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RIVM report 217617 003

Surveillance of acute respiratory infections in general practices – The Netherlands, winters 1998/1999 and 1999/2000

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Abstract

To provide insight into the virological aetiology of influenza-like illnesses and other acute respiratory infections, nose/throat swabs were taken by 30-35 general practitioners of the sentinel surveillance network of the Netherlands Institute of Health Services Research from a random selection of patients seen for such infections during the winters of 1998/1999 and 1999/2000. The swabs were analysed for respiratory viruses, *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*, at the National Institute of Public Health and the Environment using viral culture and polymerase chain reaction. At least one respiratory pathogen was detected in 56% of the 437 swabs in 1998/1999 and in 55% of the 319 swabs in 1999/2000. The most frequently detected viruses were influenza virus and rhinovirus, occurring in 26% and 15% of the swabs, respectively, combining both winters. An influenza virus was detected three times more often in swabs from patients with an influenza-like illness (i.e. in 32% of the swabs combining both winters) than in swabs from patients with another acute respiratory infection. However, in 29% of the patients with an influenza-like illness, respiratory pathogens other than influenza virus were detected, and in 44% no micro-organisms were detected. Results were compared with those of the six previous winters. Insight into the virological aetiology of acute respiratory infections obtained through this surveillance (the only one in the Netherlands carried out among general practice patients) can contribute to effective prevention and control of such infections, thus constituting a reason for continuing this surveillance.

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Samenvatting

Sinds 1970 tellen huisartsen van het peilstationnetwerk van het Nederlands Instituut voor Onderzoek van de Gezondheidszorg (NIVEL) patiënten die hen consulteren vanwege een griepachtig ziektebeeld. Deze registratie wordt gebruikt om gedurende de winter wekelijks te schatten hoeveel mensen met griep (influenza) of een daarop lijkend ziektebeeld de huisarts consulteren en hoeveel er vóórkomen in de bevolking. Om inzicht te verkrijgen in de ziekteverwekkers verantwoordelijk voor deze acute luchtweginfecties neemt sinds winter 1992/1993 ongeveer 75% van de peilstationartsen een monster uit de neus en keel bij een willekeurige selectie van de patiënten die hen consulteren vanwege een griepachtig ziektebeeld. Deze monsters worden op het Rijksinstituut voor Volksgezondheid en Milieu (RIVM) onderzocht op verschillende ziekteverwekkers. In dit rapport zijn de bevindingen van winters 1998/1999 en 1999/2000 beschreven.

In beide winters werd in ongeveer een kwart van de monsters influenza virus (griepvirus) aangetoond. Rhinovirus (een verkoudheidsvirus) werd in 15% van de monsters gevonden. In 19% van de monsters werden andere verkoudheidsvirussen aangetroffen. In bijna de helft van de monsters kon geen ziekteverwekker aangetoond worden. Een griepachtig ziektebeeld is dus niet altijd het gevolg van een infectie met influenza virus. Ook verkoudheidsvirussen kunnen een ziektebeeld veroorzaken dat op griep lijkt.

Met enkele aannames is geschat hoe vaak infecties met influenza virus voorkwamen in de Nederlandse bevolking. Sinds winter 1992/1993 werden kinderen tussen 5 en 14 jaar het vaakst getroffen, personen van 65 jaar en ouder het minst vaak.

Inzicht in het vóórkomen van griepachtige ziektebeelden en de hiervoor verantwoordelijke verwekkers kan een bijdrage leveren aan effectieve preventie en controle van luchtweginfecties. Resultaten uit deze surveillance bieden huisartsen achtergrondinformatie bij het voorschrijven van medicijnen. Daarnaast voorziet het de overheid van informatie die essentieel is voor het beslissen over de samenstelling van het influenzavaccin (de griepprik). Vandaar dat deze surveillance gecontinueerd zou moeten worden.

Summary

Since 1970, general practitioners (GP's) of the sentinel surveillance network of the Netherlands Institute for Health Services Research (NIVEL) register patients consulting them for influenza-like illnesses (ILI) to calculate the ILI incidence weekly during the winter. To provide insight in the virological aetiology of ILI and illnesses by other acute respiratory infections, about 75% of the general practitioners takes nose/throat swabs from a random selection of patients consulting them for such infections since winter 1992/1993. These swabs are analysed for respiratory viruses, *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*, at the National Institute of Public Health and the Environment (RIVM) using viral culture and polymerase chain reaction (PCR).

In winter 1998/1999, 35 general practitioners sent 437 swabs to the RIVM, 70% of which were obtained from ILI patients. In 56% at least one respiratory pathogen was detected; two pathogens were found in 5% of the swabs. Influenza virus, approximately 50% type A(H3N2) and 50% type B, and rhinoviruses were detected most frequently, namely in 24% and 15% of all swabs. It was a relatively mild influenza season with the ILI incidence in GP patients not exceeding 22 per 10 000 persons per week.

In winter 1999/2000, 30 general practitioners sent 319 swabs to the RIVM. Seventy-five percent of the swabs was obtained from ILI patients. Of all swabs 55% was positive for at least one respiratory pathogen, with 4% containing two pathogens including one triple infection. In 29% of the swabs influenza virus was detected, and in 14% rhinovirus. This winter mainly influenza type A(H3N2) circulated. The maximum ILI incidence in GP patients was 33 per 10 000 persons per week.

In both winters, ILI registration was reasonably in accordance with the number of influenza virus isolates. Looking at winters 1992/1993 until 1999/2000, the highest ILI incidence was observed in children between 0 and 4 years of age, and the lowest in people older than 64 years. Virological aetiology also differed by age category. Incidences of influenza virus infection were estimated by age, region and degree of urbanisation, based on registration of ILI and isolation of influenza virus.

Insight into the virological aetiology of acute respiratory infections obtained by this surveillance, the only one among general practice patients in the Netherlands, can contribute to effective prevention and control of such infections, thus constituting a reason for continuing this surveillance.

Abbreviations

ARI	illnesses due to acute respiratory tract infection(s)
GP	general practitioner or general practice
HSV	herpes simplex virus
ILI	influenza-like illness
m.o.	micro-organism
NIVEL	Netherlands Institute for Health Services Research
PCR	polymerase chain reaction
RIVM	National Institute of Public Health and the Environment
RSV	respiratory syncytial virus

1. Introduction

The Netherlands Institute for Health Services Research (NIVEL) has been co-ordinating a sentinel surveillance network of on average 47 general practices (GP's) since 1970. These practices care for 1% of the Dutch population, a sample representative of the national population in terms of age, sex, region and degree of urbanisation[1]. For influenza surveillance, the GP's in this network register patients who consult them for influenza-like illness (ILI) each week. Because the diagnosis "influenza" cannot be made based on clinical signs and symptoms only, the NIVEL asked the National Institute of Public Health and the Environment (Rijksinstituut voor Volksgezondheid en Milieu, RIVM) to enhance the ILI registration by virological analyses of nose/throat swabs from ILI patients. Therefore, since winter 1992/1993, GP's are asked to send nose/throat swabs from a random sample of patients who consult them for acute upper respiratory tract infections (ARI), including ILI. These swabs are sent to the RIVM for virus isolation and polymerase chain reaction (PCR) analysis.

This report describes the main findings of this NIVEL/RIVM respiratory surveillance for winters 1998/1999 and 1999/2000. It also gives an overview of the estimated incidences of influenza virus infection from winter 1992/1993 until winter 1999/2000. Results of winters 1992/1993 until 1997/1998 have been reported previously[2-7]. Since 1997 the surveillance is carried out in the framework of RIVM project number V/217617/01 'Respiratory infections: surveillance and epidemiology' and formerly in the framework of RIVM project numbers 245607 and 243614.

This report consists of four parts: (1) this introduction, (2) the methodology, (3) the results, and (4) concluding remarks and recommendations.
Chapter 2 describes the methodology used for the surveillance.
Chapter 3 presents the results of winter 1998/1999, followed by the results of winter 1999/2000 and a short overview of the findings of the winters from 1992/1993 until 1999/2000. Subsequently, the incidence of influenza virus infection is estimated by combining ILI registration and influenza virus detection rates.
Chapter 4 discusses the strengths and weaknesses of this surveillance.

2. Methods

All GP's of the sentinel surveillance network of the NIVEL registered all patients that consulted them for ILI by week and age category. The registration forms were sent to the NIVEL weekly and the incidence of ILI during the winter was calculated on a weekly basis. The criteria for ILI were acute onset (a prodromal stage of no more than 4 days), rectal temperature of at least 38ºC, and at least one of the following symptoms: cough, coryza, sore throat, frontal headache, retrosternal pain, myalgia[8]. By letter, sent in September 1998 and 1999, the GP's were asked on a voluntarily basis, to take nose/throat swabs from a random selection of the patients consulting them for ILI or an illness caused by another ARI. ARI was defined as a respiratory illness with an acute onset and at least one of the above-mentioned symptoms, excluding otitis and sinusitis. Each practice was asked to take a maximum of two swabs per week and preferably from Monday through Thursday for logistic reasons. The swabs had to be taken at random, preferably the second patient on Tuesday and the second on Thursday. As in winter 1997/1998, we asked the GP's to send only one swab per week during the influenza peak, since then an optimal number of samples would be collected and because of concern of the workload at the laboratory. The swabs were sent in GLY virus transport medium[9] to RIVM by post. Each swab was accompanied by a form on which the GP registered the following details of the patient: name, sex, date of birth, date of sampling, symptoms, duration of the symptoms, diagnosis, whether the patient was registered as an ILI patient, whether the patient had received an influenza vaccination and some possible risk factors for ARI (see appendix II). At RIVM the swabs were registered by date of sampling and subjected to virus culture and PCR. tMK cells (tertiary cynomolgus monkey kidney cells) and GaBi cells (human diploid fibroblast cells) were used for virus culture and viruses were identified using standard procedures[10]. PCR was performed for RSV[11], rhinovirus and enterovirus[12], *Mycoplasma pneumoniae*[13], and coronavirus type OC43 and 229E[14].

3. Results

Herpes simplex virus (HSV) was cultured both in winter 1998/1999 and 1999/2000, but is not likely to cause respiratory symptoms in immunocompetent persons[15]. Since there were no immunocompromised patients diagnosed with HSV in both winters, HSV was not considered to be relevant with respect to the objectives of this surveillance. Therefore, swabs in which only HSV was detected were classified as 'no micro-organism detected'.

