Metagenomics study of the microbiota of the traditional Bulgarian green cheese from Cherni Vit

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The Bulgarian green cheese is a traditional product from the village Cherni Vit near the town of Teteven in the Northern part of the Balkan mountain. It is produced from raw fats rich sheep or goat milk only within the village's area because of the specific local microclimate in combination with the lack of use of starter cultures. The metagenomics studies were accomplished on the Illumina HiSeq 2 x 250 bp paired end reads platform. In order to assess the eubacterial and the fungal microbiota of the traditional Bulgarian Green cheese the V3-V4 region of the 16S rRNA genes and the ITS2 region were targeted respectively. Between 97 and 137 eubacterial species and between 12 and 19 fungal species were observed in the four samples from 4 different cheese' batches. We found that the beneficial eubacterial and fungal species were predominant while the counts of the potentially pathogenic species were negligible. Based on these results we found that the Bulgarian green cheese is a safe for the customer dairy product, despite being prepared from raw milk.

EXPERIMENTAL WORKFLOW:

GREEN CHEESE BATCHES USED IN THIS STUDY

Cheese batch	Milk type	Type of the cheese	Aging
GC I	goat milk	hard	5 months
GC II	sheep milk	semi-hard	8 months
GC III	sheep milk	hard	7 months
GC IV	sheep milk	hard	8 months

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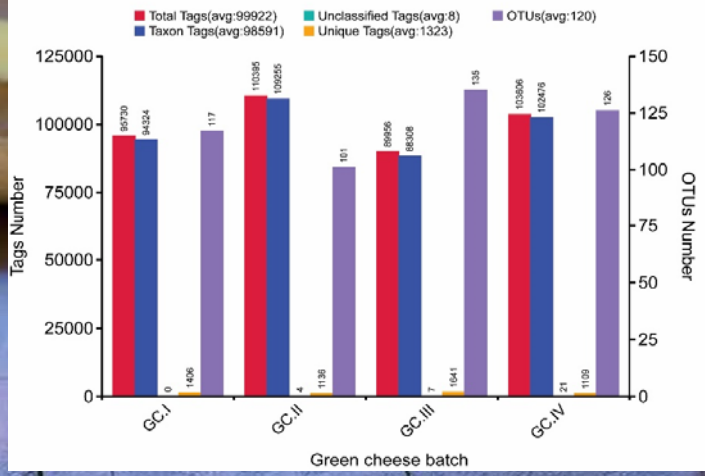
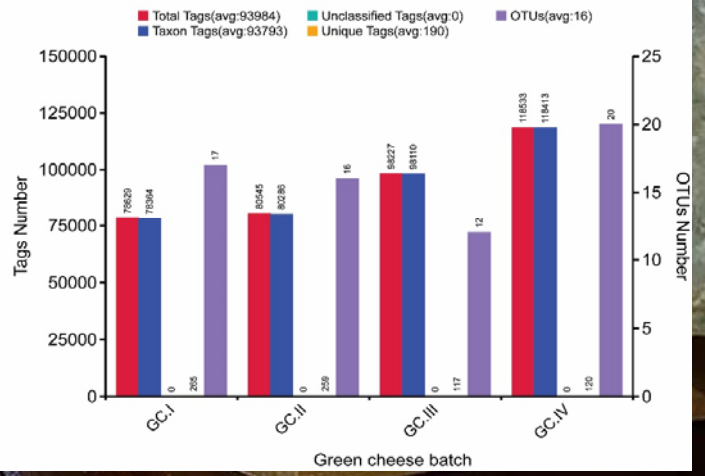
OTUs ANALYSES					
Green cheese batch	Number of the effective tags	Number of the annotated tags	Number of the unclassified tags	Number of the unique tags	Number of the OTUs
Panel A – eubacterial 16S analysis					
GC I	95730	94324	0	1406	117
GC II	110395	109255	4	1136	101
GC III	89956	88308	7	1641	135
GC IV	103606	102476	21	1109	126
Average	99922	98591	8	1323	120
Panel B – fungal ITS2 analysis					
GC I	78629	78364	0	265	17
GC II	80545	80286	0	259	16
GC III	98227	98110	0	117	12
GC IV	118533	118413	0	120	20
Average	93984	93793	0	190	16

