

SCREENING PROCEDURE FOR BIOGENIC AMINE PRODUCTION BY LACTIC ACID BACTERIA ISOLATED FROM TRADITIONALLY FERMENTED SAUSAGES

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Abstract

In order to apply certain microorganisms as starter cultures, it is necessary to carry out extensive investigations in respect to their genetic, biochemical and functional properties. Besides the exact identification on taxonomical level, pure culture of starter microorganism has to exhibit stabile and desirable physiological properties in its substrate - the meat. Above all, applied culture has to be safe for humans, i.e. it can not produce toxins, biogenic amines or other harmful substances. The aim of these investigations is to carry out adequate selection of lactic acid bacteria (LAB) species isolated from autochthonous fermented sausages in respect to the ability of these microorganisms to produce biogenic amines by amino acids decarboxylation *in vitro*. It is known that significant number of microorganisms that can be isolated from fermented products can lead to the accumulation of biogenic amines, especially tyramine, 2-phenylethylamine, tryptamine, cadaverine, putrescine and histamine. Excessive intake of these compounds poses a health risk for consumers due to their harmful effects on nervous and gastrointestinal system as well as blood pressure.

In order to investigate ability of LAB to produce biogenic amines, 50 strains of these microorganisms were used. Each strain was isolated and taxonomically identified. LAB's were isolated using conventional microbiological techniques. The LAB isolates from de Man, Rogosa, Sharpe broth (MRS broth, Oxoid) were first checked by Gram staining and catalase reaction. Gram-positive and catalase-negative isolates were then identified by a commercially available biochemical identification system API 50CHL. Genotypic identification of the LAB isolates was performed according to the manufacturer's protocol for Gram-positive bacteria. Microorganisms were isolated from "Sremska", "Levacka" and "Uzicka" sausages, manufactured in traditional manner. Qualitative and quantitative determination of biogenic amines in 24 hours old liquid cultures of LAB was carried out using high-performance liquid chromatography with UV detection. The following amines were determined: histamine (HIS), tyramine (TYR), cadaverine (CAD), putrescine (PUT), spermidine (SPD), spermine (SPE) and tryptamine (TRY). Detection limit of analytical method is 0.5mg/L. Values of biogenic amines content are expressed in ug/mL.

The results of investigation of LAB ability to produce biogenic amines as by-products of metabolism show low values of all seven biogenic amines in liquid cultures. The determined values do not exceed 20 ug/mL (HIS), 50 ug/mL (TYR) while PUT and SPD are found in negligible quantities in only a few strains. CAD and TRY were also found in traces while SPE content is below 15ug/mL of liquid culture. These findings can be compared to proposed toxic amount of e.g. histamine in foodstuffs of 100 mg/kg or 2 mg/L in alcoholic beverages or tyramine of 100 - 800 mg/kg and fenylethylamine of 30 mg/kg leading to the conclusion that found amounts are significantly below the levels that are considered as health risk.

The results of investigations showed that isolated autochthonous LAB from national fermented sausages are not significant producers of biogenic amines, tested *in vitro*. Having in mind other favorable technological properties, they can be used as potential starter microorganisms in meat industry.

Key words: Screening procedure, Biogenic amine, Lactic acid bacteria.