Il Progetto «MICROFLOWER»

Composizione e proprietà antiossidanti di microalghe ad alto valore biologico

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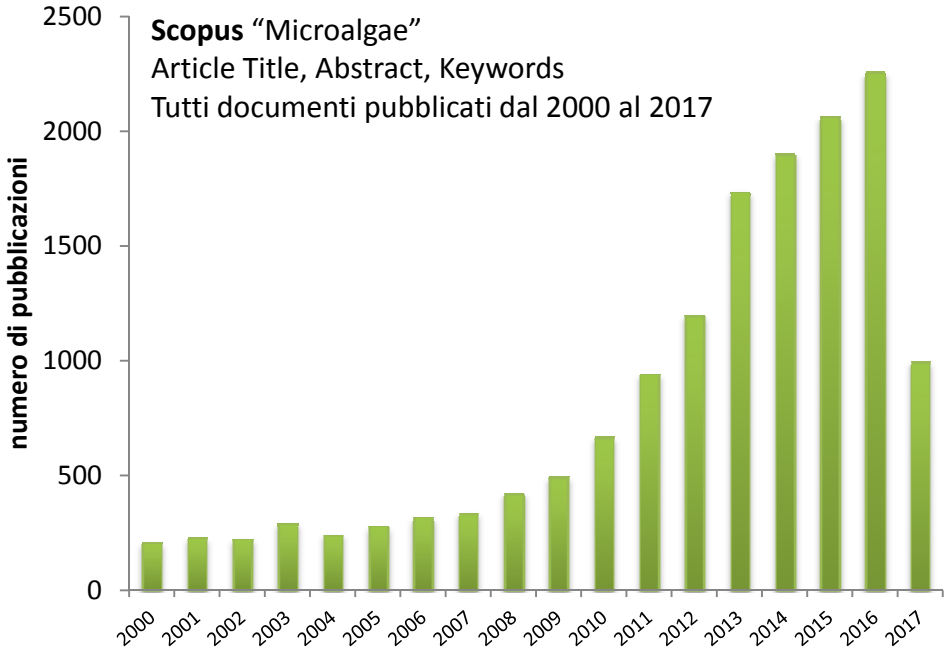
Progetto finanziato da:

Sponsor dell'evento:

Outline

- Composizione chimica delle microalghe
- Microalghe in ambito alimentare, farmaceutico e cosmetico
- Elevato valore biologico delle microalghe: la spirulina
- Risultati MICROFLOWER
- Conclusioni



Composizione chimica delle microalghe

Proteine | Carboidrati | Lipidi

Vitamine | Pigmenti | Minerali

Additivi alimentari /coloranti
Alimenti funzionali
Prodotti farmaceutici
Cosmetici

Koller et al. Algal Research 2014 6:52–63; Spolaore et al. J Biosci Bioeng. 2006 101(2):87-96

Applicazioni alimentari

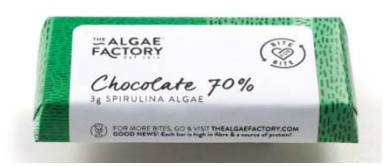


TABLE 1. General composition of different human food sources and algae (% of dry matter) (3)

Commodity	Protein	Carbo-hydrate	Lipid
Bakers' yeast	39	38	1
Meat	43	1	34
Milk	26	38	28
Rice	8	77	2
Soybean	37	30	20
Anabaena cylindrica	43–56	25–30	4–7
Chlamydomonas rheinhardii	48	17	21
Chlorella vulgaris	51–58	12–17	14–22
Dunaliella salina	57	32	6
Porphyridium cruentum	28–39	40–57	9–14
Scenedesmus obliquus	50–56	10–17	12–14
Spirulina maxima	60–71	13–16	6–7
Synechococcus sp.	63	15	11

Spolaore et al. J Biosci Bioeng. 2006 101(2):87-96

Applicazioni farmaceutiche

Omega-3 *polinsauri* (PUFA)	**Omega-6** *polinsauri* (PUFA)
Acido alpha linolenico **(ALA)** ***essensiale***	Acido linoleico **(LA)** ***essenziale***
Acido Eicosapentaenoico **(EPA)**	Acido-Gamma-linolenico **(GLA)**
Acido Docosaesaenoico **(DHA)**	Acido Arachidonico **(AA)**

Acidi grassi polinsaturi

- DHA sviluppo cerebrale e della vista
- GLA antinfiammatorio e antibatterico
- EPA antinfiammatorio e vasodilatatore

