



# Unlocking the Potential of Rice: Innovative Extraction Methods and Bioactive Compound Analysis

TRACING RICE AND VALORIZING SIDE STREAMS ALONG  
MEDITERRANEAN BLOCKCHAIN

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Veterinária, I.P.

# iBET's contribution in TRACE RICE project

## Natural Bioactives & Nutraceuticals Area

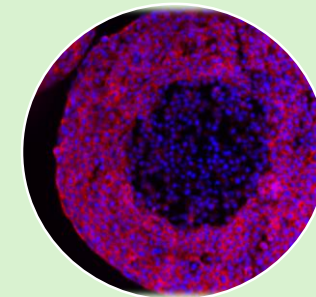
Valorizing bioactives present in food, natural matrices, and industrial by-products for a healthier and more sustainable future: from extraction to characterization and evaluation of health-promoting effects.



Extraction & Purification



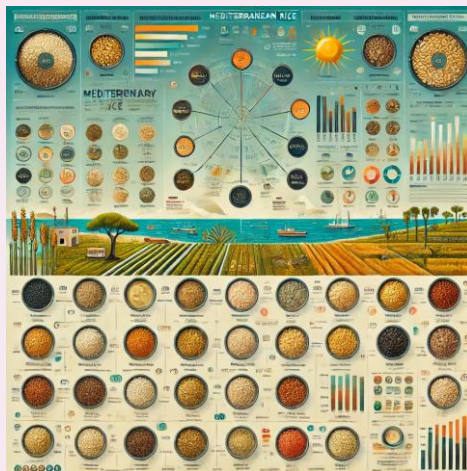
Characterization



Functionality



# Activity I: Characterization of Mediterranean rice varieties



- Chemical characterization
- Antioxidant activity

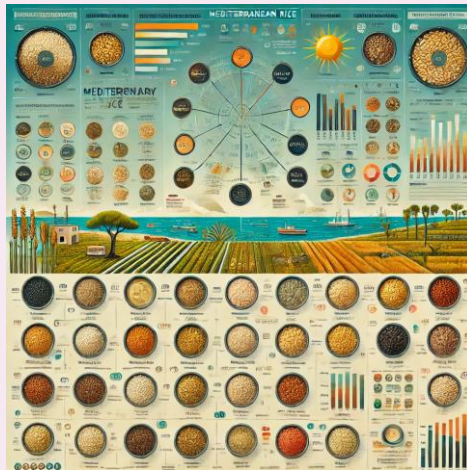
# Activity II: Valorization of rice by-products



- Green extractions & Chemical characterization
- Bioactivity: Antioxidant activity & Antiproliferative potential

# Activity I:

## Characterization of Mediterranean rice varieties



- Chemical characterization
- Antioxidant activity

### OBJECTIVE

To link the chemical profile of 22 rice varieties with nutritional value and health-promoting bioactivities