3.1 Winter 1998/1999

During winter 1998/1999, 78% of the 45 sentinel practices sent swabs to the RIVM. These 35 practices were distributed over the four regions and over the three degrees of urbanisation in the Netherlands in a similar way compared to all 45 GP's registering ILI (figures 1 and 2). Swabs were taken between week 30 in 1998 (beginning 20 July) and week 25 in 1999 (ending 27 June), a period of 49 weeks. This is longer than in winter 1997/1998, but between week 30 and week 40 only 6 swabs were sent.

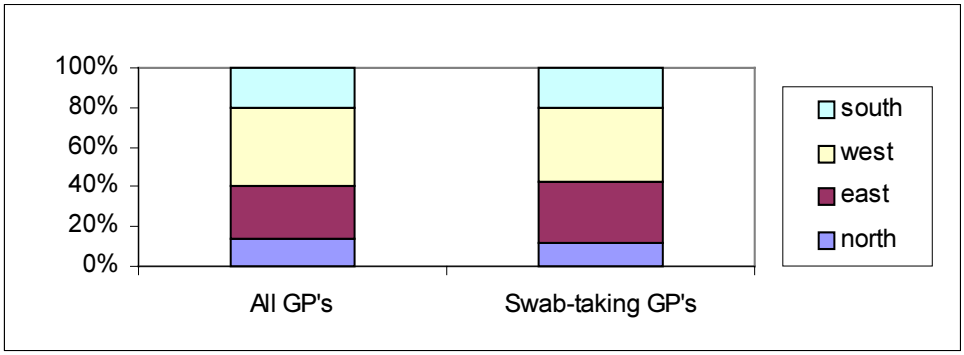

Figure 1 *Distribution over the four regions in the Netherlands of the 45 general practices (GP's) registering ILI and of the 35 GP's taking swabs too, winter 1998/1999*

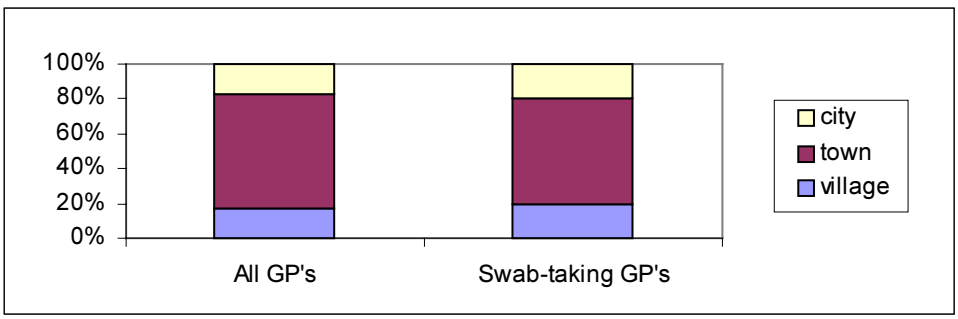

Figure 2 *Distribution over the three degrees of urbanisation in the Netherlands of the 45 general practices (GP's) registering ILI and of the 35 GP's taking swabs too, winter 1998/1999*

3.1.1 Micro-organisms detected

In winter 1998/1999 437 nose/throat swabs were sent to the RIVM. In 193 swabs (44%) no micro-organisms (m.o.'s) were detected. Of the total of 264 m.o.'s found, influenza virus was detected most often (n=106), followed by rhinovirus (n=66) and respiratory syncytial virus (RSV; n=34) (table 1). In twenty swabs (5%) two pathogens were found.
For rhinovirus, enterovirus and RSV both viral culture and PCR were used. In total 74% (79 of 107) of these respiratory pathogens were detected by PCR only. In one swab PCR turned out negative, whereas RSV was grown in culture (table 1).

Table 1 *Micro-organisms detected in 437 nose/throat swabs from GP patients with ARI (including ILI), winter 1998/1999*

Micro-organism	Number by culture	Number by PCR	Total number	Proportion of all submitted swabs (%)	Proportion of all m.o.'s[a] detected (%)
Adenovirus	1	ND[b]	1	0.2	0.4
Coronavirus	ND	30	30	7	11
Enterovirus	3	7	7	2	3
Influenza virus	106	ND	106	24	40
M. pneumoniae	ND	17	17	4	6
Parainfluenza virus	3	ND	3	0.7	1
Rhinovirus	15	66	66	15	25
RSV	9	33	34	8	13

a) m.o.'s = micro-organisms
b) ND = not determined

In total 306 (70%) swabs were taken from patients that were registered with ILI. Influenza virus was detected in 32% (97 of 306) of these swabs. Although the patients were registered as having an influenza-like illness, in 31% of the swabs, other respiratory pathogens than influenza virus were found. In 42% (128 of 306) of these swabs no m.o.'s were detected (table 2).

Table 2 *Micro-organisms detected in 306 nose/throat swabs from GP patients registered with ILI, winter 1998/1999*[a]

Micro-organism	Number by culture	Number by PCR	Total number	Proportion of all submitted swabs (%)	Proportion of all m.o.'s[b] detected (%)
Adenovirus	1	ND[c]	1	0.3	0.5
Coronavirus	ND	20	20	6.5	10
Enterovirus	2	3	3	1	2
Influenza virus	97	ND	97	32	51
M. pneumoniae	ND	11	11	3.6	6
Parainfluenza virus	1	ND	1	0.3	0.5
Rhinovirus	9	40	40	13	21
RSV	4	18	18	6	9

a) From three patients the ILI status was not registered
b) m.o.'s = micro-organisms
c) ND = not determined

The remaining 128 swabs were obtained from patients with ARI other than ILI. In these swabs rhinoviruses predominated, being detected in 20% of the swabs. In seven swabs from ARI patients that were not registered with ILI, influenza virus was found. No respiratory pathogens were detected in 62 of these swabs (48%) (table 3).

Table 3 *Micro-organisms detected in 128 nose/throat swabs from GP patients with ARI other than ILI, winter 1998/1999*[a]

Micro-organism	Number by culture	Number by PCR	Total number	Proportion of all submitted swabs (%)	Proportion of all m.o.'s[b] detected (%)
Adenovirus	0	ND[c]	0	0	0
Coronavirus	ND	10	10	8	14
Enterovirus	1	4	4	3	6
Influenza virus	9	ND	9	7	12
M. pneumoniae	ND	6	6	5	8
Parainfluenza virus	2	ND	2	2	3
Rhinovirus	6	26	26	20	36
RSV	5	15	16	13	22

a) From three patients the ILI status was not registered
b) m.o.'s = micro-organisms
c) ND = not determined

Clearly, in patients registered with ILI influenza virus was detected relatively the most. Influenza virus was detected five times more often in these swabs (32%) than in swabs from patients registered with ARI other than ILI (7%). In patients registered with ARI other than ILI rhinovirus was detected more often than in swabs from patients registered with ILI (20%

vs. 13%). The percentage of the swabs in which no respiratory pathogens were found did not differ significantly between ILI patients and patients with ARI other than ILI (figure 3).

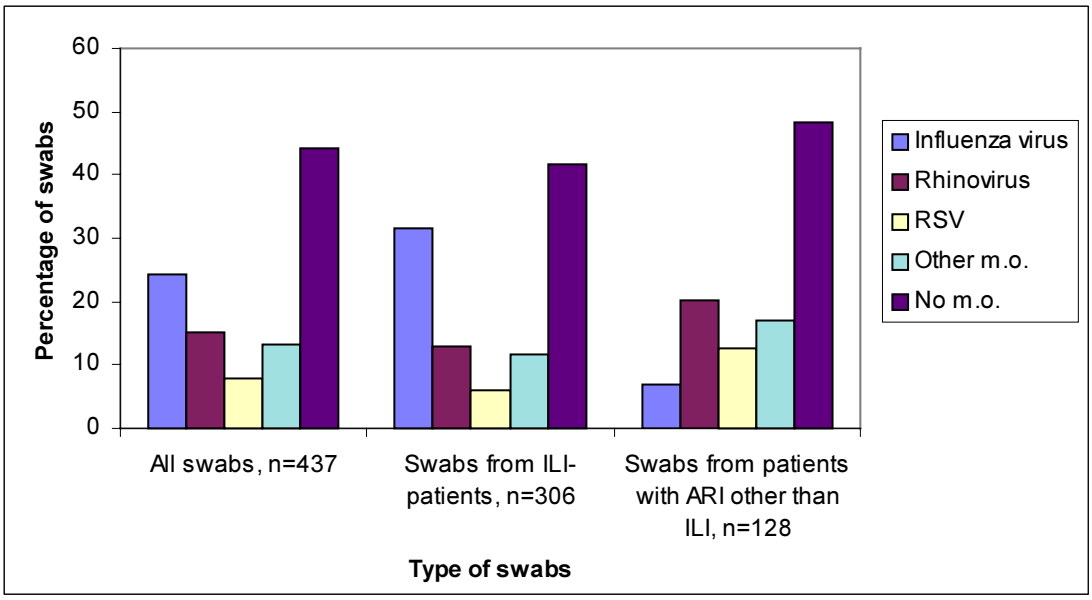

Figure 3 Results of virus culture and PCR, winter 1998/1999

Before week 38 in 1998 and after week 17 in 1999 the total number of swabs being sent in was only 10. The highest number of swabs per week was received in the period from week 50 in 1998 (starting 7 December) till week 8 in 1999 (ending 28 February). Between week 42 and week 16, the percentage of swabs in which at least one pathogen was detected varied between 46% and 68% (figure 4).

