



# CHEMICAL CHARACTERIZATION OF RED GOJI BERRY (*LYCIUM BARBARUM* L.) CULTIVATED IN REPUBLIC OF NORTH MACEDONIA

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## Good to know.....

### **GOJI BERRY CONSUMPTION SEEMS TO HAVE BENEFITS INCLUDING:**

- heart disease risk reduction,
- hypoglycaemic and hypolipidemic effects,
- anti-aging,
- anti-tumour properties,
- immune modulation,
- neuroprotective effects,
- male fertility facilitation



## Good to know.....

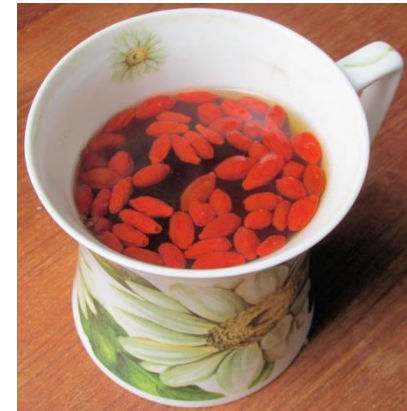
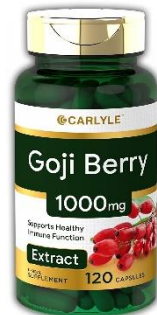
### GOJI BERRIES ARE CONSUMED:

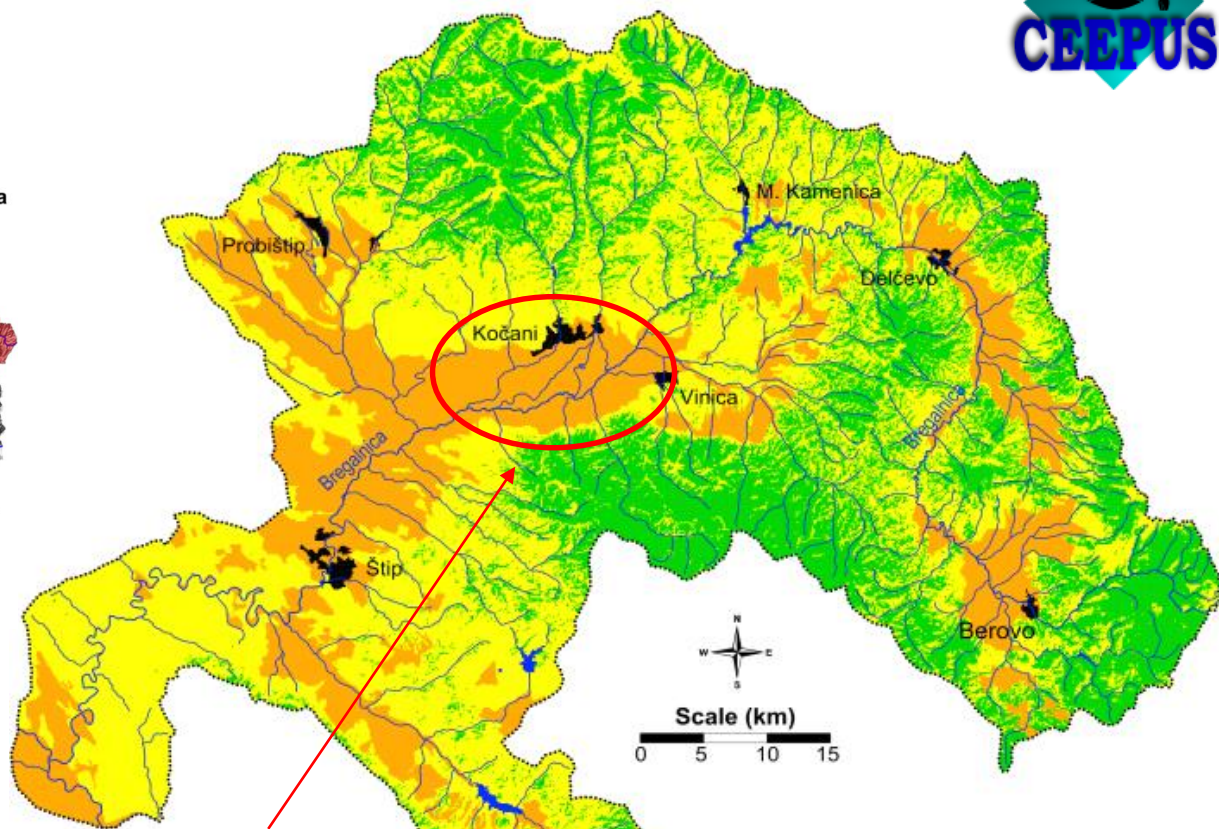
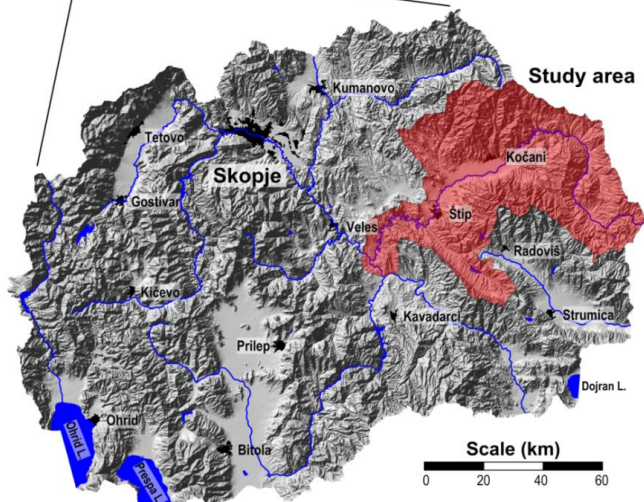
- Fresh,
- Dried,
- Processed into juice
- Capsules and concentrated extracts
- Incorporated in food....

