Analysis of the surveillance situation for viral encephalitis and meningitis in Europe

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Infective processes in the brain, spinal cord and meninges are considered to be the main causes of encephalitis, myelitis and meningitis. However, most cases remain unexplained. The incidence of different viral aetiologies (zoonotic and non-zoonotic) is especially poorly estimated, due to the lack of a standard case definition and of agreed diagnostic algorithms, including harmonised diagnostic methods and sample collection. It is important to clarify the incidence of viral encephalitis/meningitis and to optimise the diagnosis of infectious neurological illness, particularly to ensure early recognition of outbreaks or emerging infections such as West Nile encephalitis. The European Network for Diagnostics of 'Imported' Viral Diseases (ENIVD) has analysed the present surveillance situation for viral encephalitis/meningitis in Europe. Here we give an overview of the existing epidemiological sources of information in European Union (EU) Member States, mapping the laboratory capacity and identifying key requirements for a possible future surveillance study at European level. The data presented will help design a harmonised/standardised Europe-wide surveillance study investigating patients with encephalitis and/or meningitis in order to obtain more information on the role of infections in these rarely analysed syndromes, both from a clinical and an epidemiological perspective.

Introduction

Encephalitis is an irritation and inflammation of the brain parenchyma, associated with clinical evidence of brain dysfunction [1]. It often coexists with inflammation of the covering membranes of the brain and spinal cord (meningo-encephalitis). Meningeal irritation (e.g. fever, headache, general malaise, vomiting) and somnolence are signs of meningitis, while behavioural, cognitive and focal neurological symptoms and seizures are signs of the disruption of brain function. Like meningitis, encephalitis can be caused by a wide variety of infectious agents, including viruses, bacteria, fungi and parasites (Table 1). Those cases of aseptic encephalitis for which the aetiology can be determined are most often caused by viral infections: herpes simplex viruses (HSV), varicella-zoster virus (VZV), Epstein-Barr virus (EBV), mumps virus, measles virus and enteroviruses are considered to be the major causes of viral encephalitis in immunocompetent individuals worldwide [2-5]. In addition to these common pathogens, which occur throughout Europe, arthropod-borne viruses (transmitted through insect and tick bites) can cause arboviral encephalitis with similar symptoms as herpes simplex encephalitis [6]. In Europe, the most important pathogens responsible for arboviral encephalitis are tick-borne encephalitis virus (TBEV), West Nile virus (WNV) and Sandfly fever virus (SFV) [7]. Important non-arthropod-borne viral zoonotic pathogens affecting the central nervous system (CNS) are lymphocytic choriomeningitis virus (LCMV), rabies virus and Nipah virus. In regions where they are endemic, illness due to these pathogens may be correctly diagnosed because clinicians will consider them in their differential diagnosis. However, it is more than likely that incursions of these viruses (with the probable exception of rabies virus) into new regions would not be diagnosed unless the number of cases increased to unusual levels. A fact sheet concerning epidemiological, clinical, diagnostic and treatment data for the most important viruses that may cause (meningo-) encephalitis is available at ENIVD's website, http://www.enivd.org.

Despite improvements in the diagnosis of viral encephalitis, including cerebrospinal fluid (CSF) PCR [8], the aetiology of up to 75% of encephalitis cases remained unknown in recent surveys [4]. This issue is challenging when considering early detection of new and (re-) emerging pathogens such as WNV [6,9] or potential outbreaks caused by deliberate release of pathogens [10]. An accurate diagnosis is important for surveillance activities aimed at clarifying the aetiological pattern of viral encephalitis/meningitis. However, this is impossible to achieve as long as routine investigations do not include the most common pathogens in a standardised manner. Moreover, a correct (differentiated) immediate diagnosis and the introduction of symptomatic or specific therapy may have a decisive influence on survival of patients, and may reduce the extent of brain injury.

Four studies are currently being conducted in Europe, aimed at clarifying the incidence of viral encephalitis/meningitis in humans at national level and obtaining more valid clinical and epidemiological data. Details on these studies are available from the following publications and websites:

1. A multi-centre prospective study to clarify the aetiology of encephalitis in England (2005-2008): http://www.hpa.org.uk/infections/topics_az/encephalitis/study.htm

TABLE 1

The most important infections causing central nervous system disease*

Meningitis	Encephalitis/ Meningo-encephalitis
Viral (aseptic meningitis)	**Viral**
Enteroviruses	Herpes simplex virus
Tick-borne encephalitis virus and other arboviruses†	Varicella-zoster virus
Mumps virus	Epstein-Barr virus
Herpesviruses	Mumps virus
Human immunodeficiency virus	Measles virus
Influenzaviruses	Enteroviruses
Parainfluenza virus	West Nile virus
Measles virus	Tick-borne encephalitis virus
Rotavirus	Other arboviruses†
Lymphocytic choriomeningitis virus	Human immunodeficiency virus
	Rabies virus
Bacterial (septic meningitis)	**Bacterial**
Haemophilus influenzae b	*Listeria monocytogenes*
Neisseria meningitidis	*Mycobacterium tuberculosis*
Streptococcus pneumoniae	*Mycoplasma pneumoniae*
Staphylococcus spp.	*Borrelia* spp.
Streptococcus spp.	*Rickettsia* spp.
Leptospira spp.	
Treponema pallidum	
Mycobacterium tuberculosis	
Borrelia spp.	
Fungal	**Fungal**
Cryptococcus neoformans	*Cryptococcus neoformans*
Parasitic	**Parasitic**
Acanthamoeba spp.	*Acanthamoeba* spp.
Toxoplasma gondii	*Naegleria* spp.

