Characterization of the Neuroactive Compound Profile in Organic, Conventional and Processed Tomatoes

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INTRODUCTION

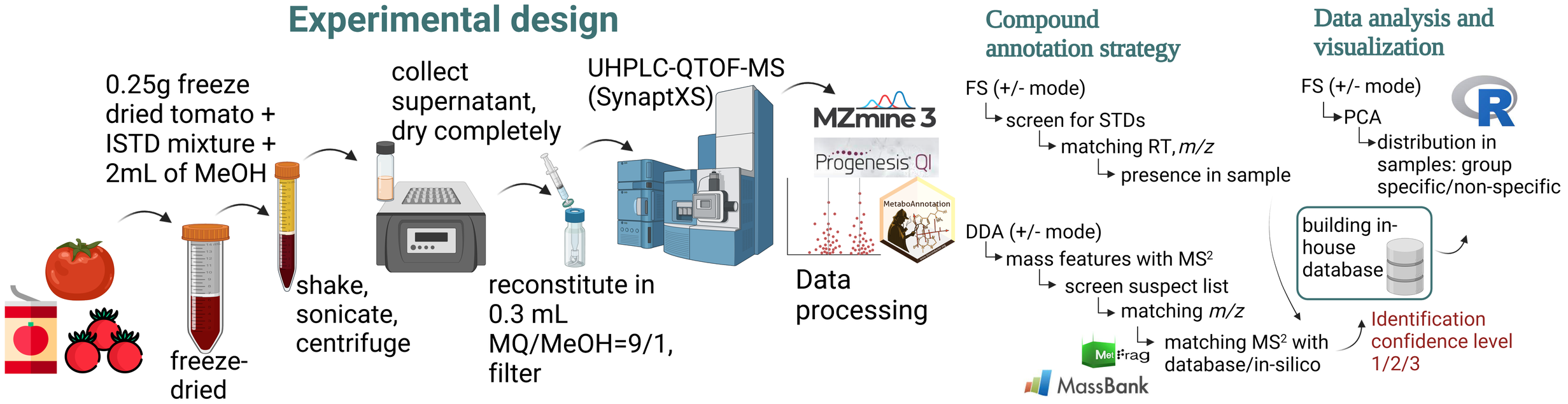
Neurodegenerative (ND) diseases are debilitating and largely untreatable conditions that affect 55+ millions of people worldwide. Despite the growing evidence of a correlation between diet, gut, and ND diseases, their connection is still unclear. Tomato is one of the most widely produced and consumed vegetables in the world and has the potential to contain neuroactive compounds.

OBJECTIVES

(1) To develope a non-target methodology using UHPLC-QToF (Synapt XS), including sample preparation, instrumental analysis and data processing;

(2) Characterize neuroactive compound profile (neuroprotective and neuro-disrupting compounds) in organic, conventional and processed tomatoes.

METHODS



Method optimization based on 27 neuroprotective and 15 neuro-disrupting chemicals (analytical standards, STD). The experimental design includes four groups of tomatoes: conventional and organic "datterini" (D and DO), conventional plump (PF), and processed tomatoes (PS). Samples were randomly analyzed in full scan (FS) and data dependent acquisition modality (DDA) in positive (+) and negative modes (-).The obtained recoveries of tested compounds were between 67-157% at higher (1.6 µg g^{-1} d.w.) and 41-130% at lower (0.1 µg g^{-1} d.w) concentration. Method and instrumental repeatability (relative standard deviation) was <20% and <15%, respectively. For most of compounds, the R^2 of linear calibration curve was >0.9 and matrix effect ±60%.

