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PS 6

Poster session and workshop 6

Analitical methods



Tuesday, September 1st
11:15h-12:45h

Occurrence of *Listeria monocytogenes* in meat products and identification by applied PCR method

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Background

Listeria belongs to the microorganisms whose monitoring deserves a great attention due to serious course of the disease. The way of transfer is of high importance in epidemiology of listeriosis. Consumption of contaminated food is the most frequent way of transfer. Fresh meat, raw meat products and ready to eat dishes are a significant source of *Listeria* spp. In case of disease the way of transfer shall be determined.

Objectives

Our study was aimed at monitoring of occurrence of *Listeria monocytogenes* in meat and meat products and at molecular identification of isolated strains with help of PCR methods.

Methods

During 1996 – 1997 we were taking samples from selected kinds of foodstuffs and monitoring occurrence of *Listeria* spp. Our attention was drawn mainly to:

- meat products without heat processing (raw salami)
- heat processed meat products
- mechanically recovered poultry meat
- soft, ripening and blue cheese

Taken samples were examined according to standard USDA methods. For certain samples of foodstuffs the quick screening test of the Clearview (Oxoid) was applied for identification of *Listeria* spp. Selected strains were identified by using the PCR method. From certain available methods the PCR method was chosen, as it is based on gene sequence for listeriolysin O. Two oligonucleotide primers LP1 and LP2 were used. PCR products were subject to electrophoretic analysis on 1% agarose gel and detected by ethidium bromide staining by applied UV transillumination.

Results and discussions

During 1996-1997 we isolated 52 *Listeria* sp. strains from foodstuffs. *Listeria* spp. were mostly isolated from meat products without heat processing (raw salami), from ripening cheese and mechanically recovered poultry meat. *L. monocytogenes* was revealed in raw meat products (meetwurst salami), dry and fermented sausage and pâté (Table 1). We succeeded to isolate 17 strains of *Listeria* spp. from 30 examined samples of mechanically recovered poultry meat. On the basis of further diagnostic tests 16 isolated strains were identified as *L. innocua*; *L. monocytogenes* was identified in one case.

As many as 65 strains of *Listeria* spp. were investigated with PCR methods. A majority of them was identified as *L. innocua* (83%). Strains *L. monocytogenes* were revealed in 17 %. The fragments yielded with help of the described PCR method reached the size of 750 pb in case of *L. monocytogenes*. The PCR method above enabled to identify *L. monocytogenes* even in the mixed culture with *L. innocua* which meat samples were contaminated experimentally by. The results reached by classical examination carried out in the time interval of several days were identical.

Conclusions

From our experiments and from literature data it follows that raw meat products represent a hazard of listeriosis for the people. It is mainly the matter of the products with a short time of ripening where the technological barriers cannot be applied, and without use of starter cultures. From our results it follows that more than 30 % of examined raw salami (meetwurst salami) contained *Listeria* spp. and three samples contained even *L. monocytogenes*.

Isolated strains of *L. monocytogenes* were identified with help of PCR method and used oligonucleotides LP1 and LP2. High sensitivity and specificity is a great advantage of the method. *L. monocytogenes* and *L. innocua* may be differentiated in the mixed samples too.

Pertinent literature

Mc Lauchlin, J. 1993. Listeriosis and *Listeria monocytogenes*. Environmental Policy and Practice 3: 201-214.

Jay, J.M. 1996. Prevalence of *Listeria* spp. In meat and poultry products. Food Control 7: 209-214.

Salamina, G., Donne E. D., Niccolini, A. and et al. 1996. A foodborne outbreak of gastroenteritis involving *Listeria monocytogenes*. Epidemiol. Infect 117: 429-436.

Rijpens, N. P., Jannes, G. and Herman L.M.F. 1997. Incidence of *Listeria* spp. and *Listeria monocytogenes* in ready-to eat chicken and turkey products determined by polymerase chain reaction and line probe assay hybridization. J. Food Protection 60: 548-550.



Table I
Frequency of *Listeria* spp. in meat products

Product	No. samples	No. (%) <i>Listeria</i> spp.	No. (%) <i>L. innocua</i>	No. (%) <i>L. monocytogenes</i>
Meetwust salami	56	18 (32)	15 (27)	3 (5)
Raw salami	20	9 (45)	7 (35)	2 (10)
Paté	60	5 (8)	3 (5)	2 (3)
Cooked Salami	19	3 (16)	3 (16)	0
Mechanically recovered poultry meat	30	17 (6)	16 (5)	1 (1)
TOTAL	185	52 (28)	44 (24)	8 (4)