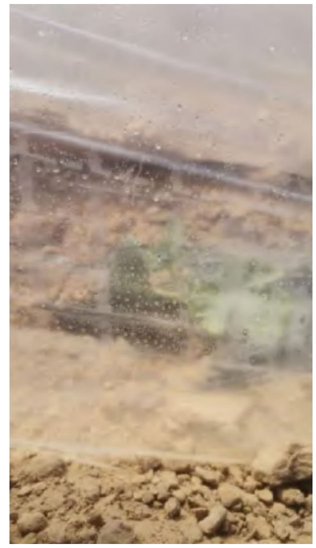
Status of implementation of the trial

Aqaba Water Company (AWC) has made available on its own cost an earth pool of the volume of 4500 m^3 equipped with a pump to pump the desalinated/ brine water to the farmers. The pump has a pumping capacity of 24 m^3/ hour.

The pool will store the desalinated water intended to be used in agriculture, AWC has started pumping quota part of the desalinated water to the farmers, the pool will be also used for the storing and later pumping of the brine.

The farmers would like to make experiments using blended desalinated water with saline water and brine in different blending ratios. Cultivation of seedlings in a 1.4 acres has started in a protected agricultural way; it is planned that 15 farmers members of Qa' Al Sa'adeen Farmers Association will benefit from the desalinated quota part and about 3000 inhabitants of Al Risha will benefit from the desalinated water for drinking and domestic use.

Farmers have opted for the cultivation of water melon (Figg. 22,23).



Figg. 22,23 Cultivation and growth of the plants irrigated by water from pilot pilot of MEDISS

It is worth mentioning that the original source of drinking and domestic water used for Al Risha population has dried/put out of service, the desalination plant is now the sole and main source of drinking and domestic water for Al Risha population.

Recommendations

The project agronomist “Timesis Company” and based on water analysis made by AWC of the desalinated water and the saline water has recommended water blending ratios, later analysis of the plants and the soil after having used desalinated water and different water blends shall be made for the sake of better calibrate the water blending ratio and for the sake of finding the most adequate plant to be irrigated with such water quality.

Results

Finally the results on pilot area had a double very important results for these environments highly sensitive to water resource scarcity:

1)Supply drinking water to a population of about 3,000 people, even in the event of an emergency;

2)Irrigate different types of crops including those with tomatoes (whose number of seedlings is being implemented within the project) and the water-melon which can give good results.

Annex - List of interviewed farmers in Al-Risha

1. Eng. Iyad al Dweak/private investor - Qaa AlSaeedin irrigation project (interviewed in site), with cultivated area of 21 hectares, currently potato;
2. Mr Mohammad AlSaeedin/private investor - Qaa AlSaeedin irrigation project (interviewed in site), with cultivated area of 41 hectares;
3. Mr Khaled Alryatie /private investor - Qaa AlSaeedin association (interviewed on phone / not available in site), with cultivated area of 30 hectares, currently watermelon;
4. Salem AlSaeedin / Farmer - Alriysha agricultural lands (interviewed on phone / not available in site), with cultivated area of 3 hectares, currently watermelon;
5. Awwad AlSaeedin/Owner - Qaa AlSaeedin irrigation project (interviewed in site).

WP3 3.4.1, 3.4.2, 3.4.3 MONITORING DATA (water, soil, crops) – JORDAN VALLEY (Palestine)

3.4.1 Water monitoring and 3.4.4 Comparing baseline value with final data

Groundwater is the main source of water for Palestinians in the occupied Palestinian territory (West Bank and Gaza Strip) and provides more than 90% of all water supplies. The main aquifer systems can be divided into four distinct units; the Western Aquifer Basin, the North-eastern Aquifer Basin and the Eastern Aquifer Basin for the West Bank, and the Coastal Aquifer for Gaza, where the groundwater is available at much shallower depth.

Following the 1967 occupation, Israel has controlled all shared water resources including surface and groundwater, and has utilized more than 85% of these resources, leaving only 15% for Palestinian use. The surface water in Palestine is represented by several seasonal wadis, as well as the Jordan River, which is currently controlled and used exclusively by the Israelis.

MEDISS project in Palestine:

Collecting surface water in Wadi Quilt, storing it in a ground pool, blending it with underground saline water and treated WW from Jericho WWTP.

MEDISS project objective in Palestine

- Increase quantity of irrigation water
- Increase quality if irrigation water

MEDISS project location in Palestine

Water monitoring progress

1- At the beginning, we identified the farms and wells from which we intend to take water samples.
2- 11 wells were selected from 11 different farms near to the pilot
3- We did the tests for the selected wells 3 times.
   1- First one at 15/12/2020 (After the rain)
   2- Second one at 11/11/2021(Before the rain)
   3- Third one at 22/6/2022 (Mid-year)
4- We did the tests for the water of some wells that were mixed with the treated water (after pilot)

Results:

Sampling date: 15/12/2020 Region: Jericho / Palestine

Test date: 15/12/2020 Weather: Sunny

Sample name ➢ ------------------- Test ⮛		Daeeq (Dahia) Farm	Daeeq (Mashro) Farm	Abu Raed Farm A1	Abu Raed Farm A2	Abu Yazan Farm	Abu Al-Ezz Farm	Musab Farm
Temperature	c	25	25	25	25	25	25	25
COD	mg/L	50	100	100	200	200	400	600
TDS	PPT	1.35	2.83	1.65	2.09	1.4	0.85	2.86
EC	mS/cm	3.6	6.18	3.33	3.82	2.81	1.7	5.38
PH	PH	7.3	7.4	7.5	7.5	7.7	7.3	7.6
Nitrate	mg/L	0.01	0.02	0.04	0.05	0.01	<<	0.03
Chloride	mg/L	18	29.5	16.5	19	14	10	26
HCO3	mg/L	100	230	90	145	145	130	105

Sampling date: 11/11/2021 Region: Jericho / Palestine

Test date: 11/11/2021 Weather: Sunny

Sample name ➢ ------------------- Test ⮛		Daeeq (Dahia) Farm	Daeeq (Mashro) Farm	Abu Raed Farm A1	Abu Yazan Farm	Abu Al-Ezz Farm	Musab Farm
Temperature	c	25	25	25	25	25	25
COD	mg/L	50	100	100	150	140	170
TDS	PPT	1.5	2.95	1.75	1.5	5.3	2.9
EC	mS/cm	4.1	6.8	4.3	3.05	11.8	6.1
PH	PH	7.3	7.5	7.9	7.8	7.2	7.6
Nitrate	mg/L	0.01	0.01	0.03	0.01	0.04	0.03
Chloride	mg/L	21	34.1	19.1	15.7	58.4	31
NaCl	mg/L	34.5	57.1	30.1	24.8	95.7	49.6
HCO3	mg/L	90	110	85	120	130	105

Sampling date: 22/6/2022 Region: Jericho / Palestine

Test date: 22/6/2022 Weather: Sunny

Sample name ➢ ------------------- Test ⮛		Daeeq (Dahia) Farm	Daeeq (Mashro) Farm	Abu Raed Farm A1	Abu Yazan Farm	Abu Al-Ezz Farm	Musab Farm	
Temperature	c	25	25	25	25	25	25	
TDS	PPT	1.41	2.82	1.71	1.43	2.3	2.9	
EC	mS/cm	3.9	6.2	3.8	2.9	11.8	5.5	
PH	PH	7.3	7.4	7.7	7.8	7.3	7.6	
Nitrate	mg/L	0.01	0.01	0.02	0.01	0.03	0.03	
Chloride	mg/L	19	32.1	17.1	14.5	50.4	27	

Sampling date: 1/11/2022 Region: Jericho / Palestine

Test date: 1/11/2022 Weather: Sunny

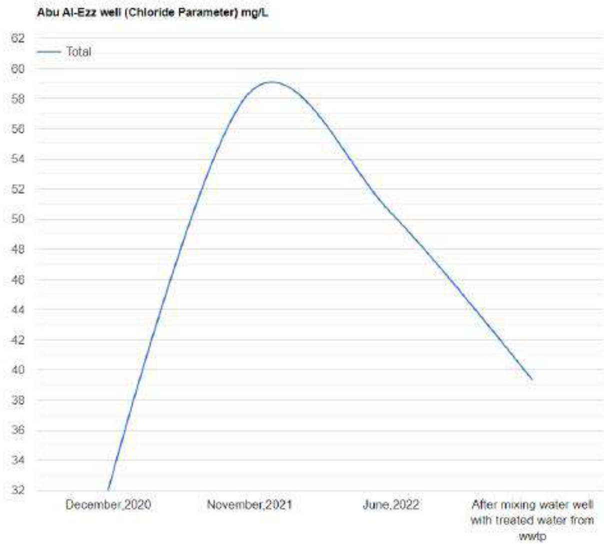
Sample name ➢ ------------------- Test ⮛		Treated water from WWTP	Daeeq (Mashro) Farm	Daeeq (Mashro) Farm Mixed with Treated water	Abu Raed Farm A1	Abu Raed Farm A1 Mixed with Treated water	Abu Al-Ezz Farm	Abu Al-Ezz Farm Mixed with Treated water
Temperature	c	25	25	25	25	25	25	25
TDS	PPT	0.8	1.75	1.5	1.75	1.5	5.3	4.2
EC	mS/cm	1.6	6.8	5.1	4.3	3.4	11.8	9.3
PH	PH	7.6	7.5	7.5	7.9	7.8	7.2	7.3
Chloride	mg/L	7.8	34.1	26.1	19.1	15.3	58.4	39.3
NaCl	mg/L	12	57.1	36.6	30.1	23.4	95.7	69.3

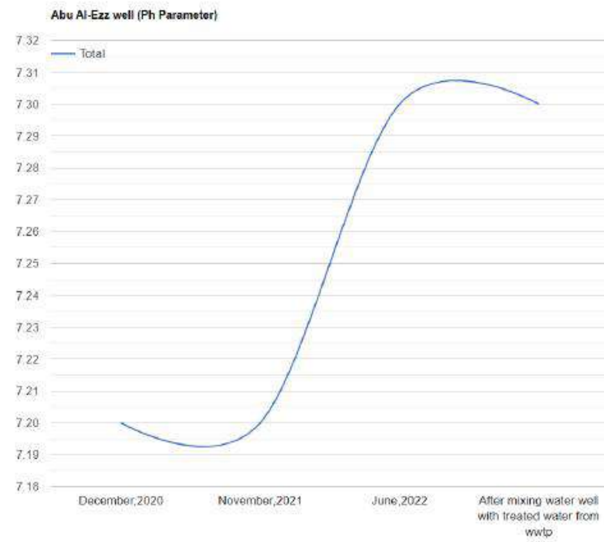
Comments: Treated wastewater was mixed with wells water at ratio 1:3

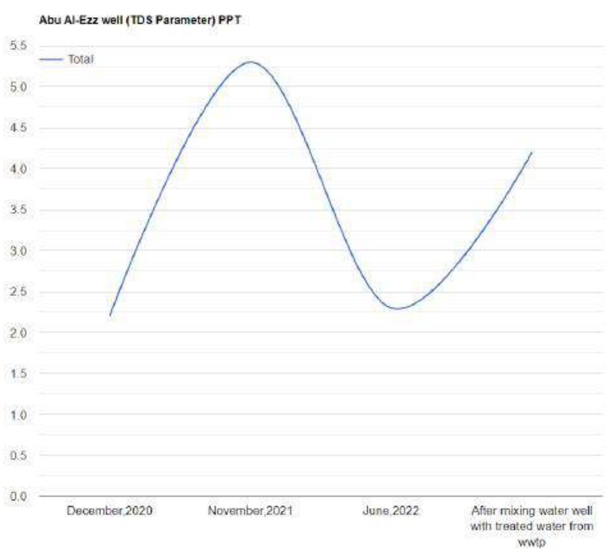
Summary

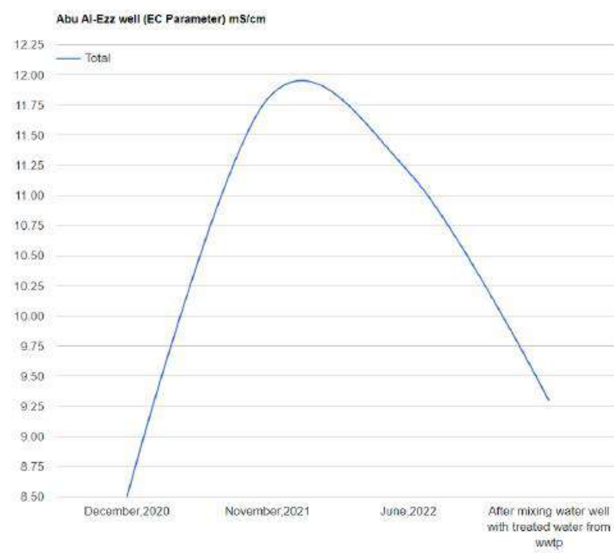
It is clear from the above analysis and comparisons that water has much better quality once is blended with treated wastewater, this is clear from all tested parameters comparing them to those of the blended water. (See below Graphs)

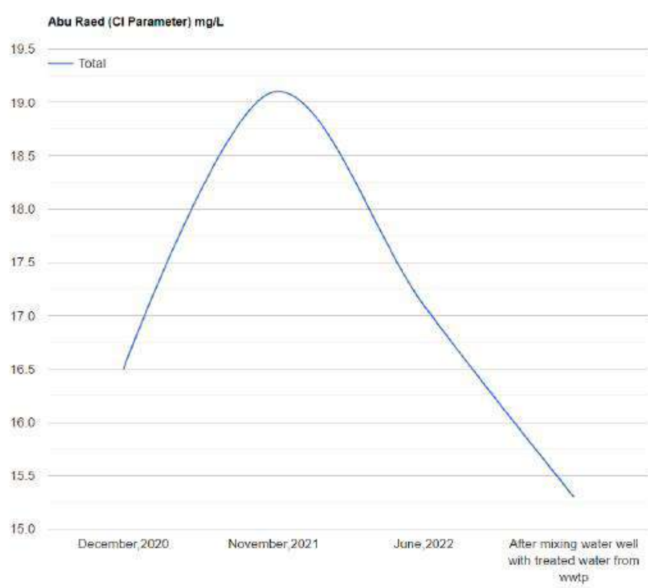
Abu Al Ezz farm is using the largest amount of treated wastewater, its well has the most saline water (about five times the salinity of the other wells), once it is blended with treated wastewater with a ratio of one to three, the water quality is significantly improved, it is recommended that he mixes it further with wadi water.