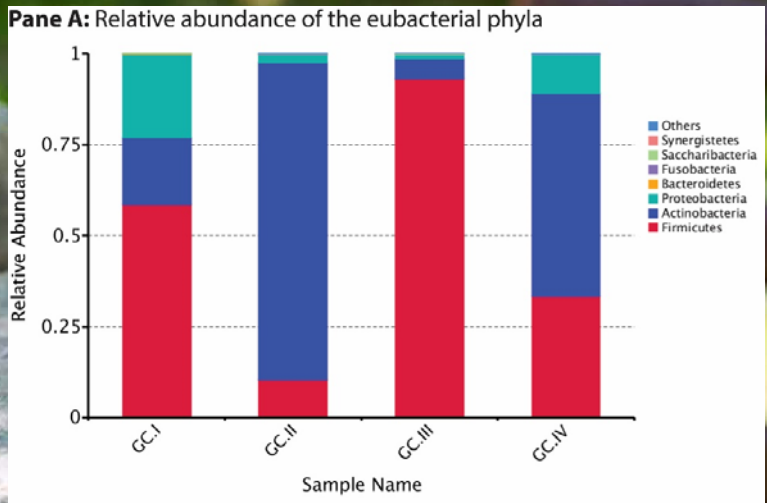
Pane A: Eubacterial OTUs based on 16S analyses



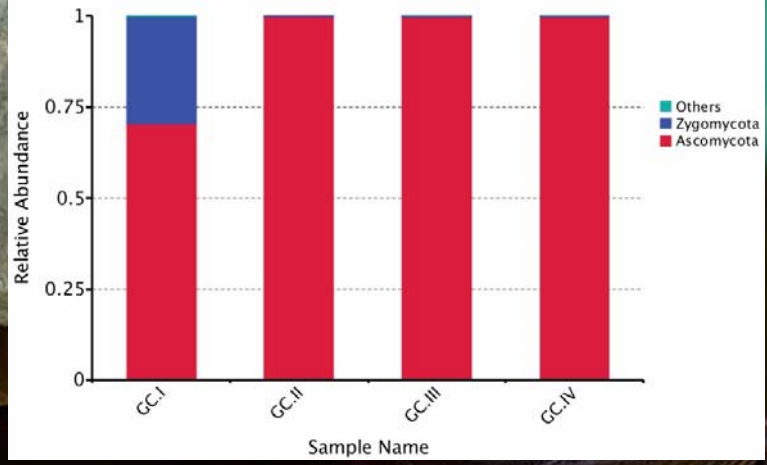
Pane B: Fungal OTUs based on ITS2 analyses



Pane A: Relative abundance of the eubacterial phyla



Pane B: Relative abundance of the fungal phyla



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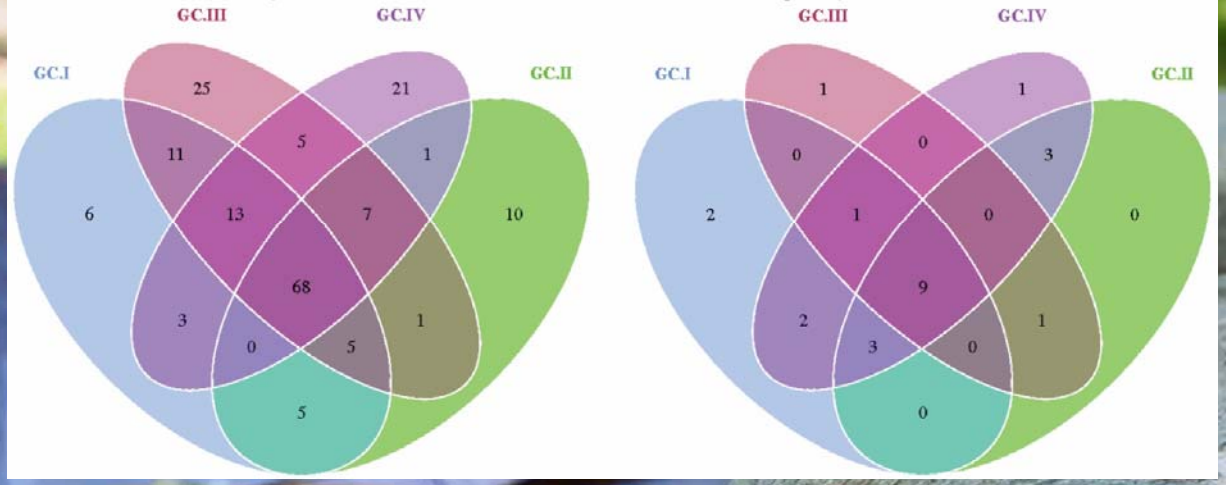
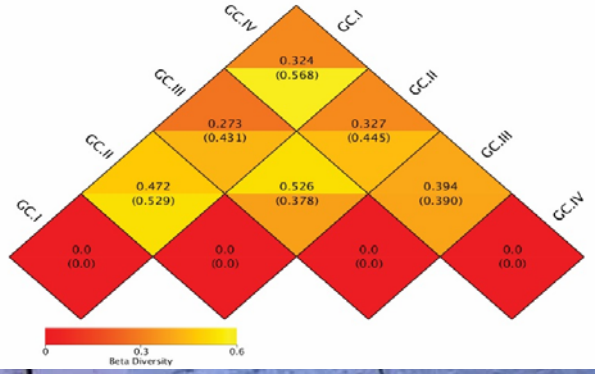
Pane A: Eubacterial species

