ADVISORY COMMITTEE ON THE MICROBIOLOGCIAL SAFETY OF FOOD

Changing pattern of human listeriosis in England and Wales 1993 - 2004

1. At the March ACMSF meeting, the ACMSF requested an update on the origin of recent increasing trends in listeriosis. In response to that request, Mr Iain Gillespie and Dr Jim McLauchlin from the Health Protection Agency have prepared the attached paper.

Secretariat
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Changing pattern of human listeriosis in England and Wales, 1993-2004.
Report for the ACMSF.
Health Protection Agency, London. September 2005.

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1. Summary.

- Microbiological and epidemiological data on 1759 cases of human listeriosis reported in the United Kingdom between 1993 and 2004 were reviewed.
- Between 2001 and 2004, the number of cases in England and Wales increased significantly. An increase in Scotland between 2000 and 2004 was not statistically significant. There was no observed trend in risk Northern Ireland.
- Seven clusters of cases were detected which were likely to represent common source foodborne outbreaks. The numbers of cases per cluster varied between two and eighteen. Hospital sandwiches were associated with four of the clusters, butter with one, and no food vehicles were identified in the remaining two clusters. Two episodes of neonatal cross infection (two cases per episode) were also detected.
- The identified clusters of cases do not explain fully the upsurge in disease incidence, which was largely confined to non pregnancy-associated cases aged >59 years from both sexes. Multiple *L.monocytogenes* strains were identified for the upsurge and the patients had diverse underlying illnesses. The upsurge occurred in almost all regions and did not show an unusual seasonal pattern. This upsurge is unlikely to be due to a common exposure to a single food source.
- Patients aged 60 years and over accounted for 76% of all listeriosis cases in England & Wales from 2000 to 2004 and this level was exceeded in the first eight month of 2005 (78%). This upsurge cannot be explained by the current knowledge of the epidemiology of listeriosis, suggesting that resources should be diverted to understanding and controlling the disease in this age group.

2. Introduction.

The bacterium *Listeria monocytogenes* and the disease listeriosis were first recognised in laboratory animals in Cambridge in 1924 (Murray *et al.*, 1926). It later became apparent that the disease also affects humans. The disease most often affects unborn and newly delivered infants as well as the immunocompromised and elderly: listeriosis also occurs in otherwise healthy individuals but is rare. Listeriosis primarily presents as abortion, septicaemia or infections of the central nervous system, with a high case fatality rate (Farber and Peterkin 1991). Although the majority of cases of listeriosis are acquired by consumption of contaminated food, the epidemiology is complex. The ubiquitous nature of the bacterium, together with a wide range in incubation periods (1->90 days), means that investigations to identify specific food vehicles are problematic (McLauchlin 1996a).

The reported annual incidence of human listeriosis in England and Wales increased from less than 75 cases between 1966 and 1981 to a maximum of 136 cases in 1985. During 1987, 1988 and 1989 the incidence almost doubled, with 238, 278 and 237 cases reported. This was probably associated with the consumption of Belgian pâté (McLauchlin et al., 1991). The numbers of cases declined following warnings for pregnant women and the immunocompromised to avoid consuming pâté and a withdrawal from retail sale of this food from a single manufacturer (McLauchlin et al., 1991).

Throughout the 1990s the incidence of listeriosis in England & Wales was stable with, on average, 110 cases reported annually (Health Protection Agency). However, the incidence has risen in recent years, with 146, 136, 237 and 213 cases reported in the years 2001, 2002, 2003 and 2004 respectively. The purpose of this report is to describe human listeriosis in England and Wales between 1993 and 2004 in order to identify possible reasons for this upsurge.

3. Materials and methods.

The routine surveillance of human listeriosis in England & Wales is co-ordinated at the Health Protection Agency Centre for Infections. Cases are ascertained by the voluntary reporting of laboratory-diagnosed cases from medical microbiology laboratories through a routine electronic reporting system. Cases are also ascertained by the submission of cultures to the national reference centre for confirmation of identity and subtyping. Epidemiological and microbiological data from both systems are combined, de-duplicated and stored in a bespoke database. Additional demographic and clinical data is sought from consultant medical microbiologists in charge of the case using a standard questionnaire.

The identity of *L.monocytogenes* isolates referred to the national reference centre are confirmed by either phenotypic tests as described previously, including the use of the API Listeria identification kit (McLauchlin 1997), or using a 5'-nuclease-based real-time PCR assay to amplify a fragment of the *L.monocytogenes* haemolysin gene (Nogva et al., 2000). All isolates of *L.monocytogenes* are characterised by serotyping (Seeliger and Hohne, 1979; Doumith et al., 2004), phage-typing up to 2003 (McLauchlin et al., 1996b), amplified fragment length polymorphism analysis from 2002 (AFLP, Guerra et al., 2002), and pulsed-field gel electrophoresis from 2003 on selected isolates (PFGE, Graves and Swaminathan 2001).

For the purposes of surveillance, a case of listeriosis is defined as a patient with a compatible illness from whom *L.monocytogenes* was isolated from a normally sterile site, usually blood or cerebrospinal fluid (CSF). Cases were categorised as pregnancy-associated (all materno-foetal, and neonatal cases; a mother baby pair constituting one case) and non-pregnant (a case aged greater than one month).

For the purposes of this paper, microbiological, clinical and demographic data for England & Wales were abstracted from the routine surveillance system. Annual totals for Scotland and Northern Ireland were obtained from the websites of the Scottish Centre for Infections and Environmental Health and the Communicable Disease Surveillance Centre Northern Ireland respectively (Scottish Centre for Infections and Environmental Health; Communicable Disease Surveillance Centre Northern Ireland).

Age-specific national and regional denominator data for the years 1993-2003 (the most recent set of data available) were obtained from the Office for National Statistics.

Data analysis was performed using Microsoft Excel, Epi Info version 6.04b and Stata version 8.2. Relative proportions, and changes in relative proportions with time, were compared using the chi-squared test and the chi-squared test for trend respectively. Point estimates of relative risks (RR), with accompanying 95% Confidence Intervals (CI) and significance tests were also calculated.

4. Results.

4.1. All reported cases.

Between 1993 and 2004, 1759 cases of human listeriosis were reported in the United Kingdom, comprising 1469, 135, 115 and 40 in England, Scotland, Wales and Northern Ireland respectively (Fig. 1). A further two cases of non-invasive human listeriosis, reported in England in 1999 and 2001, did not fulfil the case definition and were excluded from the analysis.

Having controlled for differences in total population (Fig. 2), the risk of listeriosis was higher in Wales than in any other country during the course of the study (RR 1.32; 95%CI 1.11-1.58; P=0.002). The risk of disease increased significantly in England (RR 1.69; 95% CI 1.52-1.86; P<0.001) and Wales (RR 1.57; 95% CI 1.09-2.27; P=0.015) between 2001 and 2004. A similar increase in risk in Scotland between 2000 to 2004 was not statistically significant (RR 1.35; 95% CI 0.96-1.89; P=0.08). No discernable trend in risk was observed in Northern Ireland.