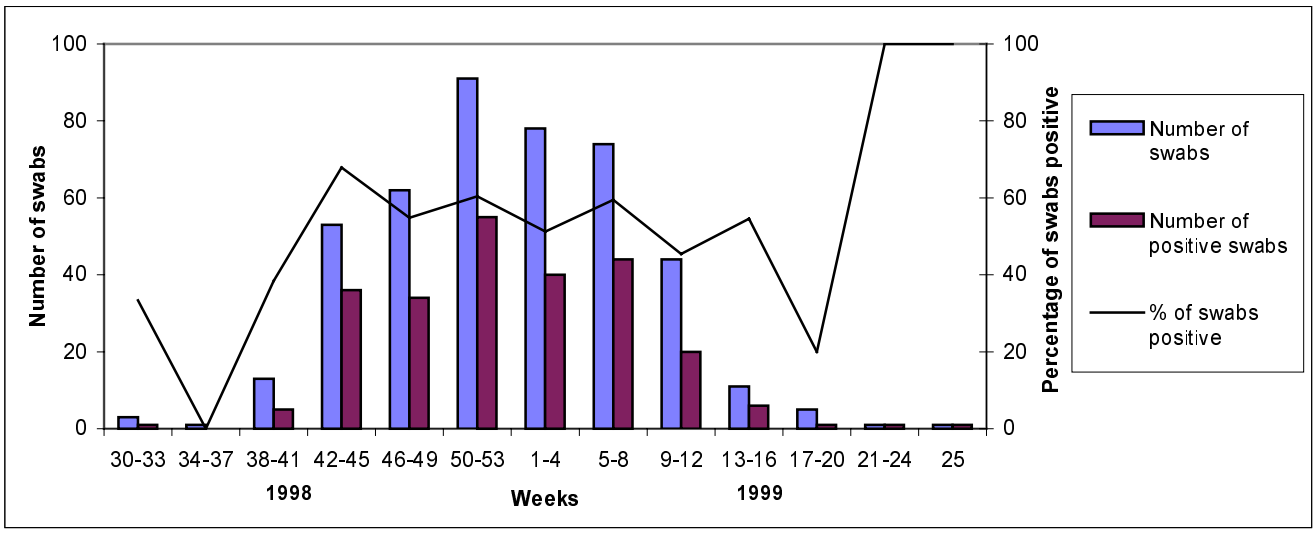

Figure 4 Number of (positive) swabs by 4-week periods, winter 1998/1999

Influenza virus was first detected in a swab taken on 20 November 1998 (week 47) and predominated from week 50 until week 8 in 1999. Rhinoviruses were found throughout the winter, with a peak between week 42 and 45 in 1998. RSV accounted for 11% - 21% of all detected pathogens between week 43 and 52 in 1998. Coronavirus type OC43 was detected mainly between week 42 and 52, accounting for 13% of all m.o.'s found in that period (figure 5). The same patterns were observed for ILI patients only (data not shown).

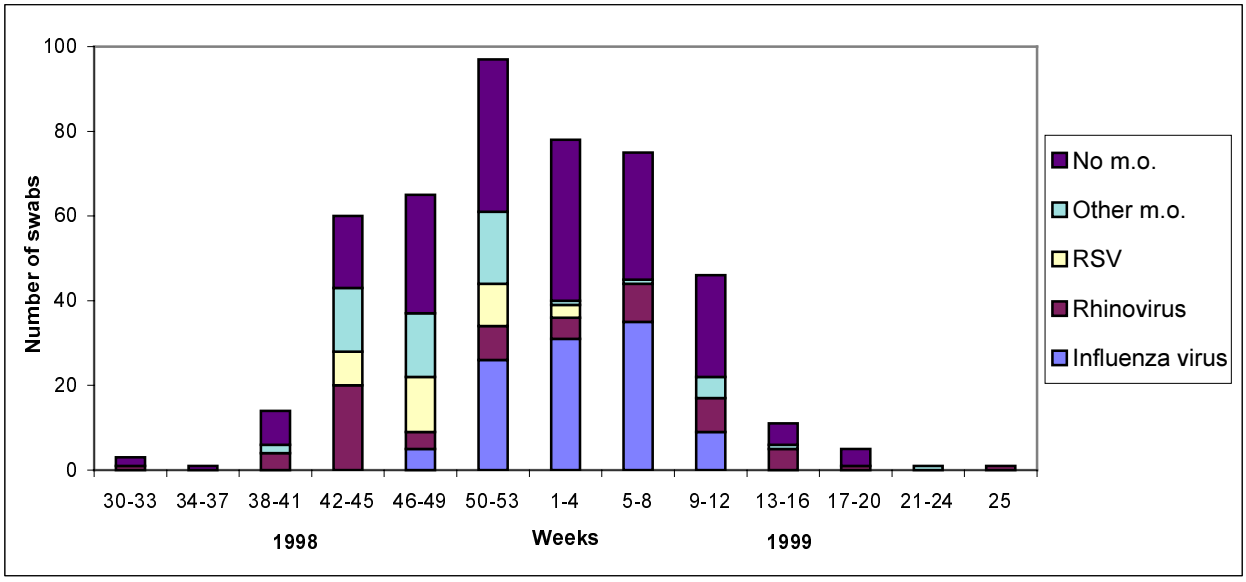

Figure 5 Results of virus culture and PCR by 4-week periods, winter 1998/1999

The results of the laboratory analyses of the swabs obtained from patients aged 0 – 4 years differed considerably from the results of the swabs of patients of 5 years and older (figure 6). In 82% of all swabs taken from young children (0-4 years old), a pathogen was found. In patients 5 to 14 years this percentage was 63% and in patients 15 years and older less than 56%. This difference in diagnostic deficit could be (partly) explained by the finding that children between 0 and 4 years old were taken to the GP sooner after onset of symptoms, leading to an increased chance of detecting a pathogen if present. 42% of these young children were sampled within 2 days after onset of symptoms versus 33% of patients of 5 years and older (see also paragraph 3.3.1). Influenza virus, rhinovirus and RSV were found in all age categories. In the age category of 5 to 14 years influenza virus predominated, being found in 44% of all swabs. RSV was found most often in patients between 0 and 4 years of age: 41% (14 of 34) of all RSV-detections were made in this age category.

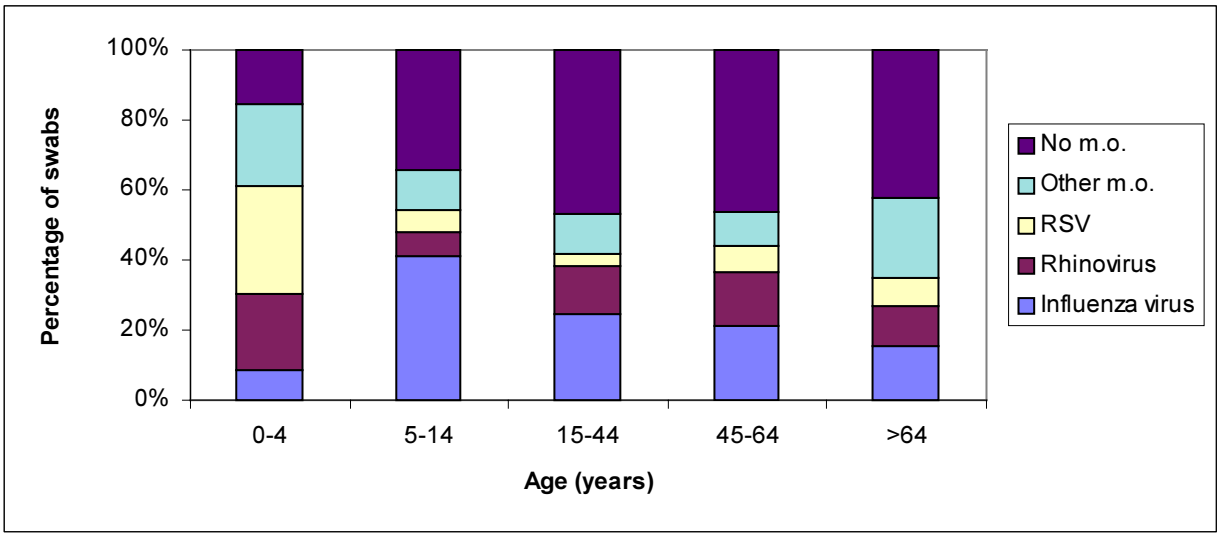

Figure 6 Results of virus culture and PCR by age category, winter 1998/1999

3.1.2 Multiple infections

In total 20 multiple infections were found, i.e. 5% of all swabs and 8% of all positive swabs. The patients with a multiple infection were 10 men, 8 women and two persons of unknown sex. Age varied between 2 months and 76 years. Rhinovirus was found in 13 combinations, mostly with RSV (n=7) (table 4).

Table 4 Combinations of micro-organisms found in multiple infections, winter 1998/1999

Micro-organism	Rhinovirus	Influenza A(H3N2) virus	RSV	Enterovirus	Total
RSV	7				7
Coronavirus type OC43	4	4	1		9
M. pneumoniae	2		1	1	4
Total	13	4	2	1	20

Thirty-one percent of the total number of times coronavirus type OC43 was found, was in combination with another m.o. (table 5). In contrast, only 8% of the total number of times influenza A(H3N2) virus detected, it was combined with another m.o.

Table 5 Micro-organisms found in multiple infections, winter 1998/1999

Micro-organism	Number of times in a multiple infection	Total number of times detected	Percentage in a multiple infection (%)
Rhinovirus	13	66	20
RSV	9	34	26
Coronavirus type OC43	9	29	31
Influenza A(H3N2) virus	4	50	8
M. pneumoniae	4	17	24
Enterovirus	1	7	14

3.1.3 Influenza virus isolation and ILI registration

Of all 106 isolates of influenza virus, 50 were of the A(H3N2) type and 56 were typed as influenza B. No A(H1N1) influenza viruses were found. In hospital patients in the Netherlands, only 3 A(H1N1) influenza viruses and relatively more A(H3N2) (n=152) than B (n=84) influenza viruses were isolated this winter[16]. Of the 106 GP patients that were diagnosed with influenza virus this winter, 10 (9%) were vaccinated against influenza. According to virus isolates, the influenza season consisted of two peaks with the first peak in weeks 50-52 and the second in weeks 6 and 7. The first peak mainly consisted of influenza B, whereas influenza A(H3N2) predominated during the second peak. Registration of ILI was reasonably in accordance with the number of isolated influenza viruses: the second peak in registered ILI was one week after the peak in isolates. The influenza season was fairly mild[17], with the overall number of ILI not exceeding 22 per 10 000 persons per week. Compared with previous winters, this influenza season was stretched out over a long period of time: the ILI incidence exceeded 5 per 10 000 persons between week 49 in 1998 and week 11 in 1999 (figure 7). In the influenza season, more ILI patients were diagnosed with influenza virus when compared to the entire winter (42% vs. 32%).

