

## **BIOGENIC AMINES IN MEAT AND FERMENTED MEAT PRODUCTS\***

Joanna Stadnik, Zbigniew J. Dolatowski

University of Life Sciences in Lublin

**Abstract.** Recent trends in food quality and safety promote an increasing search for trace compounds that can affect human health. Biogenic amines belong to this group of substances. They can cause distinctive pharmacological, physiological and toxic effects in organisms. Their amounts are usually increasing as a consequence of the use of poor quality raw materials, during controlled or spontaneous microbial fermentation or in the course of food spoilage. The origin of biogenic amines makes them suitable as chemical indicators of the hygienic quality and freshness of some foods being associated to the degree of food fermentation or degradation. The development of appropriate manufacturing technologies to obtain products free or nearly free from biogenic amines is a challenge for the meat industry. This review briefly summarises current knowledge on the biological implications of biogenic amines on human health and collects data on the factors affecting their formation in meat and fermented meat products.

**Key words:** biogenic amines, fermented meat products, food safety

### **BIOGENIC AMINES**

Biogenic amines are organic bases of low molecular weight that possess biological activity. They can be formed and degraded as a result of normal metabolic activity in animals, plants and microorganisms, and are usually produced by the decarboxylation of free amino acids or by amination and transamination of aldehydes and ketones. Removal of the  $\alpha$ -carboxyl group from a precursor amino acid leads to the corresponding biogenic amine. The names of many biogenic amines correspond to the names of their originating amino acids. For example, histidine is decarboxylated to produce histamine,

---

\*The literature review conducted within the framework of a project partially financed by the Ministry of Science and Higher Education. Project No. NN 312 275435.

tryptophane to tryptamine, tyrosine is decarboxylated to produce tyramine, lysine to cadaverine and putrescine can be produced from three amino acids: glutamine, arginine and agmatine [Bodmer et al. 1999, Halász et al. 1994, Maijala et al. 1995].

According to their chemical structure biogenic amines can either be: aliphatic (putrescine, cadaverine, spermine, spermidine), aromatic (tyramine,  $\beta$ -phenylethylamine), heterocyclic (histamine, tryptamine) [Alberto et al. 2002, Bouchereau et al. 2000, Kaniou et al. 2001, Karovičová and Kohajdová 2005, Ōnal 2007, Suzzi and Gardini 2003]. They have also been classified as mono-, di- and polyamines, or on the basis of their synthesis as “natural polyamines” and “biogenic amines” [Bardócz 1995].

## FUNCTIONS AND PHYSIOLOGICAL SIGNIFICANCE OF BIOGENIC AMINES

Biogenically active amines are compounds formed and broken down by usual metabolic processes in the cells of living organisms. In the human, endogenously synthesised biogenic amines fulfil an array of roles in cellular metabolism [Ōnal 2007].

Biogenic amines are sources of nitrogen and precursors for the synthesis of hormones, alkaloids, nucleic acids and proteins. They can also influence the processes in the organism such as the regulation of body temperature, intake of nutrition, control of blood pressure [Bouchereau et al. 2000, Jansen et al. 2003]. Tyramine and histamine also act as hormonal mediators in humans and animals. Psychoactive amines, such as dopamine and serotonin, are neurotransmitters in the central nervous system [Ōnal 2007].

Traditionally cadaverine, putrescine, spermine and spermidine have been classified within the group of biogenic amines. However, particularly due to their specific biological roles in eukaryotic cells they are now becoming set apart as a distinct group-polyamines. They are formed by *de novo* synthesis and play important roles in many human and animal physiological functions [Bardócz 1995]. Lovaas [1991] has demonstrated that polyamines inhibit the oxidation of polyunsaturated fatty acids, and its antioxidative effect is correlated with the number of amine groups in the polyamine. The participation in cell membrane stabilisation and cell proliferation and differentiation, since they are involved in DNA, RNA and protein synthesis, has been of extraordinary interest as polyamines, both formed endogenously and taken from diet, can be observed in rapidly divided cells and tissues such as tumour cells. Control of polyamine content in the diet of cancer patients is therefore very important [Kalač and Krausová 2005, Kalač et al. 2005, Latorre-Moratalla et al. 2010, Ōnal 2007, Santos 1996, Standarová et al. 2008].

As polyamines are taken up preferentially by tissues with high demands, they could be useful for post-operation patients or during wound healing and for growth and development of the neonatal's digestive system. They also seem to be necessary for the maintenance of normal growth and general properties of the adult digestive tract [Kalač and Krausová 2005, Kalač et al. 2005].

## MICROORGANISMS PRODUCING BIOGENIC AMINES

Undesired accumulation of biogenic amines in foods requires the availability of precursors (i.e. amino acids), the presence of microorganisms with amino acid decarboxylases, either derived from environmental contamination or from an added starter culture

and favourable conditions that allow bacterial growth, decarboxylase synthesis and decarboxylase activity [Alberto et al. 2002, Bodmer et al. 1999, Karovičová and Kohajdová 2005, Suzzi and Gardini 2003].

The capability to form biogenic amines is generally considered a strain specific characteristic rather than a species property. It is thus difficult to find precise correlations between biogenic amines contents and microorganism counts [Halász et al. 1994, Standarová et al. 2008, Suzzi and Gardini 2003].