4.2. Common source clusters of cases.

During the course of this study, putative *L.monocytogenes* isolates from 1327 out of 1584 cases (84%) in England and Wales were confirmed and characterised in the national reference centre. Clusters of cases (some of which were identified as common source food-borne outbreaks) were detected (Table 1).

4.2.1. Cluster 1.

A local cluster of four listeriosis cases occurred in the North East of England in May 1999. The patients ages ranged from 48 to 79, all were immunocompromised and *L.monocytogenes* was isolated from blood cultures from each patient, as well as a CSF sample from one case. A common risk factor was consumption of sandwiches sold in hospital and the same *L.monocytogens* strain was isolated from two of the patients, sandwiches bought in the hospital and the sandwich manufacturing environment.

4.2.2. Cluster 2.

Seventeen laboratory-confirmed cases of listeriosis occurred between January and July 2003. *Listeria monocytogenes* was isolates from blood in 14 cases, CSF from two and both blood and CSF from one. All cases occurred in the North East of England, 14 in the Yorkshire and Humberside region and all were due to the same *L.monocytogenes* type. Eleven cases were pregnancy-associated and two miscarriages and one stillbirth were reported. Four of the non-pregnancy associated cases were immunocompromised, all were >50 years of age and two deaths were reported.

In April 2003, routine testing identified a low level intermittent *L.monocytogenes* contamination of butter produced at a dairy in the Yorkshire and Humberside region. Initial tests of *L.monocytogenes* from different batches of butter showed levels of the bacterium at 180 cfu/g and <20 cfu/g (250g packs). In subsequent batches tested during April and May, the same strain of *L.monocytogenes* was intermittently detected from other batches, all at low levels (<20 cfu/g). A positive release system for butter production was introduced in the middle of May coupled with revised cleaning procedures. The implicated strain was isolated from drains within the factory in June and July. No *L.monocytogenes* was detected in subsequent butter production. The isolates from the butter were indistinguishable from those isolated from the 17 patients.

Interviews were conducted with cases and controls and descriptive epidemiology suggested that cases were exposed through the use of the 2Kg catering packs of butter

supplied to local sandwich outlets from the dairy rather than consuming contaminated product in the home. A Food Hazard Warning was issued by the Food Standards Agency on 15 July and a voluntary recall of 2Kg packs was instituted, together with a positive release system for product from the dairy. No further cases infected with this *L.monocytogenes* type were detected after early July 2003, and this strain was not detected in any other isolates from food.

4.2.3. Cluster 3.

A further cluster of listeriosis, which was due to a different *L.monocytogenes* strain from cluster 2, affected 18 non-pregnant individuals in North East England (11 of which were in Yorkshire and Humberside) during January to October 2003. The cases occurred in patients >45 years of age (range 45-99) and *L.monocytogenes* was recovered from blood cultures from all patients. Fourteen of the patients were known had underlying conditions that might lead to impaired immunity. No food items were associated with this cluster. Seven of the patients were reported to have died.

4.2.4. Cluster 4.

Two cases were detected in Wales in May 2003 amongst cancer patients attending the same oncology outpatient clinic. The same strain of *L.monocytogens* was isolated from blood from both patients within two days: neither patient died. Epidemiological investigations revealed that the two patients consumed either ham or tuna sandwiches from a hospital canteen. There was evidence for poor temperature control at the point of sale to the patients.

Follow-up testing of a wide range of sandwiches at the hospital and investigations at the factory revealed frequent contamination by *L.monocytogenes:* all at low levels. Isolates from the two cases were indistinguishable from isolates from 49 sandwiches and 11 environmental sites within the sandwich manufacturing environment. Two other *L.monocytogenes* strains were isolated from an additional nine sandwiches and two environmental sites within the factory. The sandwiches were widely distributed, but indistinguishable isolates were not recognised from any subsequent cases of human listeriosis in the UK.

4.2.5. Cluster 5.

Five cases of listeriosis amongst pregnant women occurred in South West England during October 2003. All were infected by the same strain of *L.monocytogenes* which was recovered from blood cultures taken from each patient. There were no fatalities. Epidemiological investigations showed a common link with consumption of sandwiches from a single hospital retailer. The implicated strain was subsequently recovered from the sandwich manufacturer (one sandwich and one environmental site). Two other *L.monocytogenes* strains were recovered from either one sandwich or three other environmental sites in the factory. All sandwiches were contaminated at low levels.

4.2.6. Cluster 6.

A cluster of six cases of non pregnancy-associated listeriosis occurred in the East Midlands in early 2004. The cases, all greater than 55 years of age, were ill within a six week period with five of the cases occurring within a four day period. Three *L.monocytogenes* subtypes were identified and three patients were infected with a single type. No common risk factor for these patients was identified.

4.2.7. Cluster 7.

Two cases of listeriosis were detected amongst two male patients aged 75 and 77 attending the same renal unit in the South East of England in September 2004. *Listeria monocytogenes* was isolated from the blood cultures taken from both patients. The isolates from the patients were of different types. Epidemiological investigations implicated the consumption of hospital sandwiches. *Listeria monocytogenes* was isolated from sandwiches collected at the hospital, sandwiches from the factory and from the factory environment. Amongst the 117 isolates recovered from sandwiches and the factory, multiple *L.monocytogenes* strains were identified: two of these were indistinguishable from those recovered from blood cultures taken from one of the patients. The strain from the other patient was not recovered from any of the sandwiches or from any other food isolates submitted to the Centre for Infections.

4.2.8. Other clusters.

Two pairs of neonatal listeriosis cases occurred in different hospitals in South East England in 1997 and 1998. Here, cross-infection is likely to have taken place from a congenitally infected infant to an apparently healthy neonate following contact within the delivery suite.

4.3. Trends amongst 'sporadic' cases of human listeriosis in England & Wales

Following exclusion of cases linked to clusters described above, there was a significant increase in sporadic listeriosis in England (RR 1.58; 95% CI 1.43-1.76; P<0.001) and Wales (RR 1.48; 95% CI 1.02-2.15; P=0.039) in 2001 to 2004 compared with 1993 to 2000.

Cases of sporadic listeriosis in England and Wales by patient type are shown in Fig 3. In 1993, non pregnancy-associated cases of listeriosis accounted for approximately 80% of the 103 cases reported. By 2004, non pregnancy-associated cases accounted for approximately 90% of the 205 cases reported (χ^2 for trend P<0.001).

The risk of sporadic non pregnancy-associated listeriosis by age group is shown in Fig. 4. between 2001 and 2004 the risk in people aged ≥60 years has increased by a third (RR 1.33; 95%CI 1.25-1.42; P<0.001). This effect has been observed in both males and females (Figs 5 and 6) and has occurred in most regions of England and in Wales (Fig 7). The seasonal pattern of infection was no different for non pregnancy-associated cases aged ≥60 years reported between 1993 and 2000 and those reported between 2001 and 2004 (Fig 8).