Carboidrati

- Polisaccaridi solfati

  immunomodulanti e antitrombotici

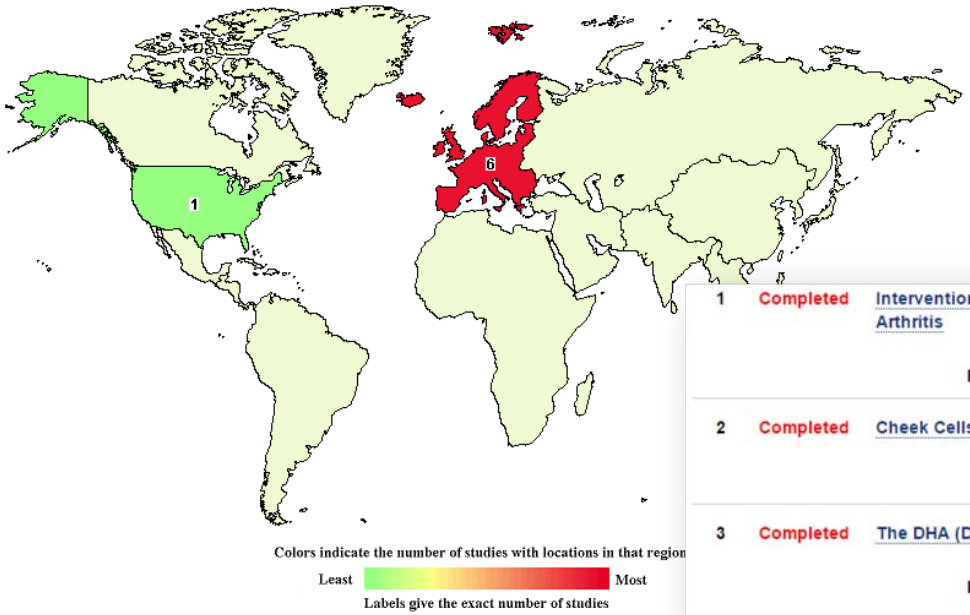
Pigmenti

- *Marker* fluorescenti
- Carotenoidi
  - β-carotene antiossidante
  - Astaxantina antiossidante

de Jesus Raposo et al. Life Sciences 2013 93:479–486; de Morais et al. BioMed Research International 2015 Article ID 835761; Odjadjare et al. Crit Rev Biotechnol 2017 37(1): 37–52

Trial clinici

ClinicalTrials.gov (ultimo accesso 12/06/2017)
"Microalgae"



1	Completed	Intervention With Long-chain n-3 Polyunsaturated Fatty Acids From Microalgae Oil in Patients With Rheumatoid Arthritis	**Condition:** Rheumatoid Arthritis **Interventions:** Dietary Supplement: long-chain n-3 PUFA; Dietary Supplement: sunflower oil
2	Completed	Cheek Cells - Non-invasive Fatty Acid Status Marker	**Condition:** Healthy **Intervention:** Dietary Supplement: DHA (docosahexaenoic acid)
3	Completed	The DHA (Docosahexaenoic Acid) Oxford Learning and Behaviour (DOLAB) Study	**Conditions:** Learning; Behaviour **Interventions:** Dietary Supplement: DHA (docosahexaenoic acid); Dietary Supplement: Sunflower oil capsules
4	Terminated	Effect of Docosahexaenoic Acid (DHA)-Enriched Human Milk in Premature Newborns	**Condition:** Premature **Intervention:** Dietary Supplement: Supplementation of lactating mothers who has delivered prematurely with DHA
5	Completed	Consumption of Eggs Rich in Lutein and Omega-3 Fatty Acids on the Macular Pigment	**Condition:** Age-related Macular Degeneration **Intervention:** Other: Nutritional study
6	Completed	Intervention With n-3 Polyunsaturated Fatty Acids (PUFA)-Supplemented Products in Moderate Hypertriglyceridemic Patients	**Condition:** Hypertriglyceridemia **Intervention:** Dietary Supplement: n-3 PUFA
7	Completed	Docosahexenoic Acid (DHA) Supplementation and Cardiovascular Disease in Men With High Triglycerides	**Condition:** Hypertriglyceridemia **Interventions:** Dietary Supplement: Docosahexenoic acid (DHA); Dietary Supplement: Olive oil

Applicazioni cosmetiche

Cute e capelli

Proprietà delle microalghe

- ❑ Anti-età
  - fotoprotettori
  - antiossidanti
  - inibitori degradazione del collagene
- ❑ Anti-macchia/Sbiancanti
- ❑ Antimicrobici

Impieghi

Idratanti
Tonici
Ristrutturanti/anti-aging
Solari

Wang et al. Bioresource Technology 2015 184:355–362

Elevato valore biologico delle microalghe

Arthrospira platensis

Content in 100 g
Proteins 35.4–70.0 g
*Amino acids*Glutamate 7.0–7.3 g
Leucine 5.9–8.4 g
Aspartate 5.2–6.0 g
Lysine 2.6–4.6 g
Tyrosine 2.6–3.4 g
Phenylalanine 2.6–4.1 g
Methionine 1.3–2.7 g
Fat 4.0–16.0 g
*% of total fatty acids*Palmitic 25.8–44.9%
Gamma-linoleic 17.1–40.1%
Linoleic 11.1–12.0%
Oleic 10.1–16.6%
Palmitoleic 2.3–3.8%
Stearic 1.7–2.2%
Carbohydrates 14.0–19.0 g
Crude fiber 3.0–7.0 g
Minerals
Potassium 2.0–2.6 g
Sodium 1.5–2.2 g
Total phosphorus 1.3–2.2 g
Iron 273.2–787.0 mg
Magnesium 330
Calcium 120–900 mg
Vitamins
B12 5.7–38.5 μg
B2 3.0–4.6 mg
B6 0.5–0.8 mg
Niacin (B3) 13–15 mg
Folic acid 0.05–9.92 mg
Carotenoids 0.3–2.6 g
Tocopherol 0.4–9.8 g