* adapted from: www.meduniwien.ac.at/hygiene; www.enivd.de/ENCDISEASES/fs_encdiseases.htm

† Arbovirus = arthropod-borne virus (e.g. Toscana virus)

2. The Meningitis/Encephalitis registration study in Lower Saxony, Germany (MERIN, 2003-open) and the German enterovirus surveillance study (2005-2007): http://www.nlga.niedersachsen.de
3. Epidemiological study to optimise the diagnosis and prognosis of encephalitis infections in France (2007): http://www.invs.sante.fr/surveillance/encephalites_2007/default.htm
4. A systematic laboratory-based surveillance of unexplained neurological illness to rule out flavivirus infection in The Netherlands [9].

ENIVD's current project involves a preliminary survey regarding the epidemiological situation of viral encephalitis in EU Member States. It is meant to identify the requirements for a possible future surveillance study at European level, as well as to improve the diagnostic methods and to carefully monitor the present situation especially regarding WNV, TBEV and SFV as potential emerging arboviral causes of encephalitis. The activities concerning the improvement of diagnostics and surveillance data planned by the individual ENIVD working groups for those arboviral pathogens will be presented in separate publications. In this study, the ENIVD working group for viral CNS diseases presents the results of a preliminary survey of the existing surveillance systems in Europe. A number of important issues are considered that will need to be addressed when designing a surveillance study on the aetiological pattern of viral encephalitis/meningitis at European level.

Methods

This preliminary data survey was performed from May 2006 to April 2007. PubMed (the United States' National Library of Medicine) was searched for relevant papers published between 1996 and 2006. The search terms selected were: "meningitis/encephalitis survey", "meningitis/encephalitis surveillance", "meningitis/encephalitis study", "meningitis/encephalitis epidemiology", and "meningitis/encephalitis diagnostics". Furthermore, epidemiological data were collected via internet searches or requested from national contact points by e-mail. The data were reported by national and/or regional public health authorities for infectious disease control (ministries of health, public health institutes and/or reference laboratories) or other organisations or networks (e.g. the International Scientific Working Group on Tick-borne Encephalitis) focussing on pathogens affecting the CNS. We decided to search/ask only for data from 2004 because this was the most recent year for which all datasets were completed and proofed. We focussed on "reported cases of bacterial meningitis/encephalitis", "reported cases of viral meningitis/encephalitis", and "reported cases of other or unknown aetiology".

The data were collected, analysed and verified by the national contact points in order to:

- gain an overview of the epidemiological situation in the EU Member States,
- identify existing resources that would be available in the event of a surveillance study (e.g. surveillance systems, public health institutes, clinical networks, hospitals, officially appointed laboratories, epidemiologists),
- review in particular the data on the causes of unknown aetiologies,
- and develop hypotheses on the reasons why these aetiologies are unknown.

Moreover, an expert meeting on diagnostics and surveillance of viral (meningo-) encephalitis held in Berlin in April 2006 provided information on previous, ongoing or planned national studies in six Member States that dealt with the incidence of the most relevant aetiologies of viral encephalitis/meningitis. The experiences gained from these studies are summarised here and should be taken into account in a possible future European surveillance study. This work included the selection of suitable partner institutions and clarification of whether samples would be available for further diagnostic investigation with special regard to the manner of sample collection. Furthermore, we defined the sample numbers necessary for such a study as well as established and evaluated diagnostic assays for the detection of different encephalitis-causing viral pathogens.

Results

The epidemiological situation of CNS diseases in Europe

The most recent epidemiological situation regarding CNS infections/syndromes in the 27 EU Member States (EU-27), based on disease notifications in 2004, is shown in Table 2. Bacterial causes of meningitis/encephalitis are thoroughly investigated in all Member States, at least judged by the presence of well-established surveillance infrastructures [11], and data were provided for all countries except for Belgium. In contrast, notification of viral meningitis/encephalitis cases differs between the countries because reporting policies are neither standardised nor rigorously enforced.

Although geographical differences in the occurrence of viral pathogens (either more common or endemic viruses) are likely to play a role, the variation in the incidence of viral meningitis/encephalitis across Europe that was seen in this survey is probably due to differences in the surveillance systems. One reason could be the lack of a Europe-wide standard case definition for viral CNS syndromes. Moreover, the spectrum of relevant viral pathogens reported in the surveillance systems depends on the spread of the diagnostic panels and/or notification regulations, and is therefore also very divergent.

The available diagnostic information was poor. Only few countries – namely Austria, the Czech Republic, Hungary, Poland, Slovakia, and Slovenia – could provide pathogen-specific data for more common (e.g. Herpesviruses) and endemic (e.g. TBEV) viral aetiologies. Those countries have or had a special endemic situation, and consequently a higher awareness of arboviral CNS diseases (in particular tick-transmitted ones). They may also be countries with a stronger interest to differentiate between more common and endemic aetiologies whose clinical pictures can be very similar.

Other countries only reported pathogen-specific data for major arboviral neurological diseases, like TBEV (e.g. the Baltic States, Germany and Finland) and WNV infections (e.g. Romania), without further differentiated reporting of other more common causes. Although endemic in several European countries, TBEV surveillance is not uniform nor always mandatory in Europe [12].

Only 15 (56%) of the 27 Member States provided some level of information on unexplained neurological illnesses of possible infectious aetiology. The lack of information on non-notifiable CNS syndromes in the other 12 countries indicates a data gap in surveillance. It is likely that more information is available on regional level or from surveys. This may be the case for enteroviruses, since all countries are obliged to document the absence of poliovirus circulation as part of the global eradication effort, but the data are not always publicly available. The Netherlands, for instance, has a continuous laboratory-based enterovirus surveillance that processes approximately 3,000 samples per from patients with meningitis per year. On average, 10% of those samples contain enterovirus. In addition, 400 cases are hospitalised with suspected viral meningitis in the Netherlands annually, 60 with suspected viral encephalitis, and 255 with encephalitis of unknown origin. "Suspected" means that the diagnosis derived from CSF could not be confirmed by virus detection or serology. However, the lack of data regarding the proportion of cases with other or unknown aetiology in the official notification report also makes a comparison among European countries difficult.

