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Determination of Escherichia coli in raw and pasteurized milk using a piezoelectric gas sensor array

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INTRODUCTION AND AIM

The importance of assessing the microbiological safety of food products is beyond doubt, which is also true for milk and dairy products. They are very important in the diet and contain nutrients necessary for the human body in well-balanced proportions and in an easily digestible form.

The aim of the research: to evaluate the changes in the composition of the gas phase over milk based on signals from chemical sensors to predict the quantity of the coliform bacteria group in the milk samples. .

The objects of the research:

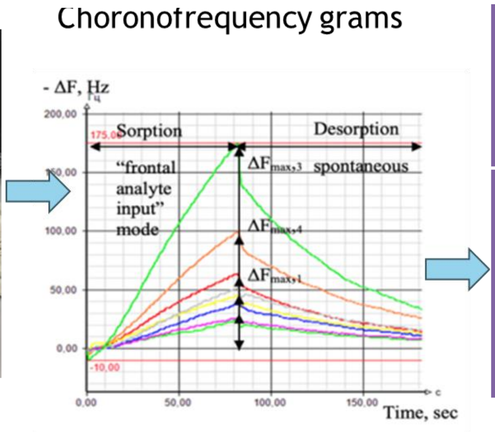
1) artificial milk as reference: sample No. 1 composed of individual components present in raw whole milk; sample No. 2 based on dry skim milk and cream
2) raw milk
3) pasteurized milk from raw milk samples

Table 1. Several physical and chemical and microbiological characteristics of the objects

Name of the indicator	Sample							
	reference No 1	reference No 2	raw cow's milk	skim milk	normalized mixture	pasteurized mixture	drinking milk (experimental)	drinking milk (control)
Milk in dry matter, %	12.3 – 12.5	12.3 – 12.5	11.36±0,30	9.71±0.17	11.50±0.38		11.58±0.33	11.53±0.41
Fat in dry matter, %	4.0 – 4.1	4.05 – 4.10	3.05±0.05	0.05±0.05	2.5±0.05			
Total protein in dry matter, %	3.0 – 3.1	3.40 – 3.45	3.46±0.1					
Lactose in dry matter, %	4.65 – 4.70	4.75 – 5.30	4.41±0.36	not determined	3.47±0.15		3.10±0.29	3.35±0.20
Titratable acidity, ^{o}T	17±0.5	18±0.5	18±0.5	19±0.5	17±0.5	18±0.5	18±0.5	18±0.5
Density, kg/m^3	1025±0.5	1025±0.5	1031±0.5	1030±0.5	1026±0.5	1026±0.5	1026±0.5	1026±0.5
QMA&OAMO, CFU/cm^3	<10^3	<10^3	5,8×10^6	not determined	5,8×10^6	<10^3	<10^3	
Coliform bacteria, CFU/cm^3	0	0	0	-	0	0	10^2 – 10^9	0

SCHEME OF MILK ANALYSIS

Diagnost-Bio-8



Number of parameters	Designation	Formula for calculation
1-8	β_i	$\beta_i=(\Delta F_{max,i}-\Delta F_{120s},i)/(120-\tau_{max,i})$, where $F_{max,i}$ and $\Delta F_{120s,i}$ are the maximum signal and signal at 120 seconds of measurement of i-th sensor, $\tau_{max,i}$ – time of achievement of maximal signal by i-th sensor.