Serotyping data were available for 774 sporadic non pregnancy-associated cases aged ≥60 years reported between 1993 and 2004 (82%; Table 2). Since 2000 there has been a slight but significant shift from serotype 1/2 to serotype 4b (χ^2 P=0.002). Amplified Fragment Length Polymorphism typing, available from 2002 onwards and used in conjunction with serotyping (Table 3), shows that multiple subtypes are responsible for the increase in cases aged 60 and over.

Data on underlying illness were available for 703 of the 949 (74%) sporadic non pregnancy-associated cases aged 60 and over. Cases reported between 2001 and 2004 were no more likely to report an underlying condition than cases reported between 1993 and 2000 (both 90%). Furthermore, classification of the conditions suggests that, with the exception of renal problems (χ^2 P=0.007), there has been little change in the underlying conditions for cases reported between 2001 and 2004 and those reported between 1993 and 2000 (Table 4).

5. Conclusions.

- The epidemiology of listeriosis is complex and, following a relatively quiescent period during the 1990s, a significant increase in the numbers of cases occurred in England and Wales from 2001. Common source foodborne outbreaks were identified, but these constituted a small proportion of the total and did not wholly account for the significant increase in cases.

- The identified food vehicles in five of the clusters were butter and sandwiches sold in hospitals. Butter has previously been epidemiologically and microbiologically associated with an outbreak of listeriosis in Finland (Lyytiksinen et al., 2000; Maijala et al., 2001) and epidemiologically associated with cases in the USA (Mascola et al., 1999). Previous investigations have shown that sandwiches were commonly contaminated with *L.monocytogenes (*Hunter et al., 1990; McLauchlin and Gilbert 1990; Wilson 1996; Willis and Greenwood 2003). There was some evidence for temperature abuse of food as a contributory factor for clusters two and four described above.

- The increase in the incidence of listeriosis from 2001 to 2004 was largely confined to sporadic cases aged >59 years. The upsurge was detected amongst both sexes, in multiple regions, amongst patients with diverse underlying illnesses and was independent of demographic changes. The upsurge is unlikely to reflect a common source foodborne outbreak confined to older members of the population since a large number of *L.monocytogenes* strains were responsible for this upsurge. Previous large foodborne outbreaks have been due to one or more rarely two *L.monocytogenes* strains. Furthermore it is unlikely that the effect is due to increased survival with chronic conditions in the last five years.

- Analysis of data from 2005 to date suggests that the changing pattern of listeriosis is continuing.

6. Current and future initiatives.

Previous strategies for the control of listeriosis remain applicable. These include:

- recognition of foodborne outbreaks and withdrawal of implicated food vehicles from retail sale;
- prevention of contamination and inhibition of the growth of the bacterium in the food chain;
- prevention of growth and cross-contamination in domestic settings;
- dietary advice to vulnerable groups to avoid high risk foods.

Health Protection Agency responses.

The HPA has put in place several initiatives to improve our understanding of listeriosis, particularly in the elderly:

Laboratory initiatives.

Improvements in molecular typing strategies will be implemented. This includes greater application of PFGE, together with investigating other rapid molecular typing methods. This will be coupled with bioinformatics approaches, which will improve data manipulation and outbreak recognition. This activity will be strengthened by an FSA-funded PhD studentship based in the HPA Centre for Infections (Colindale, London).

Epidemiological initiatives.

Since listeriosis is now predominantly a foodborne infection amongst individuals 60 years and older, efforts will be concentrated on understanding and controlling listeriosis in this age group. An extensive trawling questionnaire, seeking clinical data, travel history, food consumption and buying habits has been developed and is currently in use. Data accrued from these questionnaires will be used to generate hypotheses for *L.monocytogenes* infection in the elderly, which will then be tested using analytical epidemiological methods. This will also be used to inform on

microbiological surveillance of 'high-risk' foods more likely to be consumed by the >59 year old individuals.

Microbiological surveillance initiatives.

To address the gap in current knowledge on the prevalence, levels and types of *L.monocytogenes* in 'high-risk' foods, pan-UK studies of butter at retail sale and used in catering, as well as sandwiches provided within hospitals and residential care homes are being carried out. The study on butter has been completed with a report published, while the 12 month study on sandwiches will be completed in April 2006.

Other responses.

General information on safe food practices (particularly with respect to listeriosis), does not specifically target the older sections of the population. Efforts should be directed at the production of such advice and assessment of the effectiveness of general food safety advice.