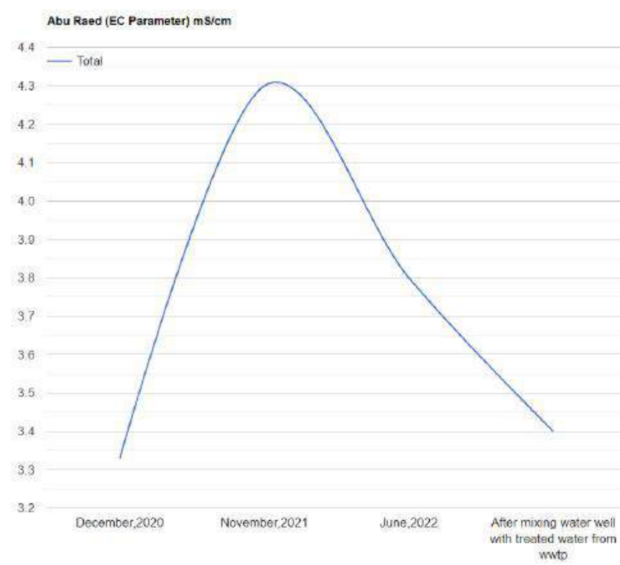

Abu Al-Ezz well (Chloride Parameter) mg/L
Total
62
60
58
56
54
52
50
48
46
44
42
40
38
36
34
32
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp

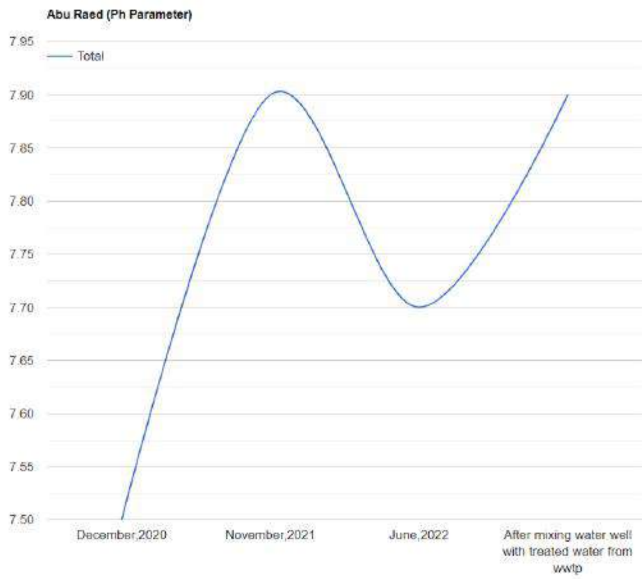

Abu Al-Ezz well (Ph Parameter)
Total
7.32
7.31
7.30
7.29
7.28
7.27
7.26
7.25
7.24
7.23
7.22
7.21
7.20
7.19
7.18
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp

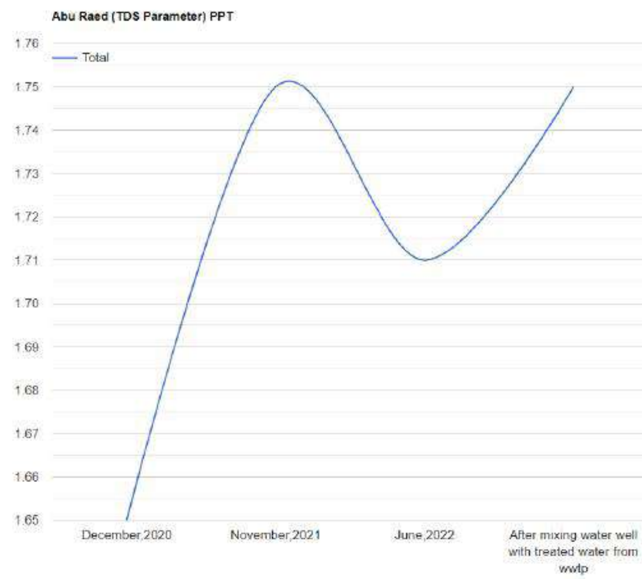

Abu Al-Ezz well (TDS Parameter) PPT
Total
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp

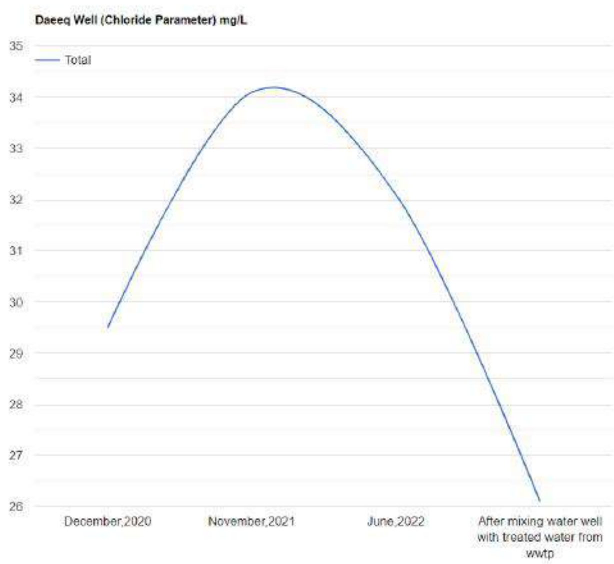

Abu Al-Ezz well (EC Parameter) mS/cm
Total
12.25
12.00
11.75
11.50
11.25
11.00
10.75
10.50
10.25
10.00
9.75
9.50
9.25
9.00
8.75
8.50
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp

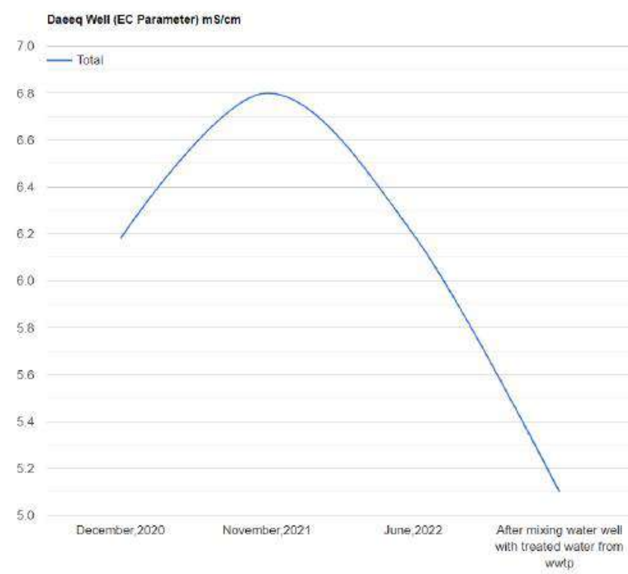

Abu Raed (Cl Parameter) mg/L
Total
19.5
19.0
18.5
18.0
17.5
17.0
16.5
16.0
15.5
15.0
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp

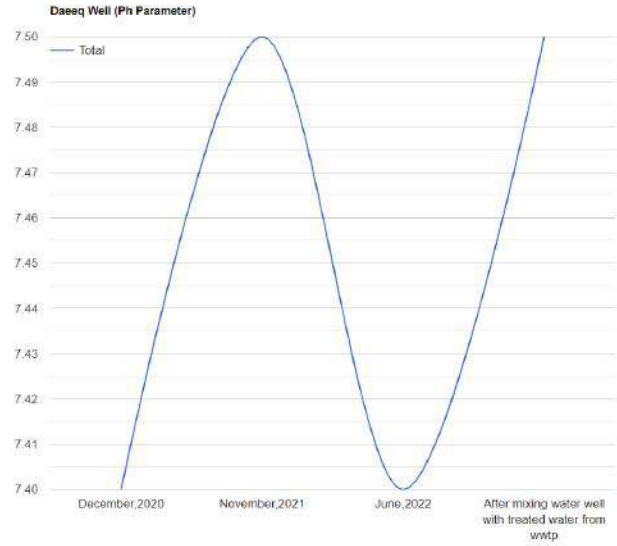

Abu Raed (EC Parameter) mS/cm
Total
4.4
4.3
4.2
4.1
4.0
3.9
3.8
3.7
3.6
3.5
3.4
3.3
3.2
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp

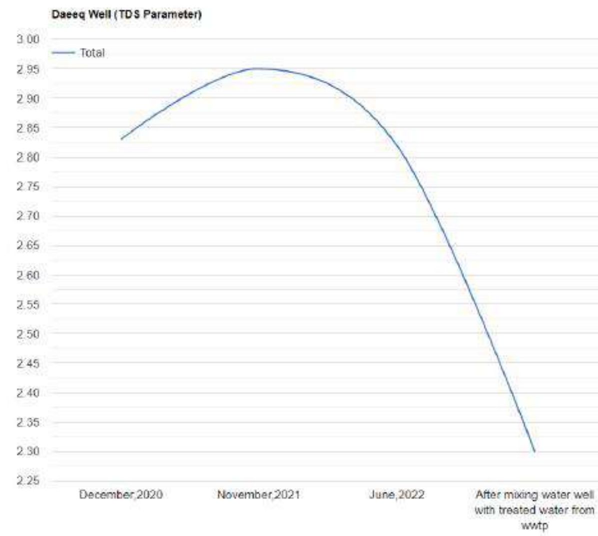

Abu Raed (Ph Parameter)
Total
7.95
7.90
7.85
7.80
7.75
7.70
7.65
7.60
7.55
7.50
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp


Abu Raed (TDS Parameter) PPT
Total
1.76
1.75
1.74
1.73
1.72
1.71
1.70
1.69
1.68
1.67
1.66
1.65
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp


Daeeq Well (Chloride Parameter) mg/L
Total
35
34
33
32
31
30
29
28
27
26
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp


Daeeq Well (EC Parameter) mS/cm
Total
7.0
6.8
6.6
6.4
6.2
6.0
5.8
5.6
5.4
5.2
5.0
December,2020
November,2021
June,2022
After mixing water well with treated water from wwtp





QGIS map shown farms location

3.4.2,3.4.3 Soil and crops monitoring and 3.4.4 Comparing baseline value with final data
Two monitoring missions have been carried out in Jericho by the agronomist and the soil scientist, from 7 to 12 February and from 19 to 23 September 2022. Seven commercial farms, growing date palms in the Jericho area (Figure 1), were studied during the monitoring period to see the effect of the project's activities on their agricultural performance and soil quality.

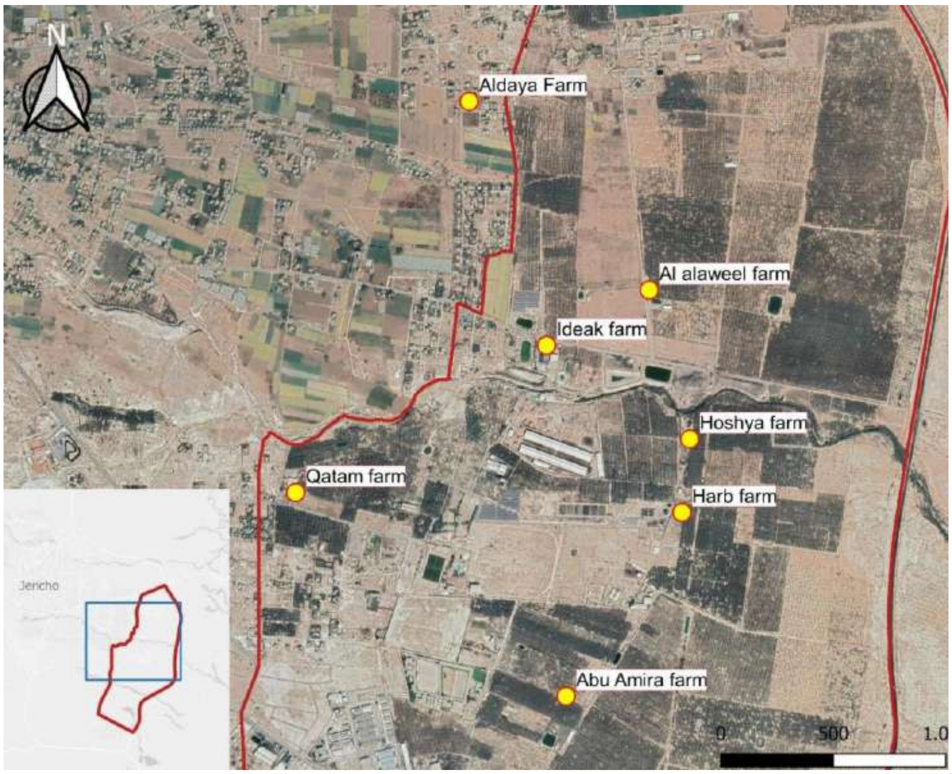


Fig. 1 The seven Farms monitored by the consultant over the period in the project area (red outline)

The soil monitoring survey

The soil monitoring campaign was realized by two missions in February and September 2022 (Fig.2) Timesis soil expert together with Jericho Governorate engineers collected soil samples in seven date farms using different irrigation water sources located in different parts of Jericho. In particular, the farms were Abu Amira, Alawaeel, Aldahya, Harb, Hoshiya, Ideak, Qatam. In each farm, four sites were chosen, and in each site 4 topsoil samples were collected around a chosen palm tree. A total of 28 topsoil samples (September) and 28 topsoil (0-10 cm) and 28 subsoil samples (10-50 cm) (July) were collected and analysed. A field characterization of the soil materials and the surface properties (salt crusts, efflorescences, humidity, etc.) was performed as well.