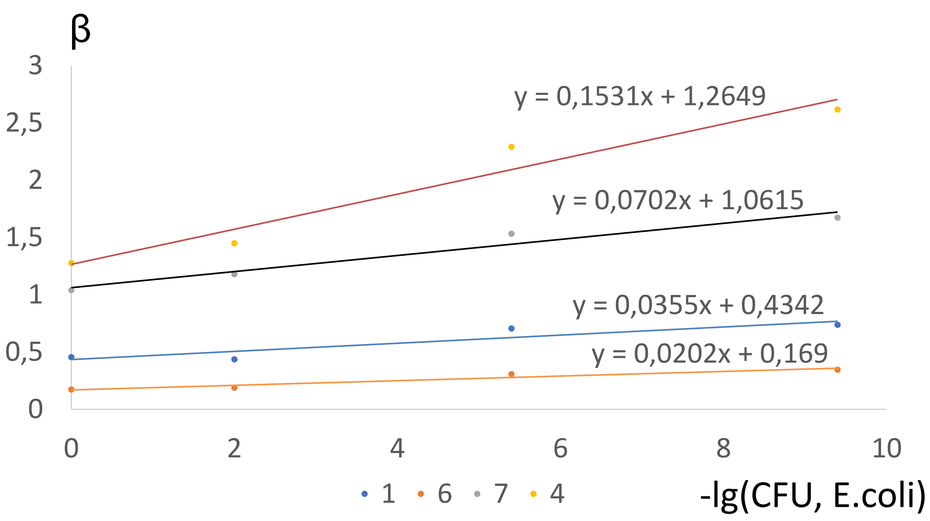


Fig. 1. Calibration graphs of the dependences of the coefficients b for sensors from lg(CFU) E.coli.

Polycomposite coatings for sensors:

1) dicyclohexyl-18-crown-6 / concentrate of micellar caseine
2) dicyclohexyl-18-crown-6 / chitosan
3) Tween 40 / Triton X-100
4) Choline +sorbitol+carbon nanotubes
5) Lanoline + silicone oxide
6) Lanoline + silicone oxide+choline +sorbitol
7) dicyclohexyl-18-crown-6 / Tritone X-100
8) sorbitol+silicone oxide

RESULTS AND DISCUSSIONS

Table 2. The results of prediction -lg(CFU/cm3) of E.coli for milk samples

Sensor number for calibration	Milk samples						
	1	2	3	4	5	6	7
1	-0,67	0,41	-1,40	3,66	0,77	4,74	2,21
4	4,47	3,04	1,79	5,56	3,88	4,89	4,47
6	0,52	1,79	-2,02	3,06	1,15	3,06	3,06
7	-1,29	1,45	-3,48	4,01	1,81	2,36	0,90
CFU(E.coli) in cm^3	0	10^2	0	0	10^2	0	10^3
Notes	Presence of other pathogenic microorganisms			High level of QMAFAnM	Artificial contaminated	Presence of mold	Artificial contaminated

To study changes in the gas phase and sensor output data over milk samples during processing, an experiment was conducted with contamination of raw milk with a pre-determined chemical composition with E.coli bacteria using a cell suspension in the amount of 1.2•10^9 CFU/cm3 after pasteurization of the normalized mixture.

Table 3. Relative changes in sensor signals (Δ, rel. units) for different stages in the technological process in relation to raw milk

Sensor number	Normalized	Pasteurized	Contaminated by E. coli
1	0,21	0	0,20
2	-0,37	-0,41	-0,79
3	-0,13	-0,09	-0,30
4	-0,15	-0,31	0,07
5	0	-0,32	0,21
6	-0,51	-0,35	-0,20
7	0,26	0,31	0,28
8	0,27	0,32	0,56

Compared with whole raw milk, during processing, depletion of the milk aroma is observed, primarily a decrease in the amount of volatile acids and an increase in the content of alcohols and ketones with a hydrocarbon radical C_5, while the strongest decrease in acids in the gas phase is observed after normalization and continues during pasteurization.

Table 4. Additional calculated parameters of sensors for milk samples

Sample	β1/β4	β1/β6	β1/β7	β4/β6	β4/β7	β6/β7
Water	0,23	2,08	0,38	9,08	1,66	0,05
1	0,21	2,29	0,42	10,86	2,00	-0,40
2	0,26	2,19	0,38	8,44	1,48	1,23
3	0,25	3,00	0,47	12,00	1,88	0,58
5	0,25	2,40	0,39	9,67	1,56	0,64
7	0,26	2,22	0,45	8,44	1,73	3,40

CONCLUSION

Thus, the proposed approaches to quantitative assessments of coliform bacteria in raw and pasteurized milk using gas-phase analysis with an array of sensors make it possible to significantly reduce the analysis time to 2-3 hours (including the sample collection and data processing) and thereby intensify the production of safe dairy products.

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