Amine production has been recognised as a defense mechanism of microorganisms against an acidic environment [Karovičová and Kohajdová 2005, Suzzi and Gardini 2003]. Tkachenko et al. [2001] suggested an interesting hypothesis on the physiological role of biogenic amines in microorganisms. Some strains, with amino acid decarboxylase activity, could overcome or reduce the effects of temperature, NaCl, and other biological and chemico-physical factors that induce stress responses in the cells, with the production of some biogenic amines.

Amino acid decarboxylases are enzymes present in many microorganisms which may be either naturally present in food products or may be introduced by contamination before, during or after food processing. They have been found in species of the genera *Bacillus*, *Clostridium*, *Pseudomonas*, *Photobacterium*, as well as in genera of the family *Enterobacteriaceae*, such as *Citrobacter*, *Klebsiella*, *Escherichia*, *Proteus*, *Salmonella* and *Shigella*, and *Micrococcaceae* such as *Staphylococcus*, *Micrococcus* and *Kocuria*. Furthermore, many lactic acid bacteria belonging to the genera *Lactobacillus*, *Enterococcus*, *Carnobacterium*, *Pediococcus*, *Lactococcus* and *Leuconostoc* are able to decarboxylate one or more amino acids [Galgano et al. 2009, Karovičová and Kohajdová 2005, Marino et al. 2000, Suzzi and Gardini 2003]. There is good evidence that in fermented meat products the contaminating microflora rather than the starter cultures is responsible for the generation of increasing biogenic amines level [Bodmer et al. 1999].

## BIOGENIC AMINES IN FOOD

Low concentrations of biogenic amines are a natural characteristic of a number of foodstuffs such as fruits and vegetables, where they are present as natural metabolic products or intermediates. In food and beverages they are formed by enzymes of raw material or are generated by microbial decarboxylation of amino acids during aging and storage [Santos 1996]. Free biogenic amines shape the typical taste of mature foods and are precursors of certain aroma compounds [Moret et al. 2005, Santos 1996]. The most important biogenic amines occurring in foods and beverages are histamine,  $\beta$ -phenylethylamine, tyramine, tryptamine, putrescine, cadaverine, spermine and spermidine [Bodmer et al. 1999, Shalaby 1996, Suzzi and Gardini 2003].

Their concentrations vary extensively not only between different food varieties but also within the varieties themselves [Bodmer et al. 1999]. In particular biogenic amines are produced in foods where high levels of protein are present. During fermentation or spoilage the protein breakdown products, peptides and amino acids, represent precursors for amine formation [Bodmer et al. 1999, Straub et al. 1993, Vinci and Antonelli 2002].

Biogenic amines are present in a wide range of food products including fish and fish products, meat and meat products, dairy products, wine, beer, vegetables, fruits, nuts and chocolate [Bardócz 1995, Bodmer et al. 1999, Santos 1996].

Cheese is the next (after fish) most frequently presented food item associated with biogenic amines poisoning. The first reported case occurred in 1967 in the Netherland and involved Gouda cheese [Stratton et al. 1991].

## BIOGENIC AMINES IN MEAT AND MEAT PRODUCTS

Meat and meat products have repetitively been reported to contain biogenic amines [Bover-Cid et al. 2006, Eerola et al. 1997, Galgano et al. 2009, Hernández-Jover et al. 1997, Kaniou et al. 2001, Maijala et al. 1995, Ruiz-Capillas and Jiménez-Colmenero 2004]. The most prevalent biogenic amines in meat and meat products are tyramine, cadaverine, putrescine, and also histamine [Ruiz-Capillas and Jiménez-Colmenero 2004]. The only amines present at significant levels in fresh meat are spermidine and spermine [Hernández-Jover et al. 1997]. High spermine contents, usually between 20 and 60 mg·kg<sup>-1</sup>, are usual in meat and meat products of warm-blooded animals. Spermidine levels in meat rarely exceed 10 mg·kg<sup>-1</sup>. Thus, an opposite relation between spermidine and spermine contents is typical for foods of animal origin as compared with plant products [Kalač and Krausová 2005].

Some amines such as tyramine, putrescine and cadaverine can be formed during storage of meat [Galgano et al. 2009, Hernández-Jover et al. 1996]. As tyramine concentrations in stored beef was found the highest on the meat surface, that it can be reduced effectively by washing [Kaniou et al. 2001, Paulsen et al. 2006].

Fermented meat products, constitute one of the foods in which considerable amounts of biogenic amines can be found as a consequence of the use of poor quality raw materials, contamination and inappropriate conditions during processing and storage. Additionally, the microorganisms responsible for the fermentation process may contribute to biogenic amines accumulation [Bover-Cid et al. 2006, Latorre-Moratalla et al. 2010]. The nonprotein nitrogen fraction which increases during fermentation includes the presence of free amino acids, precursors of biogenic amines. The major protease activity is derived from endogenous meat enzymes. Proteolysis is favoured by the denaturation of proteins as a consequence of acidity increase, dehydration and action of sodium chloride [Eerola et al. 1997, Suzzi and Gardini 2003].