Goji have been an important part of the traditional Chinese medicine for more than 2000 years.



Rapid increase in consumption in Europe is noticed at the beginning of the twenty-first century





location 1: 41°54'3.82"N, 22°22'1.34"E;  
 location 2: 41°54'14.80"N, 22°24'55.47"E;  
 location 3: 41°53'26.74"N, 22°22'45.77"E;  
 location 4: 41°53'13.20"N, 22°21'54.42"E;  
 location 5: 41°55'5.98"N, 22°26'47.71"E).



The aim of this study was to evaluate for the first time the composition, nutritional and functional properties of red Goji berry (*Lycium barbarum* L.) cultivated in the Republic of North Macedonia.

# Data set and methodology.....

**PROTEIN CONTENT** was estimated from nitrogen measured with a CHNS/O analyzer



**ANTIOXIDANT ACTIVITY** of methanolic extracts was determined using three different approaches:

- photochemiluminescence (PCL) with the ACL kit from Analytik Jena
- for antioxidative capacity of lipid-soluble compounds, DPPH (1,1-diphenyl-2-picrylhydrazyl) and ABTS (2,2'-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid) assays,



**NUTRITIVE AND TOXIC ELEMENTS** were analyzed using:

- ICP-OES (Na, Mg, K, Ca, Mn, Fe, Cu, Zn, S, P and Mo)
- GFAAS (As, Cd, Cr, Ni, Pb)
- TDAAS (Hg)

Profile of **FATTY ACIDS** (FAs) was assessed by gas chromatography coupled to flame ionization detector (GC-FID) after extraction in a chloroform:methanol mixture.