Lab analysis (Al Quds University Soil Lab in July and Milano University, Pedology Lab in September) were performed in order to define electrical conductivity (EC) and pH values which are the soil properties which can quickly react to salinity values in irrigation waters. Both EC and pH were measured on a 1:2.5 soil-water ratio.

Using the formulas shown by Sonmez et al. 2008 (Geoderma 144(1-2):361-369), the EC value for saturated paste, ECsat, and total dissolved solids (tds) were obtained, comparable to suitability data for agricultural crops. In particular, using the formula for clay-loam soils, EC in saturated paste (ECsat) is calculated as EC(1:2.5)*3.75, and tds is then obtained as EC(sat)*800.

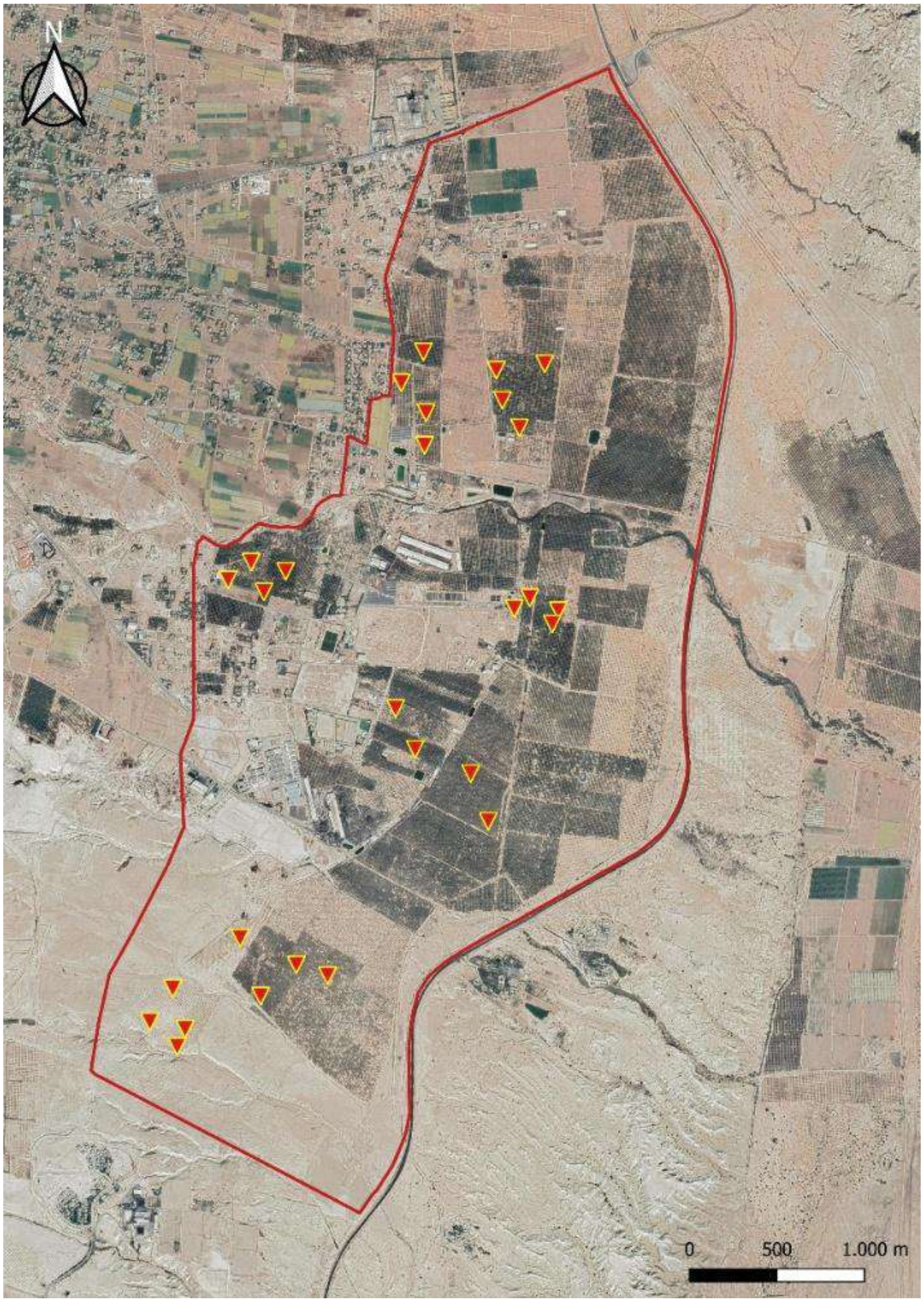


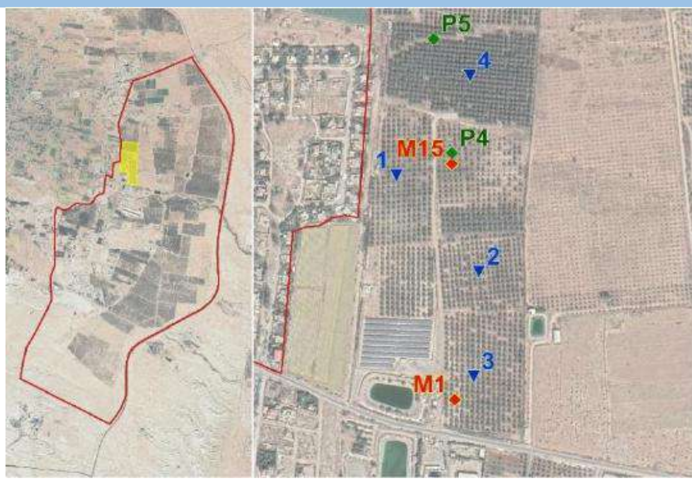
Fig. 2 Location of the monitoring soil samples in the study area

Soil salinity after irrigation

The following table 1 shows the results of lab analysis for pH and Electrical Conductivity for summer and winter samples.

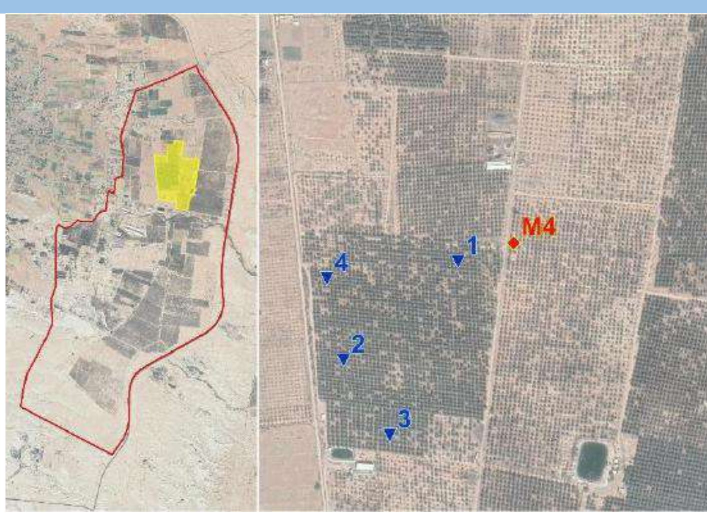
Table 1 Electrical conductivity and pH values in topsoils in monitoring samples collected in September 2022 and in older ones, collected in winter 2021-2022

ALDAHYA



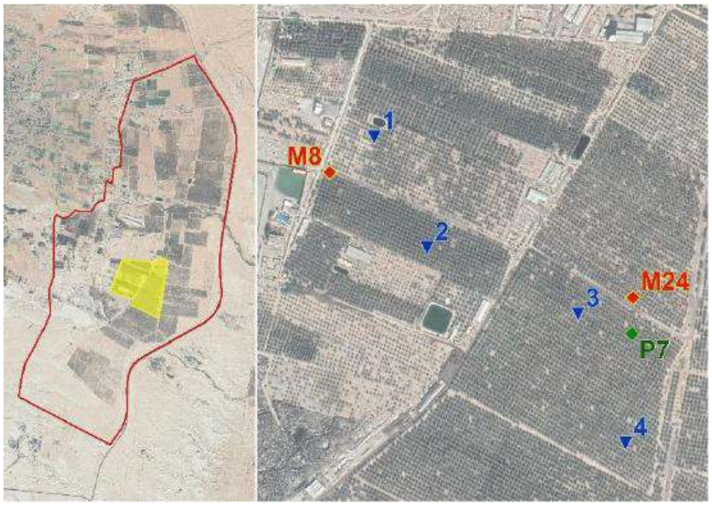
	Id sample	pH	EC (ms/cm)
Winter	M1	8,1	17,8
	M15	7,9	6,2
	P4	8,5	2,4
	P5	8,4	20,3
Summer	1	8,4	60,2
	2	8,4	36,2
	3	8,3	24,5
	4	8,6	52,6

ALAWAEEL



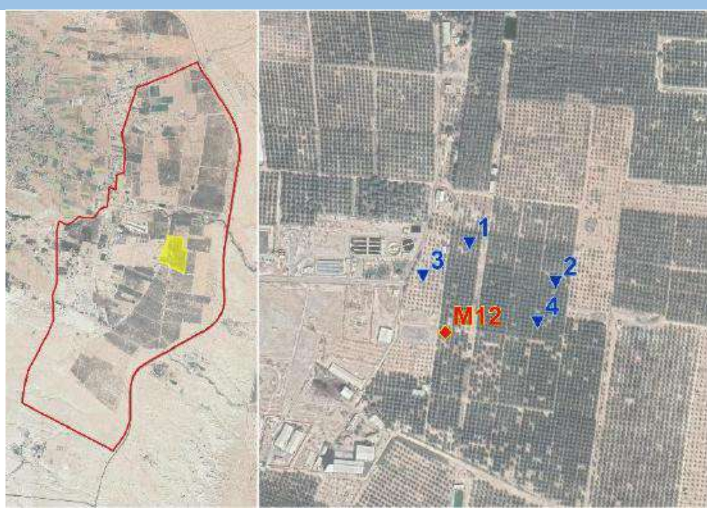
	Id sample	pH	EC (ms/cm)
Winter	M4	8,3	8,1
Summer	1	8,4	33,6
	2	9,1	114,7
	3	8,1	50,8
	4	9,3	91,6

ABU AMIRA



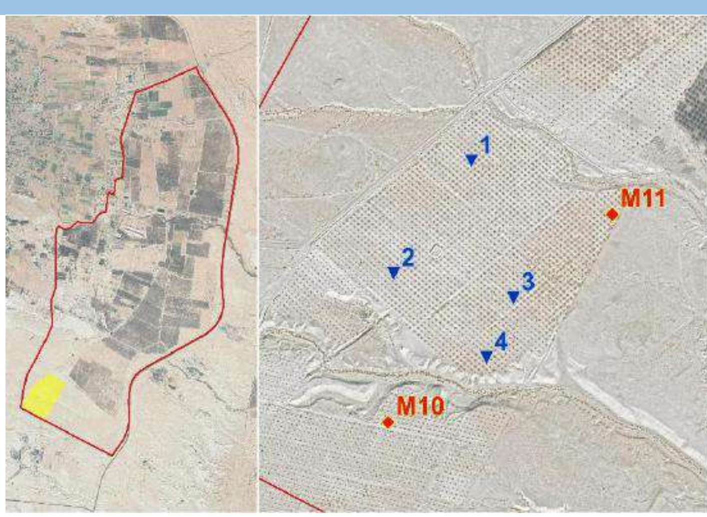
	Id sample	pH	EC (ms/cm)
Winter	M8	7,6	13,5
	P7	8,8	37,8
	M24	8,5	3,4
Summer	1	7,8	64,5
	2	8,6	31,8
	3	8,7	21,5
	4	9,1	28,8

HARB



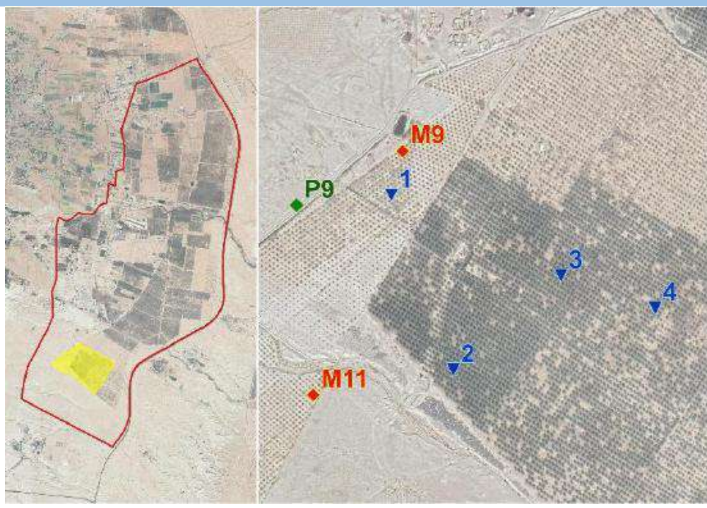
	Id sample	pH	EC (ms/cm)
Winter	M12	8,5	15,5
Summer	1	8,7	84,9
	2	9,3	33,6
	3	9,4	105,7
	4	8,4	73,1

HOSHIYA



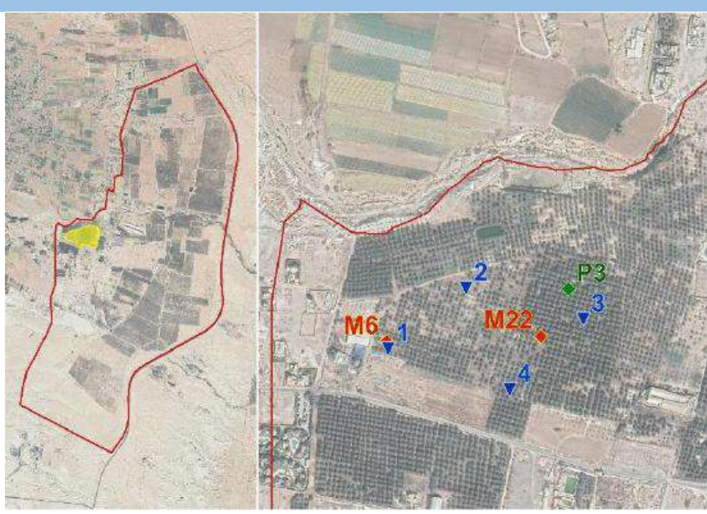
	Id sample	pH	EC (ms/cm)
Winter	M11	7,6	22,0
	M10	7,3	21,6
Summer	1	9,3	50,0
	2	8,9	36,7
	3	9,3	50,8
	4	8,6	38,8

IDEAK



	Id sample	pH	EC (ms/cm)
Winter	M9	7,5	60,5
	M11	7,6	22,0
	P9	8,5	9,1
Summer	1	7,8	34,9
	2	7,5	66,1
	3	7,8	65,1
	4	7,8	48,7

QATAM



	Id sample	pH	EC (ms/cm)
Winter	M6	7,7	18,7
	M22	7,9	134,1
	P3	8,1	19,6
Summer	1	7,9	66,9
	2	8,5	51,5
	3	8,7	36,5
	4	7,8	54,7

Comparing topsoil salinity values measured in September 2022 with nearby samples collected and analysed in February 2022, we can see a generalized increased salinity in areas where winter data were lower, while the highest values did not change significantly. A much smaller variability in the EC data has been measured in late summer (Fig.4).