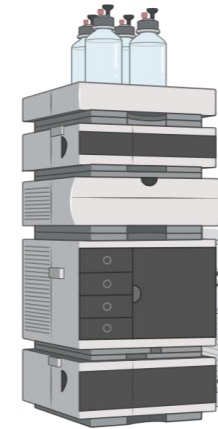


# Characterization of Mediterranean rice varieties

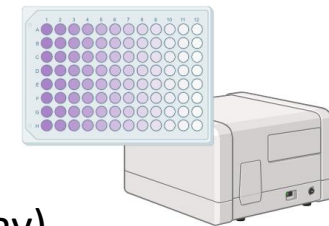


22 Mediterranean rice varieties  
(cooked brown rice)

✓ Total phenolic content (TPC)  
determined by HPLC-DAD



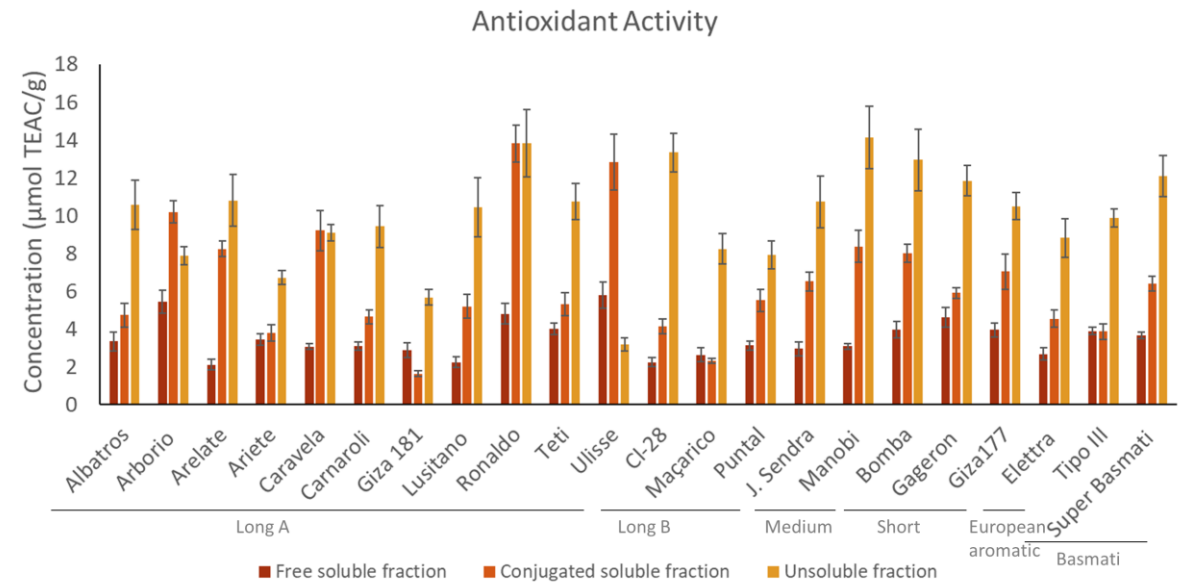
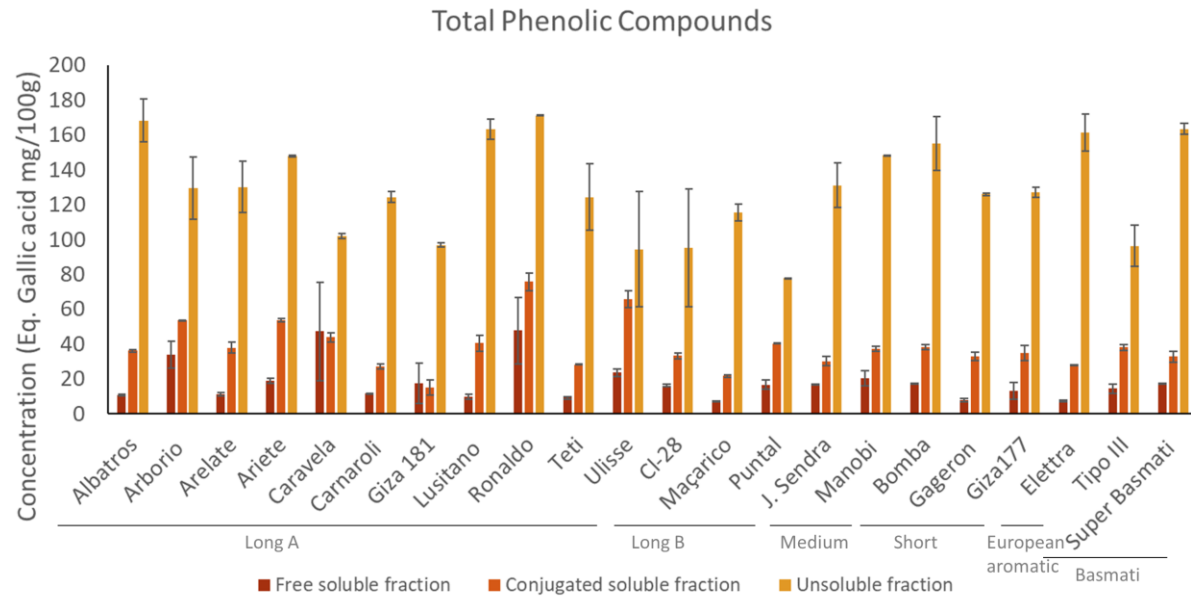
✓ Antioxidant activity (ORAC)



✓ Total saponin content (vanillin-sulfuric acid assay)

# Characterization of Mediterranean rice varieties

Phenolic compounds possess a high antioxidant and free radical scavenging potential.