In 1967 Spirulina was recognized as a 'future food source' by the International Association of Applied Microbiology

Generelly recognized as safe (GRAS) dal 1981 (FDA)

Antiossidante
Antinfiammatorio
Ipoglicemizzante
Antipertensivo
Antibatterico

Finamore et al. Oxidative Medicine and Cellular Longevity 2017, Article ID 3247528;
Lupatini et al. J Sci Food Agric 2017 97:724–732; Wu et al. Arch Toxicol 2016 90:1817–1840

MICROFLOWER

Composizione e valutazione attività antiossidante

A. platensis coltivata considerando 4 medium di coltura:

- Zarrouck: medium di riferimento
- T3.1: NH_4NO_3/KNO_3
- K1: KNO_3
- RM6: $NaNO_3$

} Medium low cost

… e considerando tre condizioni di coltura:

- Lab scale (ISILS)
- Lab scale con riciclo medium (ISILS)
- Mid scale (P680)

Confronto:
- Liofilizzazione
- Essiccazione a freddo

Proteine

Carboidrati

Lipidi

Pigmenti

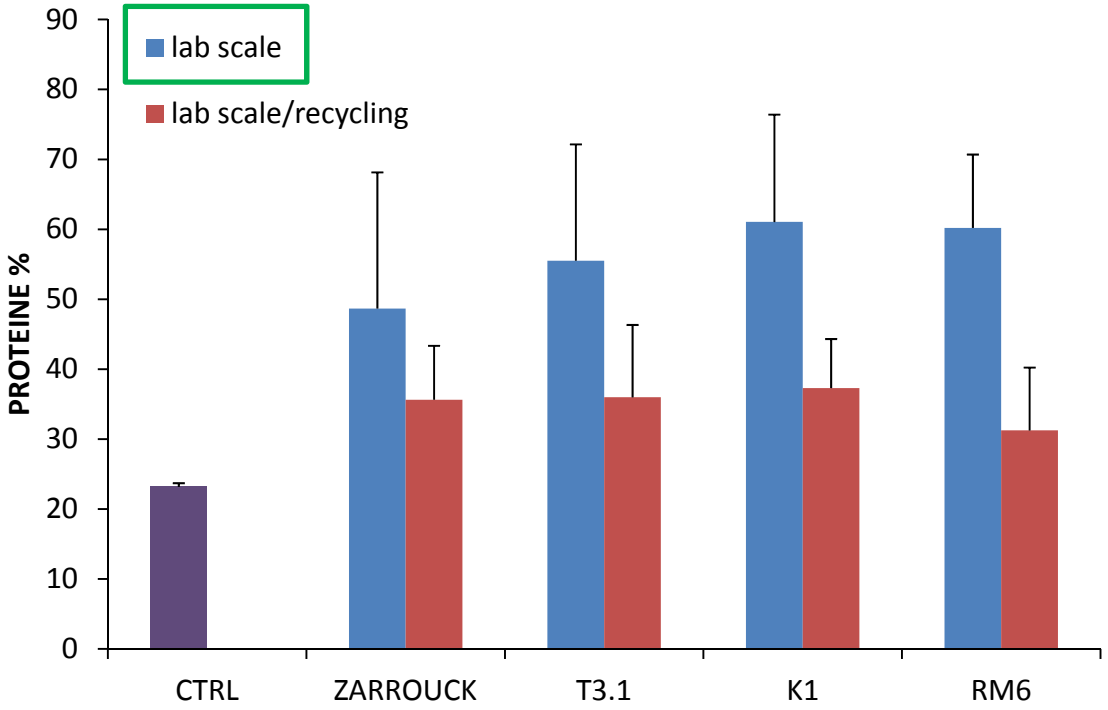
Attività antiox

Controllo:
Arthropira platensis
commerciale

Risultati MICROFLOWER *Proteine*

Materiali e metodi

Microalghe 0,1 mg/ml in H_2O
BCA-protein assay kit
Incubazione a 37°C per 2h
Centrifugazione 3000 g 10 min
λ 562 nm
Standard albumina serica bovina



Trattamento p < 0,0001
Medium n.s.