7. References.

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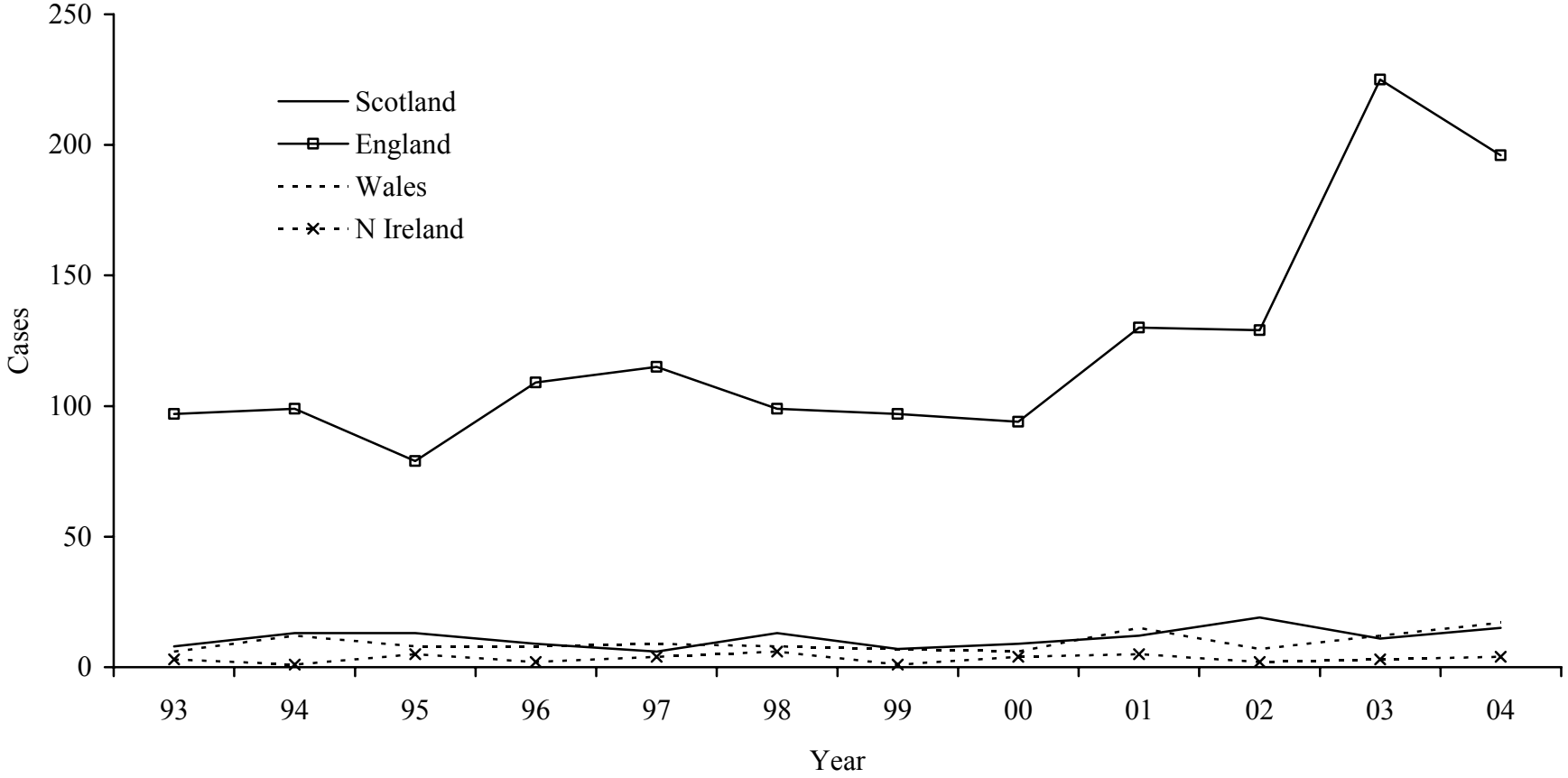
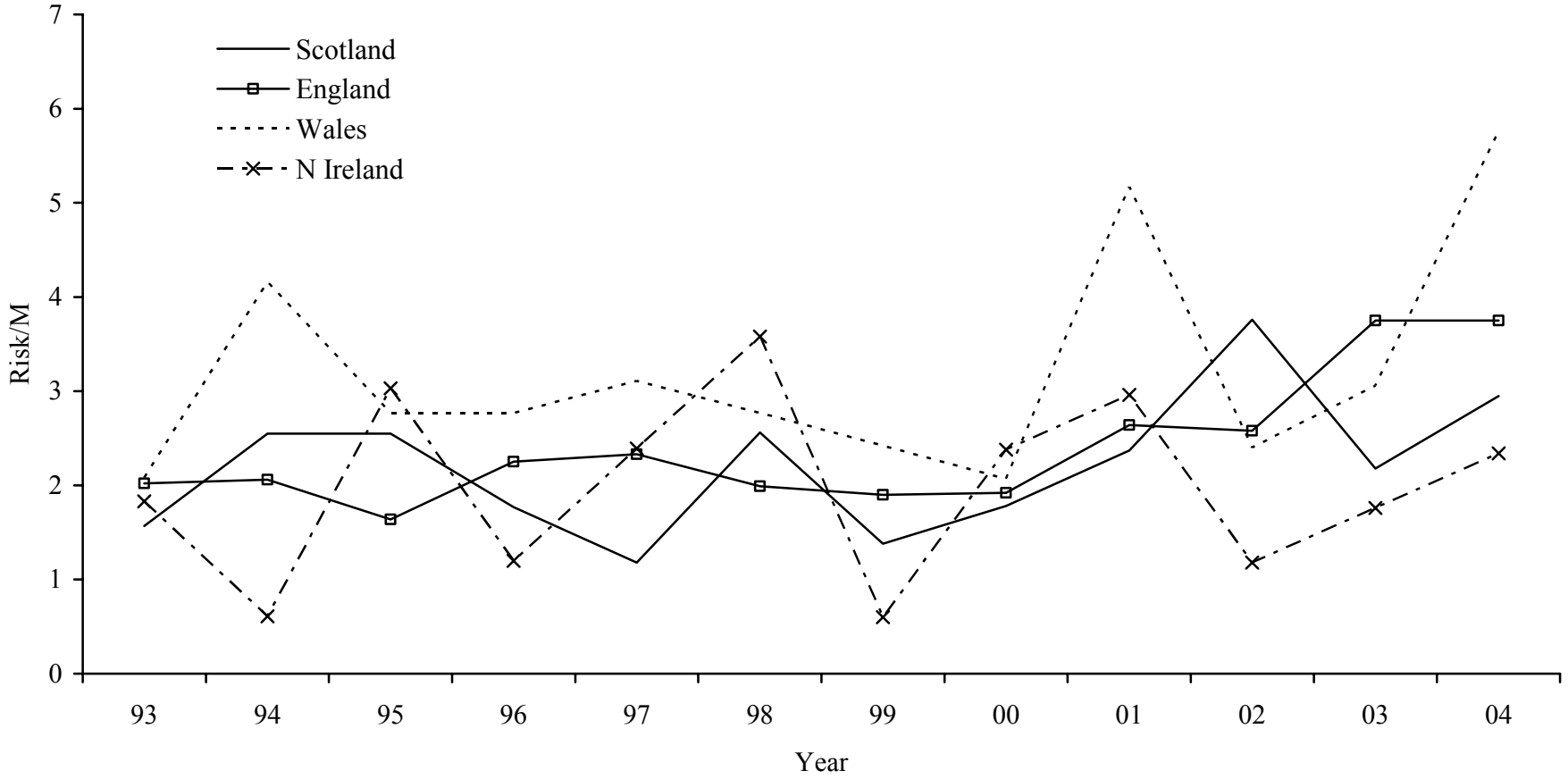
Figure 1. Reported cases of human listeriosis in the United Kingdom by country, 1993 to 2004.



Source: Health Protection Agency, Communicable Disease Surveillance Centre Northern Ireland & Scottish Centre for Infection & Environmental Health

Figure 2. Country-specific risk of all reported human listeriosis per million population, United Kingdom, 1993 to 2004.