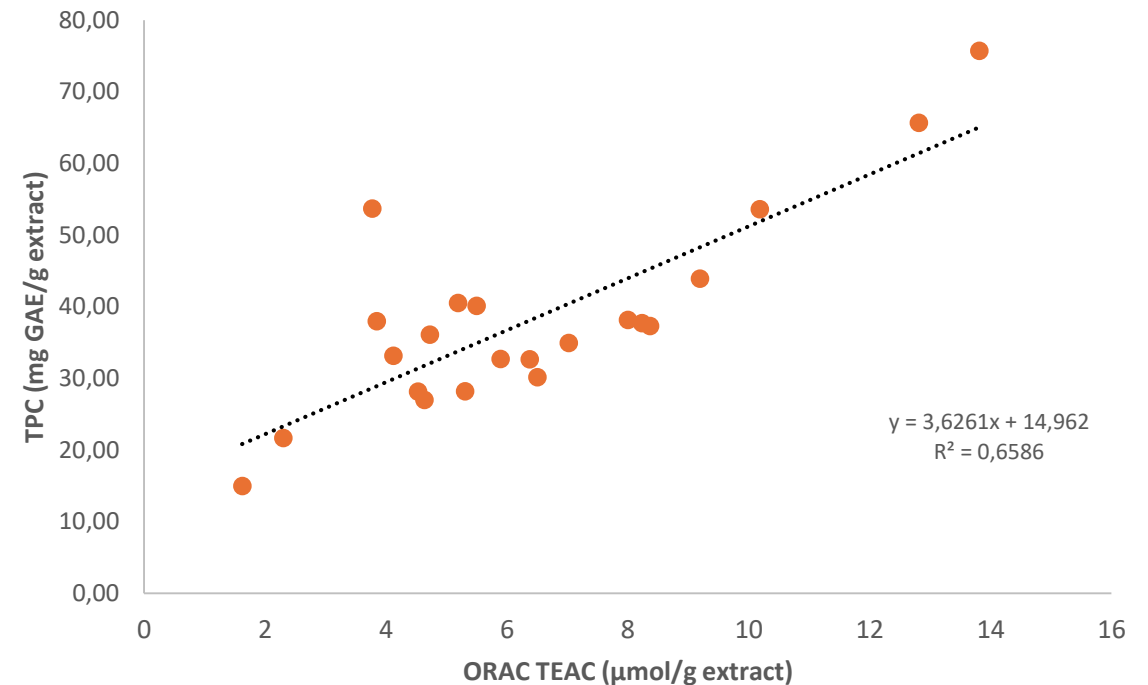


- ✓ No relation could be observed between TPC and AA and rice type
- ✓ The **insoluble phenolic fraction** consistently demonstrated **higher TPC and AA** compared to the soluble fractions
- ✓ In general, rice varieties with **higher TPC** also showed **higher AA**

# Characterization of Mediterranean rice varieties

✓ Positive correlation ( $r^2 = 0.66$ ) between TPC and AA of the conjugated soluble phenolic fraction.