Existing expertise on CNS diseases in Europe

Based on literature and internet searches, we compiled a database of the specific diagnostic and/or epidemiological capacities and functions in European institutions and microbiological reference laboratories. The database has been updated regularly since the beginning of 2006, and includes 112 reference laboratories from the 27 EU Member States, covering the main bacterial and viral aetiologies of CNS infections (Table 3).

The number of staff employed in the diagnosis and control of infectious diseases who also handle pathogens that cause CNS disease ranged from five to 419 in the different Member States (including microbiologists and epidemiologists). The size of the groups can vary, depending on whether single groups/units or whole departments were described. The people working in these departments are often responsible for more than one kind of pathogen or disease. We have compiled a contact database with postal and e-mail addresses that also includes detailed information on the groups' capacities. The information can be provided on request.

The Czech Republic and Germany have the largest number of reference laboratories for pathogens causing CNS disease, followed by France and Belgium. Cyprus, Germany, Portugal and the United Kingdom have groups specialised in the diagnostics of viral CNS infections and syndromes. Of the 112 identified laboratories, 72 (64%) provide training activities for students and/or professional personnel. Seventy-eight laboratories (70%) organise and/or participate in external quality assurance (EQA) studies. However, only 31 (28%) laboratories were involved in outbreak investigations.

The most frequently reported techniques/activities with respect to the reference pathogens/diseases are antibody detection (69% of laboratories), molecular detection (of nucleic acid) (69%), typing/

Table 2

Epidemiological data: notifications of meningitis and encephalitis in Europe (EU-27) caused by bacterial and viral agents, reported in 2004*

Member State	Total population (x1000)[a]	Reported cases of bacterial meningitis/encephalitis (incidence/100,000)	Reported cases of viral meningitis/encephalitis (incidence/100,000)	Cases of other or unknown aetiology	Reference
Austria	8,171	**total: 126** mostly MNC (1.02); PNC (0.42)	**total: 59** TBE (0.66); Herpes (0.04); Measles (0.02)	20 bacterial meningitis; 14 viral meningo-encephalitis	Federal Ministry of Health, Family and Youth, BMGFJ, Austria
Belgium	10,400	not available[$]	not available[$]	not available[$]	Scientific Institute of Public Health, IPH, Belgium
Bulgaria	7,780	**total: 95** mostly MNC[b] (0.46); PNC (0.40)	**total: 699** without further information[c]	163 bacterial meningitis	National Centre of Health Informatics, NCHI, Bulgaria
Cyprus	826	**total: 5** MNC (0.61)	**total: 20** without further information[c]	9 other bacterial meningitis	Ministry of Health, Republic of Cyprus
Czech Republic	10,229	**total: 119** MNC[b] (0.96); HIB[b] (0.21)	**total: 667** TBE (4.96) ; EV (1.56)	166 other bacterial meningitis; 668 viral meningo-encephalitis	National Reference Centre for analysis of epidemiological data, NRC/SZU, Czech Republic
Denmark	5,414	**total: 266** mostly PNC (1.87); NB (1.57)	no cases reported[c]	26 bacterial meningitis	Public Health Institute, SSI, Denmark
Estonia	1,335	**total: 18** HIB (0.97); MNC (0.38)	**total:182** TBE (13.63)	18 other and unknown aetiology	Health Protection Inspectorate, Estonia
Finland	5,235	**total: 1,117** mostly STC[b] (20.48); MNC[b] (0.84)	**total: 29** TBE (0.55)	not available	National Public Health Institute, KTL, Finland
France	60,257	**total: 1,439**[†] mostly STC (1.46); MNC (0.73)	no cases reported[c]	not available	National Public Health Institute, InVS, France
Germany	82,645	**total: 531** mostly MNC (0.46); NB (0.6)[‡]	**total: 125** TBE (0.15)	not available	Robert Koch Institute, RKI, Germany
Greece	11,098	**total: 168** mostly MNC (0.80); PNC (0.64)	**total: 199** without further information[c]	376 bacterial meningitis / encephalitis; 177 other and unspecified aetiologies	Hellenic Centre for Infectious Diseases Control, KEEL, Greece
Hungary	10,124	**total: 201** mostly PNC (0.75); MNC (0.42)	**total: 122** TBE (0.75); Herpes (0.16); EV (0.15); WNV (0.03) etc.	64 infectious encephalitis; 148 meningitis	National Centre for Epidemiology, OEK, Hungary
Ireland	4,080	**total: 225** mostly MNC[b] (4.85); PNC (0.54)	**total: 28** without further information[c]	36 other bacterial meningitis	Health Protection Surveillance Centre, HPSC, Ireland
Italy	58,033	**total: 748** mostly MNC (0.59); STC (0.58)	**total: 434** without further information[c]	236 bacterial meningitis	National Public Health Institute, ISS, Italy
Latvia	2,318	**total: 25** MNC[b] (1.04); HIB[b] (0.04)	**total: 251** TBE (10.83)	not available	State Agency "Public Health Agency", SVA, Latvia
Lithuania	3,443	**total: 101** MNC[b] (2.67); HIB[b] (0.26)	**total: 425** TBE (12.34)	not available	Centre for Communicable Disease Prevention and Control, ULPKC, Lithuania
Luxembourg	459	**total: 0** no cases in 2004, in previous years *N. meningitidis*	no cases reported[c]	not available	Ministry of Health, Health Management, Luxembourg
Malta	400	**total: 15** mostly PNC (1.75); MNC (1.00)	**total: 2** without further information[c]	5 bacterial meningitis	Ministry of Health, Public Health Department, DSU, Malta
Poland	38,559	**total: 433** mostly MNC (0.31); HIB (0.19)	**total: 308**[§] TBE (0.68)	512 bacterial, 1,119 viral meningitis/encephalitis; 353 other and unspecified aetiologies	National Institute of Hygiene, PZH, Poland
Portugal	10,441	**total: 91** MNC (0.81); HIB (0.06)	no cases reported[c]	not available	National Public Health Institute, DGS, Portugal
Romania	21,790	**total: 467** MNC (1.13)	**total: 989**[c] WN meningitis (0.01)	not available	Institute of Public Health, ISPB, Romania
Slovakia	5,401	**total: 81** MNC (0.58); PNC (0.41)	**total: 207** TBE (1.29); Herpes (0.19); VZV (0.17) etc.	103 bacterial meningitis; 40 unknown viral meningitis / encephalitis	Public Health Authority of the Slovak Republic
Slovenia	1,967	**total: 29** mostly STC (0.81); MNC (0.31)	**total: 232** mostly TBE (10.37); Herpes (0.76)	25 bacterial meningitis / encephalitis; 187 unknown viral meningitis / encephalitis	Public Health Institute of the Republic Slovenia
Spain	42,646	**total: 881** MNC[b] (2.22)	no cases reported[c]	not available	National Public Health Institute Carlos III, ISCIII-CNE, Spain
Sweden	9,008	**total: 565** mostly PNC[b] (4.66); HIB[b] (0.89)	**total: 222** without further information[c]	not available	Institute for Infectious Disease Control, SMI, Sweden
The Netherlands	16,226	**total: 297** MNC[b] (1.83)	no cases reported[c]	not available	National Institute of Health and the Environment, RIVM, Netherlands
United Kingdom	59,479	**total: 916** mostly MNC (0.93); PNC (0.29)	**total: 217** without further information[c]	294 meningitis	Health Protection Agency, HPA, UK