Source: Health Protection Agency, Communicable Disease Surveillance Centre Northern Ireland; Scottish Centre for Infection & Environmental Health & Office for National Statistics

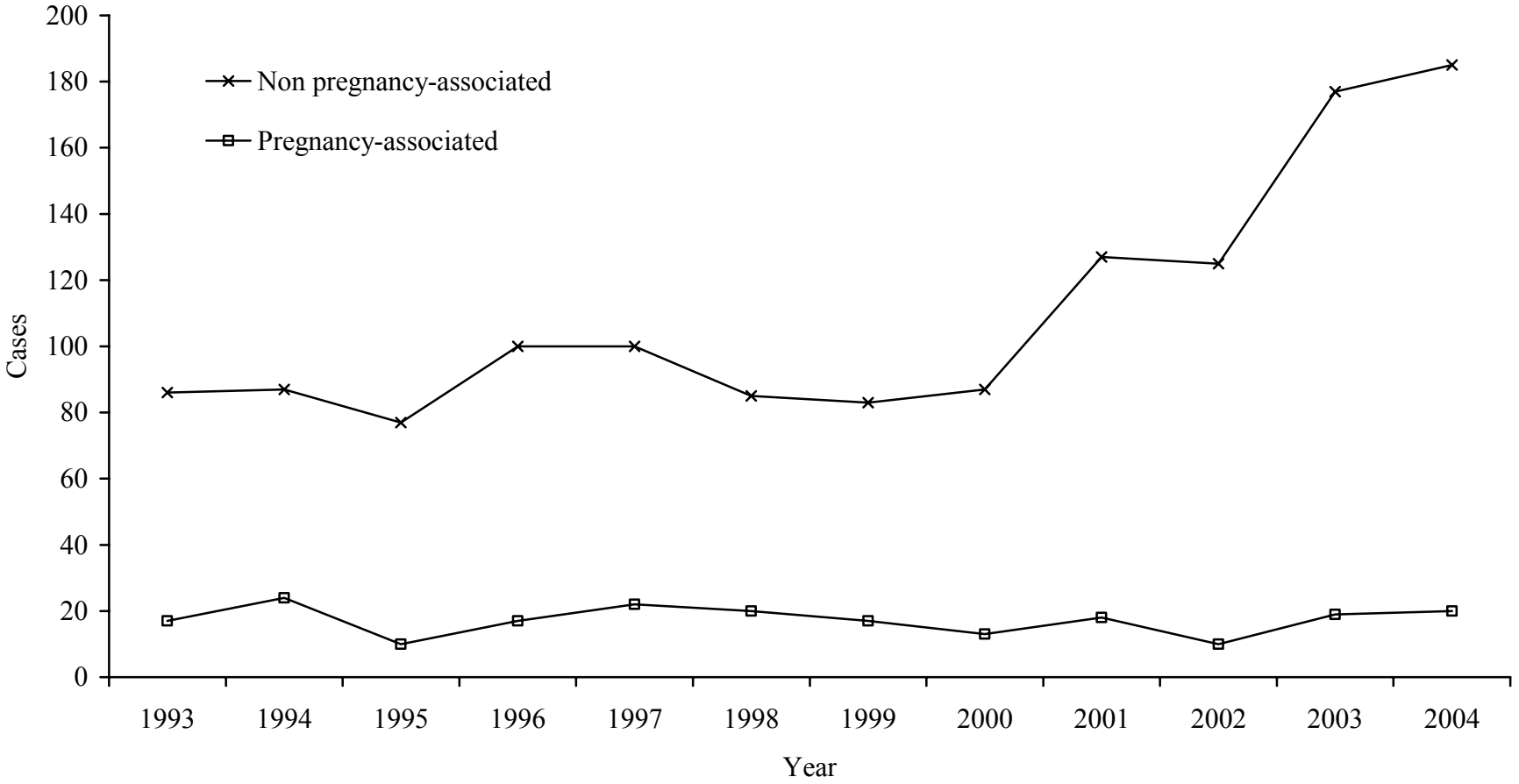
Table 1. Clusters of human listeriosis, England and Wales 1993-2004.

Cluster Number	Year	Area	Number of cases	Pregnancy associated	*L.monocytogenes* type				Vehicles of infection
					Serovar	AFLP	Phage	PFGE	
Clusters probably or likely to be common-source food-borne outbreaks									
1[a]	1999	NE England	4	0	4b	ND	NT	ND	Hospital sandwiches
2	2003	NE England	17	11	4b	V	A	2	Butter
3	2003	NE England	18	0	4b	I	G	1	None identified
4	2003	S Wales	2	0	1/2a	XI	Y	L	Hospital sandwiches
5[b]	2003	SW England	5	5	1/2a	III	Y	A	Hospital sandwiches
6	2004	E Mids	6	0	4b 4b 4b	I IV V	ND ND ND	E M J	None identified None identified None identified
7	2004	SE England	2	0	4b 4b	I V	ND ND	A B	Hospital sandwiches Not identified
Episodes of neonatal cross infection									
1	1997	SE England	2	2	4b	ND	H	ND	Contact between cases within a delivery suite
2	1998	SE England	2	2	1/2a	ND	I	ND	Contact between cases within a delivery suite

ND = not done; NT = non-typable; Two clusters have previously been described: a, Graham et al., 2002; b, Anon 2003.

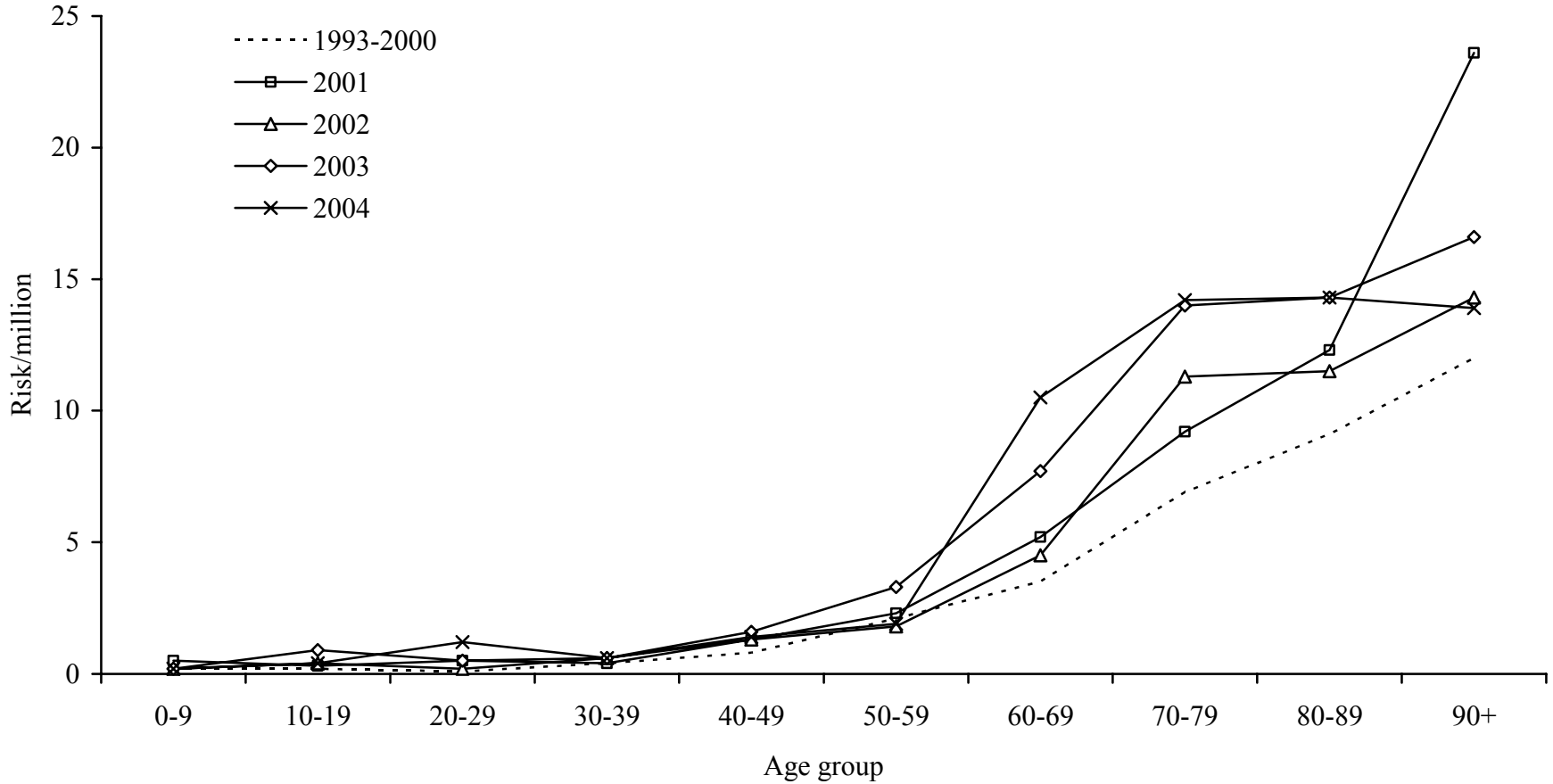
Source: Health Protection Agency Listeria database