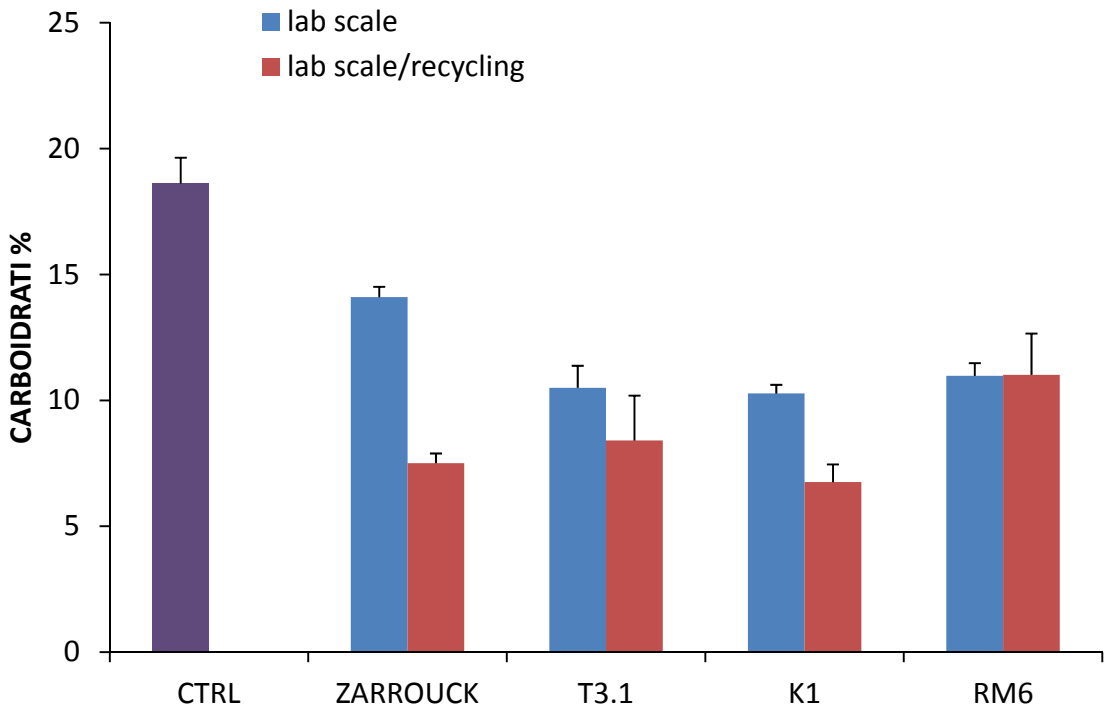
Multifactor ANOVA – Statgraphics

Risultati MICROFLOWER *Carboidrati*

Materiali e metodi

Microalghe 20 mg/ml
in HCl 2.5N 100 °C 3h
Aggiunta di Na_2CO_3 sino a fine effervescenza e H_2O
concentrazione finale 1 mg/ml
Centrifugazione 3000 g 20 min
2 ml surnatante + 1 ml fenolo +
5 ml acido solforico
Incubazione 10 min
λ 490 nm
Standard glucosio

Waghmare et al. Bioresour. Bioprocess. 2016 3:16



Trattamento p < 0,0001
Medium p < 0,0001

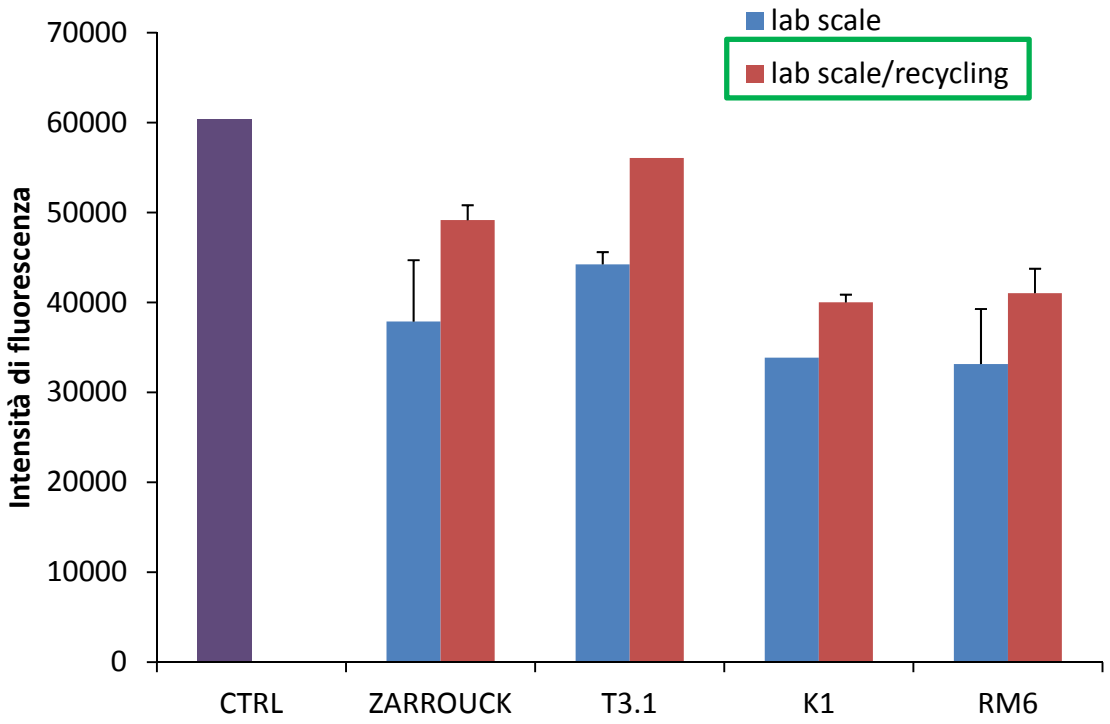
Multifactor ANOVA – Statgraphics

Risultati MICROFLOWER *Lipidi*

Materiali e metodi

Microalga 0,3 mg/ml in H_2O
Trattamento con glicerolo
Incubazione con Nile Red
(15 µg/ml) in acetone
Intensità di fluorescenza
filtri di Ecc/Em 485/590 nm

Balduyck et al. J MicrobiolMethods 2015 118:152-158



Trattamento p = 0,0275
Medium n.s.