RESULTS

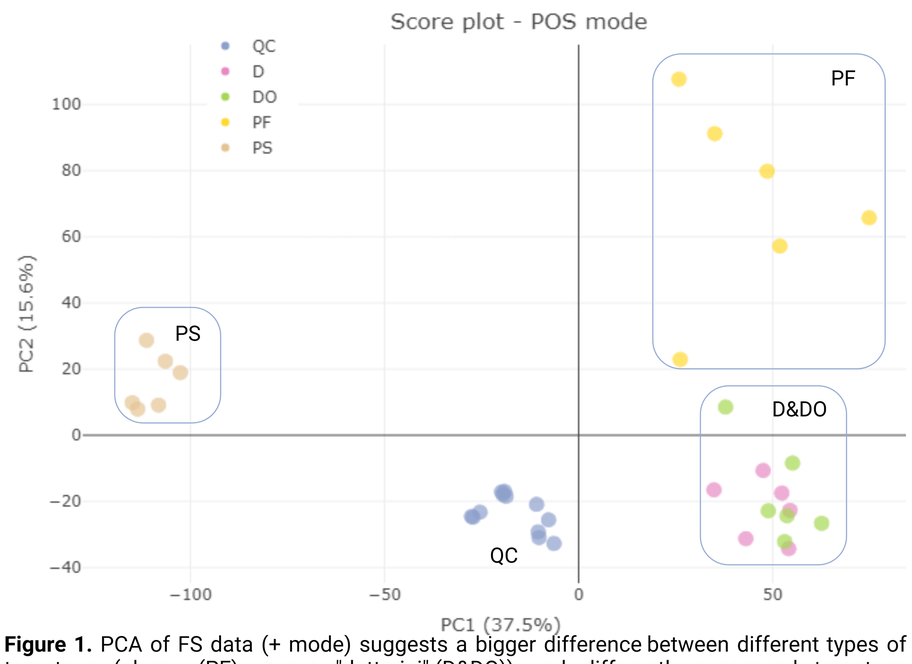


Figure 1. PCA of FS data (+ mode) suggests a bigger difference between different types of tomatoes (plump (PF) versus "datterini" (D&DO)) and differently processed tomatoes (processed (PS) versus fresh tomatoes (PF, D and DO)) than differently produced tomatoes (conventional (D) versus organic (DO)).

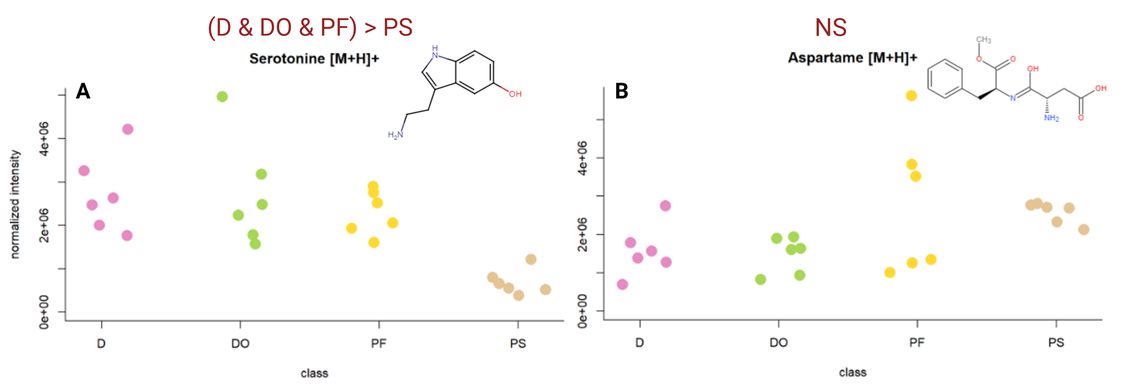


Figure 2. Examples of the distribution of two annotated compounds between different groups of tomatoes:
Figure 2A: Serotonin as a representative of **neuroprotective** and **group specific compound**, obtained in D, DO and PF ((D & DO & PF) > PS).
Figure 2B: Aspartame as a representative of **neuro-disrupting** and **non-group specific compound** (NS) with similar distribution between different group of samples.

Name	RT [min]	m/z	Δppm	Confidence level	Group
neuroprotective					
Citric acid	1.43	193.0354	6.0	2	NS
Phenylalanine	3.40	166.0861	1.2	2	NS
Chlorogenic acid (I)	5.29	355.1029	1.4	2	NS
Chlorogenic acid (II)	5.40	355.1029	1.5	1	NS
Rutin	7.89	611.1612	0.9	1	NS
L-Tyrosine	1.26	182.0816	2.5	2	PS
Dopa	1.32	198.0767	2.9	2	PS
5'-Deoxy-5'-	4.75	298.0975	2.2	2	PS
L-Valyl-L-leucine	4.90	231.1708	2.2	2	PS
Caffeic aldehyde	1.89	165.055	2.1	2	PF
Adenosine	2.50	268.1063	8.6	2	PF
Adenine	2.50	136.0623	3.6	2	PF
Guanine	2.64	152.0575	5.1	2	PF
Pantothenic Acid	4.18	220.118	0.4	2	PF
L-Tryptophan	4.51	205.0976	2.0	2	PF
Tyramine	1.95	138.0902	8.1	1	DO
Serotonine	2.74	177.1027	2.6	1	NPS
Tryptamine	4.58	161.1075	1.2	1	D & PF
neuro-disrupting					
Procainamide	4.98	236.1760	1.3	2	NS
Aspartame	5.43	295.1297	2.8	2	NS
Solasodine (I)	8.67	414.3375	2.1	2	NS
Solasodine (II)	8.99	414.3368	0.3	2	NS
Tomatine	10.13	1034.5481	4.8	1	NS
Acetaminophen	1.58	152.0709	2.1	2	PF
1-Naphthylamine	4.56	144.0812	2.6	2	PF
Amoxicillin	6.14	366.1139	5.6	1	PF
Artemisinin	7.41	283.1540	0.8	2	PF & PS
Disulfiram	3.34	297.0587	1.7	2	NPF