Fermented products with comparable microbiological profiles may differ in their biogenic amines content, indicating that the production of such compounds depends on a complex interaction of factors [Suzzi and Gardini 2003].

Meat raw material is the natural source of the substrate from which biogenic amines are produced. It also is the largest component of the matrix in which the decarboxylation reactions take place and any conditions that alter its nature and characteristics will influence the formation of biogenic amines [Ruiz-Capillas and Jiménez-Colmenero 2004].

Many authors [Eerola et al. 1997, Komprda et al. 2004, Maijala et al. 1995] observed significant differences in biogenic amines content in fermented meat products depending on raw material hygienic quality. However, the same raw material can lead to very different amine levels in final products depending on the presence of decarboxylating microorganisms, either derived from environmental contamination or from starter cultures, and the conditions supporting their growth and activity.

Amines content and profiles may vary depending on various extrinsic and intrinsic factors during the manufacturing process, such as pH, redox potential, temperature, NaCl,

the size of the sausage, hygienic conditions of manufacturing practices, and effect of starter cultures [Gardini et al. 2001, Komprda et al. 2004, Latorre-Moratalla et al. 2008].

pH is a key factor influencing the amino acid decarboxylase activity. Bacterial amino acid decarboxylases usually have acid pH optimum. Decrease of pH results in increasing decarboxylase activity of bacteria. In these conditions bacteria produce more decarboxylases as part of their protective mechanism. However, rapid and sharp reduction in pH is known to reduce the growth of the amine-positive microorganisms [Majjala et al. 1995].

The redox potential of the medium also influences biogenic amines production. Conditions resulting in a reduced redox potential stimulate histamine production, and histidine decarboxylase activity seems to be inactivated or destroyed in the presence of oxygen [Karovičová and Kohajdová 2005].

Amine formation by bacteria is decisively influenced by temperature. Temperature between 20°C and 37°C is optimal for the growth of the most bacteria containing decarboxylases, decreased temperature stops their growth [Karovičová and Kohajdová 2005].

Relatively less attention is devoted to the effect of the chemical substances added during manufacture of the fermented meat products. Despite the known antimicrobial properties of some spices, the direct evaluation of an effect of spices on microflora in relationship with biogenic amines production is not present in available literature [Komprda et al. 2004]. According to Suzzi and Gardini [2003] biogenic amines accumulation decreases markedly with the increase of NaCl concentration, while proteolytic activity is higher for intermediate concentration of salt, pointing out that there is not necessarily a correlation between these two variables. Karovičová and Kohajdová [2005] reported that presence of sodium chloride activates tyrosine decarboxylase activity and inhibits histidine decarboxylase activity.

A relationship was also found between biogenic amines content and the size of dry fermented sausages. The diameter of the sausage affects the environment in which microorganisms grow; for example, salt concentration is usually lower and water activity is higher in sausages with a larger diameter. A larger diameter may be one of the reasons for a higher production of certain amines, such as tyramine and putrescine. Generally, biogenic amines levels in the bigger diameter sausages were higher than in the thinner sausages and in the central part of the sausages than in the edge [Ruiz-Capillas and Jiménez-Colmenero 2004, Suzzi and Gardini 2003].

The importance of using measures focused on the hygienic quality of both raw material and processing units to avoid the development of aminogenic contaminant bacteria and in turn, to reduce biogenic amines content, is well known. However, proper hygiene may not be enough to avoid some biogenic amines formation and other technological measures must be applied [Latorre-Moratalla et al. 2010].

## **THE ROLE OF STARTER CULTURES IN BIOGENIC AMINES FORMATION**

The choice of suitable starter culture with aminooxidase activity is fundamental in preventing the formation of high levels of biogenic amines in fermented meat products [Karovičová and Kohajdová 2005, Suzzi and Gardini 2003].

The selection of lactic acid bacteria with application in meat fermentation has to take in consideration the various, specific requirements of the fermentation process [Roig-

-Sagués and Eerola 1997]. The inability of the culture to form biogenic amines but also its ability to grow well at the temperature intended for processing of the product and competitiveness in suppressing the growth of wild amine producing microflora should be taken into consideration in the selection of starter cultures [Suzzi and Gardini 2003]. The formation by the strains of bacteriocin (for example curvacin A) can increase their competitiveness [Hammes and Hertel 1996]. A rapid pH decrease caused by amine negative starter cultures can largely prevent biogenic amines accumulation in fermented meat products. Moreover, starter cultures able to compete with nonstarter bacteria during the later phase of ripening and throughout storage can further avoid excessive biogenic amines production [Suzzi and Gardini 2003].

Some authors cited by Lu et al. [2010] and González-Fernández et al. [2003] held that the use of starter cultures was not sufficient to avoid the presence of biogenic amines in some fermented meat products. However, other works cited by the same authors have indicated that addition of negative amine-producer starter culture to carry out a controlled fermentation could be an advisable practice to prevent excessive amine accumulation. This fact could be related with the lack of effectiveness of the starter culture depending on the hygienic quality of the raw material used.

Lactic acid bacteria, responsible for the acidification process together with micrococci and/or coagulase-negative staphylococci contribute to color formation and aroma development as a result of their proteolytic and lipolytic activities are the microorganisms widely used in the meat industry as starter cultures [Latorre-Moratalla et al. 2010, Suzzi and Gardini 2003].