**TOTAL PHENOLIC CONTENT (TPC)** was measured using the Folin-Ciocalteu assay.

## Data set and methodology.....

### DATA PROCESING

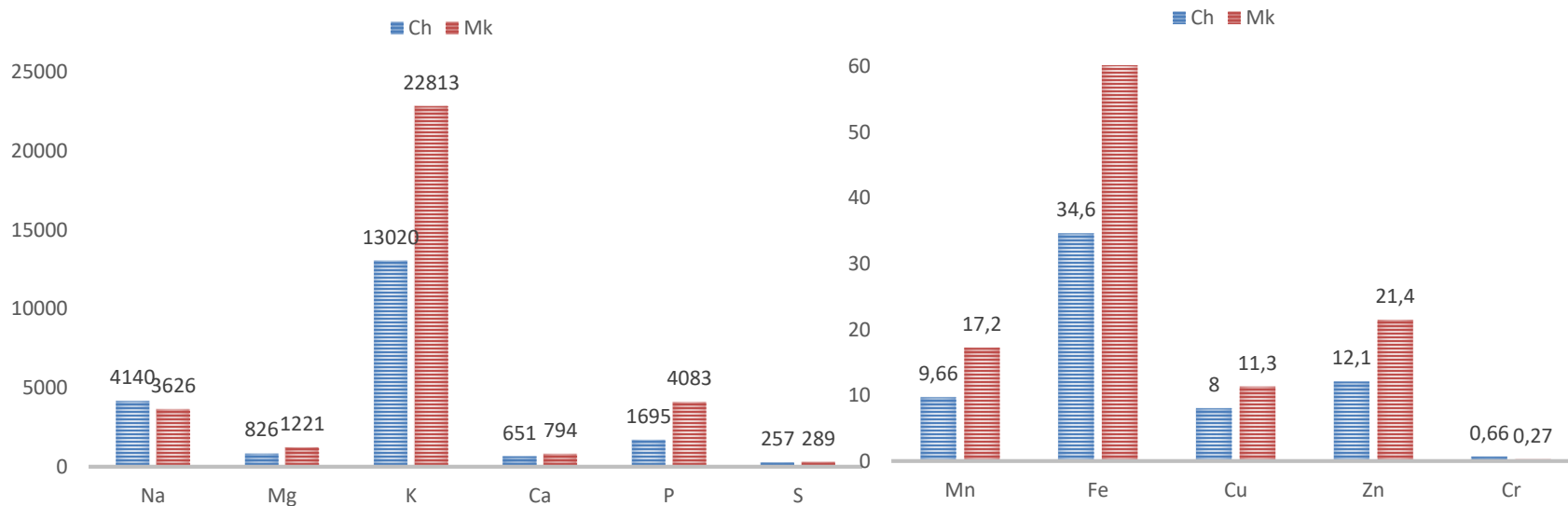
- **Comparative analysis:** Recommended Daily Allowance (RDA)
- **Multivariate analysis:** Principal Component Analysis (PCA) and Cluster Analysis (CA) using heat map

A large green curved arrow pointing from the list of methods towards the summary text.

The nutritional and functional properties of Macedonian Goji were compared to those of *Lycium Chinese L.*

	Na	Mg	K	Ca	Mn	Fe	Cu	Zn	P	S	Cr	As	Pb	Cd	Ni	Hg
<b>Min</b>	1660	1100	17600	567	12.0	51.5	10.3	18.0	3560	214	0.015	0.006	0.020	0.004	0.025	0.004
<b>Max</b>	6240	1390	28100	1010	20.5	76.9	12.4	26.0	4935	445	0.860	0.018	0.211	0.167	8.820	0.011
<b>X</b>	3626	1221	22813	794	17.2	64.0	11.3	21.4	4083	289	0.270	0.011	0.063	0.043	1.422	0.006
<b>s</b>	1344	90	3576	141	2.8	8.7	0.9	2.6	543	76	0.337	0.005	0.080	0.056	2.917	0.003

Elements content given in mg/kg dry goji berry



K>P>Na>Mg>Ca>S>Fe>Zn>Mn>Cu>Cr

	K	Cu	Mn	P	Fe	Mg	Zn	Ca	Cr
RDA (mg/day) <sup>a</sup>	2000	1	2	700	14	375	10	800	0.04
%RDA reference <sup>b</sup>	13.0	16.0	9.7	4.8	4.9	4.4	2.4	1.6	
%RDA±C.I. <sup>c</sup>	<b>22.8±9.6</b>	<b>22.5±4.7</b>	<b>17.2±7.5</b>	<b>11.7±4.2</b>	<b>9.1±3.3</b>	<b>6.5±1.3</b>	4.8±1.4	2.0±1.0	11.0±43.6

<sup>a</sup> Recommended Daily Allowance for minerals according to Regulation (EU) No 1169/2011

<sup>b</sup> *Lycium Chinese* L., RDA corresponding to consumption of 20 g/day

<sup>c</sup> *Lycium Barbarum* L. from Republic of Macedonia, RDA corresponding to consumption of 10–30 g/day

%/RDA contributions

### Total Target Hazard Quotient (TTHQ)

TTHQ of 0.087±0.036 (95% confidence level)