The developed **compound annotation strategy,** suggests the presence of group and non-group specific neuroactive compounds and identified several **neuroprotective**, e.g., polyphenols, amino acids, neurotransmitters, as well as **neuro-disrupting** compounds, e.g., alkaloids, pharmaceuticals, and food additives.

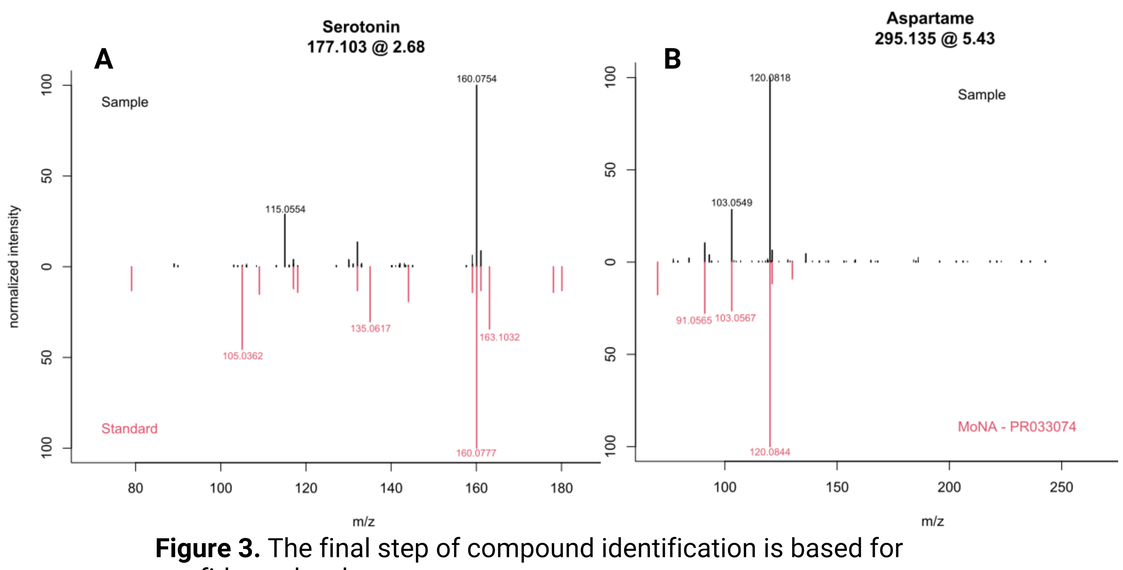


Figure 3. The final step of compound identification is based for confidence level
1. L1, e.g., serotonin (Figure 3A) on matching the RT, *m/z,* and MS^2 spectra of annotated compound in the sample (black spectra) with the corresponding analytical standard (red spectra).
2. L2, e.g., aspartame (Figure 3B) on matching the *m/z,* and MS^2 spectra of annotated compound in the sample (black spectra) with the MS^2 information available in on-line databases (red spectra).

CONCLUSIONS

(1) The developed and validated method enables the characterization of the neuroactive compound profile in organic, conventional and processed tomatoes;

(2) Results suggest a bigger difference between different types and differently processed tomatoes than differently produced;

(3) Based on the preliminary results, the developed compound annotation strategy is able to identify neuroprotective and neuro-disrupting compounds at different confidence level and suggests the presence of group and non-group specific neuroactive compounds;

(4) The annotation of compounds, using the developed strategy, is ongoing and aims to find out the most relevant neuroprotective/disrupting compounds that will be further investigated in the *in vitro* batch colon model to extend the knowledge of diet-gut microbiome-brain interactions and contribute towards preventing the burden of ND disease.

ACKNOWLEDGEMENTS

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