Correlation TPC vs Antioxidant Activity

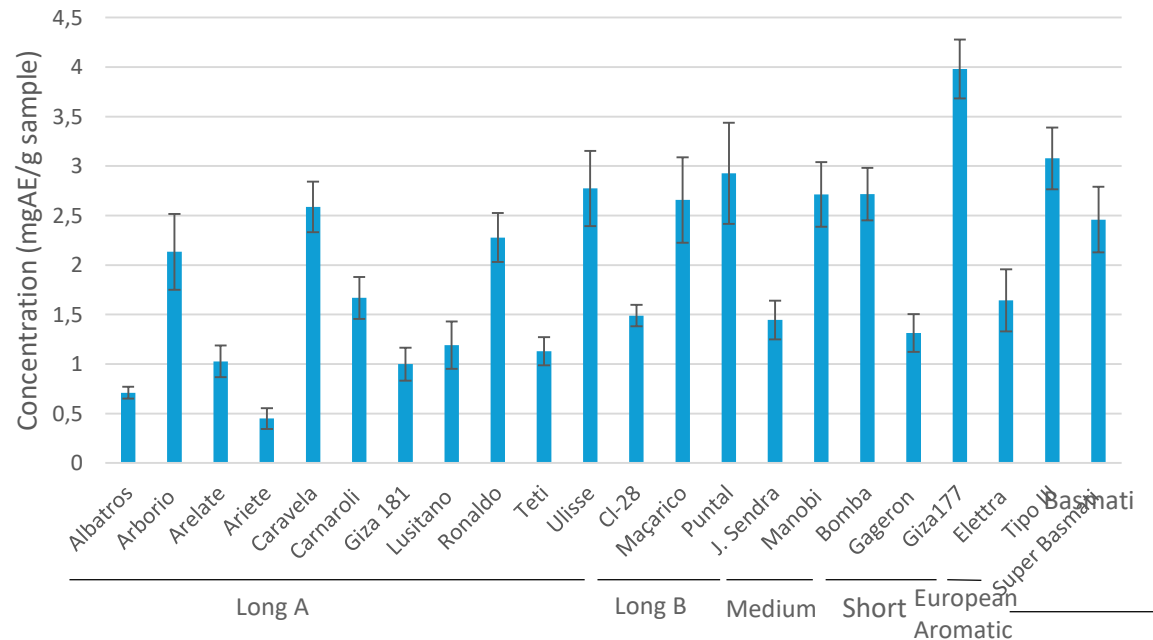


# Characterization of Mediterranean rice varieties

## Saponins:

- ☹️ Anti-nutrient, bitter, inhibits protein digestibility
- ☺️ Foaming agents, detergents, and emulsifiers; antioxidant, anti-cancer, anti-inflammatory, anti-thrombotic etc.

Total Saponin Content

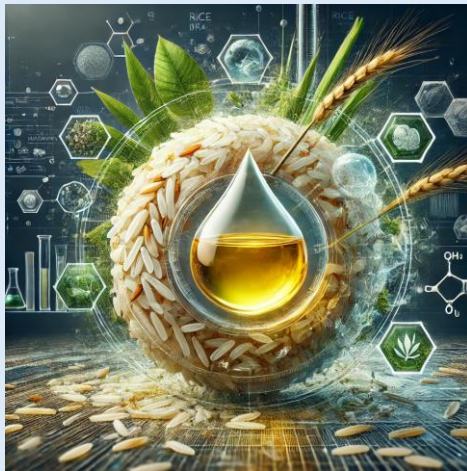


✓ No relation could be observed between total saponin content and rice type



# Activity II:

## Valorization of rice by-products



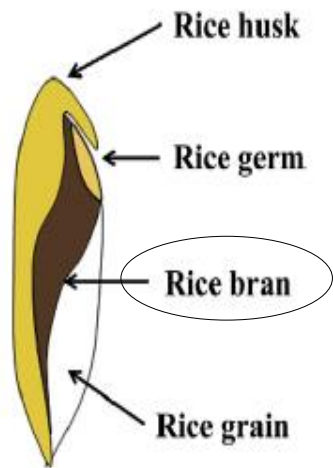
- Green extractions & Chemical characterization
- Bioactivity: Antioxidant activity & Antiproliferative potential

### OBJECTIVE

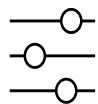
To valorize rice by-products through the development of green extraction methods to obtain bioactive rich-extracts

# Valorization of rice by-products

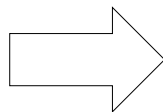
## Extraction & chemical characterization