Such data, converted into EC (sat), show that the soils sampled in September 2022 are all in the extremely saline range (ECsat>16).

The generalized increase in EC values (indicating salinity) is related to the hot and dry summer characterizing the climate in Jericho, increasing evapotranspiration and salt accumulation on the soil surface, and to the winter rains which partly removed salts from the soil surface, particularly where the soils are not extremely saline.

An effect of irrigation water is however also visible, even if precise data of the irrigation water salinity and origin are not always available. In fact, salt efflorescences and crusts were observed on wet soils quite often, close to irrigation infrastructures and in the wettest soils (fig. 3).

Fig. 3 Saline crusts and efflorescences on humid soils and irrigation waters in September 2022

Table 2 shows the origin of irrigation waters used by the seven farms. Hoshiya, for example, mostly uses water obtained from a highly saline private well, while Qadam uses mostly freshwaters collected from the Wadi during winter in large quantities, with additions of strongly saline waters from a private well. The flushing effect of salts caused by the large freshwater use in Qadam when it’s available is visible as there is no increase in soil salinity during summer compared to winter.

Nevertheless, overall, the soil salinity data are not significantly different in the different farms during summer.

Table 1 Irrigation water main sources in the seven farms, and average soil EC measured in September 2022 and in February 2022

Farm	Water sources used for irrigation	Links with MEDISS water	Average soil EC (summer, mS/cm)	Average soil EC (winter, mS/cm)
Aldahya	Underground water, plus wadi water in winter, via its own water pool, plus treated water	No	43.4	16.9
Alawaeel	Blend of underground water with the one provided by the association, plus own link to treated water and wadi water in winter, plus own well (3000 ppm)	Yes	72.7	8.1
Abu Amira	Its irrigation system is linked to that of the association. It has 2 wells; one of which very salty (11,000 ppm); the other has low discharge; the water is a blend between these water sources plus treated waters	Yes	36.7	18.2
Ideak	Spring water, not saline (1400 ppm), coming from the Betlehem aquifer	Yes	53.7	28.3
Hoshiya	3 owned wells, saline (from 3,500 to 5,000 ppm). It is linked to the palm farmers association's pipeline but the farmer does not use that water	Yes	41.6	22.0
Harb	Blend of underground water (3000 ppm, saline, but with little availability) with the one provided by the association	Yes	71.6	15.5
Qatam	Underground water (strongly saline, 5500 ppm) plus wadi water in winter, via its own water pool	No	52.4	57.5

Table 3 Surface appearance in the sampling points in the seven farms

Sample	Surface aspect
Aldahya - 1	Soft salt crust, cemented layer close to the surface locally
Aldahya – 2	Allochtonous brown soil, very few salt efflorescences
Aldahya – 3	Allochtonous brown soil, very few salt efflorescences, with natural cemented soil locally close to the surface
Aldahya – 4	White soil, no salt crust
Alawaeel - 1	
Alawaeel – 2	Strong saline crust
Alawaeel – 3	Salt efflorescences but no crust, soft and loose soil
Alawaeel - 4	Soft soil with hard salt crust on the surface
Abu Amira - 1	Strong saline crust, many efflorescences
Abu Amira – 2	Soft and weak crust, salt efflorescences below 6-8 cm
Abu Amira – 3	Hard crust
Abu Amira - 4	Hard crust, salt efflorescences on 50% of the surface
Ideak – 1	White soil
Ideak – 2	Thin brown soil layer (allochtonous) with salt efflorescences where irrigated and humid
Ideak – 3	White soil with brine on top, wet
Ideak – 4	Many efflorescences
Hoshiya – 1	Strong crust, soil is mixed brown (allochtonous) and white soil (autochtonous)
Hoshiya – 2	Soft brown soil with no salt evidences
Hoshiya – 3	Crust under the surface, salt efflorescence where wet
Hoshiya – 4	Salt crust and efflorescences where it is soft
Harb – 1	Allochtonous soil recently added on the surface, petrogypsic close to the surface locally
Harb – 2	Salt crust on the surface and very hard salty horizon below
Harb – 3	Very strong and thick salt crust on the surface
Harb – 4	Crust close to the irrigation pipeline
Qatam – 1	Salt efflorescences, wet
Qatam – 2	Very hard, almost cemented but wet
Qatam – 3	Wet
Qatam – 4	Wet

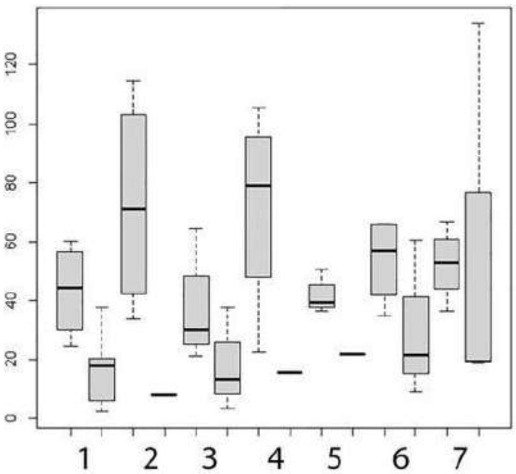


Fig.4 Electrical conductivity in soils in the 7 farms in alphabetical order; summer data on the left, winter ones on their immediate right

Soil salinity after irrigation, comparison between land unit and soil type

The effect of Land Unit and, thus, soil types is also not particularly strong. As visible in fig. 5, the highest salinity values are in Soil Unit 7 and 9 (not significantly)

Soil Unit 9 mostly includes Soil Type 3, which is a well-drained soil formed from alluvial deposits, moderately saline and strongly sodic; Soil Unit 7 includes Soil Type 6, characterized by the presence of a petrocalcic/Petrogypsic horizon at shallow depth and quite high salt contents. The two types are thus very different, and none of them is the most saline soil type in the area, verifying the hypothesis that irrigation water is the most important factor during the summer months.

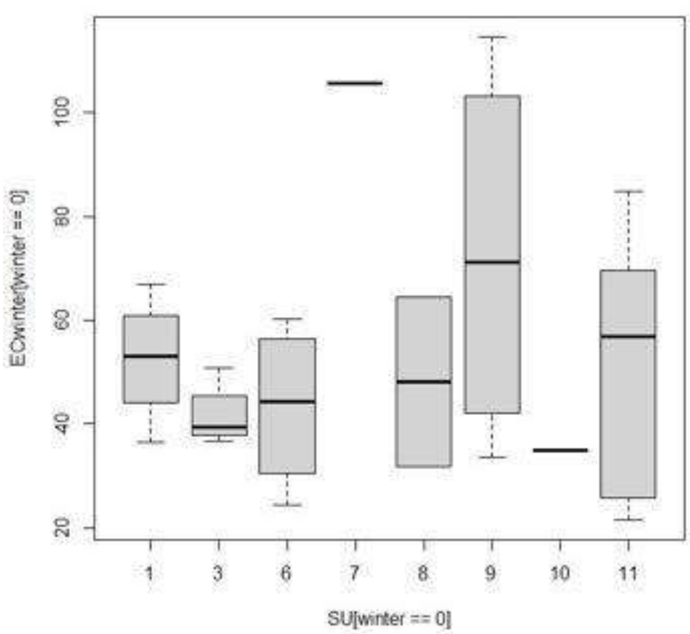


Fig. 5 Electrical conductivity in soils, measured in September 2022, in the soil map units in the 7 farms

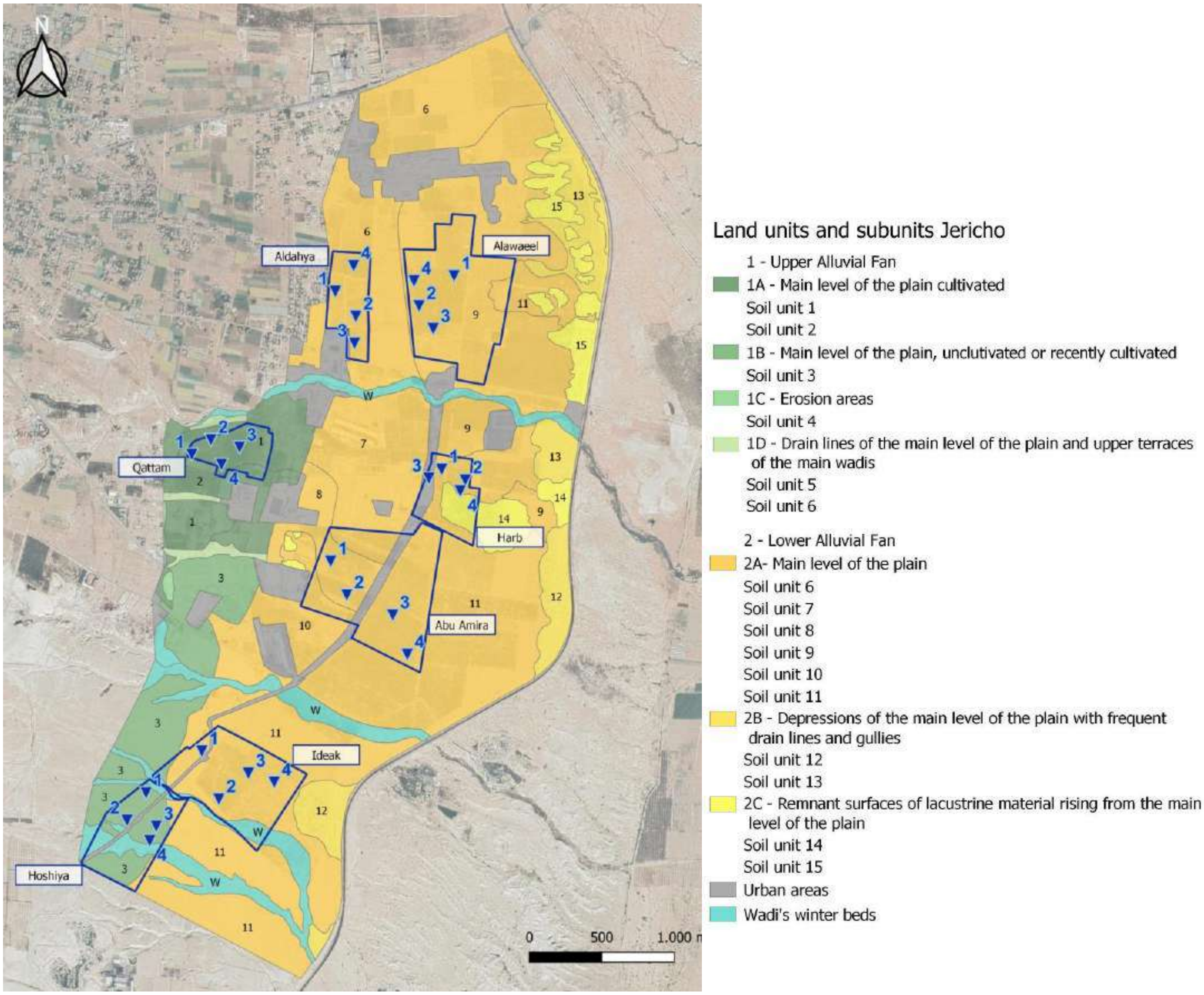


Fig. 6 The Soil map of Jericho area and the sampling points of September 2022 in the seven farms

Notes on soil management

In order to improve crop growth in saline soils, the excess salts must be removed from the root zone. For this purpose, the most effective method is leaching. Leaching is accomplished by ponding fresh water on the soil surface and allowing it to infiltrate; salt dissolves in water, becomes mobile and can be removed from the soil. Leaching is effective when the washing water is discharged out of the area under reclamation. Excess water, with dissolved salts, must be removed from the area to avoid that the salts can go back to the surface, or in any case in the rooting zone, subsequently by evapotranspiration when irrigation has ended.

it is therefore very important that the area under reclamation has a good natural drainage system. Failing that, it is strongly recommended to set up a network of surface and subsurface drains.