* As reported by the national/regional public health authorities for infectious disease control (ministries of health, public health institutes, reference laboratories). When comparing the data across countries, please note that reporting criteria may vary. Incidence rates were calculated per 100,000 inhabitants.

[a] General public health statistics: http://www.who.int/about/regions/euro/en/index.html
[b] Classified as invasive bacterial disease with a broad case definition (including cases of meningitis and septicaemia).
[c] TBE is not a notifiable disease in this country.
[$] Data are not available due to incomplete surveillance network.
[†] Number of cases adjusted for the coverage of a clinical laboratory network as well as corrected for under-notification and incidences calculated per 100,000 inhabitants for meningitides with or without bacteraemia, Epibac 2004, Metropolitan, France (http://www.invs.sante.fr/surveillance/epibac/default.htm).
[‡] Incidence is based on the notification from six federal states (Berlin, Brandenburg, Mecklenburg Western Pomerania, Saxony, Saxony-Anhalt and Thuringia) with a total of 13,433,358 inhabitants.
[§] Until 2005 meningitis in the course of other infectious diseases such as mumps were not reported.

EV: Enteroviruses; HIB: *H. influenzae* type b; MNC: Meningococci; NB: Neuroborreliosis; PNC: Pneumococci; STC: *Streptococcus* spec.; TBE: Tick-borne encephalitis; VZV: Varicella-zoster virus; WN(V): West Nile (virus).

TABLE 3

List of existing resources for the surveillance of meningitis and encephalitis in Europe (EU-27) (data as reported)*