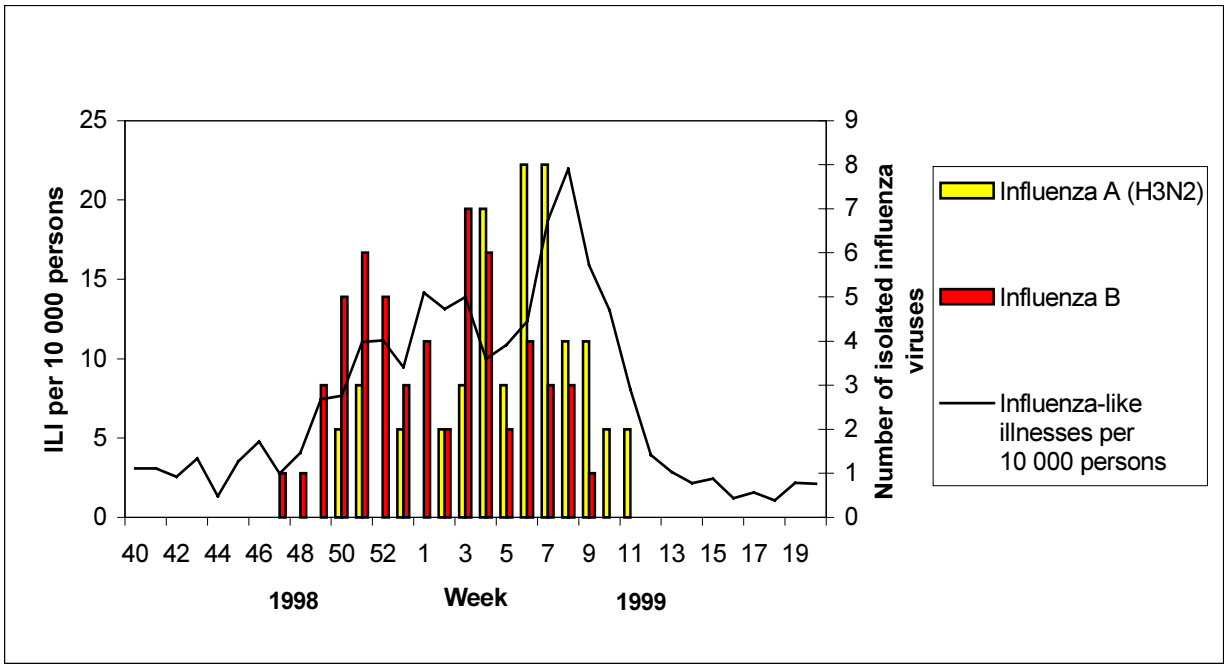

Figure 7 Isolates of influenza virus and ILI registered by week, winter 1998/1999

When categorised in different age groups, the reported incidence of ILI shows a quite erratic picture for the age group 0 to 4 years. This might be related to small outbreaks in child day care, but this is unconfirmed. ILI incidences in the remaining age categories followed approximately the same pattern with a peak between week 7 and 9. The entire season, the ILI

incidence was highest in the age group 0 to 4 years, except for week 8 when it was highest for persons between 45 and 64 years of age (figure 8).

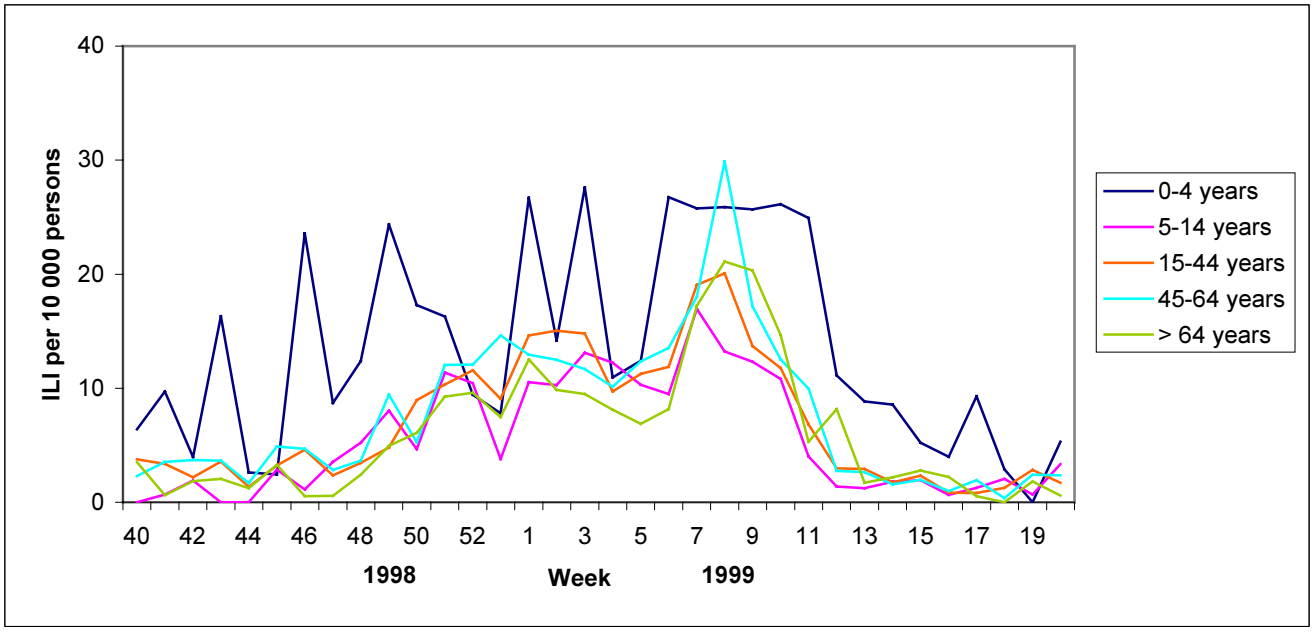

Figure 8 *Reported incidence of ILI in GP patients by age category and week, winter 1998/1999*

3.2 Winter 1999/2000

In winter 1999/2000 49 practices participated in the sentinel surveillance network, of whom 30 (61%) sent in swabs. There were slightly more GP's from the east of the Netherlands taking swabs as compared to all the GP's of the NIVEL (figure 9). In terms of degree of urbanisation, of all the GP's in the sentinel network there were relatively more GP's located in villages sending in swabs (figure 10). Swabs were sent between week 40 of 1999 (beginning 4 October) and week 30 of 2000 (ending 30 July).

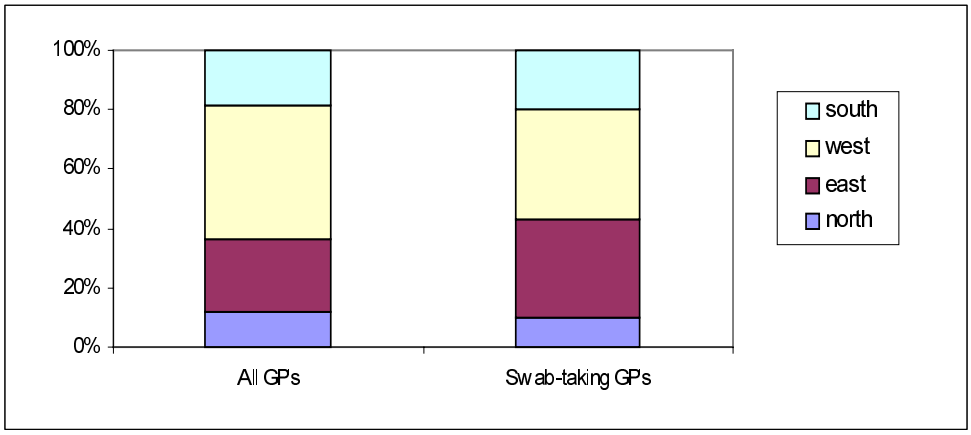

Figure 9 *Distribution over the four regions in the Netherlands of the 49 general practices (GP's) registering ILI and of the 30 GP's taking swabs too, winter 1999/2000*

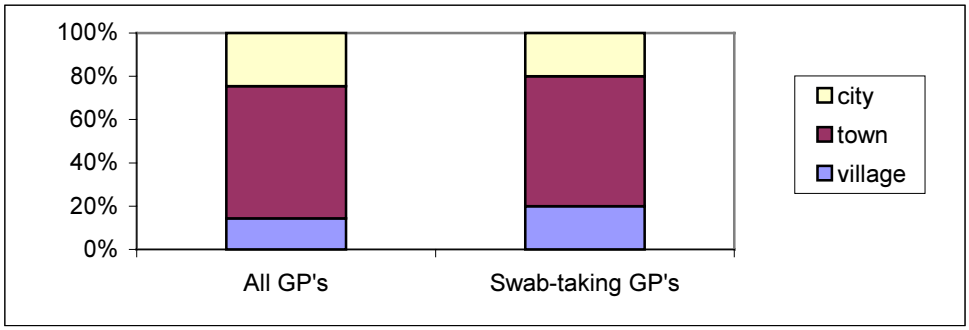

Figure 10 *Distribution over the three degrees of urbanisation in the Netherlands of the 49 general practices (GP's) registering ILI and of the 30 GP's taking swabs too, winter 1999/2000*

3.2.1 Micro-organisms detected

In winter 1999/2000 319 nose/throat swabs were sent to the RIVM. Of all m.o.'s detected, influenza virus was detected most frequently (91 of 189, 48%). Rhinovirus (n=45) and RSV (n=22) were also detected relatively often. In 13 swabs (4%) two pathogens were found, and one swab contained rhinovirus and both coronavirus type OC43 and type 229E. In 145 (45%) of the swabs no respiratory pathogens were detected (table 6).
For rhinovirus, enterovirus and RSV both viral culture and PCR were used. In total 79% (59 of 75) of these respiratory pathogens were detected by PCR only. Enteroviruses were not detected using virus culture; PCR showed to be a valuable method (table 6).

