THE ROLE OF BEZOIC ACID AS A FUNGAL DECONTAMINANTE OF BEEF CARCASSES

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SUMMARY

20 genera appertainaing 36 species and 1 species variety were found to be associated with beef carcasses of which 9 genera representing to 14 species and 1 species variety were completely disappeared by 0.7% benzoic acid treatment. Benzoic acid had an inhibitory effect on fungal counts such as *Acremonium strictum*, *Alternaria alternata*, *Aspergillus flavus* and *A. niger* from 16, 11, 642 and 384 to 9, 3, 515 and 263 respectively. In the same time, it had a complete inhibitory effect on *Alternaria tenuissima*, *A. flavus var columnaris*, *A. melleus*, *A. oryzae*, *Eurotium niveoglaucum*, *Fennellia flavipes*, *Gibberella intricans*, *Isaria* sp., *Malbranchea* sp., *Microascus cinereus*, *Nigrospora sacchari*, *Penicillium pinophilum*, *Pleospora herbarum*, *Rhizopus stolonifer* and *Torula herbarum*, however these fungi were recovered rarely before treatment. Some fungi such as *Cladosporium*, *Emericella* and *Penicillium* were resistant. The influence of benzoic acid on yeast was noted on *Candida albicans*. From these results, the use of benzoic acid to improve the quality of beef carcasses was recommended.

Introduction

The mycological status of carcass meats is highly dependant on the conditions which the animals are reared, slaughtered and processed. Fresh meat represents an environment which is ideal for the growth of many mycoflora; thus contamination can early result in spoilage or hazard to the health of the consumer. Meat hygiene aims to reduce contamination and pevent the proliferation of harmful mycoflora.

The mould affections of cattle carcasses in slaughter houses were studied by many investigators (Refai and Lott, 1969; Kotula et al. 1975; Hadlock et al. 1976; EL-Daly et al. 1988; Mansour et al. 1990, Yassien et al. 1990, Mohmmed, 1993 ; Refai et al. 1993) but nothing revealed any trial to inhibit or reduce such contamination.

Benzoic acid is the oldest food preservative in use as antimycotic agent and permitted by Food and Drug Adminstration. It is the most effective in controlling yeasts and moulds and
least in bacteria (Chistner and Tanner, 1972, Lück, 1986) while Jermini and Schmidt-Lorenze (1987) postulated that benzoate inhibits yeasts more than mould or bacteria.

The aim of the present work was to evaluate the mycological status of beef carcasses in a slaughter house and try to reduce or inhibit such contamination.

Materials and Methods

160 swabs were taken from beef carcasses at Assiut slaughter house; out of which 80 to evaluate the mycological status (before spraying) and 80 after spraying the same area swabed before by 0.7% benzoic acid for 3-5 minutes.

The swabs were transfered directly to the laboratory and cultured on dichloran rose-bengal medium according to King et al. (1979) at 25 °C for 7-10 days. The grown moulds were counted, identified macroscopically and microscopically according to Pitt, 1979, Domsch et al., 1980, Samson and Van Reenen-Hoekstra; 1988 and Kozazakiewciz, 1989.

Results and Discussion

Fungi associated with beef carcasses:

Thirty fungal species in addition to one species variety representing 20 genera were found to be associated with beef carcasses. *Aspregillus* was the most fungus contaminating 65.3% of the samples tested and accounted about 90% of the total isolates obtained. Of the eight *Aspergillus* species and one variety isolated, *A. flavus* (27.5% of the samples and 4.7% of total isolates) and *A. niger* (46.3% and 32%) were the most prevalent while other aspergilli were rarely encountered (Table 1). *A. niger* was found to be one of the most common moulds on the surface of beef carcasses (EL-Daly et al. 1988, Mansour et al. 1990). Also members of the *Aspergillus* such as *A. candidus*, *A. flavus*, *A. fumigatus* and *A. niger* were reported to be prevalent on fresh meat (Yassien et al. 1990, Mohamed, 1993).

Acremonium (represented by one species and recovered in 17.5% of the samples and 1.36% of total isolates), *Alternaria* (2 species, 7.5% and 1.02%), *Cladosporium* (2 species 22.5% and 2.56%), *Nectria* (1 species, 11.3% and 1.45%) and *Penicillium* (7 species, 18.8% and 1.88%) following *Aspergillus* in their frequencies and counts (Table 1). All the obove genera were also isolated from beef carcasses but with various incidences by EL-Daly et al. (1988), Mansour et al. (1990), Yassien et al. (1990) and Mohamed (1993). Other group of fungi were encountered in low frequencies and listed in table (1).