Leaching should preferably be done when the soil moisture content is low and the groundwater table is deep. It is also very important to use good quality water, which contains a low content of dissolved salts.

Background information on date palm cultivation and water issues in Jericho

Date palm (*Phoenix dactilyfera*) represents the most important crop in the Jordan Valley, because of its important economic return (figure 7,8). Despite this enterprise requires high investments and has relevant productions costs, it fetches the best prices, thanks to a strong demand from foreign countries.

Intensive cultivation of date palms (CV Madjool, for export and CV Barhi, for local consumption – figure 3) started in Jericho around 25 years ago. Traditionally, main crops grown in the area were vegetables, so to name Jericho as the "food basket of the West Bank". Vegetables have been always grown in the fertile land in the close surroundings of the urban area. Main/unique outlet of this produce is the domestic market, but in recent times the area grown with vegetables is being reduced more and more because the value of the land for housing is passing the value of same land when cultivated.

Fig. 7 Date palm orchard in Jericho

Fig. 8 Dates, CV Medjoul (L) and CV Barhi (R)

Date palms are grown far from the urban area, in the surrounding desert land which is affected by a certain degree of salinity. The cultivation of date palms is expanding because of its high economic value. All the first-grade production of Medjoul is exported (UK and Arab countries being the main markets). Low grade production is sold in the domestic market, also for the production of the very popular "ajua" (date paste) which is used to prepare many kinds of traditional cakes.

Soil and water salinity are a concern for the date palm growers of Jericho (figure 4). The plant is classified as "tolerant" to salinity. Indeed, according to scientific literature[1], the crop can tolerate water and soil salinity up to 2,900 and 4,300 TDS, respectively, while ensuring 90% of the maximum production. The production decreases to about 70% when water and soil salinity reaches 4,700 and 7,000 TDS, respectively. However, when salinity ranges around the highest value in the soil solution, yield and quality of the dates are reduced, according to the interviewed farmers.

Palms in Jericho need around 23,000-25,000 m3/ha/year of water, to sustain the production and the physiological needs of the crop.

Ground water wells are the only source of water available during the major part of the year when the trees are in high need of water, namely from April to September. During the winter (October to March), the water of the local river, the wadi Al Khalil, is available as well for irrigation. Farmers pump this low-saline water to the palms with the specific purpose of "washing" the root zone from the salts accumulated during the summer season (Fig.9).

Most of groundwater, which fed all the wells used by the farmers, has medium to high salinity (from 2,500 up to 10,000 TDS). Palestinian farmers are not allowed by the Israelis occupation army to dig wells deeper than 90 metres. In the palm area, many wells have been dug so the common aquifer is over exploited, causing increase of salinity.

Fig. 9 . Scattered white spots on soil surface, resembling typical salt efflorescence

Reuse of treated waste water for irrigation purpose

A limited group of farmers is using, over the whole year, the waste water treated by the waste water treatment plant (WWTP) of Jericho to irrigate their palm orchards (figure 10). Each farmer has its own link to the "irrigation tank" of the WWTP. The treated water is then pumped in the farm's water pool where it is mixed with the ground water (abstracted from private wells) in order to reduce the salinity of the latter. Quality of the treated waste water of the WWTP of Jericho is reportedly good, as it has very low salinity (ranging from 800 to around 1,000 TDS).

[1] FAO. 1985. Water quality for agriculture. By: R.S. Ayers and D.W. Westcot. FAO Irrigation and Drainage Paper 29 Rev. 1. Rome, Italy

As explained by the director of the WWTP, the plant could daily produce around 6,400 m3 of treated water. However, various neighbourhoods of Jericho city and surroundings are not yet linked to the main sewage system dumping into the WWTP. Therefore, nowadays only one of the 2 lines is working, producing 2,500 m3/day of purified water, available for irrigation. All the treated water is re-used by the palm growers, during the period April-September. Afterwards, the share reused decreases and in December and January just 50% of the total water is taken by the farmers. Farmers pay the water one year in advance to the municipality of Jericho, and the cost is 1 NIS/m3 plus the cost for the electric power of their pumps, also supplied by the municipality.

A low number of farmers use treated water because of its limited availability during spring and summer. Moreover, as the agronomist of the Palm Farmer Cooperative Association explained, many farmers are reluctant, if not opposed, to using treated wastewater, as they believe it can damage date palms and jeopardize production. Furthermore, there is mistrust towards the reuse of wastewater due to religious reasons.

Fig. 10 The "irrigation tank" at the WWTP of Jericho. Pipelines and pumps of the single farmers who buy this water are shown. The pipeline of the MEDISS project is visible as well, under the roof

Current farmers' strategy for irrigating the date palms and limiting soil salinization

Most of the farmers have in their properties water reservoirs to mix together water from different sources, such as rainwater (harvested during winter time), water from the stream (figure 11), treated waste water (if purchased from the WWTP) and ground water. However, there is no a fixed ratio of mixing the various typologies of water; each farmer follows his own "recipe", according to the available kinds of water.

Fig. 11 The water stream flowing in Jericho, the wadi Al Khalil

Ground water is often saline and majority of wells are progressively reducing the discharge, as reported by the interviewed farmers. As mentioned above, in order to limit the process of soil salination due to the irrigation with saline water, farmers use to wash the soil surrounding the palms with fresh water during wintertime. Fresh water originates from rain and/or the local stream and it is prior stored in the private reservoirs (figure 12).

Fig. 12 A water pool of a commercial farm. It optionally accumulates rain water, stream water, underground water and, in some few cases, treated waste water from the WWTP of Jericho

An interesting initiative to improve supply and quality of irrigation water for the farmers is the one being developed by the Palm Farmer Cooperative Association in Jericho and Jordan Valley – PFCA and the Date Water Users Association - DWUA, established in 2021. The 2 organisations are closely linked to each other. The DWUA presently has 24 members.

The initiative consists in building about 30 km of a common water line, U-shaped, to send the water abstracted from 5 wells (belonging to members of the Association) to all the members of the Association. However, the 5 wells abstract water which has various degrees of salinity (figure 13), as almost all the wells of the area.

جمعية مستخدمي ري النخيل
Dates Water Users Association

التاريخ: 16.6.2022

المهندس معتز الحروب المحترم
محافظة اريحا والأغوار

تحية طيبة وبعد...

الموضوع: نتائج فحص الملوحة لمياه الآبار

بالإشارة إلى الموضوع أعلاه، تشهد إدارة جمعية مستخدمي ري النخيل بأن النتائج التالية هي نسبة الملوحة في آبار المياه المزودة للخط الناقل التابع لجمعية مستخدمي ري النخيل والذي يقوم بتزويد لأعضاء الجمعية.

الرقم	البئر المزود	نسبة الملوحة PPM
1	A 001	1,450
2	A 002	1,830
3	A 003	1,940
4	A 004	1,790
5	A 005	2,670

مع فائق الاحترام والتقدير

جمعية مستخدمي ري النخيل

Hazard	TDS (ppm or mg/L)	dS/m or mmhos/cm
None	<500	<0.75
Slight	500-1000	0.75-1.5
Moderate	**1000-2000**	**1.5-3.00**
Severe	>2000	>3.0

The current price of the water charged to the members of DWUA is 1.6 NIS/m3, including the water cost paid to the owners of the 5 wells and maintenance of the line. So far, a segment of 16 km of the target length has been realised. The remaining part will be reportedly completed by 2023.

Fig.13 Salinity of ground water of the 5 wells which feed the strategic water project of DWUA. All 5 wells show from moderate to severe salinity

The MEDISS Project and the improvement of the water strategic plan of DWUA

The MEDISS Project has implemented a new line to pump the waste water from the WWTP to one big reservoir with a capacity of 80,000 m3 (figures 14, 15, 16 and 17). This reservoir is currently being enlarged. In this pond, the water from the local stream (during winter time), the ground water from the 5 wells plus the treated waste water are mixed in adequate proportions to obtain a resulting water with acceptable salinity. This water is then diverted in the main pipeline and used by all the DWUA's members willing to pay for it (about 0,50 ILS/m3).

Fig.14 The reservoir used by the MEDISS Project to mix together the waters of different sources

Fig.15 The MEDISS reservoir: detail of the pumping station built by the project

Fig.16 The pumping station set up by MEDISS at the WWTP of Jericho

The effect of the MEDISS Project's action on dates production

The effect of the blended water on growth and production of the palms of seven farmers have been monitored by the consultant during the growing season, in the period from February to September 2022 (harvest time). Table 30 provides locations and water information on the seven concerned farms. Those farmers grow from 1,500 to 10,000 palm trees in their farms.

Not all the seven farmers had the chance, however, to use the water from the DWUA's new pipeline because of some technical problems which occurred during the starting phase of the action. Nevertheless, the consultant took into account the opinions of the farmers on the innovative project's approach and on the effect of it on the performance of their palm orchards.

Table 4 Characteristics of the monitored date palm farms

Farm	Coordinates	Water sources for irrigation	Notes
Aldaya farm	31°51'22.35"N 35°29'29.22"E	Underground water + stream water in winter, accumulated via its own water pool + treated waste water	Not yet linked to the DWUA's pipeline. Link to the WWTP.
Alawaeel farm	31°50'55.97"N 35°30'0.62"E	Blend of underground water with the one provided by the DWUA + treated waste water + stream water in winter	The farm's irrigation system is linked to that of DWUA + own wells (3,000 TDS) + link to the WWTP
Abu Amira Farm	31°49'56.96"N 35°29'48.85"E	Blend of treated water with underground water	The farm's irrigation system is linked to that of DWUA. It has 2 wells; one of which very salty (11,000 TDS); the other has too low discharge
Ideak farm	31°50'47.49"N 35°29'43.62"E	Fresh water from a mountain spring, (1,400 TDS)	No link to the association's pipeline. Water from the aquifer of Bethlehem
Hoshiya farm	31°50'34.65"N 35°30'8.26"E	Exclusive use of underground water from 3 owned wells, saline (from 3,500 to 5,000 TDS)	It is linked to the DWUA's pipeline but it does not use that water, at present. It fully relies on the water abstracted by his own 3 wells, although saline
Harb farm	31°50'24.02"N 35°30'7.35"E	Blend of underground water with the one provided by the association	The farm's irrigation system is linked to that of DWUA. The well is moderately saline (3,000 TDS), but low availability of water
Qatam farm	31°50'25.10"N 35°29'2.14"E	Underground water + stream water in winter, accumulated via its own water pool	The farm's irrigation system is not linked to that of DWUA, yet. The farm has 1 well, with salty water (5,500 TDS). In the next future, it will link its system to the pipeline of the association

Farmer evaluation of the MEDISS's approach to irrigation water improvement

The general problem raised by almost all the interviewed farmers is about the bad quality of the underground water and its progressive decline in quantity. As mentioned above, Palestinian farmers are not allowed by the Israeli government to dig deep wells in their own properties. Such limitation, in conjunction with the growing need of water for irrigation by the expanding cultivation of date palms in the surrounding of the city of Jericho, is causing the depletion of groundwater and its salinization.

Because of this now structural problem, farmers are increasingly resorting to the use of alternative water, such as treated water. Along with this, farmers are more and more aware of the importance of recycling rainwater in an efficient manner, through its harvest and storage during the winter months. All farmers recognise the problem of salt accumulating in the soil, due to continuous use of groundwater. To overcome this problem, they practice the technique of washing the soil during the winter, irrigating with fresh water from the rain and the Al Khalil stream. However, the amount of underground water available for irrigation during spring and summer remains low and often insufficient to match the crop requirements, therefore treated water is becoming more and more important, as further resource. But treated water presents two specific problems. The first is objective, in that it is available in limited quantities. The second is subjective, as the vast majority of farmers interviewed, despite using it, believe that it is not good for either the palms or the soil when used as the sole source of irrigation water, although its salinity is not significant. Added to this is a certain reluctance to use treated water for religious reasons.

All farmers have recognised that the result achieved by the MEDISS Project, namely the preparation of a sustainable blend of different alternative kinds of water for irrigating the date palms, is quite positive. This outcome, associated to the strategic water project which the DWUA is completing, can concretely contribute to come with the problem of water scarcity to sustain date palm production. The system will be further improved and strengthened in the following months. Results in the field are promising. The photos taken by the consultant during harvest September 2022 show that dates production, obtained through the sustainable blend of waters, is of satisfactory quality with high percentage of fruits which can be classified as "super-jumbo", namely fetching the highest price in the international market (Fig. 18,19,20,21).