Table 6 Micro-organisms detected in 319 nose/throat swabs from GP patients with ARI (including ILI), winter 1999/2000[a]

Micro-organism	Number by culture	Number by PCR	Total number	Proportion of all submitted swabs (%)	Proportion of all m.o.'s[b] detected (%)
Adenovirus	1	ND[c]	1	0.3	0.5
Coronavirus	ND	15	15	5	8
Enterovirus	0	8	8	3	4
Influenza virus	91	ND	91	29	48
M. pneumoniae	ND	1	1	0.3	0.5
Parainfluenza virus	5	ND	5	2	3
Rhinovirus	14	45	45	14	24
RSV	2	22	22	7	12

a) One swab was sent to the Erasmus University in Rotterdam where it was typed as a newly discovered pneumovirus[18]
b) m.o.'s = micro-organisms
c) ND = not determined

Seventy-five percent of the swabs were obtained from patients registered with ILI (n=240). Influenza virus accounted for 56% of all m.o. detected, with RSV and rhinovirus (both 14%) being the other respiratory pathogens detected relatively often. No m.o.'s were found in 111 (46%) of these swabs (table 7).

Table 7 *Micro-organisms detected in 240 nose/throat swabs from GP patients registered with ILI, winter 1999/2000*[a]

Micro-organism	Number by culture	Number by PCR	Total number	Proportion of all submitted swabs (%)	Proportion of all m.o.'s[b] detected (%)
Adenovirus	1	ND[c]	1	0.4	0.7
Coronavirus	ND	10	10	4	7
Enterovirus	0	7	7	3	5
Influenza virus	79	ND	79	33	56
M. pneumoniae	ND	1	1	0.4	0.7
Parainfluenza virus	4	ND	4	2	3
Rhinovirus	7	19	19	8	14
RS-virus	2	20	20	8	14

a) From one patient the ILI status was not registered
b) m.o.'s = micro-organisms
c) ND = not determined

Of all respiratory pathogens found in swabs from patients with ARI other than ILI, rhinovirus was detected most often (26 of 48, 54%) (figure 11, table 8). Although the patients were not diagnosed as having ILI, influenza virus accounted for 25% of all m.o.'s detected. Coronavirus accounted for another 10% of all m.o.'s detected. In 42% (33 of 78) of the swabs from patients with ARI other than ILI, no m.o.'s were detected (table 8).

Table 8 *Micro-organisms detected in 78 nose/throat swabs from GP patients with ARI other than ILI, winter 1999/2000*[a, b]

Micro-organism	Number by culture	Number by PCR	Total number	Proportion of all submitted swabs (%)	Proportion of all m.o.'s[c] detected (%)
Adenovirus	0	ND[d]	0	0	0
Coronavirus	ND	5	5	6	10
Enterovirus	0	1	1	1	2
Influenza virus	12	ND	12	15	25
M. pneumoniae	ND	0	0	0	0
Parainfluenza virus	1	ND	1	1	2
Rhinovirus	7	26	26	33	54
RS-virus	0	2	2	3	4

a) From one patient the ILI status was not registered
b) One swab was sent to the Erasmus University in Rotterdam where it was typed as a newly discovered pneumovirus[18]
c) m.o.'s = micro-organisms
d) ND = not determined

The detection rate of influenza virus was twice as high in swabs from ILI patients than in swabs from patients registered with an ARI other than ILI (33% versus 15%). In swabs from patients registered with ARI other than ILI rhinovirus was detected four times more often than in swabs from ILI patients (33% vs. 8%). As in winter 1998/1999, the percentage of the swabs in which no respiratory pathogens were found did not differ significantly between ILI patients and patients with ARI other than ILI (figure 11).

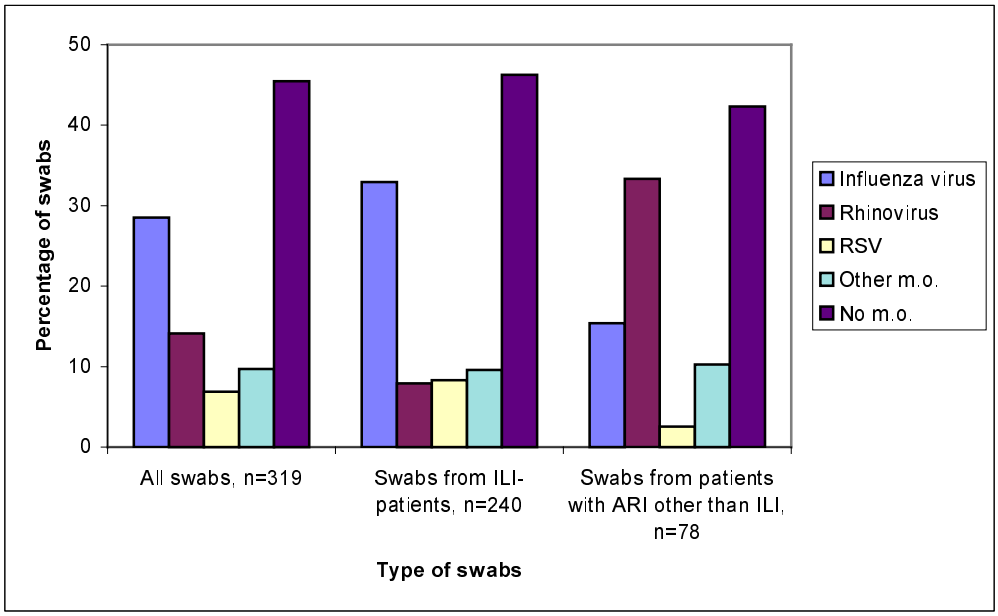

Figure 11 Results of virus culture and PCR, winter 1999/2000

The GP's were asked to send in swabs starting in week 40 (beginning 4 October 1999). The highest number of swabs received per week was between week 1 and 4 of 2000 (from 3 January until 30 January). Between week 45 in 1999 and week 16 in 2000 (8 November and 23 April) the percentage of the swabs with at least one respiratory pathogen found was more than 50%. The total number of swabs from week 13 until week 16 was only 7 (figure 12).

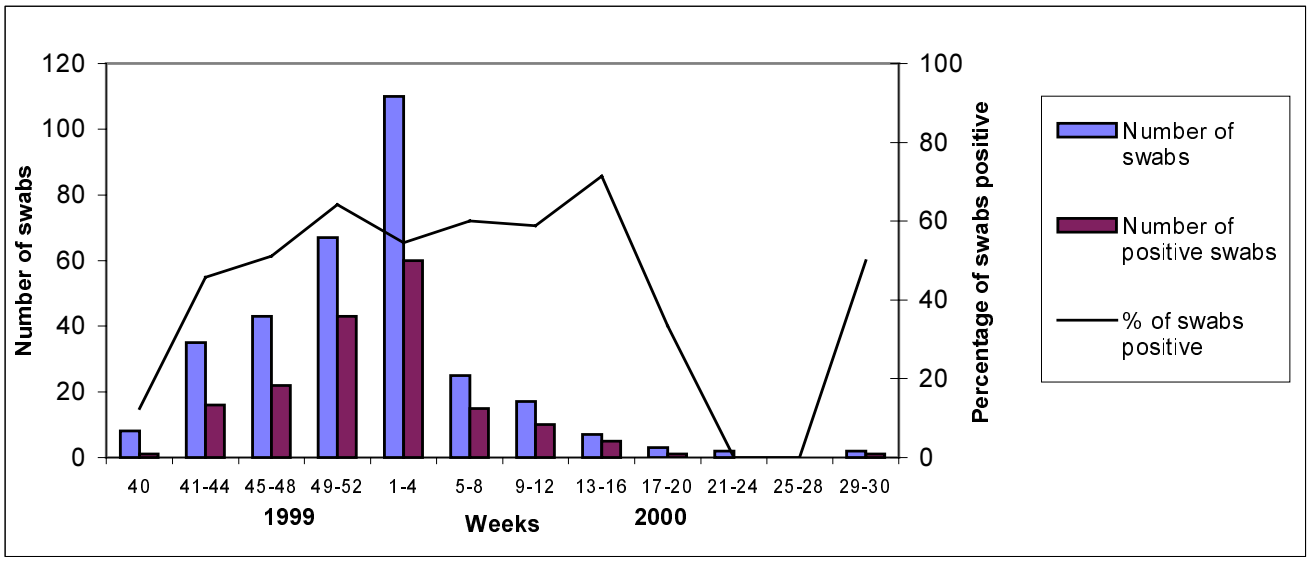

Figure 12 Number of (positive) swabs by 4-week periods, winter 1999/2000

The first swab with influenza virus was taken on 7 November (week 45). Influenza virus predominated from week 49 in 1999 until week 4 in 2000. Rhinovirus was found from the beginning of the surveillance (week 40 1999) until week 16 (19 April 2000). Of the 22 times RSV was detected, 10 findings occurred between week 1 and 4 (figure 13). The same patterns were observed for ILI patients only (data not shown).