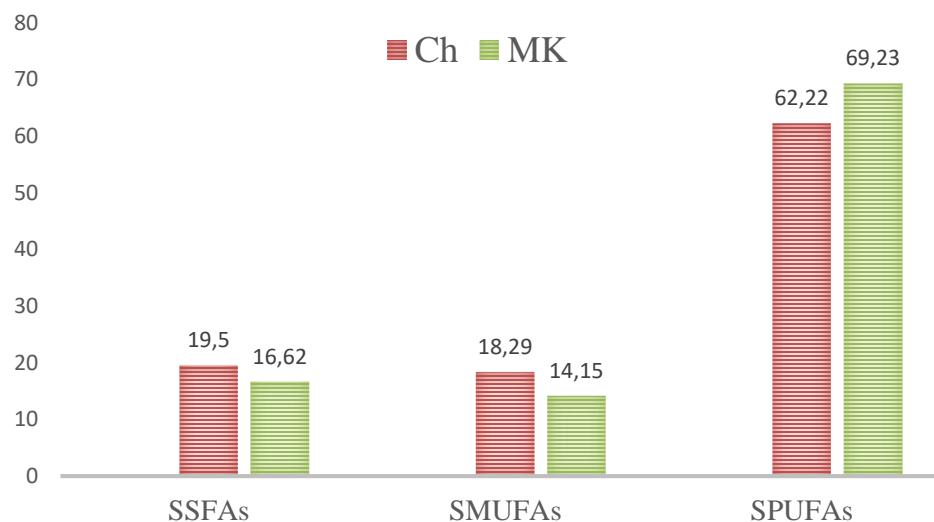
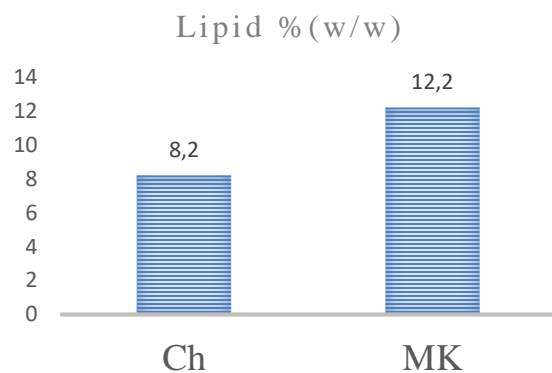
The calculus was based on 30 g/day ingested dried Goji berry, 365 days/year exposure frequency, 60 kg average b.w., 70 exposure years and oral reference doses (RfDs) in mg/kg-day of 0.001 (Cd), 0.0003 (As), 0.0003 (Hg), 0.004 (Pb) and 0.02 (Ni).

# FATTY ACIDS profile (FAs) was assessed

by gas chromatography coupled to flame ionization detector (GC-FID) after extraction in a chloroform : methanol mixture.



Lipid	C8:0	C10:0	C12:0	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	C20:0	C22:0	ΣSFAs	ΣMUFAs	ΣPUFAs	
% (w/w)	Caprylic	Capric	Lauric	Myristic	Palmitic	Palmitoleic	Stearic	n9 Oleic	n6 Linoleic	n3 Linolenic	Arachidic	Behenic		As	s	
Lycium Chinese	8.2	0.99	0.85	0.81	0.08	9.93	0.19	4.00	18.10	59.22	3.00	1.59	1.26	19.50	18.29	62.22
Min	10.4	0.05	ND	ND	0.09	8.31	ND	2.87	12.75	63.43	0.04	0.59	0.01	13.02	13.00	65.91
Max	13.8	1.23	1.16	1.28	0.19	11.79	0.32	4.13	15.23	70.98	3.53	1.54	0.24	19.76	15.43	73.98
Average	12.2	0.61	0.39	0.46	0.12	10.51	0.20	3.49	14.02	67.64	1.59	1.12	0.11	16.62	14.15	69.23
<b>C.I. (t<sub>critical value</sub>, 95%, n=8 =2,365)</b>	1.2	0.43	0.32	0.41	0.03	1.16	0.09	0.31	0.75	2.41	1.07	0.33	0.07	2.23	0.77	2.30
<b>t<sub>calculated</sub>, 95%, n=8</b>	8.241	2.055	<b>3.340</b>	1.978	<b>3.981</b>	1.181	0.438	<b>3.874</b>	<b>12.910</b>	<b>8.245</b>	<b>3.115</b>	<b>3.356</b>	<b>38.373</b>	<b>3.04</b>	<b>12.746</b>	<b>7.198</b>



**FATTY ACIDS profile (FAs)** was assessed by gas chromatography coupled to flame ionization detector (GC-FID) after extraction in a chloroform : methanol mixture.

Human health risk?