Effect of benzoic acid on the fungi associated with beef cacasses:

From the results shown in table (1), it could be noted that benzoic acid affected, to some extent, on the total counts as well as the compositions of the fungi, due to its inhibitory effect on some fungal species such as *Acremonium strictum*, *Alternaria alternata*, *Aspergillus flavus*, *A. niger* and *Nectria haematococca*. *A. flavus* that was inhibited by about 10.8% in the present study , is well known as a mycotoxin producer. Fortunately, it was previously reported that growth and aflatoxin production was reduced by benzoate treatment (Uraih and Chipley, 1976, Uraih et al. 1977, Masimango et al. 1979). Bandelin (1958) also found *A. niger* and other species belonging to *Alternaria* and *Penicillium* to be inhibited by 0.15% benzoate. However, some fungi were promoted by benzoic acid treatment such as *A. terreus*, *Cladosporium cladosporoides*, *C. sphaerospermum*, *Penicillium chrysogenum* and *P. islandicum*. Other group of fungi, that were mostly encountered rarely were not recovered after treatment with benzoic acid: *Alternaria tenuissima*, *Aspergillus flavus* var. *columnaris*, *A. melleus*, *A. oryzae*, *Eurotium niveoglaucum*, *Fennellia flavipes*, *Gibberella inricans*, *Isaria* sp., *Malbranchea* sp., *Microascus cinereus*, *Nigrospora sacchari*, *Penicillium pinophilum*, *Pleospora herbarum*, *Rhizopus stolonifer* and *torula herbarum*.

Yeast associated with beef carcasses:

Three species (*Candida albicans*, *Geotrichum candidum* and *Trichosporon cuteanum*) in addition to some unidentified yeasts species were recovered. *C. albicans* was the most frequent yeast (recovered in 27.5% of the samples and accounted 20.2% of the total yeasts). These yeasts were also reported from meat products but with different incidences (Abdel-Rahman et al. 1984).

Effect of benzoic acid on yeast associated with beef carcasses:

Benzoic acid at o.7% reduced the count of yeasts by about 7%. *Candida albicans* count was reduced while *Trichosporon cuteanum* count was promoted (Table 2). The obtained results (Tables 1&2) indicated that benzoic acid as an antimicrobial agent is more effective on fungi followed by yeasts while bacteria not be affected.

In conclusions: The contamination of meat with fungi, yeasts and bacteria may lead to spoilage and render it to inferior quality for consumption (Mossel, 1977). The incidence of such microorganisms in meat indicates bad hygienic measures during slaughtering. Schmidt-Lorenz (1977) considered all moulds in relation to foodstuffs as potential pathogens. Moreover, among the moulds isolated in the current study , some were previously recorded to constitute a public health hazard due to production of mycotoxins such as *Alternaria alternata* (tenuazonic acid), *A. flavus* (aflatoxins), *A. niger* (oxalic acid), *A. sydowii* and *Emericella nidulans* (Sterigmatocystin), *Epicoccum nigrum* (flavipin), *Fusarium*, *Gibberella* and *Nectria* spp. (naphthoquinones,

zearalenone, trichothecenes and fusaric acid), *Penicillium aurantiogriseum* (cyclopiazonic acid, penicillic acid and penitrem A), *P. islandicum* (islanditoxin and luteoskyrin), *P. oxalicum* (roquefortine c), *P. purpurogenum* (purpurogenone and rubratoxins) and *Rhizopus stolonifer* (toxic cyclic peptide) (Sutic *et al.* 1972, Wu *et al.* 1974, Kamel *et al.* 1976, Leistner and Eckarde, 1981, Mirocha, 1983; Frisvad, 1988, Northolt and Soentoro, 1988).

Fortunately the presence of spores or even growth of particular fungi in not alaways followed by toxin production (Frisvad, 1988). However, prevention of fungal growth is necessary because of the rise in contamination with mycotoxins. In the current study, promising results with the use of benzoic acid could be obtained more valuabe with fungi followed by yeasts. Therefore, the sanitary instructions in slaughter houses, and the use of a preservatives such as benzoic acid are recommended to improve the quality of meat.

References

Abdel-Rahman, H. A., YOUSSEF, H. and HEFNAWY, Y. (1984). Mycological quality of meat products in Egypt. Assiut Vet. Med. J. 12, 24, 153-159.

Bandelin, F. J. (1958). The effect of pH on the efficiency of various mould inhibiting compounds. J. Am. Pharm. Assoc. Sci. Ed. 47, 691-694.

Chistner, D. F. and TANNER, F. W. (1972). Antimicrobial food additives. In: Euria, T. Handbook of Food Additives. 2nd Ed. p.115 Chem. Rubber Pub. Co. Cleveland, Ohio.

Domsch, K. W., Gams, W. and Anderson, T. H. (1980). Compendium of soil fungi, Academic Press, London.

EL-Daly, E., Abdel-Galil, Y. and Saleh, E. (1988). Studies on food borne fungi on carcase surface and air in Zagazig abattoir. The Bull. of the High Institute of Public Health, 18, 1065-1072.