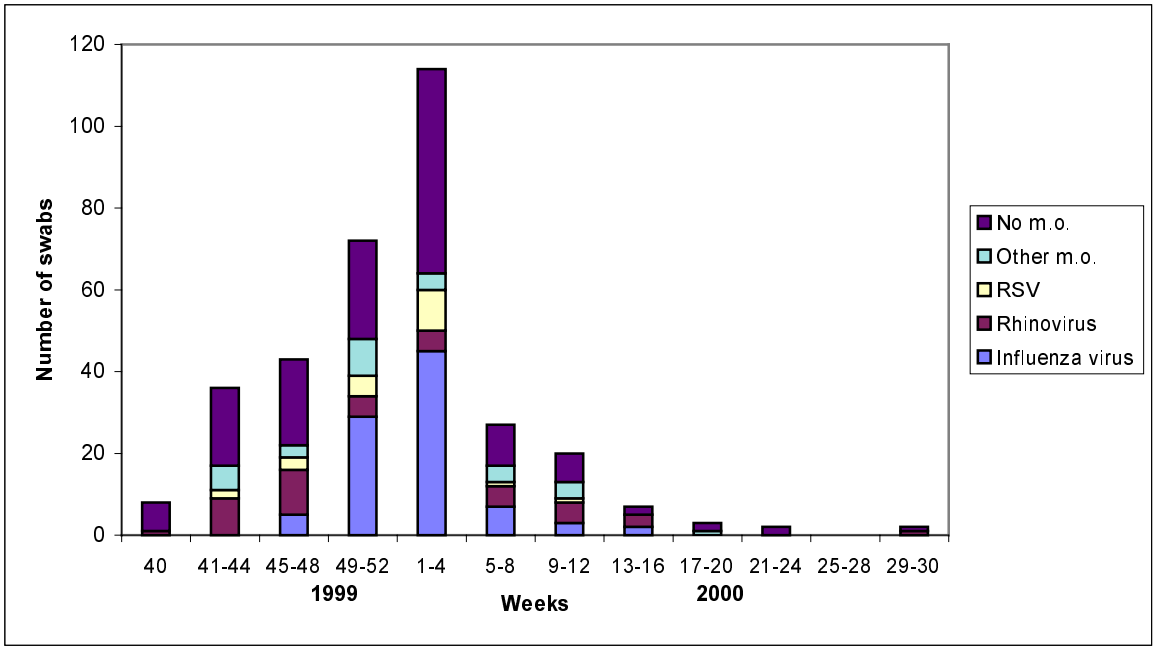

Figure 13 Results of virus culture and PCR by 4-week periods, winter 1999/2000

In 78% of all swabs taken from young children (0-4 years old), a pathogen was found. In older patients this percentage was less than 54%. The finding that children between 0 and 4 years old were taken to the GP sooner after onset of symptoms might be related to this difference in diagnostic deficit. 44% of these children were sampled within 2 days after onset of symptoms versus 39% of patients of 5 years and older (also see paragraph 3.3.1). The percentage of swabs in which influenza virus was found, was approximately the same in all age groups, ranging between 27 and 33%. Strikingly, rhinovirus was not detected in patients between 5 and 14 years old and in patients 65 years and older. As in winter 1998/1999, RSV was detected often in the youngest age group (6 times of the 22 RSV-isolates in total). This winter however, RSV was also detected often in swabs from patients between 15 and 44 years old (n=8). Sixty-four percent (14 of 22) of all RSV-isolates were found in these two age groups (figure 14).

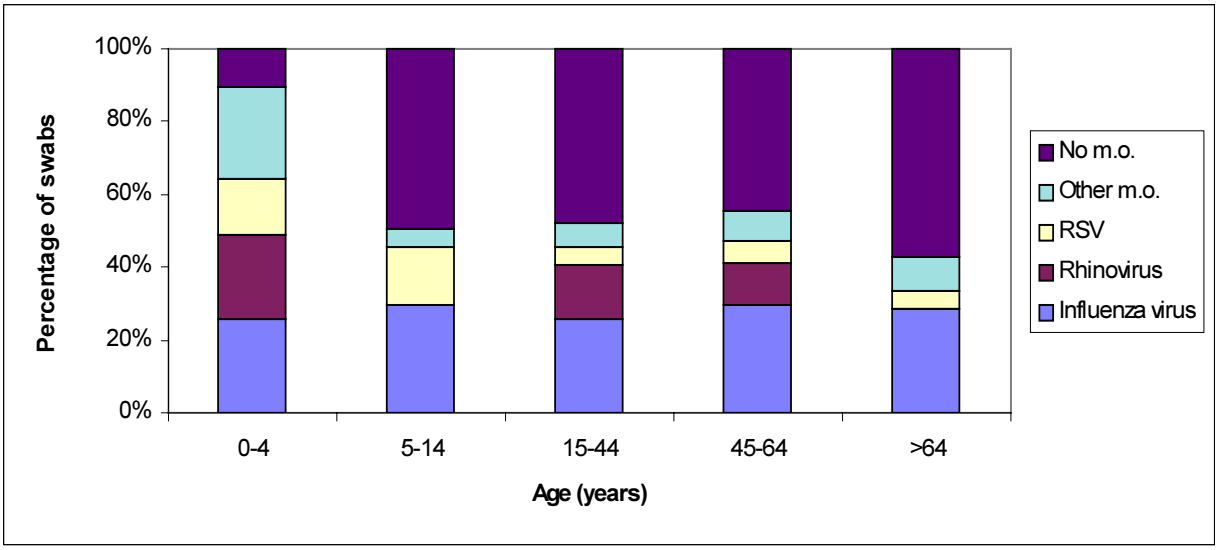

Figure 14 Results of virus culture and PCR by age category, winter 1999/2000

3.2.2 Multiple infections

In total 13 double infections and one triple infection were found. This is 4% of all swabs, and 8% of all positive swabs. The double infections occurred in 10 men and 3 women, with age varying between 11 months and 62 years. Influenza A(H3N2) virus was found in 7 combinations. Fifty percent of the total number of times coronavirus type OC43 was found, it was in combination with another m.o. (table 9 and 10).
In a six-months-old boy a triple infection was found: rhinovirus combined with coronavirus type OC43 and type 229E.

Table 9 Combinations of micro-organisms found in double infections, winter 1999/2000

Micro-organism	Influenza A(H3N2) virus	RS-virus	*M. pneumoniae*	Influenza A(H1N1) virus	Total
Rhinovirus	1	2		1	4
Enterovirus	2		1		3
Coronavirus type OC43	2	2			4
RSV	2				2
Total	7	4	1	1	13

Table 10 Micro-organisms found in multiple infections, winter 1999/2000

Micro-organism	Number of times in multiple infections	Total number of times detected	Percentage in multiple infection (%)
Influenza A(H3N2) virus	7	87	8
RSV	6	22	27
Rhinovirus	5	45	11
Coronavirus type OC43	5	10	50
Enterovirus	3	8	38
Coronavirus type 229E	1	5	20
Influenza A(H1N1) virus	1	3	33
M. pneumoniae	1	1	100

3.2.3 Influenza virus isolation and ILI registration

In total influenza virus was detected 91 times. Of these, 87 were of the A(H3N2) type, 3 of the A(H1N1) type and 1 was influenza B (week 12). This is in accordance with the influenza virus types found in hospital patients in the Netherlands this winter[19]. Of the 91 patients that were diagnosed with influenza virus, 14 (15%) were vaccinated against influenza.
In contrast to winter 1998/1999, there was only one peak of influenza this winter, namely in week 1 of 2000 (figure 15). Registration of ILI was in accordance with the number of influenza isolates, with the peak of the registered ILI cases in weeks 1 and 2 of 2000. Between weeks 49 of 1999 and week 5 of 2000, ILI incidence per 10 000 persons per week exceeded 5, with a peak incidence of 33 per 10 000 in week 1 of 2000. Compared with previous winters[17], the influenza season was of short duration, with a peak incidence close to the average of the previous winters. In the influenza season, more ILI patients were diagnosed with influenza virus when compared to the entire winter (44% vs. 33%).

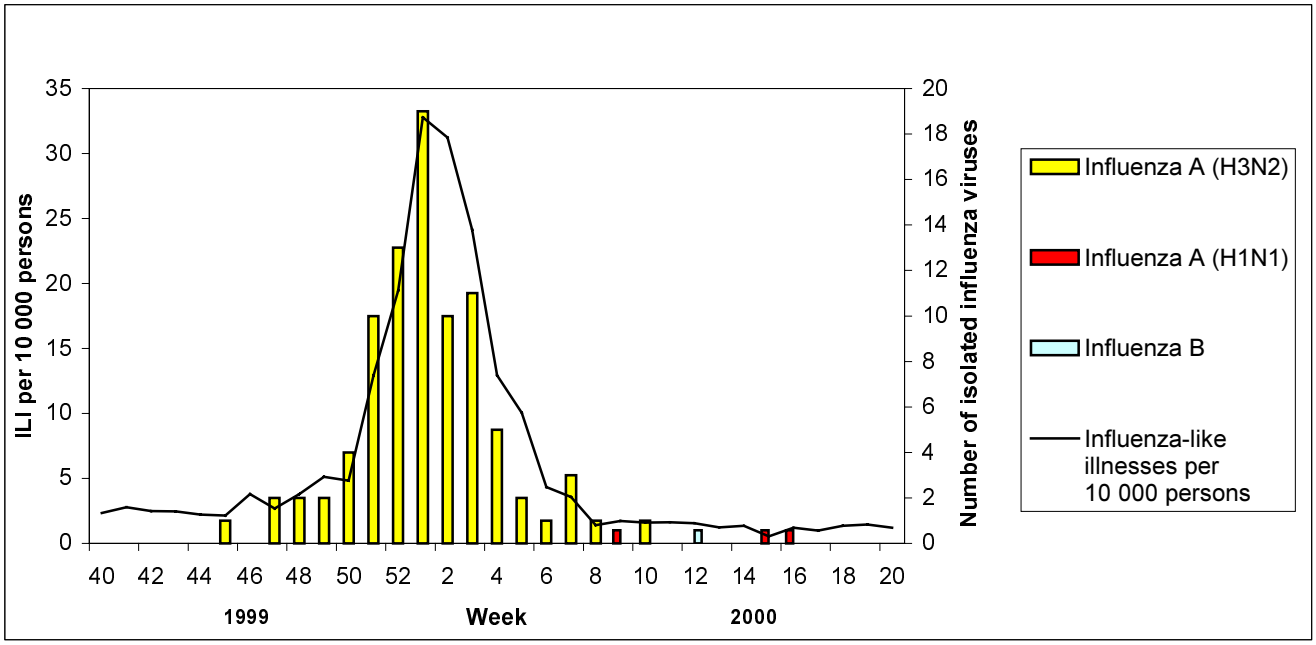

Figure 15 Isolates of influenza virus and ILI registered by week, winter 1999/2000

The ILI incidence had the same course over time for all age categories. Throughout the winter, the reported incidence of ILI was highest for patients between 0 and 4 years of age. At the peak of the influenza season the reported incidence was lowest for patients between 5 and 14 years of age (figure 16).