In Europe strains of *Lactobacillus curvatus* and *Lactobacillus sake* are usually used. However, some strains of *Lactobacillus curvatus* used as starter cultures form up to four different biogenic amines and should not be used. Strains of *Lactobacillus sake* are free of this potential [Kołozyn-Krajewska and Dolatowski 2009, Vinci and Antonelli 2002]. This indicates that *Lactobacillus sake* may be more suitable than *Lactobacillus curvatus* for use as a starter culture to prevent the formation of biogenic amines [Roig-Sagués and Eerola 1997].

## DETOXIFICATION OF BIOGENIC AMINES

Under normal conditions, during the food intake process in the human gastrointestinal system, low amounts of exogenous biogenic amines absorbed are metabolized to physiologically less active degradation products. This detoxification system providing protection from small amounts of biogenic amines normally presented by foods includes specific enzymes: monoamine oxidase (MAO, EC 1.4.3.4), diamine oxidase (DAO, EC 1.4.3.6) and polyamine oxidase (PAO, EC 1.5.3.11). However, in the case of allergic individuals or upon intake of high loads of biogenic amines in foods, the detoxification system is unable to eliminate these biogenic amines sufficiently. If detoxification is inefficient, biogenic amines are readily absorbed and get into systemic circulation, leading to toxic effects [Bardócz 1995, Bodmer et al. 1999, McCabe-Sellers et al. 2006, Önal 2007, Standarová et al. 2008].

Individuals with respiratory and coronary problems or those with hypertension or vitamin B<sub>12</sub> deficiency are sensitive to lower doses of biogenic amines [Bardócz 1995]. People with gastrointestinal problems (gastritis, irritable bowel syndrome, Crohn's

disease, stomach and colonic ulcers) are also at risk because the activity of oxidases in their intestines is usually lower than that in healthy individuals. Patients, who are taking medicines with inhibiting effect to MAO, DAO and/or PAO such as painkillers, psychopharmaceutics, and drugs used for the treatment of Alzheimer's and Parkinson's diseases might have a changed metabolism of biogenic amines, which can cause healthy problem [Bouchereau et al. 2000, Hernández-Jover et al. 1997, Kalač and Krausová 2005, Latorre-Moratalla et al. 2008, Moret et al. 2005]. Other compounds, like alcohol and acetaldehyde, also can augment the toxic potential of biogenic amines, since they promote the transportation of these through the intestinal wall [Ruiz-Capillas and Jiménez-Colmenero 2004]. The toxic potential of these dietary amines is even more alarming if we consider that approximately 20% of the European population regularly consumes MAOI drugs as antidepressants, which inhibit aminooxidase activity [Ruiz-Capillas and Jiménez-Colmenero 2004].

Polyamines, such as putrescine, cadaverine, spermidine and spermine, although not exerting a direct toxic effect, inhibit histamine or tyramine detoxifying enzymes and thus act as potentiators of their toxicity. These amines in the intestinal tract compete for the detoxifying enzymes that tend to increase the level of histamine and tyramine in blood [Al Bulushi et al. 2009, Bodmer et al. 1999, Eerola et al. 1997, Kaniou et al. 2001, Karovičová and Kohajdová 2005, Majjala et al. 1995, Moret et al. 2005, Önal 2007, Standarová et al. 2008].

## TOXICOLOGY OF BIOGENIC AMINES

Due to their psychoactive and vasoactive properties, some biogenic amines introduced with the diet can cause distinctive pharmacological and toxic effects [Hernández-Jover et al. 1997].

The most conspicuous symptoms of consumption of high doses of biogenic amines are vomiting, respiratory difficulties, perspiration, palpitation, hypo- or hypertension and migraine [Kordiovská et al. 2006].

Psychoactive amines, such as histamine, putrescine and cadaverine can cause some neurotransmission disorders due to their action as false neurotransmitters. Some aromatic amines (tyramine, tryptamine, and  $\beta$ -phenylethylamine) show a vasoconstrictor action while others (histamine and serotonin) present a vasodilatador effect in blood vessels, capillaries and arteries, causing headaches, hypotension, flushing, gastrointestinal distress and oedemas [González-Fernández et al. 2003, Komprda et al. 2004, McCabe-Sellers et al. 2006, Önal 2007, Standarová et al. 2008, Vinci and Antonelli 2002].

The vasoconstrictor amines, largely tyramine, have been proposed as the initiators of hypertensive crisis in certain patients and of dietary-induced migraine. The physiological effects of tyramine include: peripheral vasoconstriction, increased cardiac output, increased respiration, elevated blood glucose, and release of norepinephrine [McCabe-Sellers et al. 2006, Önal 2007]. Tyramine may also induce brain haemorrhage and heart failure [Standarová et al. 2008].

Adverse reactions as a result of biogenic amines are often classified as intolerance reactions, also called pseudoallergy or false food allergy. Intolerance is a form of hypersensitivity, which is not mediated by the immune system, in contrast to allergic reactions [Jansen et al. 2003].

The most frequent foodborne intoxications caused by biogenic amines involve histamine, which is also referred to as scombroid fish poisoning because this illness is usually related to the consumption of scombroid fish, such as tuna, mackerel, and sardines [Santos 1996].