Member State	Number of ref. labs	Pathogens-	Number of staff involved	Number of labs providing training	Number of labs involved in EQA	Number of labs involved in outbreak situation
Austria	5	LB; HIB; MMR; MNC; PNC; Polio virus; TBEV; other relevant viruses	60	5	4 (1)[a]	2
Belgium	10 (2)[a]	LB; EV; HIB; Morbilliviruses; MNC; Polio virus; Rabies virus; PNC; TBEV; WNV	> 98	6 (2)[a]	6 (4)[a]	1 (2)[a]
Bulgaria	9	LB; MNC; STC; EV; MMR; Herpesviruses; other pathogens	56	3	7 (2)[a]	1
Cyprus	2[†]	EV, Herpesviruses (viral meningitis); other relevant pathogens (MNC, PNC, HIB)	> 5	1	not reported	not reported
Czech Republic	14	STC; MNC; HIB; Herpesviruses; MMR; EV; LB; Arboviruses (incl. TBEV); other relevant pathogens	172	11	12 (2)[a]	1
Denmark	2	EV; MMR; other relevant viruses; PNC; other relevant bacteria	241	2	2	1
Estonia	3 (1)[a]	MNC; HIB; LB; other bacteria; TBEV; EV; other viruses	25	not reported	2 (1)[a]	not reported
Finland	4	PNC; MNC; HIB; EV; MMR; *M.tuberculosis*; Arboviruses; other relevant pathogens	375	1	4	1
France	10	Arboviruses; LB; EV; HIB; *Listeria*; Measles virus; MNC; PNC; Rabies virus; STC	154	5	5 (5)[a]	3
Germany	11	LB; MNC; STC; MMR; EV; HIB; Herpes virus; VZV; Rabies virus; TBEV; viral CNS infections	175	7	6 (5)[a]	4
Greece	3 (1)[a]	MNC; STC; HIB; other relevant viruses	> 16	1(1)[a]	2(1)[a]	2(1)[a]
Hungary	2	MNC; EV; other relevant pathogens	49	2	2	not reported
Ireland	2 (1)[a]	MNC; HIB; other relevant viruses	> 17	1(1)[a]	1(1)[a]	1(1)[a]
Italy	2	LB; EV; HIB; MNC; STC; other relevant pathogens	419	2	1(1)[a]	1
Latvia	1	TBEV; LB; Herpes virus; CNS bacterial infections	> 35	1	1	not reported
Lithuania	1	LB; EV; TBEV; Herpes virus; Measles virus; other relevant viruses	24	not reported	1	not reported
Luxembourg	1	MNC; HIB; Measles virus; other relevant pathogens	25	1	not reported	not reported
Malta	1	PNC; HIB; MNC; other relevant pathogens	11	1	not reported	1
Poland	3	STC; Herpesviruses, EV; Arboviruses; bacterial meningitis (incl. MNC and HIB)	> 4	3	2 (1)[a]	not reported
Portugal	3	MNC; HIB; Viral CNS infections; vector-borne pathogens (i.e. *Borrelia*, WNV); other relevant pathogens	144	3	3	3
Romania	1	LB; STC; MNC; HIB; vector-borne diseases; other relevant pathogens	31	1	1	not reported
Slovakia	3	MNC; HIB; Arboviruses; other relevant pathogens	152	1	2(1)[a]	not reported
Slovenia	2 (1)[a]	MNC; HIB; Arboviruses; other relevant pathogens (incl. TBEV, STC)	> 92	1(1)[a]	1(1)[a]	not reported
Spain	6	MNC; PNC; HIB; Herpes-, Entero-, and Arboviruses; other relevant viral pathogens	18	6	6	6
Sweden	3	MNC; other relevant pathogens	404	2	3	not reported
The Netherlands	2	Bacterial meningitis; other relevant pathogens (incl. arboviruses)	> 3	1	1(1)[a]	1
United Kingdom	6 (1)[a]	LB; MNC; STC; HIB; viral CNS infections; other relevant pathogens	> 100	4	3 (3)[a]	3

* Represents laboratories officially designated as reference laboratories (Ref. labs) for the specific pathogens/diseases, or laboratories that act as national reference centres without being officially recognised as such. Even though these laboratories are considered as a resource at national/international level by their national public health authorities, the definition of "laboratory" can vary across countries as it can include groups of different size. The number of laboratories *per se* should therefore be read with caution and additional information should be sought.

[a] Not all laboratories have presented data regarding their capacities/activities. The number in brackets shows the number of laboratories without further information (partial/complete).

[†] Reference laboratory services for MNC are done by a group in another Member State.

EQA: external quality assurance; CNS: Central nervous system; EV: Enteroviruses; HIB: *H. influenzae* type b; LB: Lyme borreliosis; MMR: Measles, Mumps and Rubella; MNC: Meningococci; PNC: Pneumococci; STC: *Streptococcus* spec.; TBEV: Tick-borne encephalitis virus; VZV: Varicella-zoster virus; WNV: West Nile virus

TABLE 4

Overview of six different studies at national level to clarify the aetiology of viral encephalitis/meningitis

Country	Status of study	Number of cases	Pathogens	Type of samples	Applied diagnostic	Unknown aetiology	Further investigation
Finland	Recently completed† Period: 1995-1996	3,231	VZV HSV Enteroviruses Influenza A virus HHV-6 TBEV Puumala virus Inkoo ortho-bunyavirus	CSF Sera	• CSF-PCR • Intrathecal antibody screen by EIA • Systemic sero-conversion measure • Multiplex-PCR and oligionucleotide microarray (new)	30-40 %	• aetiology of aseptic meningitis in an adult population • viral aetiology of CNS infections in children • viral CNS infections in adults
United Kingdom	Ongoing Period: 2005-2008	100 / year	HSV VZV EBV Mumps virus Measles virus Enteroviruses Arboviruses	CSF Blood Throat/naso-pharyngeal swab Faeces Post-mortem tissue	• CSF-PCR • Serology • Intrathecal antibody screen • Generic amplification for unknown and unrecognised infections (e.g. SISPA)	60%	• implement a special pathogen branch
The Netherlands	Recently completed‡ Period: 1999-2003	1,276	Herpesviruses Enteroviruses Adenovirus Measles virus Mumps virus	CSF	Broad variability of lab tests	59%	• enhanced ongoing surveillance for WNV (and other arboviruses) with approx. 300 CSF samples per year
Germany	Ongoing MERIN: 2003-open National Enterovirus-Surveillance: 2005-2007	1,191 (2003-2006) 514 (April 2006)	*Borrelia* Enteroviruses HSV VZV Adenoviruses Influenza A/B virus EBV CMV Mumps virus others on special request (e.g. TBEV)	CSF Faeces Sera Throat swab	• PCR • Serology • Intrathecal antibody screen • CFT • Virus isolation/ typing	66% (MERIN)	• further promotion of the MERIN project • further romotion of the national project
France	Planned: 2006-2009 (incl. follow-up)	ca. 600 /year	HSV VZV EBV HHV-6 Enteroviruses Adenoviruses CMV Influenza A/B virus Measles/ Mumps virus Arboviruses (WNV, Toscana virus, TBEV)	CSF Blood Sera Throat swab Urine	• PCR • Serology	80 %§	• project started 2007 • promotion open
Spain	Planned: for one year	600 (adults) 400 (children)	Herpesviruses Enteroviruses Adenoviruses Measles/ Mumps virus Toscana virus WNV and other flaviviruses LCMV and rabies virus	CSF Sera	• Generic PCR • Serology • Intrathecal antibody screen	unknown rate	• project not fixed

† Ref. [5]; ‡ ISIS database, RIVM, The Netherlands; § Data from PMSI and InVS, France.

