ROLE OF STARTER CULTURE IN FORMATION OF BIOGENIC AMINES IN PORK MEAT

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S-VIA.49

SUMMARY

Biogenic amine production by *Lactobacillus jensenii* was investigated in M.R.S. broth that was supplemented with different amino acids and in pork meat inoculated with the bacteria. Biogenic amines were determinated with BIOTRONIK LC 2000 amino acid analyzer and the appropriate precursor amino acids were determinated parallel.
Lactobacillus jensenii showed lysine and ornithine decarboxylase activity in the M.R.S. broth that was proved with increasing content of cadaverine and putrescine during incubation period. These results are in good agreement with the observations of pork meat because cadaverine and putrescine levels markedly increased in meat inoculated with *Lactobacillus jensenii*. The free precursor amino acid contents (lysine and ornithine) correlated with the content of cadaverine and putrescine.

INTRODUCTION

Biogenic amines in foods are of concern in relation to both food spoilage and food safety. They are produced by specific amino acid decarboxylases which either originate from the raw material or the growth of decarboxylase positive microorganisms under favourable conditions. These conditions include the particular substrate amino acid which might be present in free form or produced by the action of proteinases. The application of starter cultures can affect the production of biogenic amines, either directly or indirectly through interaction with the association/contamination flora (ten BRINK et al., 1990). In general investigations to date have suggested that elevated histamine levels in most food and beverages are due to microbiological contamination, while increases in putrescine, cadaverine and tyramine concentrations are considered to correlate with bacterial starter culture activities (and counts) in particular fermented products (SAYEM EL DAHER et al., 1984). As histamine is not the only amine is harmful to sensitive individuals and since some biogenic amines have synergistic effect (SMITH, 1980/81 and ten BRINK et al., 1990), food producers should optimize the technology and storage conditions used to secure low amine levels in foods. Foods with predictably low levels of specified amines is a goal that needs to be addressed in the future by the food industry. The purpose of this study was to investigate the role of starter culture in the production biogenic amines in pork meat.

MATERIALS AND METHODS

1./ Bacterium: *Lactobacillus jensenii* used as starter culture in Hungarian meat industry.
2./ Media: M.R.S. broth as described by De MAN et al.(1960) formulated as the Oxoid recipe CM 359.
Bacterium was activated at 30°C by repeated transfer in M.R.S. broth allowing 24h between each transfer.
M.R.S. broth was supplemented with 0.1-0.1% lysine, histidine and ornithine for investigation of biogenic amines production by *Lactobacillus jensenii*. Medium was inoculated to give an initial count of ca. cfu/ml and incubated for 6 days.
3./ Meat: Freshly slaughtered pork; Longissimus dorsi of normal pH (5.5-6). The samples were freed of visible fat and meat slices (ca. 5x6x1 cm) were placed into sterile Petri dishes and inoculated with *Lactobacillus jensenii* ($10^5/cm^2$) and incubated at +5°C and +15°C for 12 days.

4./ Determination of biogenic amines
Samples (culture fluid after centrifugation and 0.6 M $HClO_4$ extract of meat) were preseparated on a DOWEX W50x8 resin packed column (4x1 cm I.D.). The analysis was carried out with BIOTRONIK LC 2000 amino acid analyzer using 12x0.4 cm I.D. resin bed, packed with BTC 3118 (11 μm) cation-exchanger resin. Three K-citrate buffers were required to eluate the amines using a gradient of K^+ ion concentration and detection reagent was ninhydrin.
5./ Determination of free amino acids

Free amino acids were determinated from $HClO_4$ extract of samples with BIOTRONIK LC 3000 amino acid analyzer.

RESULTS AND DISCUSSION

As Fig 1. shows, the cadaverine content increased in 0.1% lysine supplemented M.R.S. broth, consequently the *Lactobacillus jensenii* is lysine decarboxylase positive. Histamine concentration didn't increase during growth of *Lactobacillus jensenii* in 0.1% histidine supplemented M.R.S. broth (Fig 2.). Only a few *Lactobacillus* species are known to be histidine decarboxylase positive. Probably *Lactobacillus jensenii* has no histidine decarboxylase enzyme activity, as there is no formation of histamine in the culture medium. Investigating the synthesis of biogenic amine in culture medium (M.R.S.) supplemented with 0.1% ornithine increased putrescine and spermidine concentrations (Fig 3.) were detected. Spermidine formation from ornithine is a result of combined activities of ornithine decarboxylase and propyl-amine transferase. Fig 4. shows biogenic amine content of pork meat inoculated with *Lactobacillus jensenii*, incubated at 5°C. Data show remarkable increase of cadaverine and spermidine content in pork meat during incubation period. These results are in good agreement with the observations of *Lactobacillus jensenii* in culture medium. Similar changes in the amine content of stored pork meat were reported by HALÁSZ et al., (1994), however changes were much more intensive in the presence of the starter culture. Tendency of changes in biogenic amine content are similar at the two investigated incubation temperatures but at the higher temperature concentrations of cadaverine and putrescine were higher (Fig 5.). This is agreement with the higher enzyme activities at elevated temperature. Determination of biogenic amines was carried out parallel with the appropriate precursor amino acids. Results are shown in Fig 6. The free precursor amino acid contents (ornithine and lysine) showed correlation with contents of putrescine and cadaverine.

CONCLUSIONS

Putrescine and cadaverine content increased in meat inoculated with *Lactobacillus jensenii* because it showed ornithine and lysine decarboxylase activity in culture medium (M.R.S.) also. This increasing of putrescine and cadaverine content was much more intensive than during storage of uninoculated meat. These results are supported with decreasing of the appropriate precursor amino acids (ornithine and lysine) in inoculated meat during incubation period. The investigations confirm that amino acid decarboxylase negative starter cultures should be used in the production of fermented meat products.

REFERENCES

ten BRINK, B., DAMINK, C., JOOSTEN, H.M.L.J. and HUIS in't VELD, J.H.J., (1990): Occurrence and formation of biologically active amines in foods. Int. J. Food Microbiol., 11: 73-84.

HALÁSZ, A., BARÁTH, Á., SIMON-SARKADI, L. and HOLZAPFEL, W.H., (1994): Biogenic amines and their production by microorganisms in food. Trends in Food Sci. & Technol., 5 (2): 42-49.

SAYEM EL DAHER, N., SIMARD, R.E., FILLION, J. and ROBERGE, A.G., (1984): Extraction and determination of biogenic amines in ground beef and their relation to microbial quality. Lebensmitt. Wiss. Technol., 17: 2023-2029.

SMITH, T.A., (1980/81): Amines in food. Food Chem., 6: 169-200.