**Supercritical CO<sub>2</sub> extraction**

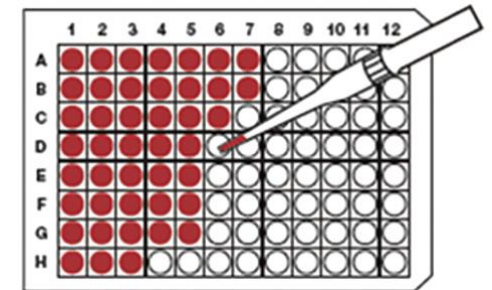
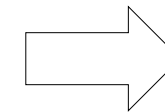
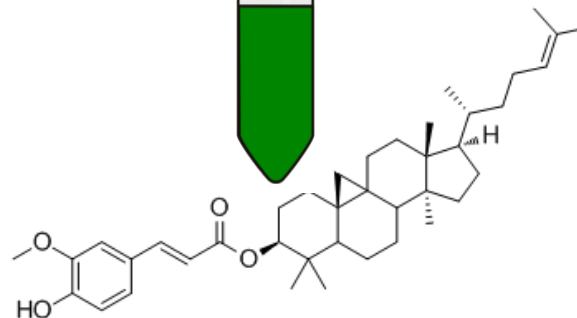


- ✓ Temperature (°C)
- ✓ Pressure (bar)



**Extract rich in:**

- ✓  $\gamma$ -oryzanol
- ✓ Nutritious unsaturated fatty acids (linolenic, linolenic and oleic acid)



**Bioactivity:**

- ✓ Antioxidant effect
- ✓ Antiproliferative potential

**$\gamma$ -Oryzanol:** Anticarcinogenic, anti-inflammatory, antidiabetic and neuroprotective, which are mainly attributed to its antioxidant capacity

# Valorization of rice by-products

## Extraction & chemical characterization

Raw material: rice bran of japonica (RB)

Fixed parameters:

Flow rate | 20 g/min

Time | 180 min

Raw material | 20 g



Exp No	T (°C)	P (bar)	Extraction yield g (%)	mg/g extract	
				Total FA	γ-Oryzanol
1	40	200	3.01 (15.1 %)	757.64	14.49
2	80	200	2.94 (14.7 %)	772.31	8.94
3	40	500	2.51 (12.6 %)	716.35	24.89
4	80	500	3.64 (18.2 %)	707.41	22.72
5	40	350	2.76 (13.8%)	752.64	20.62
6	80	350	3.53 (17.6 %)	787.22	17.56
7	60	200	3.18 (15.9 %)	723.50	15.4
8	60	500	3.66 (18.3 %)	746.41	18.78
9	60	350	3.36 (16.8 %)	784.49	15.38
10	60	350	3.16 (15.8 %)	782.52	16.75
11	60	350	3.33 (16.7 %)	745.21	19.41

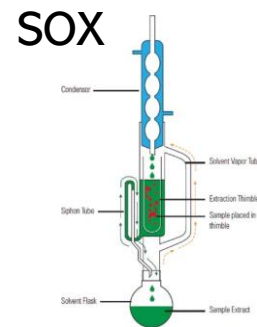
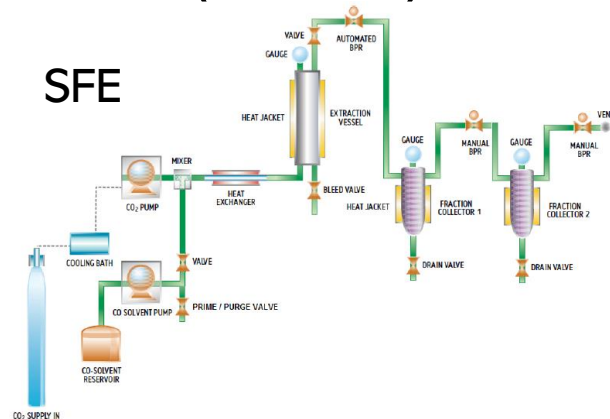
SFE Optimized conditions

Pressure: 500 bar | Temperature : 62 °C

# Valorization of rice by-products

## Extraction & chemical characterization

Optimized SFE conditions (SFE-opt) vs conventional extraction using Soxhlet with n-hexane (SOX-Hex)