CMV: Cytomegalovirus; EBV: Epstein-Barr virus; HHV-6: Human herpes virus 6; HSV: Herpes simplex virus; LCMV: Lymphocytic choriomeningitis virus; TBEV: Tick-borne encephalitis virus; VZV: Varicella-zoster virus; WNV: West Nile virus.

CFT: Complement fixation test; CNS: Central nervous system; CSF: Cerebrospinal fluid; EIA: Enzyme immunoassay; SISPA: Sequence independent single primer amplification.

subtyping (64%), antibiotic resistance/immunity testing (55%), isolation of reference pathogens (53%), microbiological analyses (46%), antigen detection (40%), providing reference material, e.g. diagnostic reagents (23%) and electron microscopy (11%) (data not shown).

Key conditions for a future surveillance study at the European level

Based on the expert meeting in Berlin in 2006, data were obtained from previous, ongoing or planned studies in six Member States in order to clarify the incidence of the most relevant aetiologies of viral encephalitis/meningitis at national level. Table 4 shows a broad variability among these studies concerning the pathogens they focussed on, the type of samples they used, the diagnostic methods they applied and the determined or calculated rate of unknown aetiology.

The consensus was that comparative data for the incidence of most viral agents of human (meningo-) encephalitis is missing. The proportion of cases with unknown aetiology ranged from 30% to 80% in the presented studies. The reasons for unknown diagnosis could be traced to either a failure of the diagnostic tests or an inappropriate case definition, resulting in under-ascertainment of both known viruses and "new" viruses.

The following issues were considered during the expert meeting in order to design a possible future surveillance study for viral (meningo-) encephalitis at the European level:

Case definition

The diagnosis of encephalitis is often difficult to establish, since many other clinical conditions may mimic encephalitis. In addition, several arboviruses can cause a range of neurological syndromes, including meningitis or paralytic illness. Therefore, a broad case definition will be necessary in order to capture all relevant cases of acute and suspected CNS diseases (meningitis, encephalomyelitis and encephalitis) in the first stage of a study. To date, there is no standard clinical case definition that includes all relevant types of infectious CNS diseases, although this would be practical from a clinical perspective. A limited case definition (e.g. one that excludes signs of aseptic meningitis) could lead to under-ascertainment of relevant cases. A distinction between the different disease types with the final goal of identifying a specific aetiological agent could be achieved in following processes. To harmonise the clinical and diagnostic approaches in a European study, all personnel involved in case notification should be informed of such a standard case definition and should be provided with a protocol for stringent data management and diagnostic algorithms. It may become necessary to adapt specific case definitions to the situation in different countries, for example if certain pathogens are endemic in some but not other areas. Therefore, the EU case definitions currently being finalised by the European Centre for Disease Prevention and Control (ECDC) should be taken into account.

Sample collection and storage

Regarding the collection of samples, it should be considered that other types of samples besides CSF (e.g. sera, faeces, throat swabs) are also important when trying to detect a broad spectrum of relevant pathogens. Basic clinical information should always be provided with the sample material. A minimum dataset should include: age, gender, domicile of the patient, date of onset/duration of the complaints, type of complaints, travel history, vaccination history (e.g. against yellow fever, Japanese encephalitis) and context of the current epidemiological situation (e.g. outbreak, cluster).

Follow-up studies may become necessary, for example if clinical intervention measures become available in the future or new pathogens are discovered. Therefore, samples of selected cases should be shipped to a central archive, aliquoted and stored at -70°C to avoid damage of the material by frequent thawing. Sample collection along with the recommended minimum dataset will be a valuable resource for later analysis of patients' and diagnostic profiles.

Diagnostic issues

A three-step model is suggested for diagnostic procedures in order to ensure comprehensive diagnostic investigation. The first step should include the local medical investigation and usual analysis (PCR and serology) of acute cases by clinical laboratories. Clinical and epidemiological features (e.g. occupation, travel history or animal contact) should be collected at this level for differential diagnostic approaches. The second step comprises the extended analysis of suspected cases by reference laboratories for commonly recognised causes of (meningo-) encephalitis, and of less commonly recognised and travel-related causes when indicated. The third step includes the identification of specific pathogens (e.g. by new typing methods) in cases of unknown aetiology, as well as the collection of selected samples by reference laboratories and storage in a centralised archive for future use.

Standard operating procedures for testing should be shared among all participating laboratories and regularly monitored by EQA programmes to ensure diagnostic consistency. In its current project, the ENIVD has begun EQA studies for the diagnostics of TBEV and WNV [13-15], and further studies on arboviral aetiologies of CNS diseases are planned [16]. Moreover, it might be advantageous to consider the new multiplex-microarray technology presented in the Finnish study (see Table 4) [17,18] in a broad European study on viral CNS diseases. This would guarantee a unique analysis platform for all participating laboratories by including the more common pathogens of viral CNS diseases (e.g. HSV, VZV, enteroviruses) as well as relevant viral zoonotic agents (e.g. TBEV, WNV, rabies virus) according to the regional/endemic situation of the European countries or on special request.

Data management

A prerequisite for a surveillance study at European level – in which data from numerous countries are pooled – seems to be the establishment of a central hub recording and managing the entire study data (patients' clinical and epidemiological data, and CNS diagnostic data). To ensure standardised reporting, ICD-10 coding is recommended in addition to the broad case definition.