Figure 3. Sporadic listeriosis in England & Wales by patient type, 1993 to 2004.



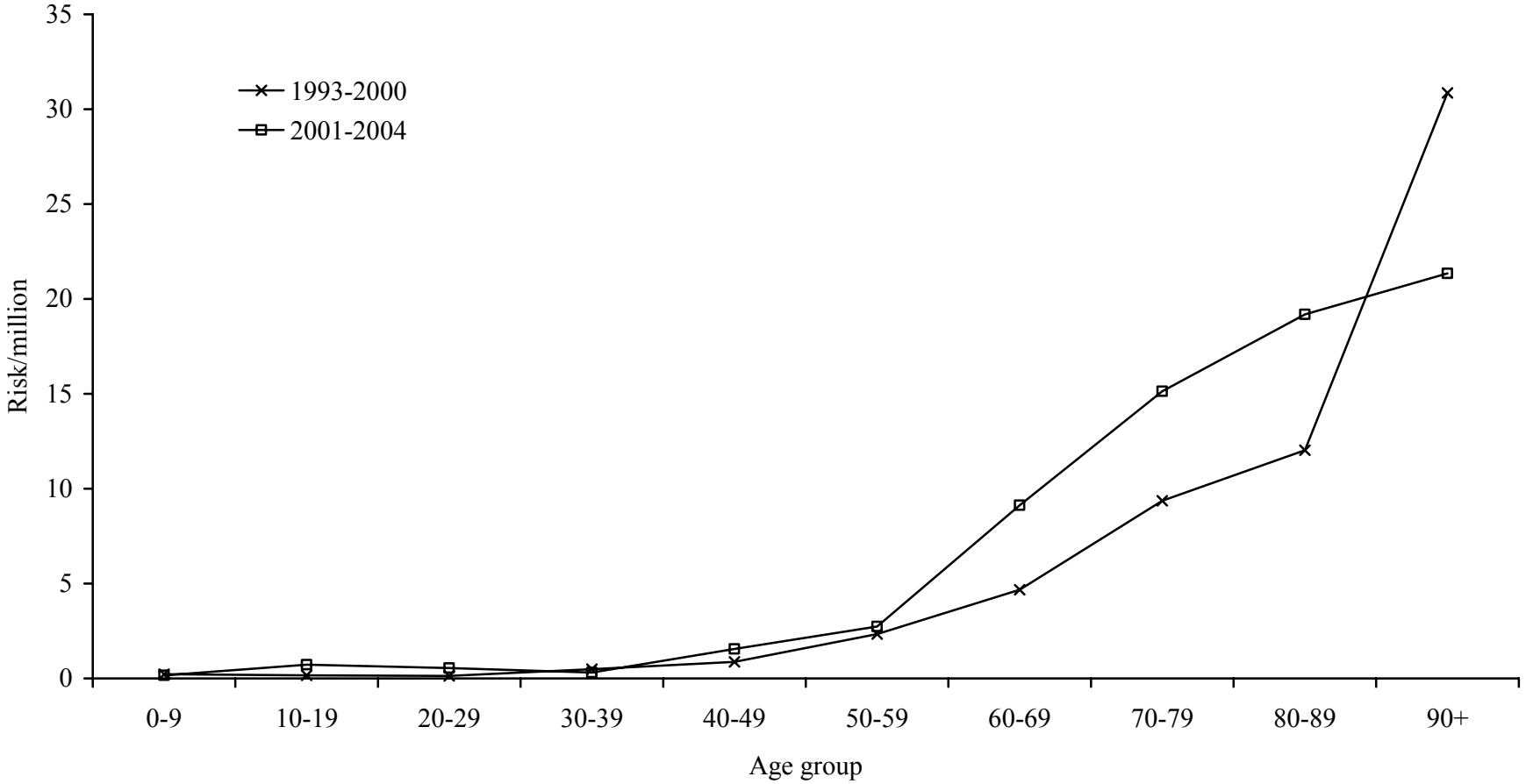
Source: Health Protection Agency Listeria database

Figure 4. The risk of non pregnancy-associated sporadic listeriosis by age group, England & Wales, 1993 to 2004 (showing 2001 to 2004 in detail).