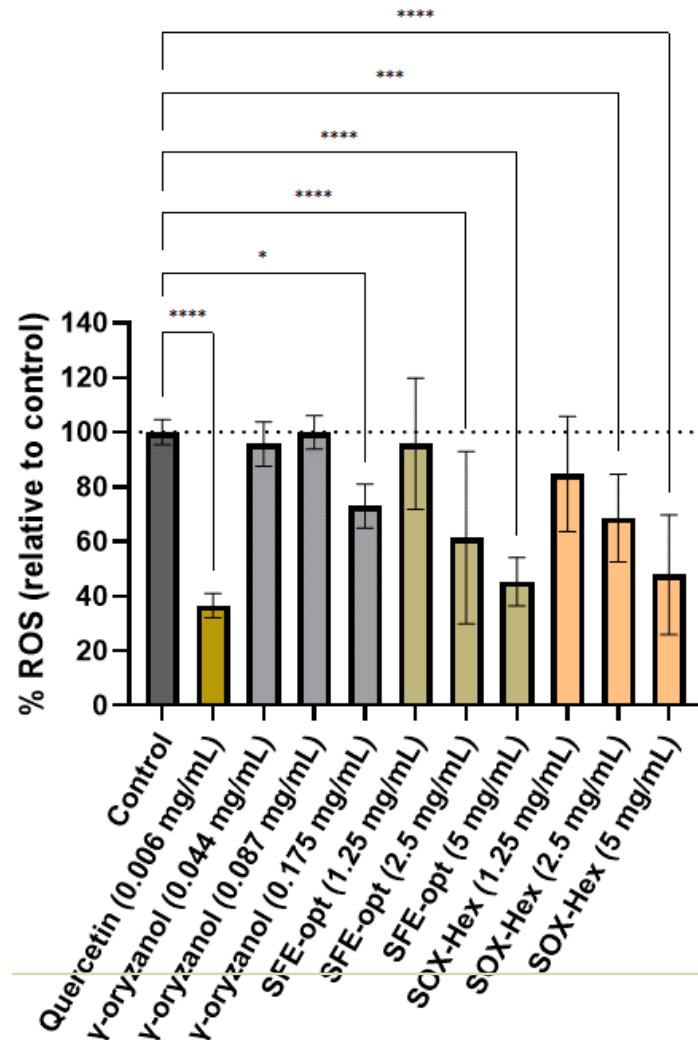


Extract	Extraction yield (%)	Total FA (mg FA/g extract)	$\gamma$ -Oryzanol (mg/g extract)
SFE-opt	17.3	784.4	36.4
SOX-Hex	18.0	723.3	18.3

- ✓ SFE-opt and SOX-Hex extraction mass yields and FA content are comparable.
- ✓ SFE revealed as the most selective solvent for  $\gamma$ -Oryzanol.

# Valorization of rice by-products

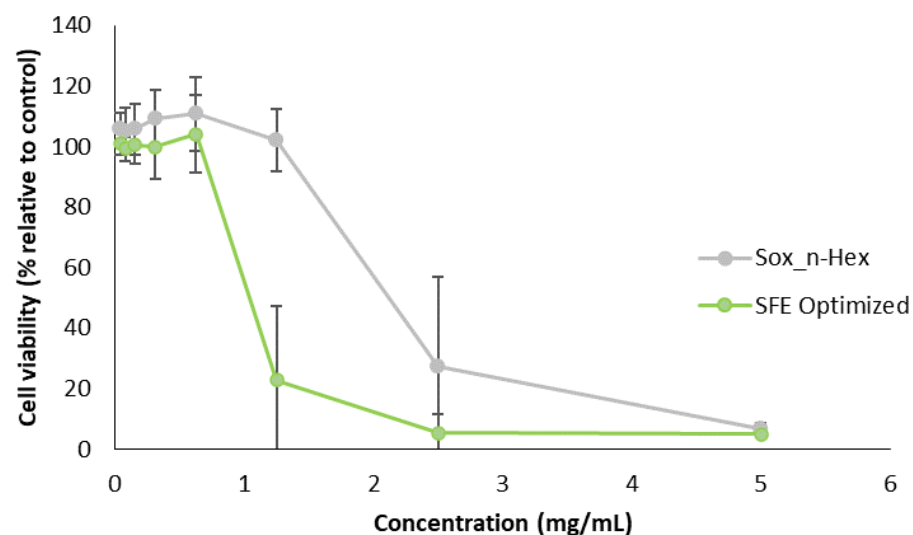
## Bioactivity – intracellular antioxidant activity (Caco-2)