High histamine consumption causes life threatening intoxication, lower amounts can lead to headache, nausea, hot flushes, skin rashes, sweating, respiratory distress, cardiac and intestinal problems. Another phenomenon is the hypertensive crises called “cheese reaction”, following consumption of food rich in tyramine such as aged cheese, wine, beer and yeast extracts but also vegetables such as sauerkraut, broad bean, banana peel and avocado [Alberto et al. 2002, Bodmer et al. 1999, Hernández-Jover et al. 1997, Karovičová and Kohajdová 2005, Ōnal 2007, Ruiz-Capillas and Jiménez-Colmenero 2004].

As far as possible negative aspects of the consumption of meat and meat products are concerned, growing attention is devoted to the polyamines which have no adverse health effect, but have been described as potential precursors of stable carcinogenic *N*-nitrosamines and to enhance the growth of chemically induced aberrant crypt foci in the intestine. Formation of *N*-nitroso compounds, constitutes an additional toxicological risk associated to biogenic amines, especially in meat products that contain nitrite and nitrate salts as curing agents [Karovičová and Kohajdová 2005, Komprda et al. 2004, Ōnal 2007, Santos 1996].

The reaction of nitrosating agents with primary amines produces short-lived alkylating agents that react with other components in the food matrix to generate products (mainly alcohols) devoid of toxic activity in the relevant contents. Secondary amines are known to form carcinogenic *N*-nitrosamines by reaction with nitrosating compounds, while tertiary amines produce a range of labile *N*-nitroso products. However, as primary biogenic amines can convert to secondary amines, not only on heating but also during storage at room temperature, and further reaction with nitrite can occur. In fatty foods, such as bacon, at high temperature and in the presence of water, the carcinogen *N*-nitrosopyrrolidine and *N*-nitrosopiperidine can be formed from secondary amines such as putrescine or spermidine [Eerola et al. 1997, González-Fernández et al. 2003, Karovičová and Kohajdová 2005, Ōnal 2007, Santos 1996].

## LEGAL LIMITS

Although the toxicity of biogenic amines is beyond all doubt, determination of the exact toxicity threshold of biogenic amines in a given food product is extremely difficult, because their effect does not depend on their presence alone, but is also influenced by other compounds and by the specific efficiency of the detoxifying mechanisms in different individuals. Hence, the toxicity of biogenic amines will depend on factors associated with the food itself (quantitative and qualitative) and also on factors associated with the consumer (individual susceptibility and state of health) [Eerola et al. 1997, Gardini et al. 2001, Halász et al. 1994, Ruiz-Capillas and Jiménez-Colmenero 2004].

Histamine is the amine most studied with regard to its toxicological effects. An intake of 5-10 mg of histamine can be considered as defecting to some sensitive people, 10 mg is considered as tolerable limit, 100 mg induce a medium toxicity and 1000 mg is highly toxic [Karovičová and Kohajdová 2005, Santos 1996]. Other authors [Eerola et

al. 1997, Gardini et al. 2001, Hernández-Jover et al. 1997, Santos 1996] suggested following toxic limits of histamine: 8-40 mg causes slight poisoning, 40-100 mg intermediate poisoning and over 100 mg can cause intensive poisoning. On the basis of data from food intoxication outbreaks a legal upper limit of 100 mg histamine·kg<sup>-1</sup> food and 2 mg·l<sup>-1</sup> of ethanol has been suggested.

The European Union has established regulations according to which histamine level should be below 100 mg·kg<sup>-1</sup> in raw fish, and below 200 mg·kg<sup>-1</sup> in salted fish for species belonging to the Scombridae and Clupeidae families [Karovičová and Kohajdová 2005].

Less is known about the toxic doses of other amines. The recommended maximum level of tyramine has been proposed variously to be in the range of 100-800 mg·kg<sup>-1</sup> of food. Value of 30 mg·kg<sup>-1</sup> for β-phenylethylamine has been reported as toxic dose in food [Gardini et al. 2001].

## ANALYTICAL METHODS FOR DETERMINATION OF BIOGENIC AMINES

There are two reasons for the determination of amines in foods: the first is their potential toxicity; the second is the possibility of using them as food quality markers [Önal 2007].

The main drawbacks in the analysis of biogenic amines in food are the complexity of matrix sample (e.g. presence of free amino acids), the presence of potentially interfering compounds, the low concentration levels at which the compounds are present in the samples and the occurrence of several biogenic amines simultaneously [Alberto et al. 2002, Karovičová and Kohajdová 2005, Önal 2007, Shalaby 1996].

For separation and quantitative determination of biogenic amines in foods several methods have been developed [Alberto et al. 2002, Karovičová and Kohajdová 2005, Kovács et al. 1999]. The extraction of amines represents the critical step of the process and it affects negatively the analytical recoveries. After extraction, the determination of biogenic amines is most commonly performed by means of chromatographic methods: gas chromatography, thin-layer chromatography, and high-performance liquid chromatography (HPLC). Biochemical assays and capillary electrophoresis have also been described [Önal 2007].

Detection by means of UV absorbance is only possible for the heterocyclic and aromatic amines. As most aliphatic amines show neither natural UV absorption nor fluorescence, most methods require that amines should be derivatised before detection, or indirect detection can be used [Kovács et al. 1999, Krischbaum et al. 1997, Straub et al. 1993].