Frisvad, J.C. (1988). Fungal species and their specific production of mycotoxins. Pp. 249. In : R. A. Samson, R. A. and Van Reenen-Hoekstra, E. S.eds. Introduction to food borne fungi,. Centraalbureau voor Schimmelculture, Baarn, The Netherlands.

Hadlock, R., Samson, R. A., Stolk, A.C. and Schipper, M. A. A. (1976). Schimmelpilze und Fleisch. Kontaminations flore. Fleishwirtsch. 56: 372-374.

Jermini, M. F. G. and SCHMIDT-LORENZ, W. (1987). Activity of Na benzoate and ethyl-paraben against osmotolerant yeasts at different water activity values. J. Food Protec. 50, 920-927.

Kamel, S. H., REFAI, M. and LOTT, A. (1976). Studies on the toxins of *Aspergillus niger* isolated from meat. Castellania 4, 159 -161.

King, A. D., Hocking, A. D. and Pitt, J. I. (1979). Dichloran- rose bengal medium for the enumeration and isolation of molds from foods. Appl. Environ. Microbiol. 37, 959-964.

Kotula, A. W., Lusbry, W.R. and Couse, J. D. (1975). Variability in the microbiological counts of beef carcasses. J. Animal Science 40, 834-837.

Kozakiewicz, Z. (1989). *Aspergillus* species on stored products. Mycol. Pap. 161. C.A.B. International Mycological Institute, Kew, Surrey, UK.

Leistner, L. and Eckardt, C. (1981). Schimmelpilze und mycotoxin in Fleischerzeugnissen, In Mycotoxine in Lebesmitteln. Reiss, J., Stuttgart, Germany.

Lück, E. (1986). Chemische Lebensmittelkonservierung. Stoffe, Wirkungen, Methoden. Springer-Verlag. Berlin, Germany.

Mansour, N., Hamdy, M., Yassien, N. and Refai, M. (1990). Dematiaceous hyphomycetes in slaughtered camels, cattles and surroundings at Cairo abattoir. Assiut Vet. Med. J. 24, 145- 152.

Masimango, N., Ramout, J. L. and Remacle, C. L. (1979). Contribution a L'etude du role des additifs chemiques dans lutte Contre L'aflatoxine Revue des fermentations et des Industries Alimentaries. 33 (4): 116-123.

Mirocha, J. (1983). *Fusarium* toxins. Proc. 1st Symp. Mycotoxin, National Res. Centre, Cairo, Egypt.

Mohamed, M. M. (1993). Mycological contamination of fresh meat in shops and Markets. M.V.Sc., Cairo, Egypt.

Mossel, A. A. (1977). Microbiology of Foods. The University of Utrecht, The Netherlands.

Northolt, M. D. and Soentoro, P. S. S. (1988). Fungal growth on foodstuffs related to mycotoxin contamination. In: Samson, R. A. and VAN REENEN-HOEKSTRA, E. S.: Introduction to foodborne fungi. Centraalbureau voor Schimmelcultures, Baarns, The Netherlands. Pp. 231-238.

Pitt, J. I. (1979). The genus *Penicillum* and its teleomorphic states Eupencillium and Talaromyces. Academic Press, London.

Refai, M. and LOTT, A. (1969). Studies on meat contamination of meat in slaughter houses, butcher shops and in cold stores. Mycosen, 12, 10, 621-624.
Refai, M., Mansour, EL-Nagar, N. and Abdel-Aziz, A. (1993). Fungal flora in Egyptian modern abattoirs. Fleischwirtsch. 73, 2, 172- 174.
Samson, R. A. and Van Reenen-Hokstra, E. S. (1988). Introduction to foodborne fungi. Centraalbureau voor Schimmelcultures, Baarn, The Netherlands (Pp. 299).
Schmidt-Lorenz; W. (1977). Allegemine Bedutung von Pilzen und Hefen in Lebensmitteln. SGHL, 6, 9-14
Sutic, M. T., Ayres, J. C. and Koehler, P. E. (1972). Identification and aflatoxin production of moulds isolated from country cured hams. Appl. Microbiol.23, 6, 656-568.
Uraih, N. and Chipley, J. R. (1976). Effect of various acids and salts on growth and aflatoxin production by *Aspergillus flavus*. NRRL 315. Microbios, 17, 51-59.
Uraih, N., Cassity, T. R. and Chiply, J. R. (19779. Partial characterization of the mode of benzoic acid on aflatoxin biosynthesis. Can J. Microbiol. 23, 1580-1584.
Wu, M. T., Ayres, J. C. and Koehler, P. E. (1974). Toxigenic Aspergilli and Penicilli from aged cured meats. Appl. Microbiol. 28, 1094-1096.
Yassien, N., MANSOUR, N., EL-DALY, E. and DARWISH, A. (1990). Contamination of slaughtered camels, cattle and its surroundings with mould in Urban abattoir. Alexandria J. Vet. Sci. 5, 2, 185-193.