Multifactor ANOVA – Statgraphics

Risultati MICROFLOWER *Pigmenti*

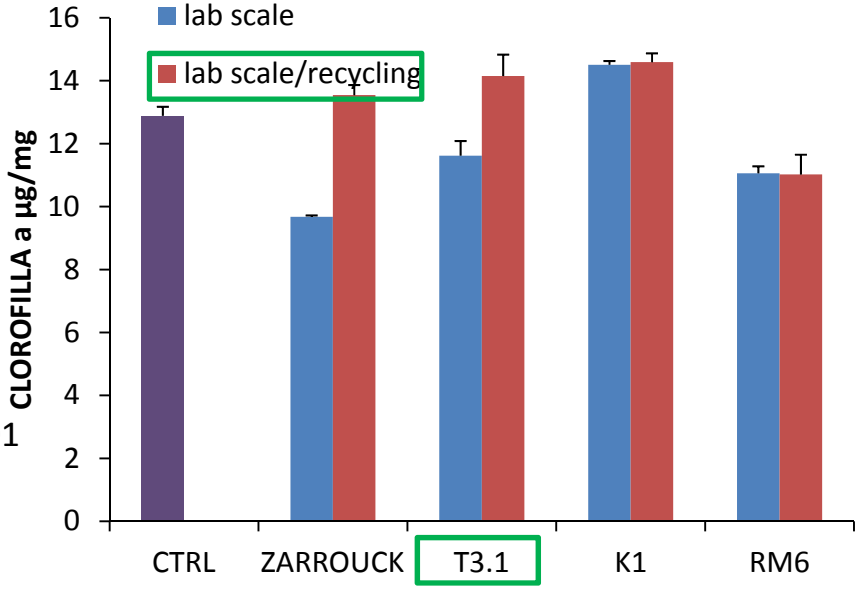
Materiali e metodi

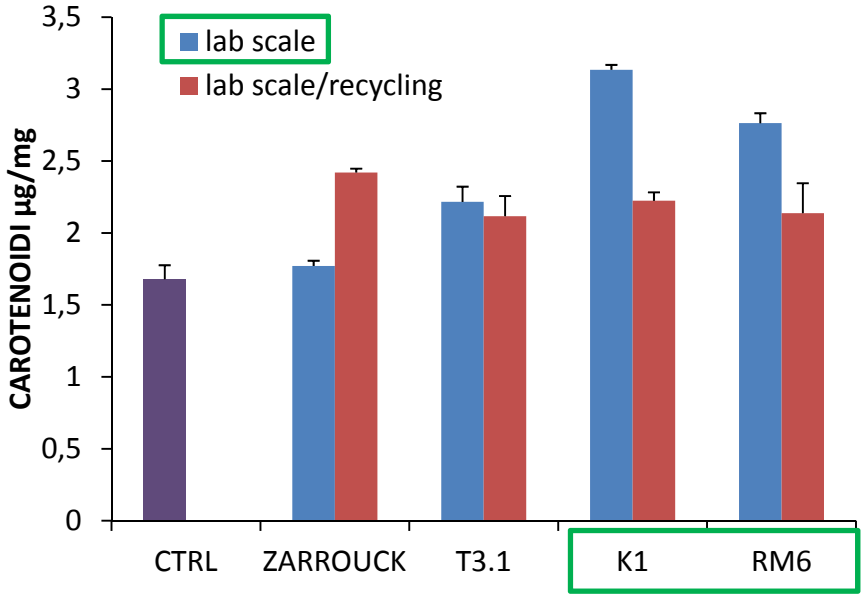
Microalghe 1 mg/ml in EtOH
3 cicli
Sonicazione 20 minuti
Centrifugazione 3000 g 10 min
λ 470, 648 e 664 nm

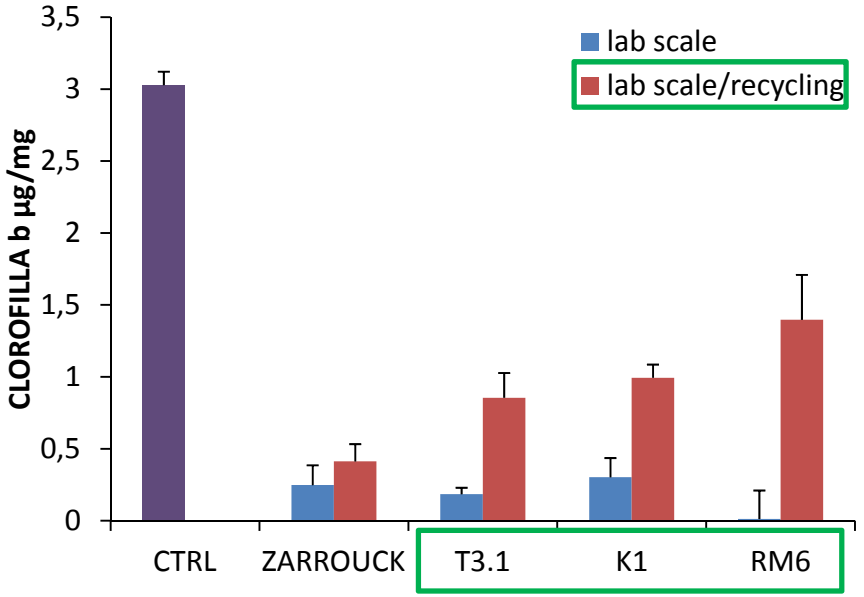
Chl a → $y=(13,36*A_{664})-(5,15*A_{648})$
Chl b → $y=(27,43*A_{648})-(8,12*A_{664})$
Carot. → $y=[(1000*A_{470})-(1,63*Chla)-(104,96*Chlb)]/221$