Fig. 18 Medjoul dates about to be harvested (L) and (R) dates just harvested, under selection process

Fig.19 Nasser Hismail Abu Qatam, the owner of the Qatam Farm

Fig.20 Phases of dates sorting to divide the fruits in quality classes

Fig.21 The business area of the Hoshiya farm, an important exporter of Medjoul dates

Annex - List of interviewed farmers in Jericho

Name	Occupation	Organization	Location
Eng. Ahmad Al Fares	Head of Department of Agriculture Jericho and JV	MoA	Jericho
Eng. Omer Sawaftah	Agriculture control Head of Division	MoA	Jericho
Eng. Amin Daraghmeh	Extension Protected Vegetables Head of Division	MoA	Jericho
Awad Daraghmeh	Extension Date Palm Head of Division	MoA	Jericho
Eng. RaieK Bsharat	Plant protection Head of Division	MoA	Jericho
Eng. Ibrahim Seibah	Jericho effluent water treatment plant Manager	MoJ	Jericho
Mr. Judeh Iseed	Palestinian Paysants Union Head of Jericho branch	PPU	Jericho
Ayman Al Taweel	Farmer- date Palm	Private	Jer- Target area
Ismael Daik	Farmer- date Palm	Private	Jer- Target area
Jamal Jarrar	Farmer- date Palm	Private	Jer- Target area
Omer Bsharat	Arab Development Project Society GM	ADPS	Jer- Target area
Ramadan Hmaidat	farmer- vegetables and date Palm	Private	Jericho
Harb Farhan	Farmer dates	Private	Jer- Target area
Imran Al Walagi	Farm Manager- ADPS	ADPS	Jer- Target area
Faisal Abu Sakker	Farmer dates and watermelon	Private	Jer- Target area
Belal Eseed	Farmer dates	Private	Jer- Target area
Eisa Eisawi	Farmer dates	Private	Jer- Target area
Mosaab Ameereh	Farmer dates	Private	Jer- Target area

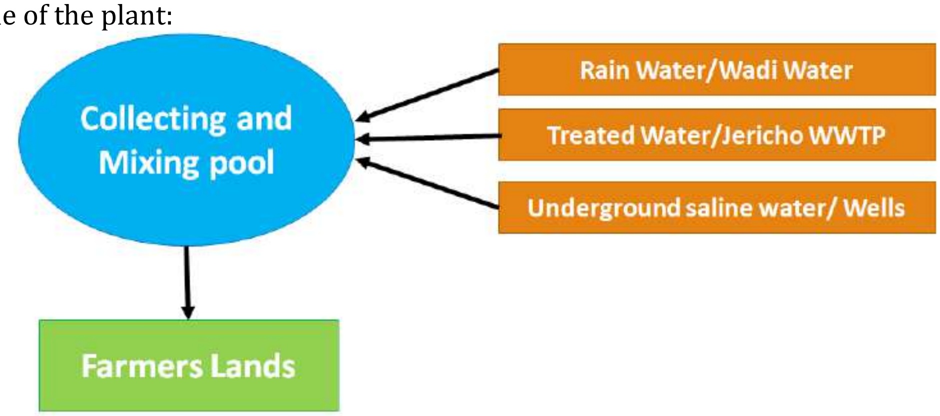
PILOT PLANTS

WP4 – Outlines of Pilot Plant – JERICO (PALESTINE)

The project concerns collecting surface water in Wadi Quilt, storing it in a ground pool, blending it with underground saline water and treated WW from Jericho WWTP.
The pilot area is located

The challenge is to increase quantity and quality of irrigation water to implement old and new crops.

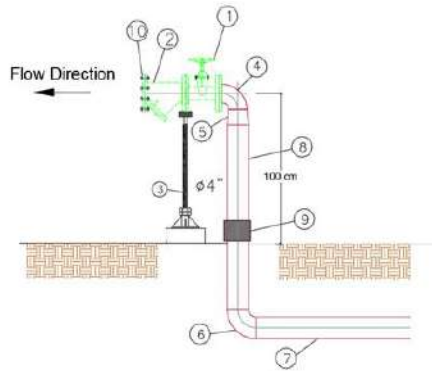
The scheme of the plant:



To connect the pool with farms a system of pipeline according to the following scheme was realized:

To connect farmers land pipe line with the main project pipe line

Connection point :
Contain valves , Strainer and Water Meter



In these photos you can find the different steps of the built:



The pilot system is coming and the results of water utilisation is presented in the chapter of this report.

WP4 – Outlines of Pilot Plant – AL RISHA (JORDAN)

Project Goal:

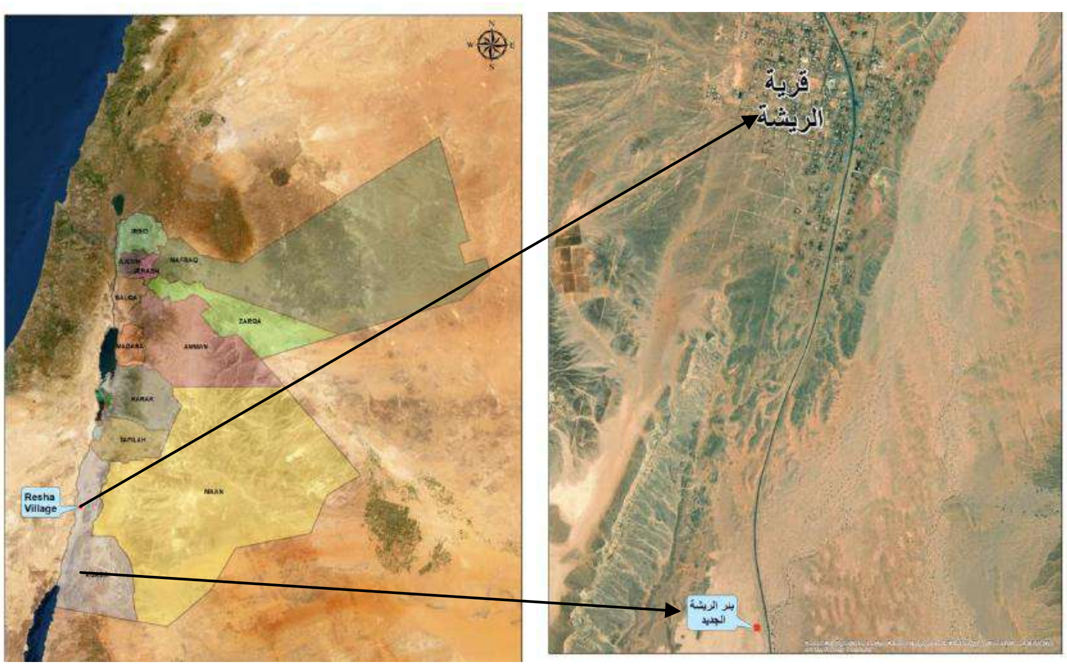
The project aims to desalinate the water of The new Well No. (9), which will provide the people of the region with water suitable for human consumption , the production capacity of up to 48 m3/h, the people of the region will benefit from the water returned from the desalination process by producing some agricultural crops, which will help them feed their livestock, and contributes to the development of remote and rural areas , Al Risha village is considered one of the pockets of poverty in Jordan.

- • **Contractor:** Gama Engineering Contracting Est.)
- • **Supervisor:** Aqaba Water Company
- • **Project Funder:** 90% from ENI CBCMED, 10% Aqaba Water
- • **Contractual value:** JD 244,130.

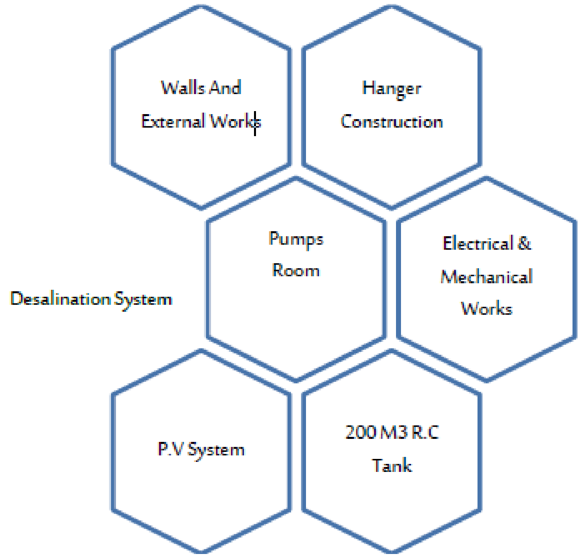
The value allocated from the grant for the project: 199,000 euros equivalent to 165,170 JD.

- **Order to proceed date:** 2021/03/01
- **Contract project duration:** (150) calendar days
- **Expected completion Date:** 15/12/2021

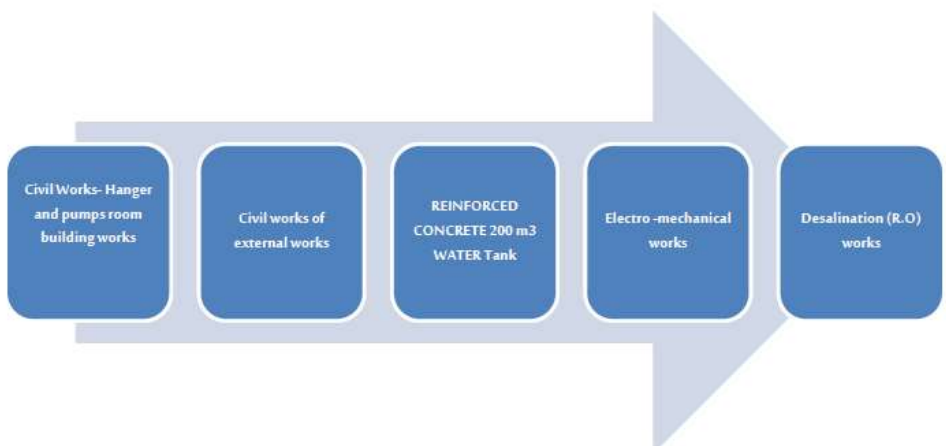
Location area



Project Main Components:



Activities Completed:
View of Completed Electro-Mechanical Works Activities:



View of Completed Electro-Mechanical Works Activities:

All materials motioned above are approved by Aqaba Water and installed on site.

Updates & Completed Works:

Since the last meeting held in Aqaba in November 2021, there wase a long hold for the project due to waiting of the approvals from the electricity company to connect the plant on the electricity grid, approvals were given on May 2022.

There was a delay in customs exemptions approvals, some of the electromechanical equipment were delayed being supplied and installed (such as crane and some equipment)

After connecting the plant to the electricity line, the contractor after an operational preparation meeting proceeds and finished the following:

- Install electrical panels and electrical boards
- Install R.O membranes
- Conduct operational tests (electrical, mechanical, treatment)
- We are now waiting the official approval from the ministry of health and ministry of water and irrigation to start pumping from the plant and produce desalinated water for drinking and brackish water for irrigation.
- We conducted a meeting with the agricultural association to discuss their preparations for receiving the water for their agricultural use (1 donum at least to cultivate tomatoes).

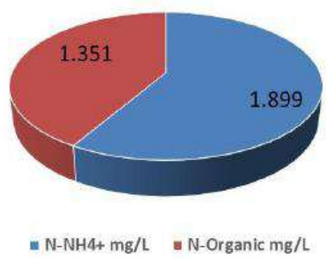
In these photos you can find the different steps of the built:








Recovery of ammoniacal nitrogen from various waste streams, like centrate from digestion processes, is becoming more and more important. On the one hand, this element is a valuable resource but on the other end, nitrogen is ranked in the top three to global issues way beyond any reasonable boundaries of sustainability.
The experimental study is focused on the plain of Arborea, that is a very extended area devoted to intensive cattle farming and agricultural activities and represents an excellence in the Sardinian agro-livestock system.
Due to the intensive agricultural practices, and the huge volumes of digestate (particularly rich in Nitrogen content) produced in the plain of Arborea, this area is a Nitrate Vulnerable Zone (NVZ). In this context, the management of digestate in the Arborea plain is a critical issue because the manure surpluse that is beyond the limit of 170 kg N per hectare cannot be distributed on the ground. Locally, the surpluses are treated by anaerobic digestion processes which deals with the disposal of the digestate on lands outside the vulnerable area.
As illustrated in the following figure, the physical chemical quality of the digestate shows a high content of ammoniacal nitrogen.
About 42% of the nitrogen forms in the digestate is in ammoniacal form which is the fraction of the total nitrogen that can be eliminated by the technology that will be tested.



Azoto Totale: 3'281 mg/L

Qualità del digestato

parametri	unità	Valore
pH	pH units	7,64
Total Suspended Solids	g/L	32,10
N-NH4+	**mg/L**	**1.899**
N-Organico	**mg/L**	**1.351**
Ca++	mg/L	993
Mg++	mg/L	355
k+	mg/L	1.605
Na+	mg/L	773
PO4--	mg/L	987
Cl-	mg/L	1.684
SO4--	mg/L	181
Dry residual (105°C)	%TS	3,21
fixed Residual (550 °C)	%TVS	12,80
Volatile Suspended Solids	g/L	19,30
COD	mg/L	30.493
Conductivity	mS/cm	17,65
TDS	g/L	9,44
Total Nitrogen	**mg/L**	**3.281**
Total P	mg/L	414
Total Alcalinity	mg/L	
Bicarbonates	mg/L	
NO3--	mg/L	
NO2--	mg/L	
Dry residual (180°C)	mg/L	
Total Hardness	°F	

The study aims to evaluate a technology for removing Ammonia Nitrogen from the centrate produced by the anaerobic digester used for the treatment of manure produced in the plain of Arborea and recovering Ammonia as ammonium sulphate solution the can be used as a fertilized or amendment.
The adopted technology consists in a membrane gas extraction technique suitable for the recovery of high-purity concentrated ammonia solution.

The principle of the technique is to apply a difference in vapour pressure for ammonia over a hydrophobic gas permeable membrane to transfer gaseous ammonia from the feed digestate solution into a stripping solution in order to produce the concentrated ammonium sulphate solution. The separation of gases from liquids by gas permeable membranes is known for some time.

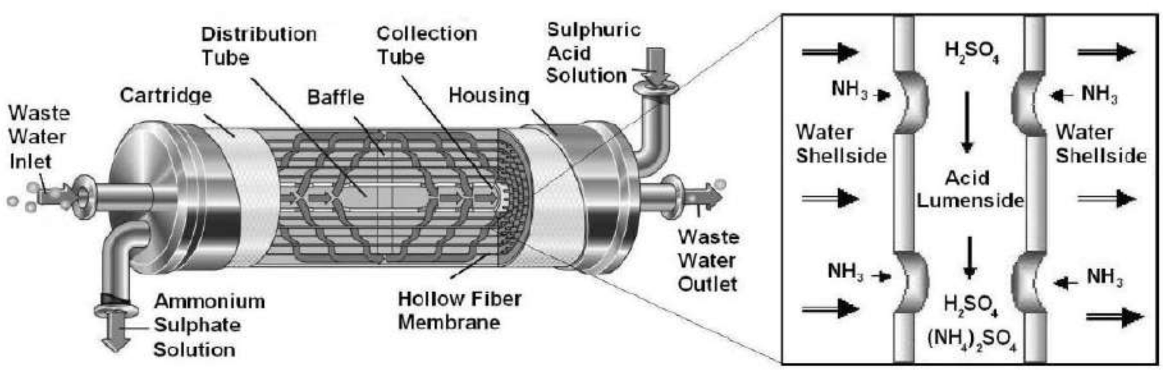
In particular, free ammonia gas content in the digestate diffuses across the micro porous hydrophobic membrane at high pH values and temperatures up to 45°C, conditions. To keep an high driving force, on the other side of the membrane circulates an acid solution where the diffused ammonia is dissolved as ammonium sulphate.