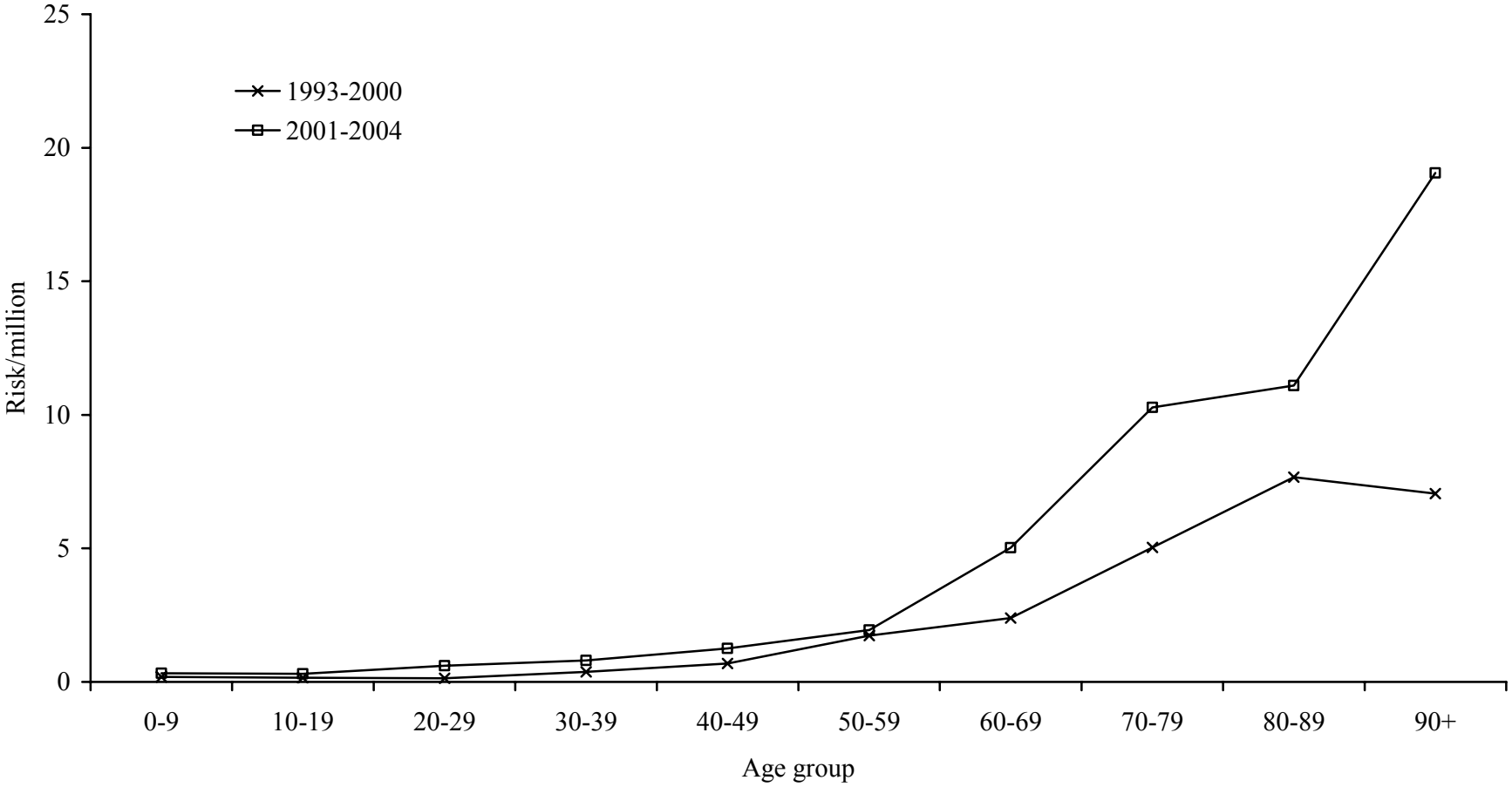
Source: Health Protection Agency Listeria database & Office for National Statistics

Figure 5. The risk of non pregnancy-associated sporadic listeriosis by age group in <u>males</u>, England & Wales, 1993 to 2004



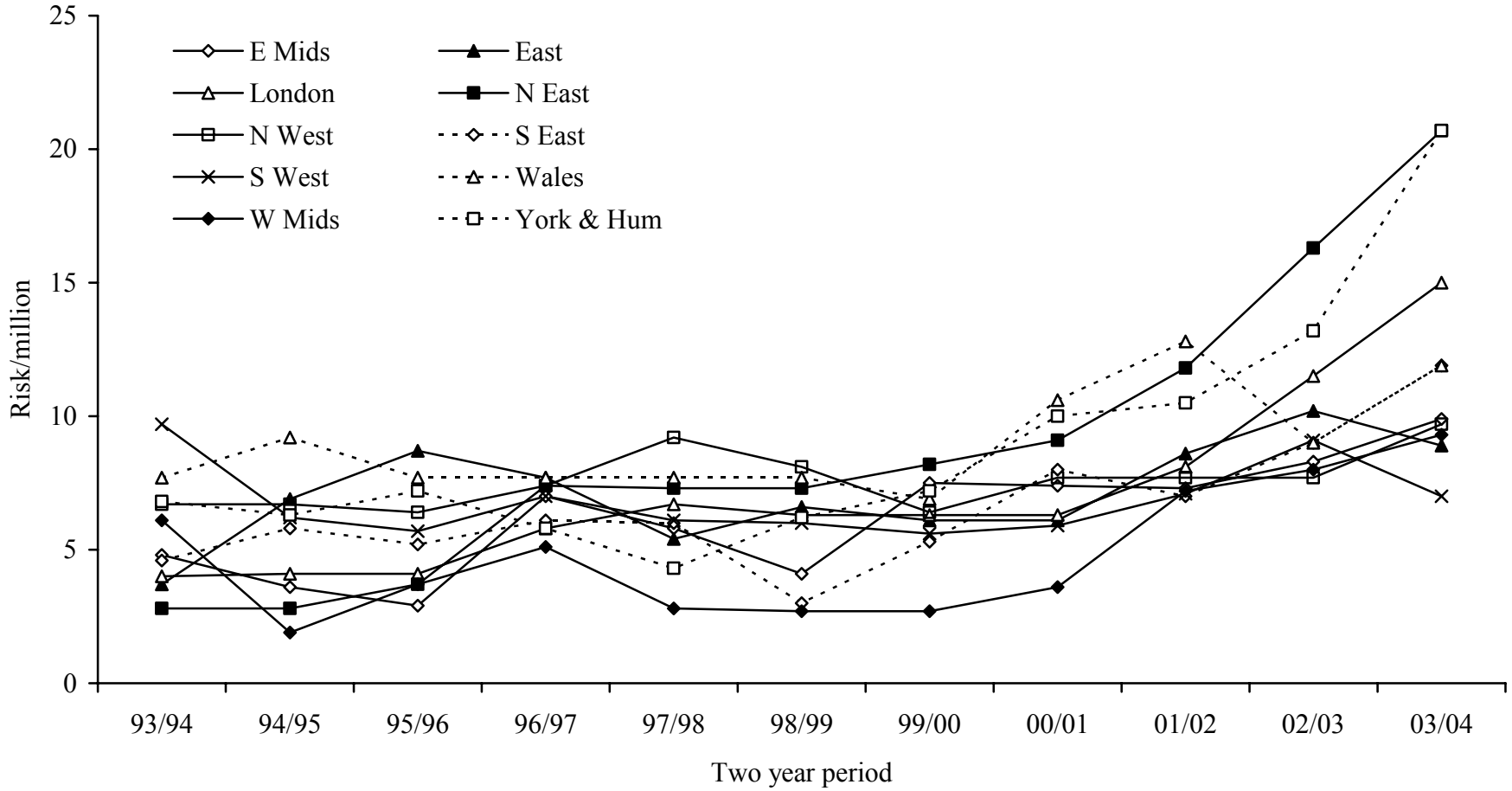
Source: Health Protection Agency Listeria database & Office for National Statistics