## BIOGENIC AMINES AS A QUALITY INDEX

The concentrations of some biogenic amines (tyramine, putrescine, and cadaverine) normally increase during the processing and storage of meat and meat products, whereas others (spermidine and spermine) decrease or remain constant [Bardócz 1995, Halász et al. 1994, Ruiz-Capillas and Jiménez-Colmenero 2004]. Therefore their amounts and ratios have been proposed as an index of the hygienic conditions of raw

material and/or manufacturing practices since their amount increase during microbial fermentation or spoilage [Alberto et al. 2002, Latorre-Moratalla et al. 2008, McCabe-Sellers et al. 2006].

The usefulness of biogenic amines as a quality index will depend on the nature of the product; results tend to be more satisfactory in fresh meat and heat-treated products than in fermented products. In the latter, they seem to be of very limited use [Ruiz-Capillas and Jiménez-Colmenero 2004].

The so-called "biogenic amine index" (BAI) introduced by Mietz and Karmas [1977] for fish and seafood was calculated according to the equation:

$$\text{BAI} = \frac{c\text{Him} + c\text{Put} + c\text{Cad}}{1 + c\text{Spd} + c\text{Spm}}$$

with Him – histamine, Put – putrescine, Cad – cadaverine, Spd – spermidine, Spm – spermine (c – concentration,  $\text{mg}\cdot\text{kg}^{-1}$ ).

Meat freshness should be evaluated by considering an amine index, which includes all the biogenic amines related to meat spoilage. As tyramine increases considerably during meat storage, this biogenic amine should also be included in a BAI. This is the case of the BAI of putrescine + cadaverine + histamine + tyramine, proposed by Wortberg and Woller [1982] and Hernández-Jover et al. [1996]. Wortberg and Woller [1982] established  $500 \text{ mg}\cdot\text{kg}^{-1}$  as the limit for Bologna sausage, minced beef and pork. Hernández-Jover et al. [1996] suggested the following limits: BAI  $<5 \text{ mg}\cdot\text{kg}^{-1}$  for good quality fresh meat; between 5 and  $20 \text{ mg}\cdot\text{kg}^{-1}$  for acceptable meat, but with initial spoilage signs; between 20 and  $50 \text{ mg}\cdot\text{kg}^{-1}$  for low meat quality; finally,  $>50 \text{ mg}\cdot\text{kg}^{-1}$  for spoiled meat.

Not all spoilage or starter microorganisms can decarboxylate free amino acids. Even within the same species, not all strains develop the same decarboxylating capacity, so that a low biogenic amine concentration does not always signal good microbiological quality. There is, therefore, no simple matter to establish a biogenic amine index that reliably predicts quality for products of this kind [Ruiz-Capillas and Jiménez-Colmenero 2004].

## SUMMARY

The biogenic amines content of various foods has been widely studied because of their potential toxicity. Any foodstuffs produced by fermentation or exposed to microbial contamination during processing or storage may contain biogenic amines. Monitoring of biogenic amines levels in fresh and processed foods and beverages is of great interest not only for their toxicological risk, but also because in non-fermented food these compounds can be a useful indicator of undesired microbial activity.

Today, consumers increasingly prefer high-quality products that are minimally processed, safe, etc., and the meat industry is, therefore, looking for emerging technologies that can achieve this in processing and storage. There is a clear interest in the study of toxicity of biogenic amines and the factors determining their formation in the context of meat processing conditions and preservation.

There are a few common parameters which can be defined for low-biogenic amines technology. First, high quality raw materials, free of biogenic amines, must be used. Additionally, proper and careful treatment of raw materials until processing is abso-

lutely crucial. Growth and activity of decarboxylase-positive microorganisms must be avoided, and activity of endogenous proteases and amino acid decarboxylases have to be inhibited by technological measures and proper and hygienic production conditions. If microbial transformations are a part of the production process, use of carefully selected, decarboxylase negative starter cultures for these fermentations is important, as well as processing conditions which favour the growth of the starter strains.