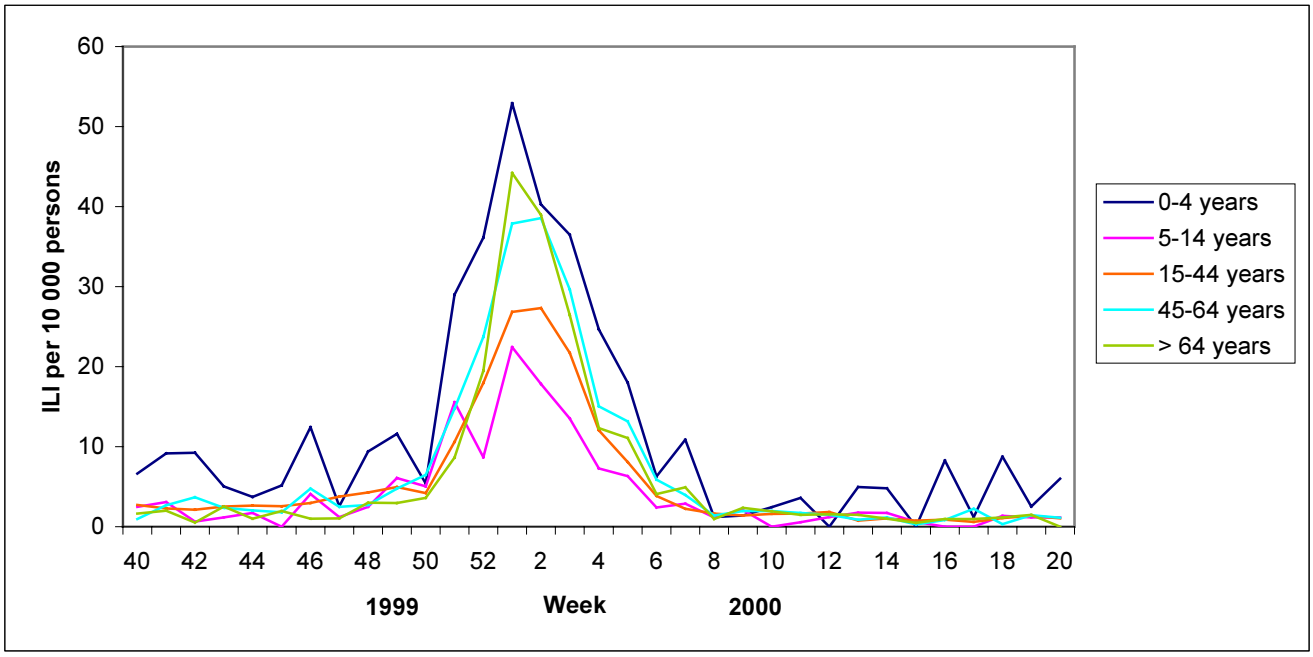

Figure 16 Reported incidence of ILI by age category and week, winter 1999/2000

3.3 Winters 1992/1993 until 1999/2000

Over the eight winters from 1992/1993 until 1999/2000, the number of GP's taking swabs varied from 30 to 37 (table 11). The number of swabs taken per winter varied from 293 to 551. In approximately 55% of the swabs at least one respiratory pathogen was detected during the last four winters. On average 65% of the swabs was obtained from ILI patients, the remainder from patients with an illness due to another acute respiratory tract infection. Approximately half of the swabs were obtained from men. The number of swabs per GP was relatively constant over the years (on average 13) but the variation between GP's was huge (table 11).

Table 11 Some key characteristics of the NIVEL/RIVM respiratory surveillance by winter

	92/93	**93/94**	**94/95**	**95/96**	**96/97**	**97/98**	**98/99**	**99/00**
Virus culture	yes	yes	yes	yes	yes	yes	yes	yes
PCR	yes	no	yes	no	yes	yes	yes	yes
Bacteriology	no	no	no	no	yes	no	no	no
Period (week number)	41-20	39-29	37-25	31-23	30-29	36-27	30-25	40-30
Number of weeks	32	43	41	45	52	44	49	43
Number of swab-taking GP's	35	36	37	31	36	30	35	30
Number of swabs	396	293	551	483	541	363	437	319
Number of follow-up swabs	0	0	72 [a]	0	55 [a]	1 [a]	0	0
Positive swabs	32%	31%	47%	35%	56% [b]	52%	56%	55%
Number of swabs taken from ILI patients (%)	213 (54%)	202 (69%)	344 (62%)	336 (70%)	306 (57%)	251 (69%)	306 (70%)	240 (75%)
Positive ILI swabs	35%	35%	47%	40%	59%	54%	58%	54%
Mean number of swabs per GP	11	8	15	16	15	12	12	11
Range	1-35	1-25	1-62	2-44	1-69	1-65	1-53	1-46
Mean number of ILI swabs per GP	6	6	9	11	9	8	9	8
Range	0-28	0-18	1-41	2-37	0-28	1-42	0-38	1-25
Male gender	52%	49%	48%	49%	51%	48%	48%	47%

a) In these winters a follow-up of some of the patients with a positive PCR-test result of the first swab took place

b) 64% with bacteriology

We chose to look for viruses and *M. pneumoniae* but not for bacteria because it has been estimated that 70% of all ARI in the community is caused by viruses and only 8% by bacteria[20]. In the winter of 1996/1997 some of the swabs were analysed for bacteria too: in only 9% of the swabs a bacterium was the only potentially pathogenic micro-organism found[6]. Compared to winter 1997/1998 *Chlamydia pneumoniae* was no longer sought for, because this pathogen was detected seldom (less than 6 times per winter). Instead, we used PCR to test for coronaviruses types OC43 and 229E (table 12).

Table 12 Micro-organisms sought for and diagnostic methods used in the NIVEL/RIVM respiratory surveillance by winter

Micro-organism	Method	92/93	93/94	94/95	95/96	96/97	97/98	98/99	99/00
Adenovirus	Culture	*	*	*	*	*	*	*	*
C. pneumoniae	PCR			*		*	*		
Coronavirus	PCR			*		*		*	*
Enterovirus	Culture	*	*	*	*	*	*	*	*
	PCR			*		*	*	*	*
Herpes simplex virus	Culture	*	*	*	*	*	*	*	*
Influenza virus	Culture	*	*	*	*	*	*	*	*
	PCR	*							
Parainfluenza virus	Culture	*	*	*	*	*	*	*	*
M. pneumoniae	PCR			*		*	*	*	*
Rhinovirus	Culture	*	*	*	*	*	*	*	*
	PCR			*		*	*	*	*
RSV	Culture	*	*	*	*	*	*	*	*
	PCR	*		*		*	*	*	*

3.3.1 Micro-organisms detected

Of all m.o.'s detected in the winters since 1992/1993, influenza virus was detected most often. In 13% (winter 1994/1995) to 29% (winter 1999/2000) of all swabs, influenza virus was detected. Rhinovirus is the second most frequently detected pathogen. From winter 1992/1993 until 1995/1996 in 4% to 9% of all swabs rhinovirus was detected. In winter 1994/1995 and from winter 1996/1997 onwards PCR was used for detection of rhinovirus[12]. This resulted in an increase in the detection of rhinovirus: in 14% to 22% of all swabs rhinovirus was detected. RSV was found in 4% of the swabs on average (range: 1% - 8%). In winters 1994/1995, 1996/1997, 1998/1999 and 1999/2000 PCR was used to search for coronavirus, resulting in 79 coronaviruses found (4% of all swabs taken in those four winters). Furthermore, six other pathogens were sought for and were detected in approximately 6% of all swabs (figure 17).

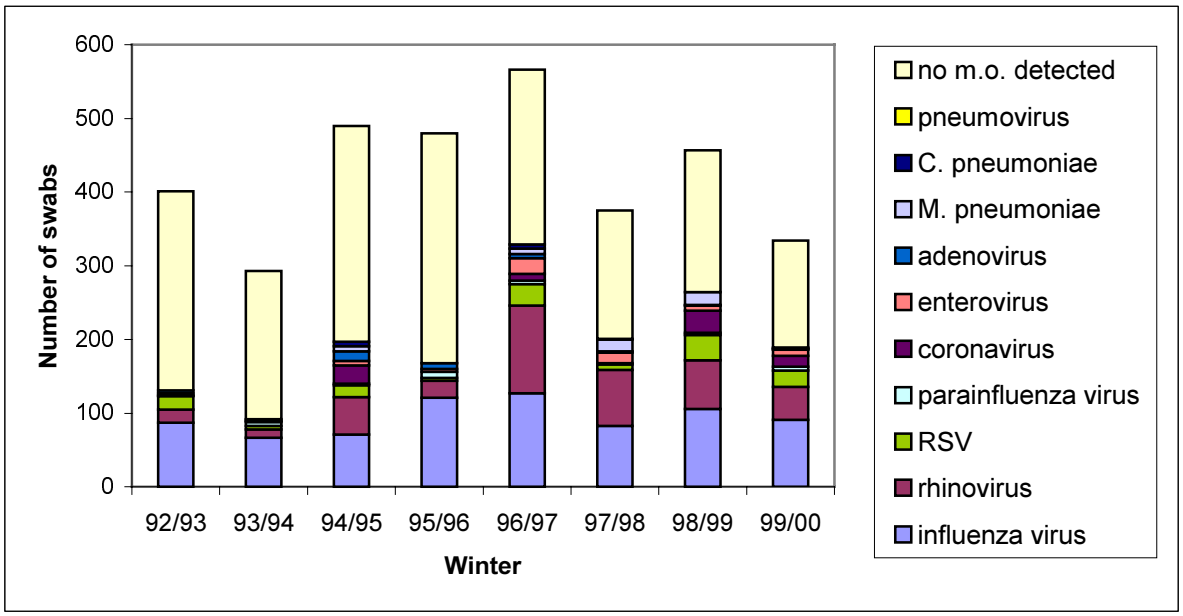

Figure 17 *Results of virus culture and PCR from swabs from GP patients with ARI (including ILI), by winter*

When looking only at patients registered with ILI, a similar picture emerged. The percentage of swabs in which influenza virus was detected ranged from 17% to 33%. Although still the second most often detected pathogen, rhinovirus was less often found. In the winters with PCR-tests for rhinovirus between 8% and 16% of all swabs were positive for rhinovirus (figure 18).