ARTHEROGENIC INDEX (AI)  
THROMBOGENIC INDEX (TI)  
OXIDISABILITY VALUE (COX)  
OXIDATIVE SUSCEPTIBILITY (OS)

$$AI = (4 \times C_{14:0} + C_{16:0} + C_{18:0}) / (\sum MUFA + \sum \omega 6 PUFA + \sum \omega 3 PUFA)$$

$$TI = [(C_{14:0} + C_{16:0} + C_{18:0}) / (0.5 \times MUFA + 0.5 \times \omega 6 PUFA) + 3 \times \omega 3 PUFA + (\omega 3 / \omega 6) PUFA]$$

$$C_{ox} = (C_{18:1} + 10.3 \times C_{18:2} + 21.6 \times C_{18:3}) / 100$$

$$OS = MUFA + 45 \times C_{18:2} + 100 \times C_{18:3}$$

Goji berry of Macedonian origin has health benefits

Sample	PUFAs/SFAs	n-3 <sup>c</sup>	n-6 <sup>c</sup>	n-3/n-6	AI	TI	Cox	OS
<i>Lycium Chinese L.</i>	3.19	3.00	59.2	0.050	0.13	0.29	6.93	2983
<b>Goji berry from Macedonia</b>	4.29	1.59	67.6	0.024	<b>0.13</b>	<b>0.31</b>	<b>7.45</b>	<b>3217</b>
<b>C.I. (t<sub>95%, n=8</sub> = 2.365)</b>	0.74	1.07	2.41	0.016	0.02	0.04	0.28	125
<b>t<sub>calc, 95%, n=8</sub></b>	3.48	3.12	8.25	3.96	0.73	1.18	4.48	4.42

Expressed in % (w/w)



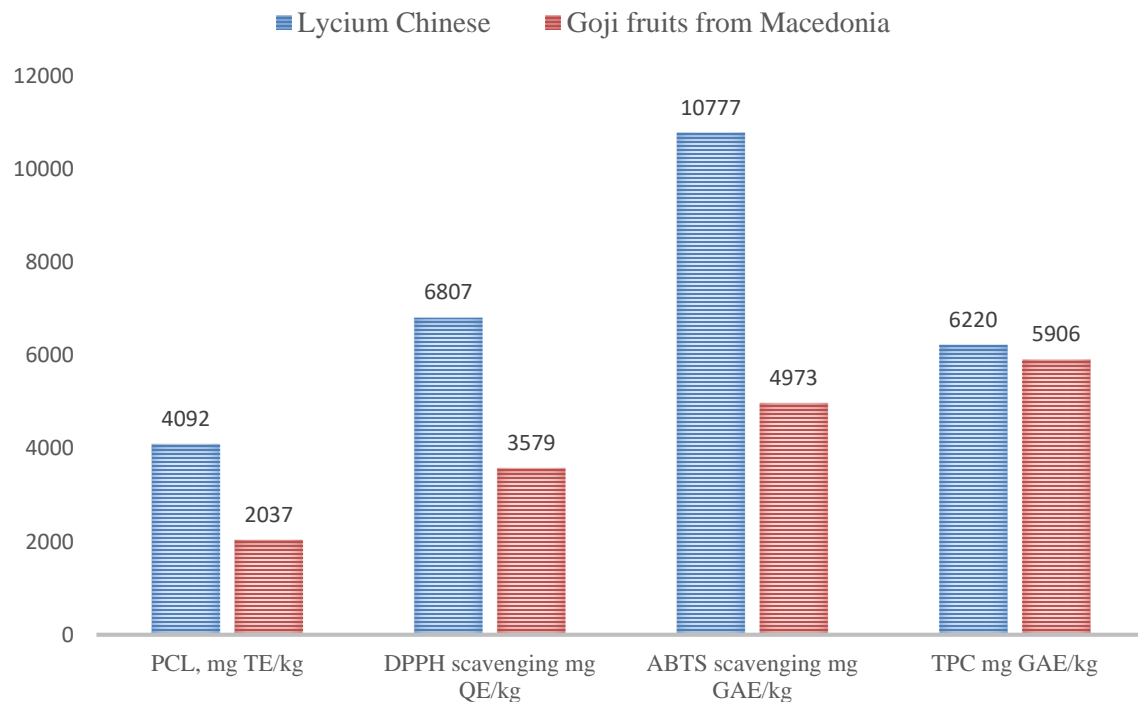
## ANTIOXIDANT ACTIVITY

of **methanolic extracts** was determined using three different approaches:

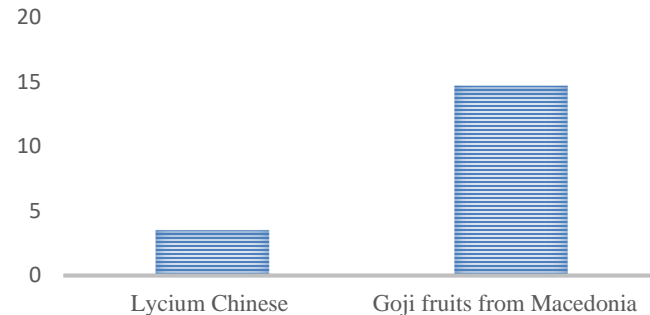
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- for antioxidative capacity of lipid-soluble compounds, DPPH (1,1-diphenyl-2-picrylhydrazyl) and ABTS (2,2'-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid) assays

## PROTEIN CONTENT

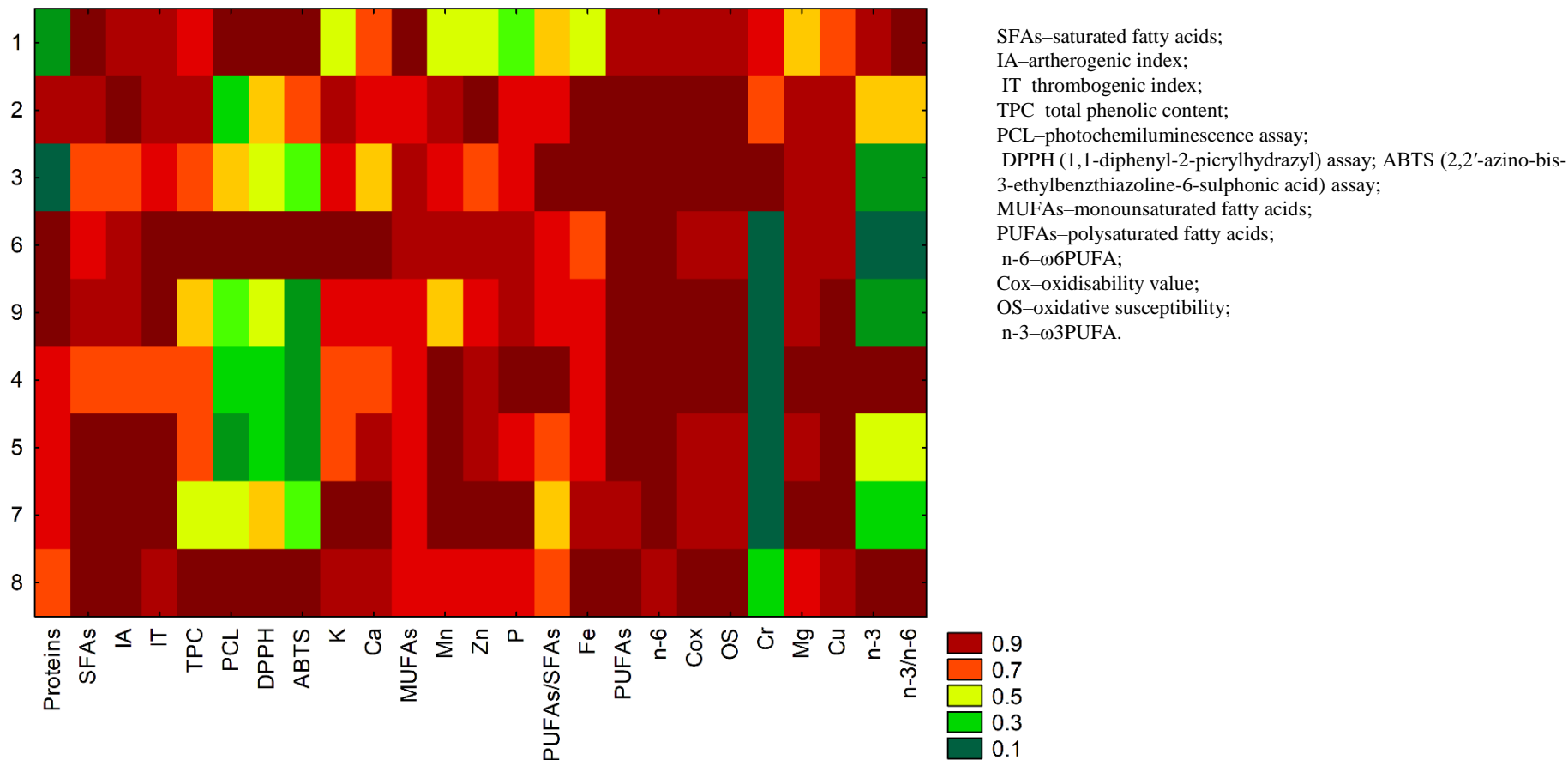
was estimated from **nitrogen measured** with a CHNS/O analyzer