A general problem in most of the national studies presented here was the failure of clinicians to report clinical cases. The contribution of the individual clinicians regarding the provision of clinical data and material varied greatly depending on hospital and medical branch (paediatricians, for example, seemed to be more cooperative than neurologists). Efforts to reach a final aetiological diagnosis are not always considered essential, for instance when all patients diagnosed with viral encephalitis are routinely given the same antiviral therapy. The success of a study depends on the voluntary cooperation of hospitals and clinicians. One of the important issues is to motivate them, for example by offering clinicians and public health officers open access to evaluated and updated study data on the internet or free diagnostic tests.

Ethical and data protection issues

Ethical issues regarding patient data will be especially relevant in follow-up studies. These concerns might not be important during the first contact when diagnostic analysis for the aetiology of CNS disease is requested by clinicians. Nevertheless, all further analysis will require a patient agreement. It became clear during the expert meeting that this is handled quite differently in the European countries. This aspect therefore needs special attention in planning a European study and should be discussed with the different European public health authorities.

Number of samples necessary for a European study

According to the data available, the UK study recorded approximately 100 cases of viral encephalitis per year. This covers an estimated 60% of all cases of viral CNS disease in England. The experts give a ratio of 1:2:0.5 cases for encephalitis:meningitis:encephalomyelitis. The true number of all cases of viral CNS disease, based on a broad case definition as recommended above, is therefore five- to six-fold higher than the number of recorded cases. For the UK study, this was calculated to be approximately 700 cases. Based on this calculation, the expected number of viral CNS disease cases in a given country can be estimated by taking

Table 5

Estimated number of cases for a possible future study on viral central nervous system diseases in Europe*

Country	Population (x1000)[a]	Total expected number of cases per year	Number of cases per 10^5 inhabitants	Number of cases for 60 % coverage
England	50,431[†]	700	1.39	420
Austria	8,189	340[b]	4.15	200
Belgium	10,419	145	1.39	90
Bulgaria	7,726	990[b]	12.81	590
Cyprus	835	80[b]	9.58	50
Czech Republic	10,220	3,780[b]	36.97	2,270
Denmark	5,431	80	1.47	50
Estonia	1,330	250[b]	18.79	150
Finland	5,249	1,750[‡]	33.34	1,050
France	60,496	4,170[§]	6.89	2,500
Germany	82,689	1,800	2.18	1,080
Greece	11,120	275[b]	2.47	165
Hungary	10,098	370[b]	3.66	220
Ireland	4,148	100[b]	2.41	60
Italy	58,093	1,150	1.98	690
Latvia	2,307	310[b]	13.44	190
Lithuania	3,431	530[b]	15.45	320
Luxembourg	465	7	1.50	4
Malta	402	6	1.50	4
Poland	38,530	2,500[b]	6.49	1,500
Portugal	10,495	150	1.43	90
Romania	21,711	930[b]	4.28	560
Slovakia	5,401	430[b]	7.96	260
Slovenia	1,967	1,000[b]	50.84	600
Spain	43,064	850	1.98	510
Sweden	9,041	180	1.99	110
The Netherlands	16,299	1,330[$]	8.16	800
				Expected total: ~ 15.000

* Including encephalitis, meningitis, encephalomyelitis.

[a] With the exception of England, data adapted from: http://www.who.int/about/regions/euro/en/index.html (actual numbers)

[b] Estimation adapted to data from existing infectious disease reports of last years ('04, '05 and/or '06), as available

[†] Source: http://www.statistics.gov.uk/CCI/nugget.asp?ID=6 (last mid-year population estimates from UK)

[‡] Estimation based on data from Ref. [5].

[§] Estimation based on data as presented for the French study.

[$] Estimation based on data as presented for the Dutch study.

into account the respective population (with or without adaptation to existing differentiated epidemiological data) (Table 5).

The calculated numbers are only rough estimations, but can be used as a general indication of how many samples, material and work would eventually be necessary to cover approximately 60% of all cases in the respective countries. Thus, these estimates may have a limited relevance to the actual incidence of viral CNS diseases in any country.

However, the data in Table 5 show that a possible future study on viral CNS diseases at European level could be extensive regarding samples, logistics, material and costs, if all cases were analysed for all relevant aetiologies including differential diagnostics.

Discussion

The incidence of most viral agents of human (meningo-) encephalitis is not estimated well by the surveillance systems of the various European countries. This harbours the risk that potential emerging infectious diseases, such as West Nile fever, will not be recognised in time by the existing surveillance infrastructures [19]. Pooling data from several countries may help identify and monitor emerging problems more quickly. Establishing a European surveillance system for viral encephalitis/meningitis by bundling the existing resources and introducing a harmonised/standardised reporting and diagnostic system will be challenging, but is essential, and not only for future preparedness and response issues. With more specific treatments or vaccines becoming available [1,20], it will also be of interest for pharmaceutical and vaccine-producing companies and public health institutions to carefully analyse the epidemiological situation, and to adapt therapeutic interventions as well as prevention strategies accordingly. A broad standard case definition and harmonised/standardised diagnostic algorithm using a multiplex-microarray system validated for a wide range of viruses may help to discover the true incidence and aetiological pattern of viral encephalitis/meningitis within each country. This would guarantee high performance and comparability of the results consistent with EQA programmes. To improve surveillance, it is also important to quantify the extent of cases of unknown aetiology, in order to allow a comparison of the data from each country and to identify possible weaknesses in the surveillance data. Therefore, clinicians must be motivated to report all cases of viral encephalitis/meningitis and to reach a definitive aetiological diagnosis.