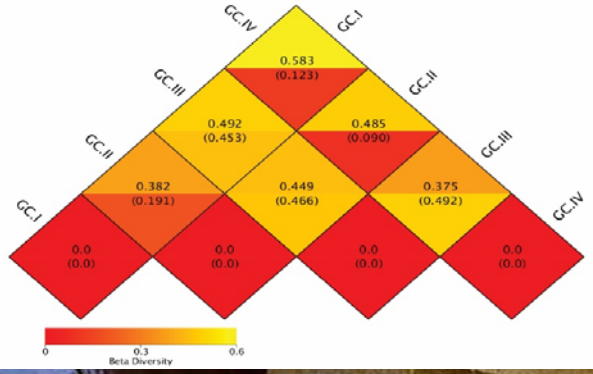


Pane B: Fungal species

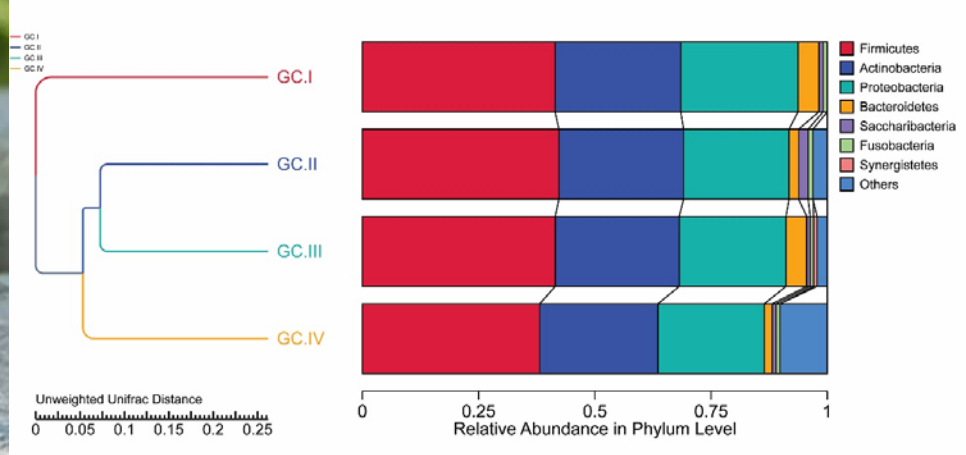
Pane A: Beta diversity heatmap for the eubacterial eubacterial communities



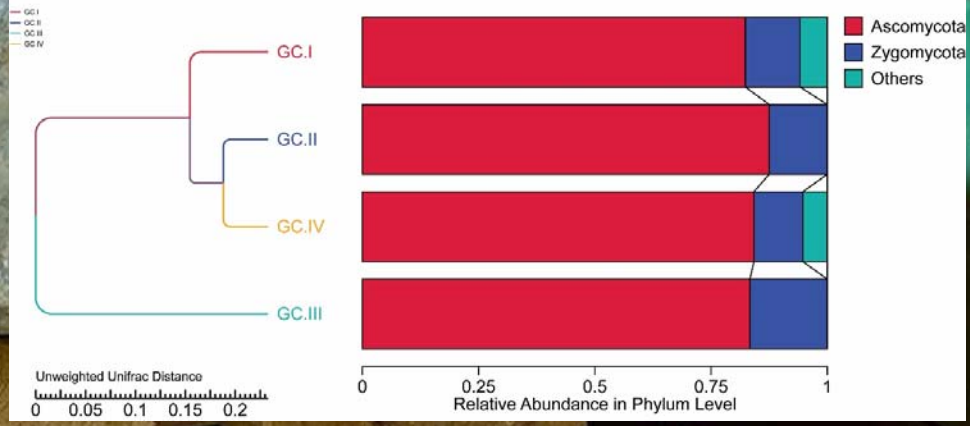
Pane B: Beta diversity heatmap for the fungal communities



Pane A: Eubacterial UPGMA tree



Pane B: Fungal UPGMA tree



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