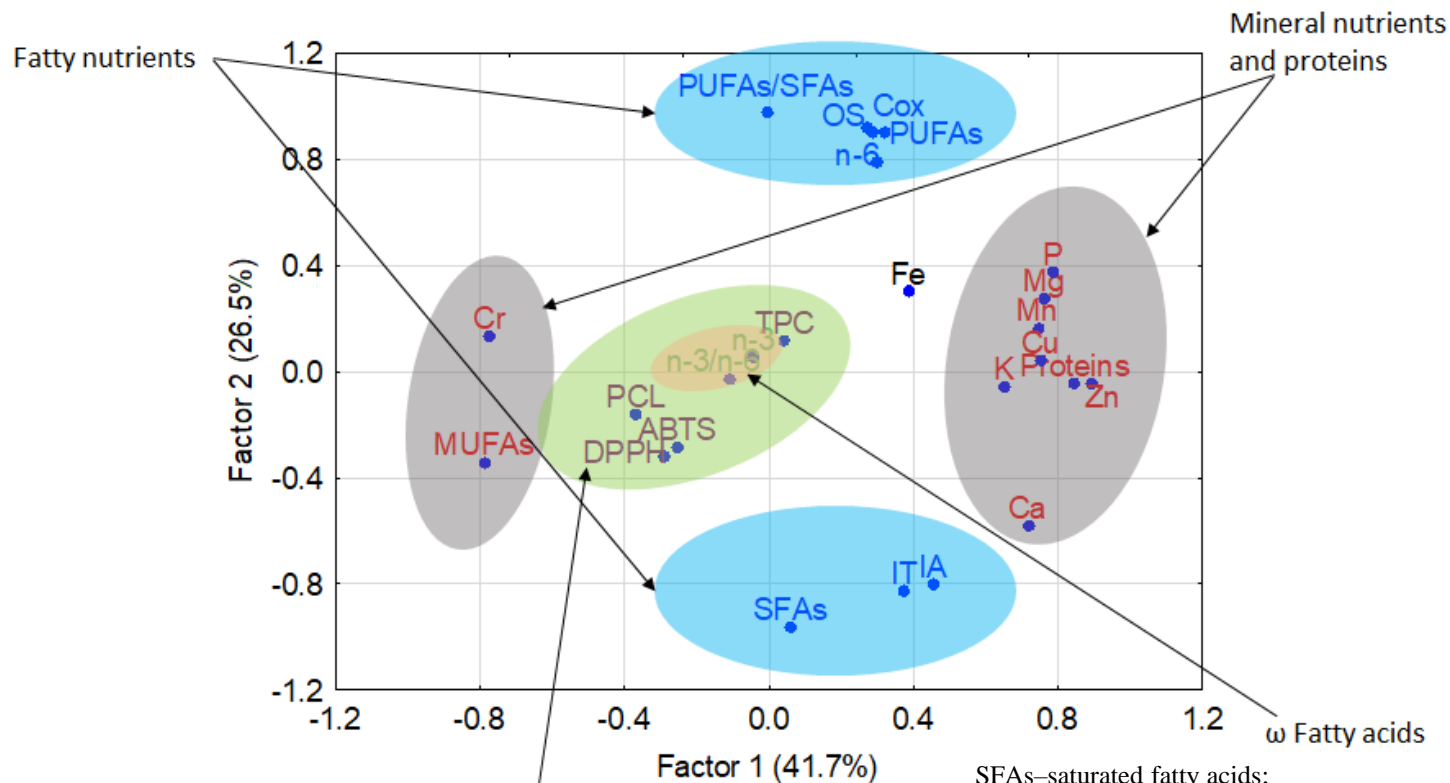
## Protein content (%)