A future study on viral CNS diseases at European level could be extensive in work load and costs; an alternative is a survey including only a small number of countries with experts willing to cooperate and to set up such a study, in order to improve the awareness and ascertainment of viral CNS diseases. Partners interested in collaborating in a European survey network on viral CNS diseases have already been identified in 13 countries in different European regions (the Czech Republic, Poland, Russia, Slovakia, Slovenia, France, Spain, Switzerland, Denmark, Finland, Germany, the Netherlands and United Kingdom). It is advisable to use the experience and knowledge of recently completed or ongoing studies at national level (see Table 4) to allow more detailed planning of a prospective European study on viral encephalitis/meningitis.

A European study based on a close cooperation between clinicians, epidemiologists and microbiologists will provide more accurate and timely data on viral CNS diseases which are of public health interest. Such an initiative could help increase case ascertainment, reduce the rate of unknown aetiologies, develop and validate new diagnostic methods, improve recommendations and guidelines, and gain more valuable clinical and epidemiological data for research purposes.

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References

1. Steiner I, Budka H, Chaudhuri A, Koskiniemi M, Sainio K, Salonen O, et al. Viral encephalitis: a review of diagnostic methods and guidelines for management. Eur J Neurol. 2005;12(5):331-43.
2. Glaser CA, Honarmand S, Anderson LJ, Schnurr DP, Forghani B, Cossen CK, et al. Beyond viruses: clinical profiles and etiologies associated with encephalitis. Clin Infect Dis. 2006;43(12):1565-77.
3. Davison KL, Crowcroft NS, Ramsay ME, Brown DWG, Andrews NJ. Viral encephalitis in England 1989-1998: what did we miss? Emerg Infect Dis. 2003;9(2):234-40. Available from: http://www.cdc.gov/ncidod/eid/vol9no2/02-0218.htm
4. Glaser CA, Gilliam S, Schnurr D, Forghani B, Honarmand S, Khetsuriani N, et al. In search of encephalitis etiologies: diagnostic challenges in the California Encephalitis Project, 1998-2000. Clin Infect Dis. 2003;36(6):731-42.
5. Koskiniemi M, Rantalaiho T, Piiparinen H, von Bonsdorff CH, Farkkila M, Jarvinen A, et al. Infections of the central nervous system of suspected viral origin: a collaborative study from Finland. J Neurovirol. 2001;7(5):400-8.
6. Whitley RJ, Gnann JW. Viral encephalitis: familiar infections and emerging pathogens. Lancet. 2002;359(9305):507-13.
7. Kallio-Kokko H, Uzcategui N, Vapalahti O, Vaheri A. Viral zoonoses in Europe. FEMS Microbiol Rev. 2005;29(5):1051-77.
8. DeBiasi RL, Tyler KL. Molecular methods for diagnosis of viral encephalitis. Clin Microbiol Rev. 2004;17(4):903-25.
9. Rockx B, van Asten L, van den Wijngaard C, Godeke GJ, Goehring L, Vennema H, et al. Syndromic surveillance in the Netherlands for the early detection of West Nile virus epidemics. Vector Borne Zoonotic Dis. 2006;6(2):161-9.
10. Bossi P, Tegnell A, Baka A, Van Loock F, Hendriks J, Werner A, et al. BICHAT guidelines for the clinical management of bioterrorism-related viral encephalitis. Euro Surveill. 2004;9(12):1-9. Available from: http://www.eurosurveillance.org/em/v09n12/0912-240.asp
11. Trotter CL, Chandra M, Cano R, Larrauri A, Ramsay ME, Brehony C, et al. A surveillance network for meningococcal disease in Europe. FEMS Microbiol Rev. 2007;31(1):27-36.
12. Günther G, Lindquist L. Surveillance of tick-borne encephalitis in Europe and case definition. Euro Surveill. 2005;10(1):2-3. Available from: http://www.eurosurveillance.org/em/v10n01/1001-221.asp
13. Niedrig M, Linke S, Zeller H, Drosten C. First international proficiency study on West Nile virus molecular detection. Clin Chem. 2006;52(10):1851-4.

14. Donoso Mantke O, Aberle SW, Avsic-Zupanc T, Labuda M, Niedrig M. Quality control assessment for the PCR diagnosis of tick-borne encephalitis virus infections. J Clin Virol. 2007;38(1):73-7.

15. Niedrig M, Avsic T, Aberle SW, Ferenczi E, Labuda M, Rozentale B, et al. Quality control assessment for the serological diagnosis of tick borne encephalitis virus infections. J Clin Virol. 2007;38(3):260-4.

16. Niedrig M, Donoso Mantke O, Schädler R. The European Network for Diagnostics of Imported Viral Diseases (ENIVD) – 12 years of strengthening the laboratory diagnostic capacity in Europe. Euro Surveill. 2007;12(4):E070419.5. Available from: http://www.eurosurveillance.org/ew/2007/070419.asp#5

17. Jääskeläinen AJ, Piiparinen H, Lappalainen M, Koskiniemi M, Vaheri A. Multiplex-PCR and oligonucleotide microarray for detection of eight different herpesviruses from clinical specimens. J Clin Virol. 2006 Oct;37(2):83-90. Epub 2006 Jul 26.

18. Jokela P, Joki-Korpela P, Maaronen M, Glumoff V, Hyypiä T. Detection of human picornaviruses by multiplex reverse transcription-PCR and liquid hybridization. J Clin Microbiol. 2005 Mar;43(3):1239-45.

19. Zeller H, Schuffenecker I. West Nile virus: an overview of its spread in Europe and the Mediterranean basin in contrast to its spread in the Americas. Eur J Clin Microbiol Infect Dis. 2004;23(3):147-56.

20. Chang GJ, Kuno G, Purdy DE, Davis BS. Recent advancement in flavivirus vaccine development. Expert Rev Vaccines. 2004;3(2):199-220.

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