The small pore size and the hydrophobic nature of the membrane prevent the liquid phase from entering into the pores or flowing through the porous wall due to the surface tension effect. Because of the very low Henry constant and high solubility of NH3 compared to other dissolved gases in water (e.g., CO2 or O2) free ammonia is the unique gas that is removed from the digestate giving it an high purity quality.

Figure below shows a schematic drawing of a membrane contactor module with a hydrophobic hollow fibre membrane bundle.

The right picture shows schematically the chemo sorption process across a single hollow fibre. The sludge liquid flows through the shell-side of the membrane module (outside of the membrane), while the acid solution (sulphuric acid) is circulating on the lumen-side.

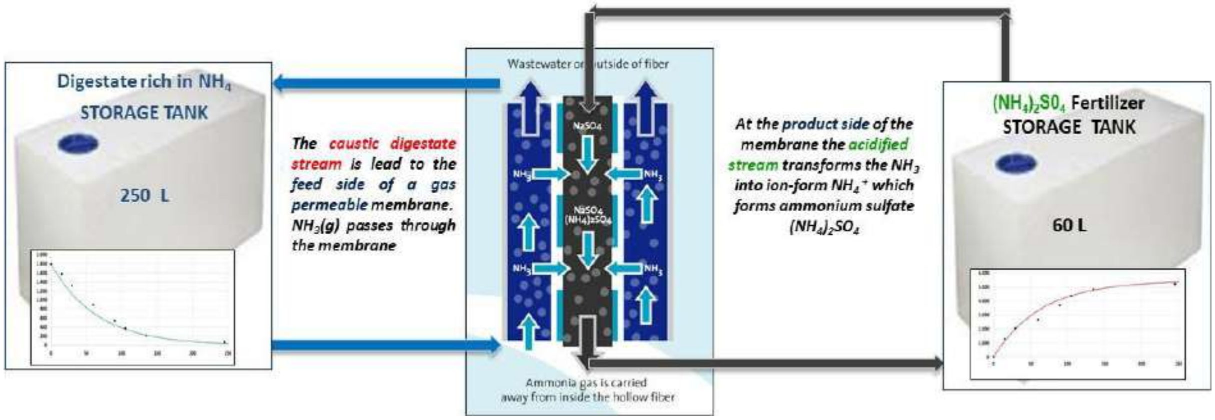
Equation 1: $2\,NH_3 + H_2SO_4 \rightarrow (NH_4)_2SO_4$



A low-pH sulphuric acid solution will instantly react with ammonia gas according to Equation 1 to form ammonium sulphate.

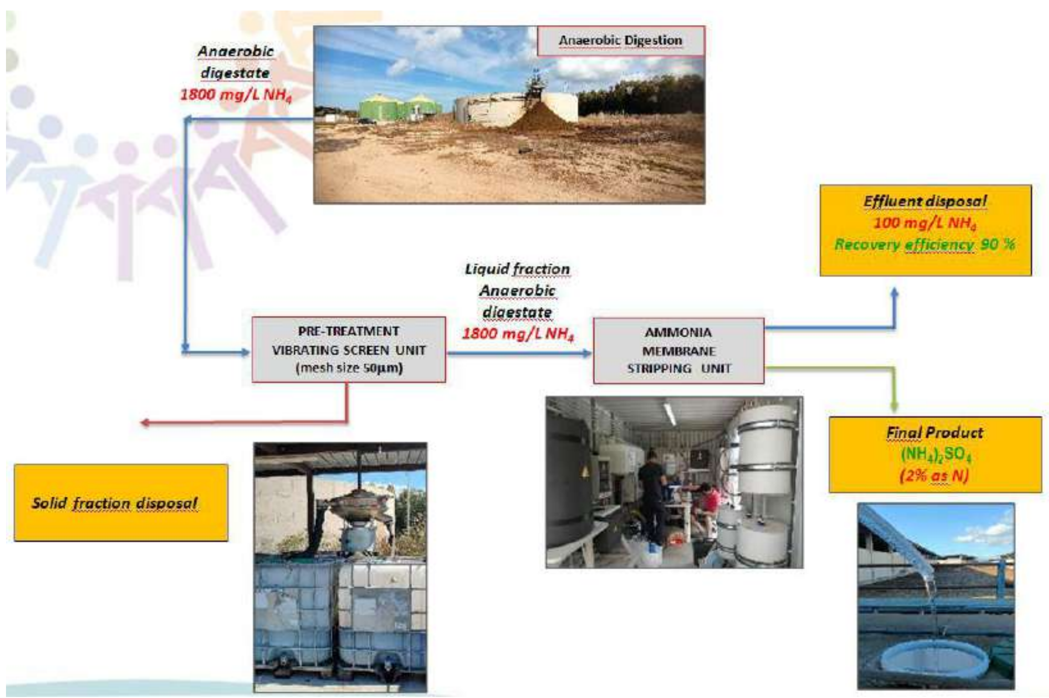
The stripping and recovery plant for ammonia is located at the fattening center of the breeders cooperative of Arborea and is housed in a standard size maritime container 20 ft.

The stripping process is depicted below:



The recovery process of the plant is schematized in the following figure. It consists in the derivation of the digestate from the circular storage tanks located close to the anaerobic digestion plant. The digestate (centrate) is clarified by an additional solid/liquid separation that takes place via a vibrating pre-screened unit of 0,45 micron meters of mesh size. The solid fraction is accumulated to be reused as a soil improver while the liquid fraction, rich in ammonia, is sent to the stripping treatment plant.

Two effluent products are obtained from the stripping unit: the treated digestate, free of ammoniacal nitrogen, and the ammonium sulphate solution, the concentration of which can be modulated according to the number of planned treatment cycles.

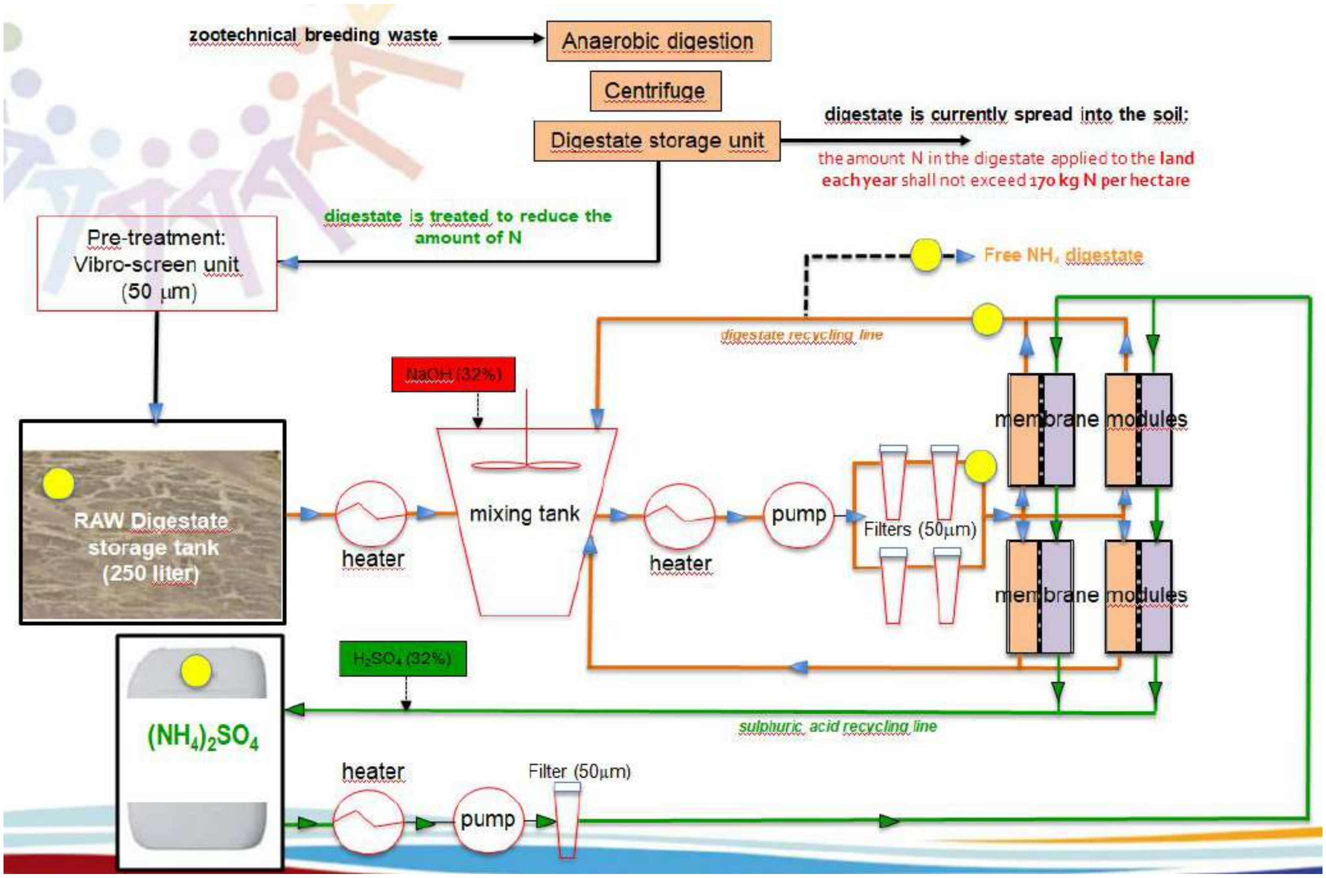


The stripping process is of a batch feature. In fact, it is a discontinuous operation by which a volume of digestate of about 250 liters is managed.

The stripping unit is equipped with four membrane modules having a specific area of 6 square meters, each.

Prior the stripping process, the digestate is subjected to a pH adjustment to the value of 10 by caustic soda and further filtration. A 60 liter diluted sulphuric acidic stripping solution is prepared to extract ammonia and converts it into ammonium sulfate solution (NH4)2SO4 that is continuously enriched at each treatment cycle.

The process scheme is reported in the following figure.



The adopted methodology for Ammonia Analysis in situ is of spectrophotometric type. Ammonium ions react alkaline environment in presence of sodium nitroprusside (catalyst) with sodium salicylate and a chlorinating agent to form a colored derivative of indophenol determined by spectrophotometric reading at 690 nm.

A volume of 0,1 mL of diluted sample was put into the vials and wait 15 minutes. The liquid inside the vial tooks on a green color and, based on the intensity of the color, the spectrophotometer calculated a final ammonia value. The same analyzes were conducted in the ENAS laboratories, adapting the EPA method used for brackish water. The data were confirmed using a professional bench spectrophotometer.
The equipment used are shown in the below figures:

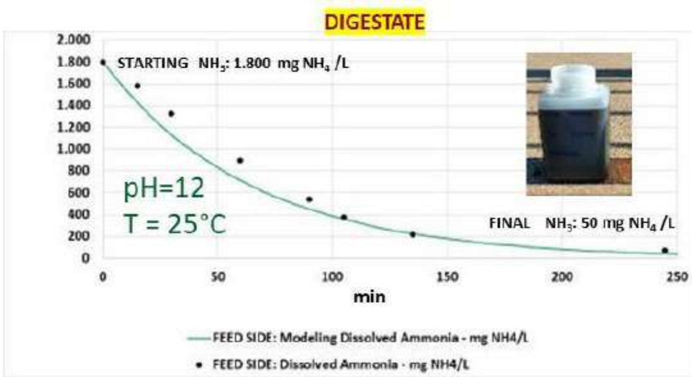
NH4 spectrophotometer (690 nm)

Kit measurement range:
12.5 - 130 mg/L of ammonia
Several preliminary dilutions has done in order
to read the final value

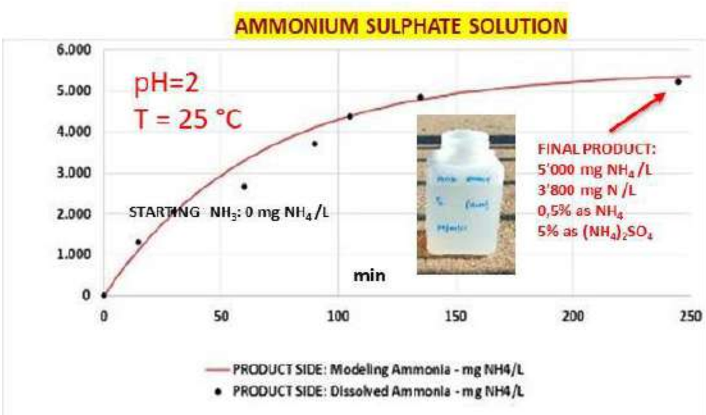
pH, T, conductivity

The starting ammonia concentration in the digestate was about 1'800 mg/L of total ammoniacal nitrogen. During each cycle some main process parameters have been frequently monitored on the both streams such as pH, conductivity, temperature and ammonia concentration.

The following figure shows the variation with the time of the ammonia concentration in the digestate recirculation basic stream and in the acid stream that is used to enrich the ammonium sulphate content during a treatment cycle.