Figure 6. The risk of non-pregnancy-associated sporadic listeriosis by age group in <u>females</u>, England & Wales, 1993 to 2004



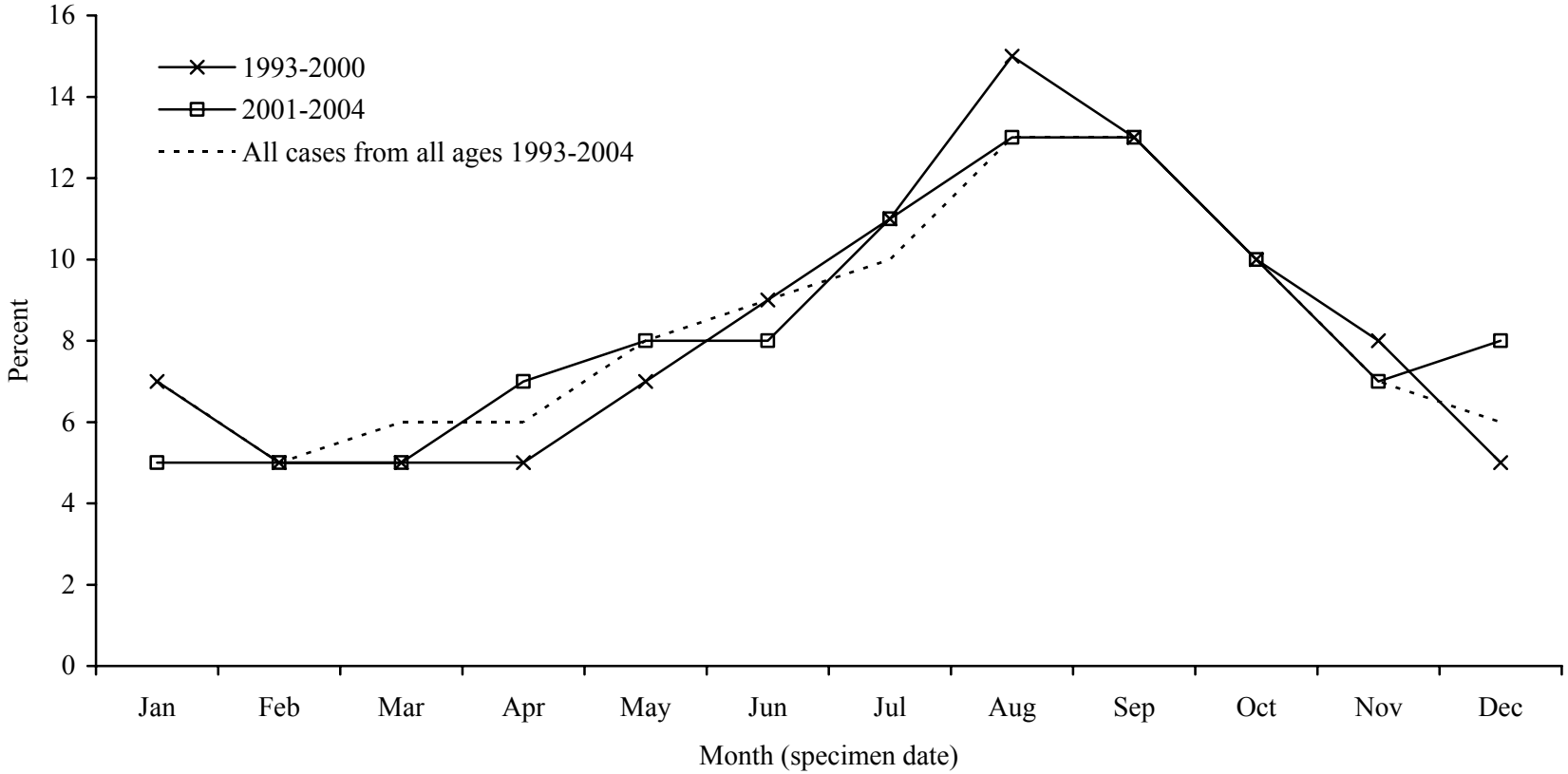
Source: Health Protection Agency Listeria database & Office for National Statistics

Figure 7. The risk of non pregnancy-associated sporadic listeriosis in people ≥60 years by Region, England & Wales, 1993 to 2004



Source: Health Protection Agency Listeria database & Office for National Statistics

Figure 8. The seasonal pattern of all cases of listeriosis and that amongst non pregnancy-associated sporadic cases aged ≥60 years, England & Wales, 1993 to 2004.



Source: Health Protection Agency Listeria database

Table 2. Serotyping data for sporadic non pregnancy-associated listeriosis cases ≥60 years, England & Wales, 1993-2004.

Serotype	Number of cases by time period (column %)		Total
	'93-'00	'01-'04	
1/2	221 (52)	155 (45)	376
4b	192 (45)	184 (53)	376
Other including non typable	15 (4)	7 (2)	22
Total	428	346	774

Source: Health Protection Agency Listeria database

Table 3. Serotype and AFLP type for sporadic non pregnancy-associated listeriosis cases ≥60 years, England & Wales, 2002-2004.

Sero AFLP type	Number of cases per year (column %)			Total
	2002	2003	2004	
4b I	17 (22)	18 (17)	16 (20)	51
4b IV	11 (14)	22 (20)	12 (15)	45
1/2 VII	15 (19)	12 (11)	17 (21)	44
1/2 II	3 (4)	17 (16)	7 (9)	27
4b V	5 (6)	6 (6)	10 (12)	21
1/2	7 (9)	6 (6)	0	13
4b	8 (10)	4 (4)	1 (1)	13
1/2 IX	0	6 (6)	5 (6)	11
1/2 III	4 (5)	3 (3)	4 (5)	11
1/2 XI	0	3 (3)	2 (2)	5
Others	9 (11)	11 (10)	8 (10)	28
Total	79	108	82	269

Source: Health Protection Agency Listeria database

Table 4. Underlying conditions reported for sporadic non pregnancy-associated listeriosis cases ≥60 years, England & Wales, 1993-2004.

Classification	Number of cases by time period (column %)		Total
	'93-'00	'01-'04	
Cancers	150 (42)	122 (44)	272
Autoimmune disorders	55 (15)	33 (12)	88
Cardiovascular disorders	47 (13)	34 (12)	81
Alcohol-related disorders	14 (4)	9 (3)	23
Renal disorders	6 (2)	16 (6)	22
Diabetes	5 (1)	9 (3)	14
Hepatic and biliary disorders	7 (2)	4 (1)	11
Immunosuppressed	1 (<1)	4 (1)	5
Post operative	3 (1)	2 (1)	5
Multiple pathologies	48 (13)	32 (12)	80
Other pathologies	14 (4)	10 (4)	24
Not specified	7 (2)	1 (<1)	8
Total	357	276	633

Source: Health Protection Agency Listeria database