## REFERENCES

- Al Bulushi I., Poole S., Deeth H.C., Dykes G.A., 2009. Biogenic amines in fish: Roles in intoxication, spoilage, and nitrosamine formation—a review. *Crit. Rev. Food Sci.* 49, 369-377.
- Alberto M.R., Arena M.E., Manca de Nadra M.C., 2002. A comparative survey of two analytical methods for identification and quantification of biogenic amines. *Food Control* 13, 125-129.
- Bardócz S., 1995. Polyamines in food and their consequences for food quality and human health. *Trends Food Sci. Tech.* 6, 341-346.
- Bodmer S., Imark C., Kneubühl M., 1999. Biogenic amines in foods: Histamine and food processing. *Inflamm. Res.* 48, 296-300.
- Bouchereau A., Guénot P., Larher F., 2000. Analysis of amines in plant materials. *J. Chromatogr. B*, 747, 49-67.
- Bover-Cid S., Miguelez-Arrizado M.J., Latorre Moratalla L.L., Vidal Carou M.C., 2006. Freezing of meat raw materials affects tyramine and diamine accumulation in spontaneously fermented sausages. *Meat Sci.* 72, 62-68.
- Eerola S., Roig Sagués A.-X., Lilleberg L., Aalto H., 1997. Biogenic amines in dry sausages during shelf-life storage. *Z. Lebensm. Unters. Forsch. A*, 205, 351-355.
- Galgano F., Favati F., Bonadio M., Lorusso V., Romano P., 2009. Role of biogenic amines as index of freshness in beef meat packed with different biopolymeric materials. *Food Res. Int.* 42, 1147-1152.
- Gardini F., Martuscelli M., Caruso M.C., Galgano F., Crudele M.A., Favati F., Guerzoni M.E., Suzzi G., 2001. Effects of pH, temperature and NaCl concentration on the growth kinetics, proteolytic activity and biogenic amine production of *Enterococcus faecalis*. *Int. J. Food Microbiol.* 64, 105-117.
- González-Fernández C., Santos E.M., Jaime I., Rovira J., 2003. Influence of starter cultures and sugar concentrations on biogenic amine contents in *chorizo* dry sausage. *Food Microbiol.* 20, 275-284.
- Halász A., Barath A., Simon-Sarkadi L., Holzapfel W., 1994. Biogenic amines and their production by micro-organisms in food. *Trends Food Sci. Tech.* 5, 42-49.
- Hammes W.P., Hertel C., 1996. Selection and improvement of lactic acid bacteria used in meat and sausage fermentation. *Lait* 76, 159-168.
- Hernández-Jover T., Izquierdo-Pulido M., Veciana-Nogués M.T., Mariné-Font A., Vidal-Carou M.C., 1997. Biogenic amine and polyamine contents in meat and meat products. *J. Agric. Food Chem.* 45, 2098-2102.
- Hernández-Jover T., Izquierdo-Pulido M., Veciana-Nogués M.T., Vidal-Crou M.C., 1996. Biogenic amine sources in cooked cured shoulder pork. *J. Agric. Food Chem.* 44, 3097-3101.
- Jansen S.C., van Dusseldorp M., Bottema K.C., Dubois A.E.J., 2003. Intolerance to dietary biogenic amines: a review. *Ann. Allerg. Asthma Im.* 91, 233-241.
- Kalač P., Krausová P., 2005. A review of dietary polyamines: Formation, implications for growth and health and occurrence in foods. *Food Chem.* 90, 219-230.
- Kalač P., Křížek M., Pelikánová T., Langová M., Veškrna O., 2005. Contents of polyamines in selected foods. *Food Chem.* 90, 561-564.

- Kaniou I., Samouris G., Mouratidou T., Eleftheriadou A., Zantopoulos N., 2001. Determination of biogenic amines in fresh unpacked and vacuum-packed beef during storage at 4°C. *Food Chem.* 74, 515-519.
- Karovičová J., Kohajdová Z., 2005. Biogenic amines in food. *Chem. Pap.* 59 (1), 70-79.
- Kołożyn-Krajewska D., Dolatowski Z.J., 2009. Probiotics in fermented meat products. *Acta Sci. Pol., Technol. Aliment.* 8 (2), 61-74.
- Komprda T., Smělá D., Pechová P., Kalhotka L., Štencl J., Klejdus B., 2004. Effect of starter culture, spice mix and storage time and temperature on biogenic amine content of dry fermented sausages. *Meat Sci.* 67, 607-616.
- Kordiovská P., Vorlová L., Borkovcová I., Karpišková R., Buchtová H., Svobodová Z., Křížek M., Vácha F., 2006. The dynamics of biogenic amine formation in muscle tissue of carp (*Cyprinus carpio*). *Czech J. Anim. Sci.* 51, 262-270.
- Kovács Á., Simon-Sarkadi L., Ganzler K., 1999. Determination of biogenic amines by capillary electrophoresis. *J. Chromatogr. A*, 836, 305-313.
- Krischbaum J., Busch I., Brückner H., 1997. Determination of biogenic amines in food by automated pre-column derivatization with 2-naphthylloxycarbonyl chloride (NOC-Cl). *Chromatographia* 45, 263-268.
- Latorre-Moratalla M.L., Bover-Cid S., Talon R., Garriga M., Zanardi E., Ianieri A., Fraqueza M.J., Elias M., Drosinos E.H., Vidal-Carou M.C., 2010. Strategies to reduce biogenic amine accumulation in traditional sausage manufacturing. *LWT-Food Sci. Technol.* 43, 20-25.
- Latorre-Moratalla M.L., Veciana-Nogués T., Bover-Cid S., Garriga M., Aymerich T., Zanardi E., Ianieri A., Fraqueza M.J., Patarata L., Drosinos E.H., Lauková A., Talon R., Vidal-Carou M.C., 2008. Biogenic amines in traditional fermented sausages produced in selected European countries. *Food Chem.* 107, 912-921.
- Lovaas E., 1991. Antioxidative effects of polyamines. *J. Am. Oil Chem. Soc.* 68, 353-358.
- Lu S., Xu X., Zhou G., Zhu Z., Meng Y., Sun Y., 2010. Effect of starter cultures on microbial ecosystem and biogenic amines in fermented sausage. *Food Control* 21, 444-449.
- Maijala R., Nurmi E., Fischer A., 1995. Influence of processing temperature on the formation of biogenic amines in dry sausages. *Meat Sci.* 39, 9-22.
- Marino M., Maifreni M., Moret S., Rondinini G., 2000. The capacity of *Enterobacteriaceae* species to produce biogenic amines in cheese. *Lett. Appl. Microbiol.* 31, 169-173.
- McCabe-Sellers B.J., Staggs C.G., Bogle M.L., 2006. Tyramine in foods and monoamine oxidase inhibitor drugs: A crossroad where medicine, nutrition, pharmacy, and food industry converge. *J. Food Compos. Anal.* 19, S58-S65.
- Mietz J.L., Karmas E., 1977. Chemical quality index of canned tuna as determined by high-pressure liquid chromatography. *J. Food Sci.* 42 (1), 155-158.
- Moret S., Smela D., Populin T., Conte L.S., 2005. A survey on free biogenic amine content of fresh and preserved vegetables. *Food Chem.* 89, 355-361.
- Önal A., 2007. A review: Current analytical methods for the determination of biogenic amines in foods. *Food Chem.* 103, 1475-1486.
- Paulsen P., Hagen U., Bauer F., 2006. Changes in biogenic amine contents, non-protein nitrogen and crude protein during curing and thermal processing of *M. longissimus, pars lumborum* of pork. *Eur. Food Res. Technol.* 223, 603-608.
- Roig-Sagués A., Eerola S., 1997. Biogenic amines in meat inoculated with *Lactobacillus sake* starter strains and an amine-positive lactic acid bacterium. *Z. Lebensm. Unters. Forsch. A*, 205, 227-231.
- Ruiz-Capillas C., Jiménez-Colmenero F., 2004. Biogenic amines in meat and meat products. *Crit. Rev. Food Sci.* 44, 489-499.
- Santos S.M.H., 1996. Biogenic amines: their importance in foods. *Int. J. Food Microbiol.* 29, 213-231.
- Shalaby A.R., 1996. Significance of biogenic amines to food safety and human health. *Food Res. Int.* 29 (7), 675-690.