# Heat map after two-way joining analysis

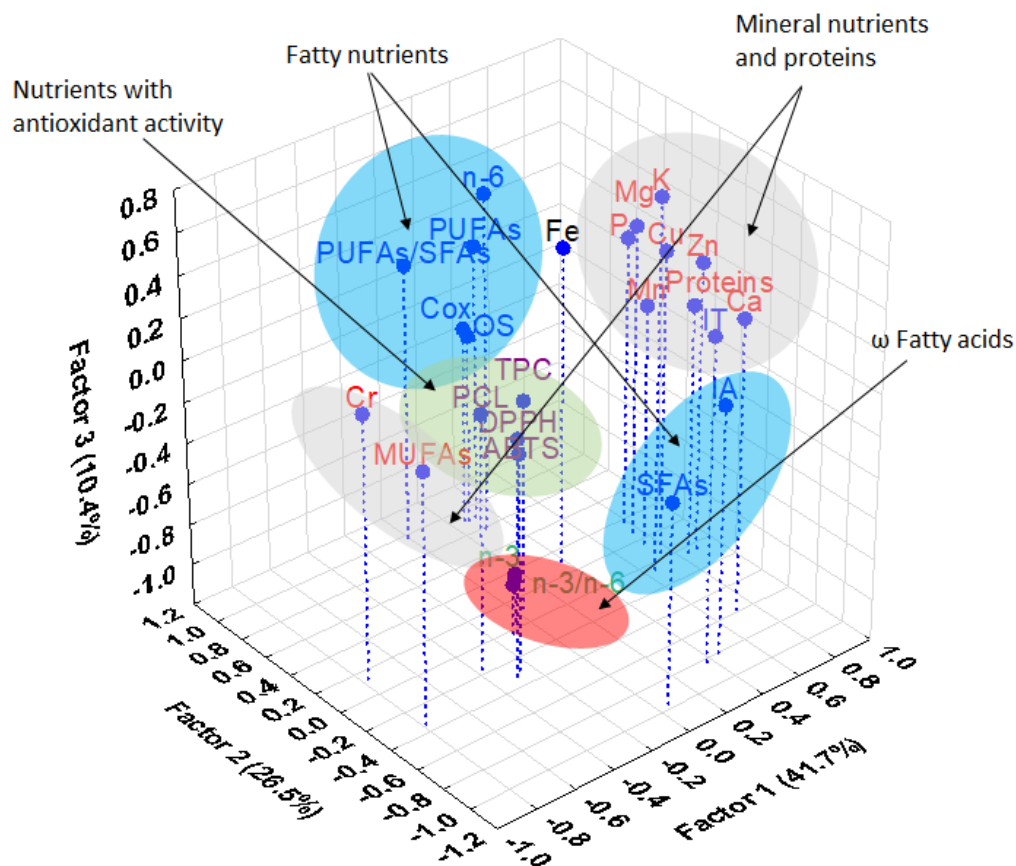


# The 2D scatterplot of the first two varimax-rotated PCA factors



SFAs—saturated fatty acids;  
 IA—atherogenic index;  
 IT—thrombogenic index;  
 TPC—total phenolic content;  
 PCL—photochemiluminescence assay;  
 DPPH (1,1-diphenyl-2-picrylhydrazyl) assay;  
 ABTS (2,2'-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid) assay;  
 MUFAs—monounsaturated fatty acids;  
 PUFAs—polysaturated fatty acids;  
 n-6— $\omega$ 6PUFA;  
 Cox—oxidisability value;  
 OS—oxidative susceptibility;  
 n-3— $\omega$ 3PUFA.

# The 3D plot of the first two varimax-rotated PCA factors



SFAs—saturated fatty acids;  
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 PUFAs—polysaturated fatty acids;  
 n-6— $\omega$ 6PUFA;  
 Cox—oxidisability value;  
 OS—oxidative susceptibility;  
 n-3— $\omega$ 3PUFA.

# CONCLUSIONS

The lipid profile of FAs dominated by **PUFAs**

PCA allowed grouping nutritional components and those with bioactive properties in 4 groups:

- (1) minerals and proteins
- (2) fat nutrients
- (3)  $\omega$  fatty acids
- (4) components with antioxidant activity.



Macedonian Goji berry is a **rich source** of mineral nutrients (K, Cu, Mn, P, Fe, Mg and Zn) and proteins.

**Very low concentrations of toxic elements** compared to allowable values in berry fruits set by European Commission and TTHQ index **demonstrated no risk for human health.**

Antioxidant activity was associated to **non-polar lipid soluble antioxidants, polyphenols and n-3 PUFA**

M. Senila, V. Ivanova-Petropulos, B. Balabanova et al. (2019) Composition and evaluation of nutritional and functional properties of red goji berry (*Lycium barbarum* L.) cultivated in Republic of Macedonia. *Food Analytical Methods* (submitted for publication)



***Thank you for your attention!***

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