Lichtenhaler, F4.3.1-F4.3.8, Wiley & Sons, Inc., 2001

Trattamento $p < 0,0001$
Medium $p < 0,005$







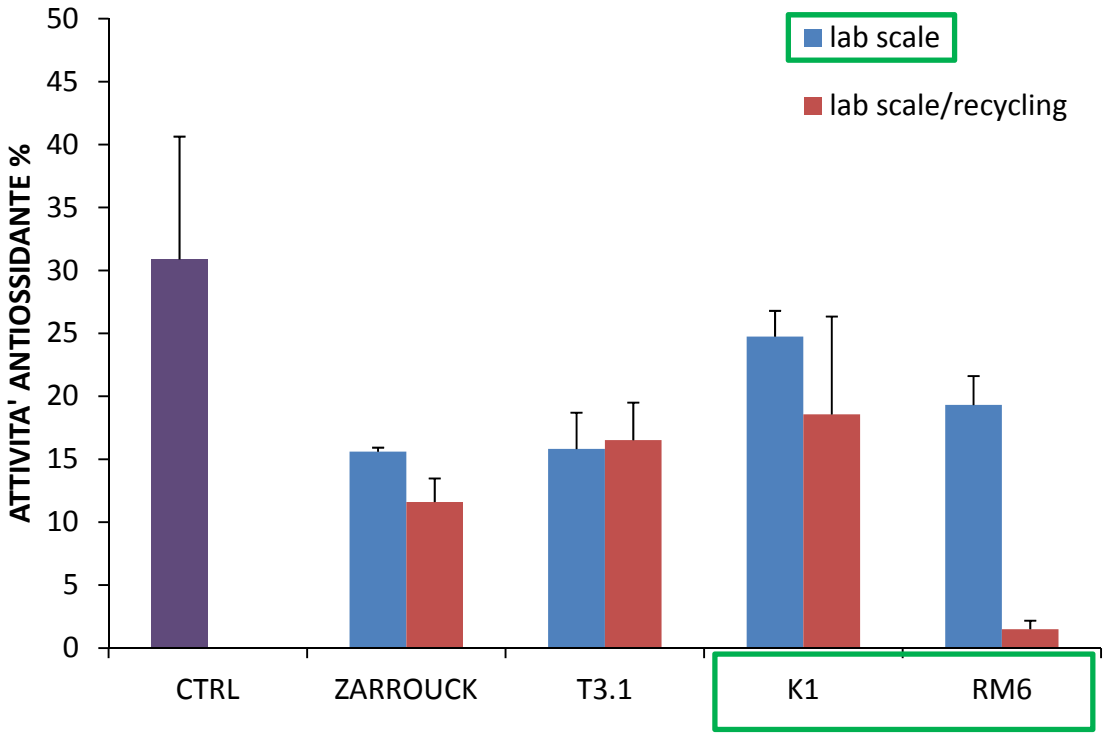
Multifactor ANOVA – Statgraphics

Risultati MICROFLOWER *Attività antiossidante*

Materiali e metodi

Microalga 25 mg/ml
MeOH 70% v/v
Soluzione DPPH (2,2-diphenyl-2-picrylhydrazyl hydrate)
Incubazione 20 minuti
Centrifugazione 3000 g 10 minuti
λ 515 nm
Ac. ascorbico controllo positivo
$Att\% = (A_{ctr} - A_{camp})/A_{ctr} * 100$
A_{ctr} assorbanza controllo negativo
A_{camp} assorbanza campione