- ✓ Inhibition of ROS formation in dose-dependent effect
- ✓ Both extracts presented higher antioxidant capacity when compared with  $\gamma$ -oryzanol  $\rightarrow$  other compounds may be scavenging ROS

# Valorization of rice by-products

## Bioactivity – antiproliferative effect (HT29)



Extract	EC50 (mg/mL)
SFE-opt	0.9±0.04
SOX-Hex	1.5±0.19

- ✓ Both extracts inhibited HT29 cell proliferation in a dose-dependent manner.
- ✓ SFE-opt extract showed higher antiproliferative effect



# Conclusions

- **No relation** could be observed between total phenolic compound, antioxidant activity and saponin content and rice type or variety.
- SFE is a plausible technology to extract bioactive-rich extracts. Compared to conventional extraction;
  - SFE-opt and SOX-Hex **extraction mass yields** and **FA content** are comparable.
  - Supercritical CO<sub>2</sub> was revealed as the most **selective** solvent for **γ-Oryzanol**.
  - Both extracts presented higher **antioxidant capacity** when compared with γ-oryzanol.
  - SFE-opt extract showed higher **antiproliferative effect**.

# Acknowledgements

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**iNOVA4Health**  
-Advancing Precision Medicine-

**LS4FUTURE**

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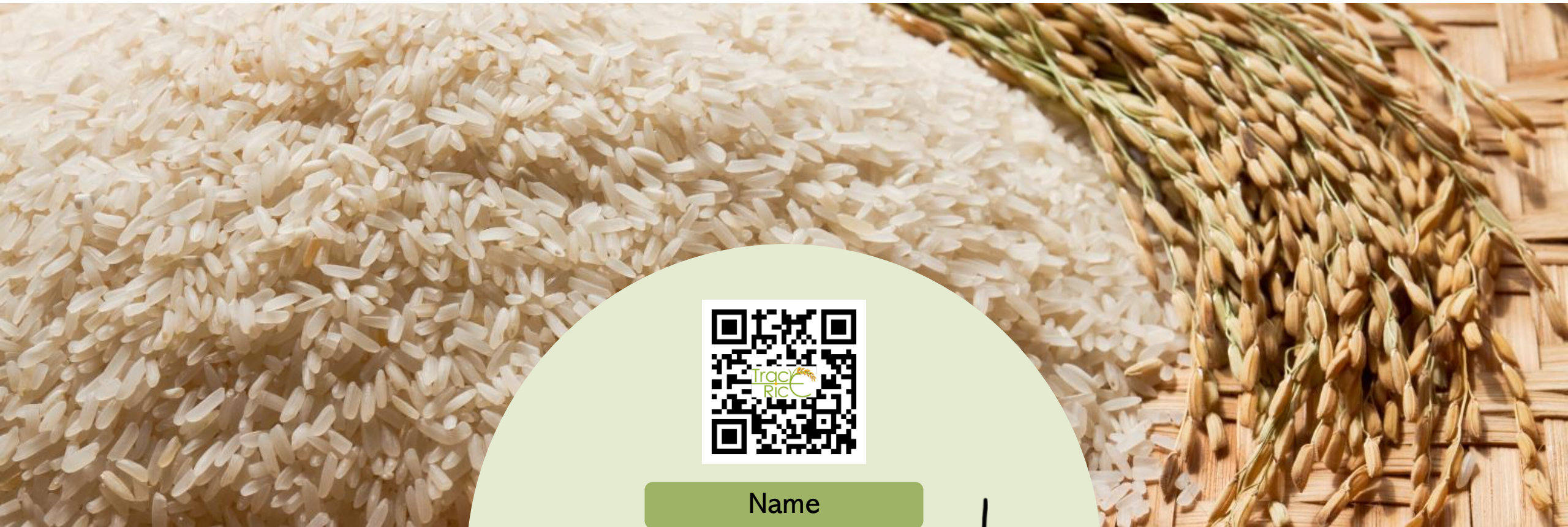




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thank you!