	CAUSTIC SIDE			
time (min)	pH	cond (mS/cm)	T (°C)	CAUSTIC SIDE: Ammonia - mg NH_4/L
0	12,00	37,80	22,00	1.800
15	12,29	39,30	21,90	1.580
30	12,62	47,80	22,00	1.320
60	12,87	63,50	22,50	890
90	12,86	67,80	23,70	540
105	12,76	67,50	24,90	370
135	12,72	67,00	25,80	215
245	12,71	65,90	27,90	66

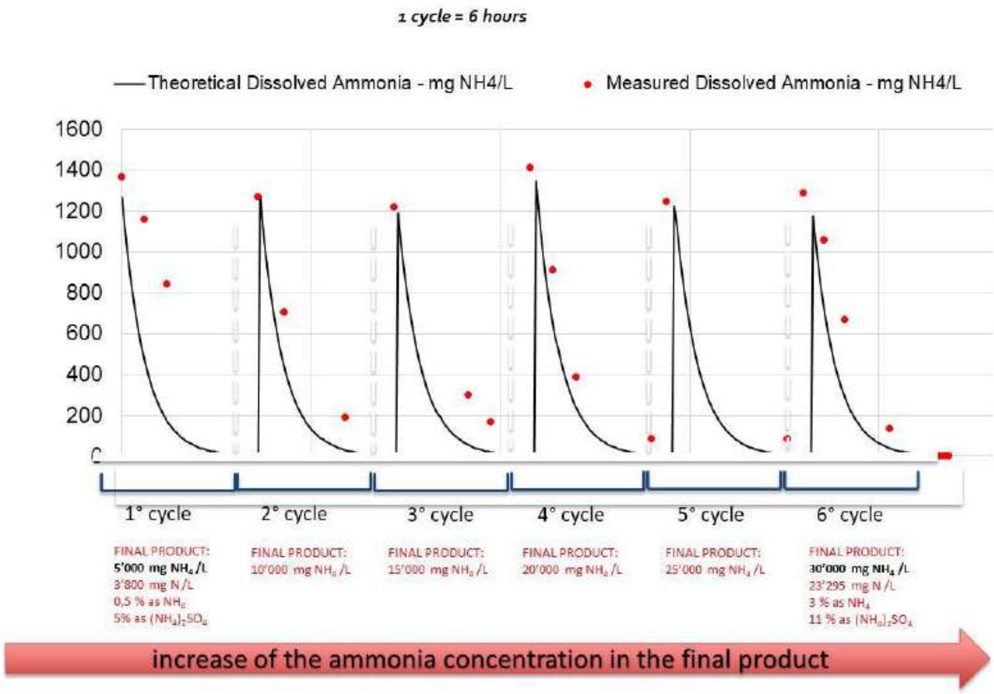


	ACID SIDE			
time (min)	pH	cond (mS/cm)	T (°C)	ACID SIDE: Ammonia - mg NH_4/L
0	1,39	58,90	18,50	0
15	1,84	38,10	31,30	1.300
30	2,33	34,30	21,80	2.050
60	2,28	39,00	23,10	2.650
90	2,48	40,00	24,00	3.700
105	2,33	40,80	24,90	4.360
135	2,37	43,00	25,60	4.840
245	3,15	41,10	28,10	5.220

It can be noted that the concentration of ammonia in the basic stream decreases as the concentration of ammonia in the acid stream increases. These trends demonstrate a good functioning of the stripping process through the semi-permeable membranes.

The complete removal of ammonia in the basic stream was accomplished by recirculating the stream for about five hours. This was the duration of each cycle.

As depicted in the figure below, the enrichment of ammonia in the ammonium sulphate solution was achieved by approximately six successive cycles during which a total of 1,500 liters of digestate were processed. At the end of the six cycles the ammonium sulphate concentration was about 30,000 mg/L of ammonia. (23,295 mg N/L - 3% as NH4 - 11% as ammonium sulphate)



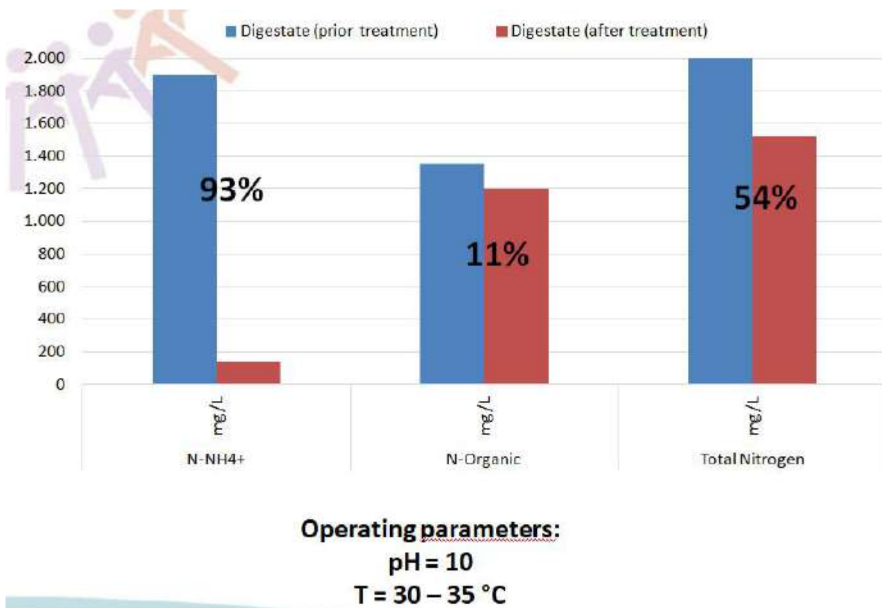
Throughout the experimental study the plant has recovered about 40 kg of NH_4, stored in 2'000 liters of ammonium sulfate solution to 8% in $(NH_4)_2S0_4$ (20'000 $mgNH_4/L$).
The table below summarizes the quality of the ammonium sulphate solution obtained after one cycle.

parameters	unit	Ammonium Sulphate product
pH	pH units	1,16
Total Suspended Solids	g/L	
N-NH4+	mg/L	6'207
N-Organic	mg/L	
Ca++	mg/L	38,3
Mg++	mg/L	12,4
k+	mg/L	7,95
Na+	mg/L	120
PO4--	mg/L	
Cl-	mg/L	203
SO4--	mg/L	33'134
Dry residual (105°C)	%TS	
fixed Residual (550 °C)	%TVS	
Volatile Suspended Solids	g/L	
COD	mg/L	
Conductivity	mS/cm	60,9
TDS	g/L	
Total Nitrogen	mg/L	
Total P	mg/L	<LQ
Total Alcalinity	mg/L	<LQ
Bicarbonates	mg/L	<LQ
NO3--	mg/L	0,79
NO2--	mg/L	<LQ
Dry residual (180°C)	mg/L	39'723
Total Hardness	°F	16,5

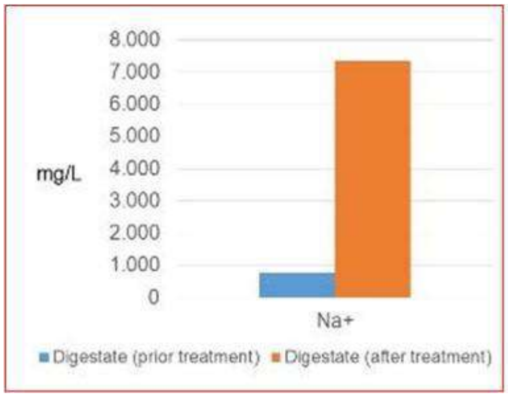
The extremely acid pH value has been neutralized at the end of the enrichment process.
The table below summarizes the quality digestate, that has been processed by the stripping unit, compared to the initial quality before the treatment.

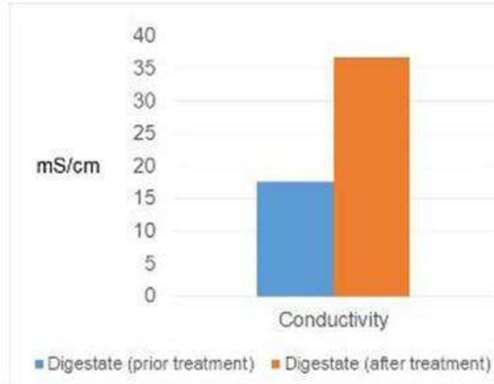
parameters	unit	Digestate (prior treatment)	Digestate (after treatment)
pH	pH units	7,64	14,00
Total Suspended Solids	g/L	32,10	39,30
N-NH4+	mg/L	1.899	142
N-Organic	mg/L	1.351	1.198
Ca++	mg/L	993	601
Mg++	mg/L	355	208
k+	mg/L	1.605	1.438
Na+	mg/L	773	**7.338**
PO4--	mg/L	987	761
Cl-	mg/L	1.684	1.484
SO4--	mg/L	181	330
Dry residual (105°C)	%TS	3,21	3,93
fixed Residual (550 °C)	%TVS	12,80	28,00
Volatile Suspended Solids	g/L	19,30	11,30
COD	mg/L	30.493	20.090
Conductivity	mS/cm	17,65	36,70
TDS	g/L	9,44	14,90
Total Nitrogen	mg/L	3.281	1.523
Total P	mg/L	414	276
Total Alcalinity	mg/L		
Bicarbonates	mg/L		
NO3--	mg/L		
NO2--	mg/L		
Dry residual (180°C)	mg/L		
Total Hardness	°F		

Results show that the stripping unit has an average ammonia removal efficiency of about 93 %. The figure below highlights this point.

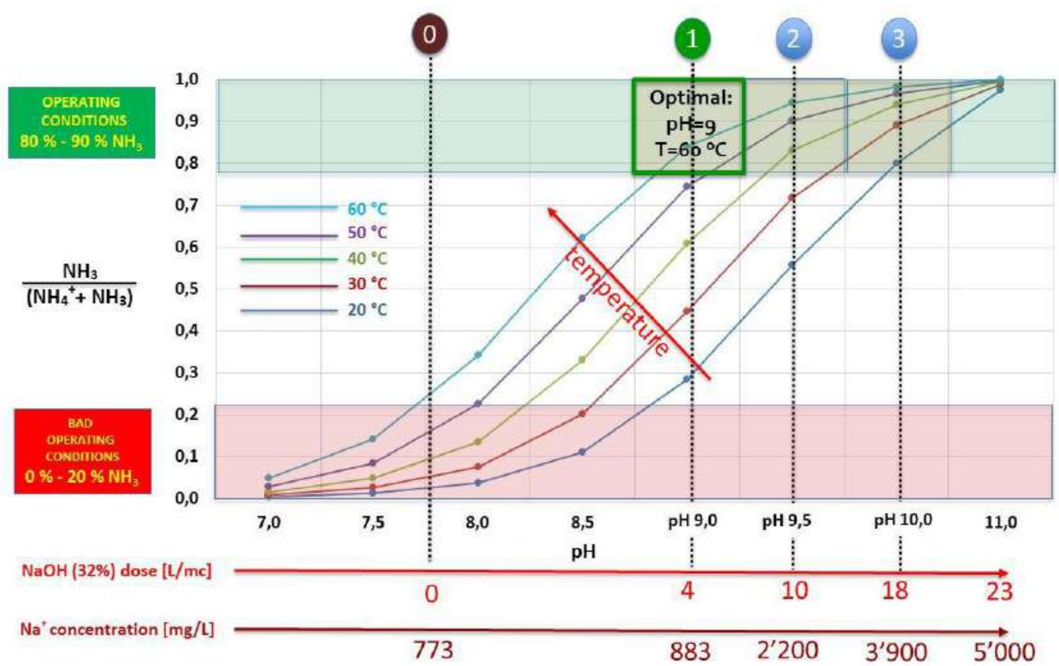


The analysis of the treated digestate focus on a critical aspect that is the increase of sodium concentration, and consequently the conductivity, in the digestate after the stripping treatment. The figure below highlights this point.





The high sodium content in the digestate is due to the high amount of soda needed to raise the pH of the digestate in order to shift the balance between ammoniacal nitrogen in ionic form to ammoniacal nitrogen in gaseous form.
Because of the high buffering power, high amounts of sodium hydroxide are required, which consequently provokes an excessive increase in sodium in the digestate.
This aspect can hinder the spreading practice of the treated digestate on the soils and therefore the optimal process conditions have been defined in order to minimize the addition of sodium hydroxide for the pH adjustment.
At first, before the stripping process, it will be recommended to implement a carbon dioxide stripping section in order to reduce the very high buffer power of the digestate. Secondary, it will be appropriate to adjust the pH of the digestate no more than 8-9 units. Tertiary it will be necessary to increase the digestate temperature to about 60°C in order to reach the ammoniacal balance in the digestate toward the optimal value of 0,8 – 1.
The following table shows the optimal conditions for stripping process management should be reached at the point n.1. In particular, at these conditions the concentration of sodium present in the treated digestate can be about 800 mg/L. Point n.0 represents the starting conditions of the digestate before whatever treatment. Points n.2 and n.3 represents operating conditions that are not recommended due to the excess sodium hydroxide doses.



In the course of the experimental study, the high suspended solid content present in the digestate, although the very efficient pre-screening unit, became the most critical aspect to be solved in designing a full-scale stripping unit.
Due to the solid still present in the digestate, the pre and post heating units very frequently were malfunctioning because of the thermal resistances that produced a "cooked digestate" that clogged the filters positioned before the membrane units.
With regard to filters, 50 micronm wire-wound filters were replaced with washable cage filters of the same size, but with no success.
To overcome this drawback a frequent manual back washing of the cage filter has been done.
The picture below illustrates the some encountered drawbacks.

Vibroscreen replacement

Pump replacement

Test with several kinds of filters