Lucconi et al. Pharm Dev Technol 2013 19:65-72

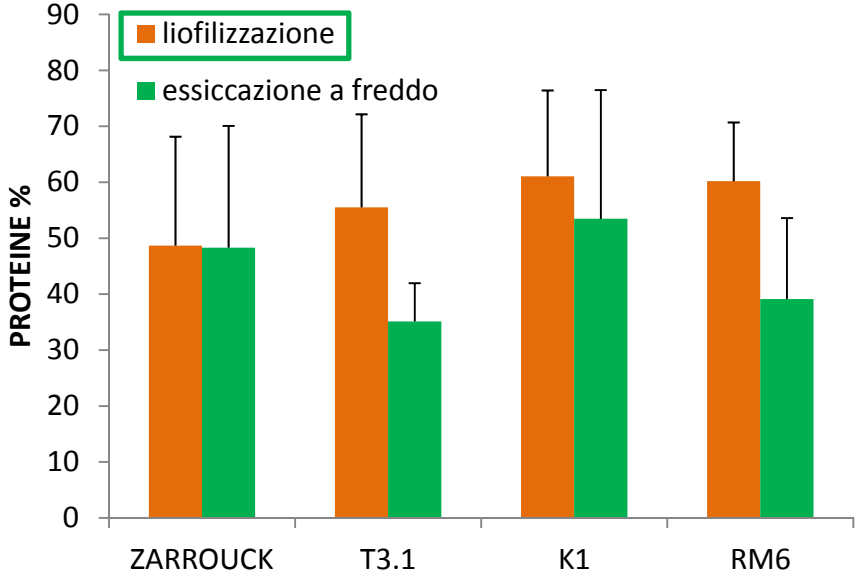


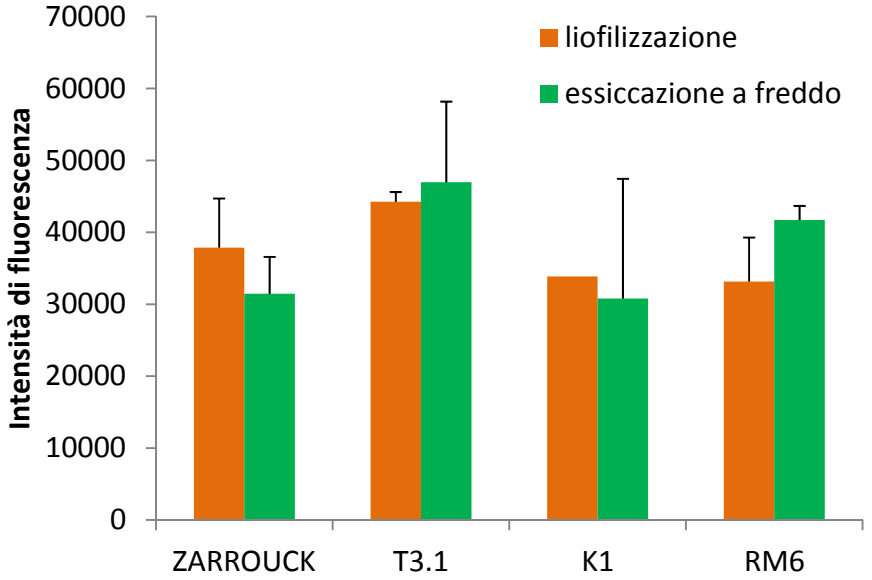
Trattamento $p < 0,0001$
Medium $p < 0,0001$