- Standarová E., Borkovcová I., Vorlová L., 2008. The occurrence of biogenic amines in dairy products on the Czech market. *Acta Sci. Pol., Med. Veter.* 7 (4), 35-42.
- Stratton J.E., Hutkins R.W., Taylor S.L., 1991. Biogenic amines in cheese and other fermented foods: a review. *J. Food Protect.* 54, 460-470.
- Straub B., Schollenberger M., Kicherer M., Luckas B., Hammes W.P., 1993. Extraction and determination of biogenic amines in fermented sausages and other meat products using reversed-phase-HPLC. *Z. Lebensm. Unters. Forsch.* 197, 230-232.
- Suzzi G., Gardini F., 2003. Biogenic amines in dry fermented sausages: a review. *Int. J. Food Microbiol.* 88, 41-54.
- Tkachenko A.G., Nesterova L.Y., Pshenichnov K., 2001. Role of putrescine in the regulation of the expression of the oxidative stress defence genes of *Escherichia coli*. *Microbiology* 70, 133-137.
- Vinci G., Antonelli M.L., 2002. Biogenic amines: quality index of freshness in red and white meat. *Food Control* 13, 519-524.
- Wortberg B., Woller R., 1982. Quality and freshness of meat and meat products as related to their content of biogenic amines. *Fleischwirtschaft* 62, 1457-1463.

## AMINY BIOGENNE W MIĘSIE I FERMENTOWANYCH PRODUKTACH MIĘSNYCH

**Streszczenie.** Obecne tendencje w zapewnianiu jakości i bezpieczeństwa żywności koncentrują się na poszukiwaniu substancji śladowych, których obecność w żywności może niekorzystnie oddziaływać na zdrowie człowieka. Do substancji tych należą aminy biogenne. Ich spożycie z żywnością może mieć w organizmie charakterystyczne skutki farmakologiczne, fizjologiczne i toksyczne. Zawartość amin biogennych jako metabolitów bakteryjnych wzrasta wskutek złej jakości higienicznej użytego surowca, w czasie kontrolowanej bądź spontanicznej fermentacji oraz w wyniku zepsucia żywności. Ze względu na mechanizm powstawania, stężenie amin biogennych może być wskaźnikiem jakości higienicznej i świeżości wynikającej ze stopnia zaawansowania procesów fermentacyjnych lub rozkładczych produktów spożywczych. Wyzwaniem dla przemysłu mięsnego jest opracowanie technologii produkcji służących minimalizacji zawartości amin biogennych w wyrobach mięsnych. Celem opracowania było podsumowanie aktualnego stanu wiedzy o wpływie amin biogennych na zdrowie człowieka oraz czynnikach warunkujących ich tworzenie w mięsie i fermentowanych produktach mięsnych.

**Słowa kluczowe:** aminy biogenne, fermentowane produkty mięsne, bezpieczeństwo żywności

*Accepted for print – Zaakceptowano do druku: 5.07.2010*

*For citation – Do cytowania: Stadnik J., Dolatowski Z.J., 2010. Biogenic amines in meat and fermented meat products. Acta Sci. Pol., Technol. Aliment. 9(3), 251-263.*