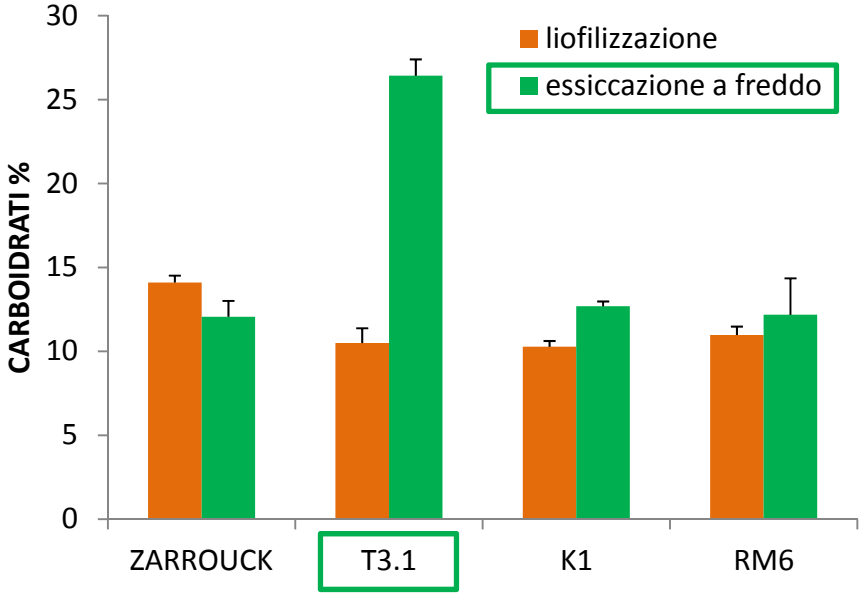
Multifactor ANOVA – Statgraphics

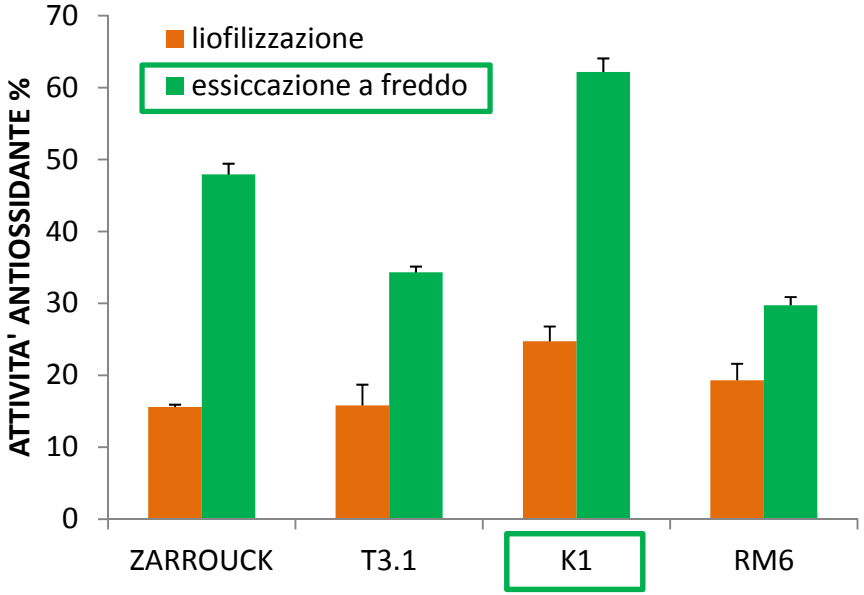
Risultati MICROFLOWER

Liofilizzazione Vs Essiccazione a freddo

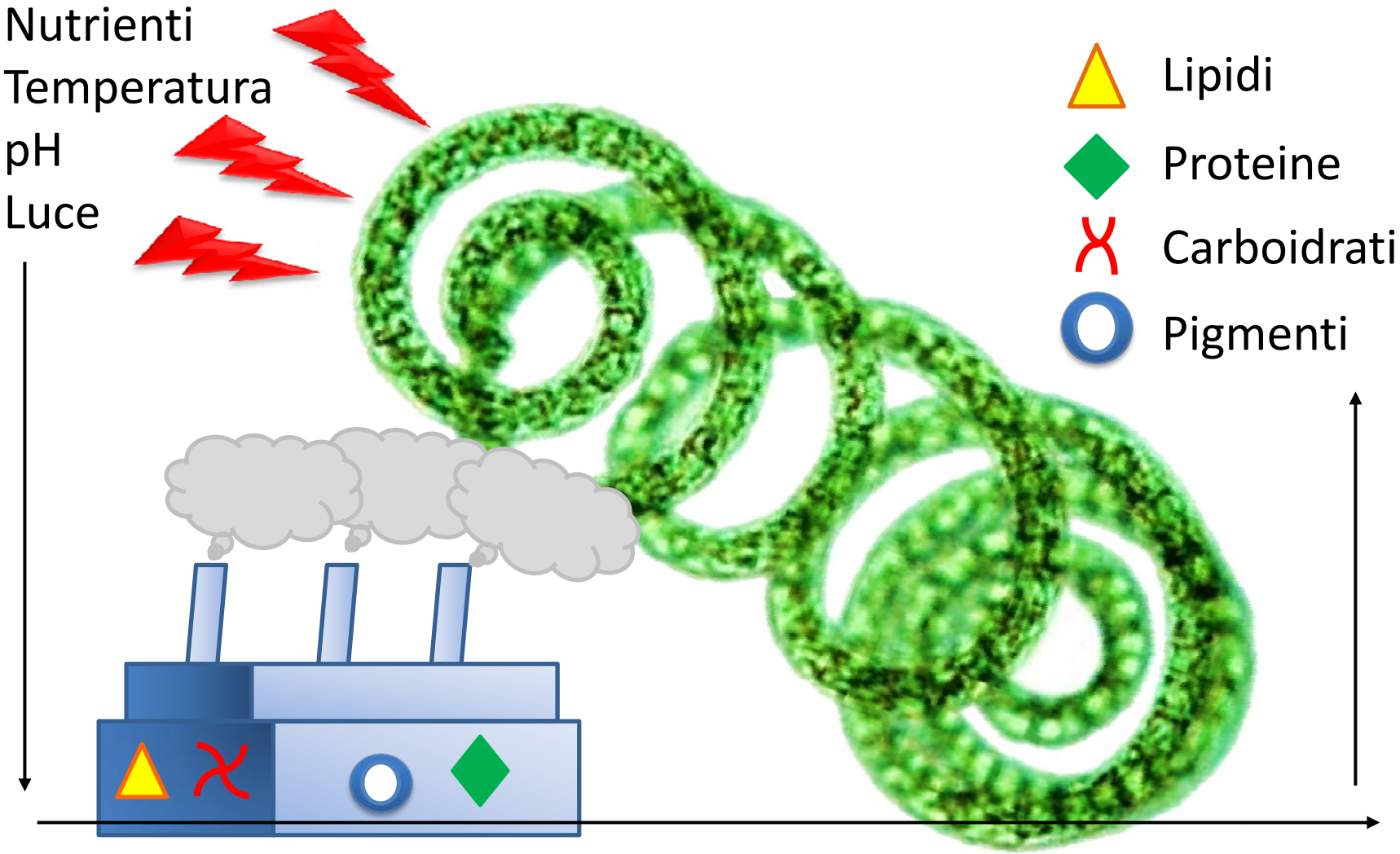








Conclusioni



Microalgae as Drug Factory

UNIVERSITA' DEGLI STUDI DI PAVIA
DIPARTIMENTO DI SCIENZE DEL FARMACO
Cell Delivery System Laboratory

Grazie dell'attenzione!

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Sponsor dell'evento:

LUMSON Cosmetic Packaging Industries

GRAFICHE CAM Pandino (CR)