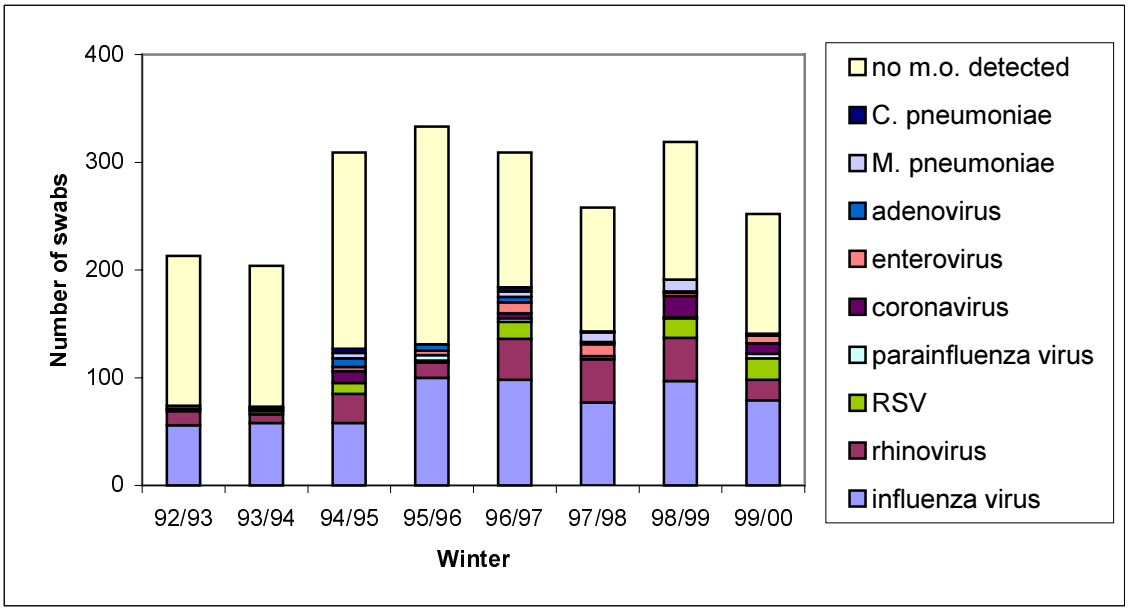

Figure 18 *Results of virus culture and PCR from swabs obtained from GP patients registered with ILI, by winter*

Although all swabs were taken from people consulting their GP for ARI, still in approximately half of the swabs (range: 44% - 69%) no respiratory pathogen was found (figure 17). This diagnostic deficit could have been caused by a number of aspects:

a) the patient did not suffer from an infection (the ARI signs and symptoms were caused by e.g. allergy);
b) the time of swab-taking was too late after the onset of symptoms to be able to detect a pathogen;
c) the time between swab-taking and analyses was too long to be able to detect a pathogen;
d) limitations of the diagnostic methods used;
e) the presence of respiratory pathogens not searched for.

To deal with item b and c, the GP's were asked to only take swabs from patients within 5 days after onset of symptoms, to send the sample immediatley after obtaining it, and preferably not on Friday, because of possible problems with the post. For winter 1998/1999 and winter 1999/2000, 36% of the swabs were taken ≤ 2 days after onset of symptoms, 92% ≤ 5 days after onset of symptoms. The maximum time between onset of symptoms and time of swab-taking was 30 days (figure 19).

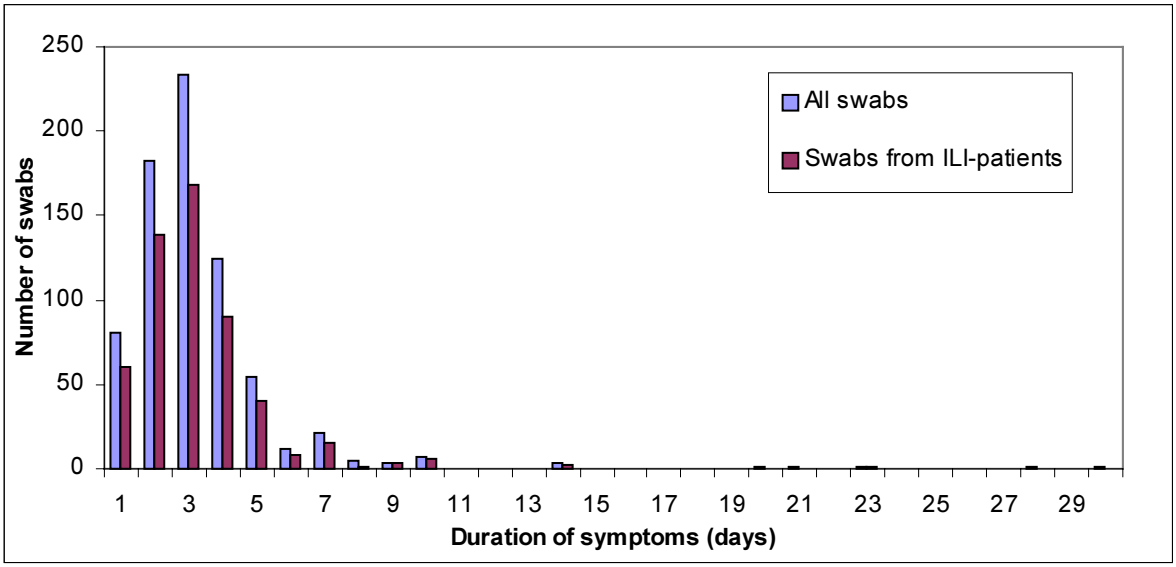

Figure 19 *Duration of symptoms at the time of swab taking, for all swabs and for those from patients registered with ILI, winters 1998/1999 and 1999/2000. Duration of symptoms was unknown for 22 swabs, 13 of which were from ILI patients*

Further, in these two winters there was some variation in the time the swab was on the way from the GP to the laboratory; in both winters combined, 65% of the swabs was 1 day on the way and 87% 1 or 2 days. The maximum transport time was 8 days (figure 20).

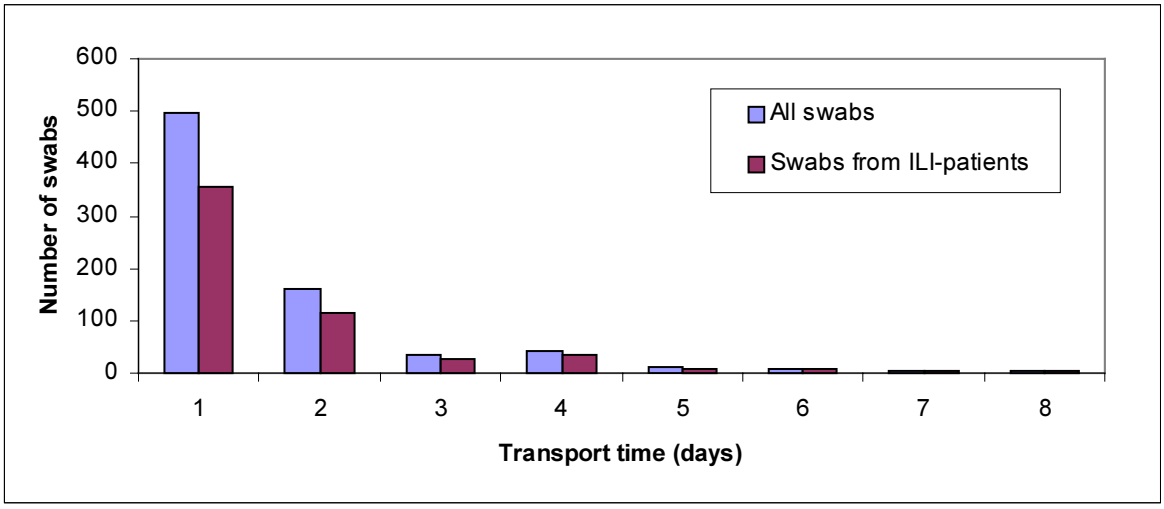

Figure 20 *Transport time of all swabs and of those from patients registered with ILI, winters 1998/1999 and 1999/2000*

Combining duration of symptoms and transport time, 82% of the swabs were analysed within 6 days after onset of symptoms (figure 21). The time between onset of symptoms and analyses was negatively associated with the number of pathogens detected. When laboratory analyses were started 6 days or more after onset of symptoms, the percentage of swabs in which one or more pathogen was detected was less than 50%. When the laboratory analyses

were started within 5 days after onset of symptoms, the percentage of positive swabs varied between 52% and 70%.

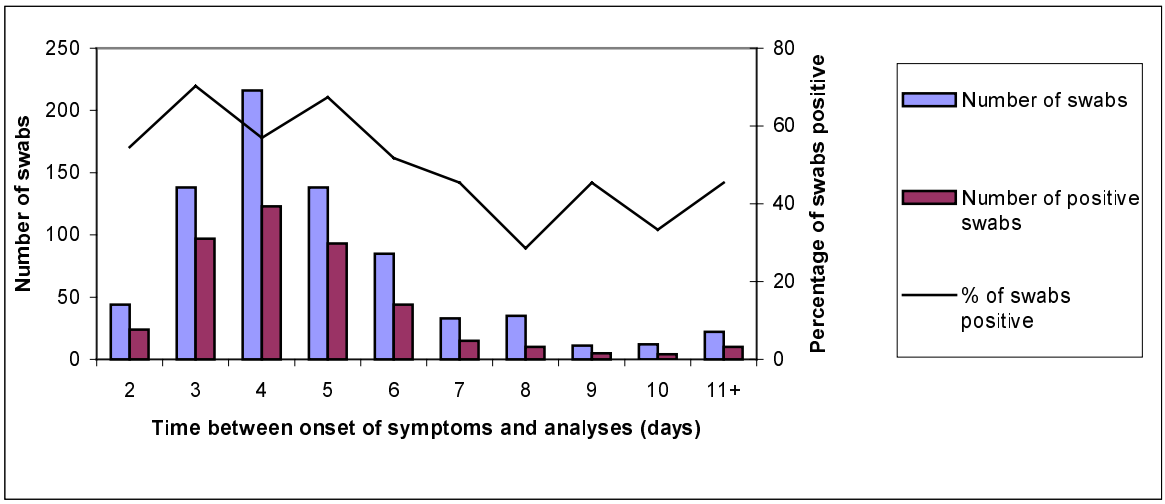

Figure 21 *Number of (positive) swabs by time between onset of symptoms and analyses, winters 1998/1999 and 1999/2000. Time between onset of symptoms and analyses was unknown for 22 swabs*

The age distribution of the patients from whom a swab was taken during winter 1998/1999, winter 1999/2000 and during winters 1992/1993-1999/2000 in total, is comparable with the age distribution of the Dutch general population. There is some over-sampling of patients between 15 and 44 years, especially in winter 1998/1999. Furthermore, there are relatively fewer swabs taken from patients between 5 and 14 years and from patients over 64 years of age (figure 22).

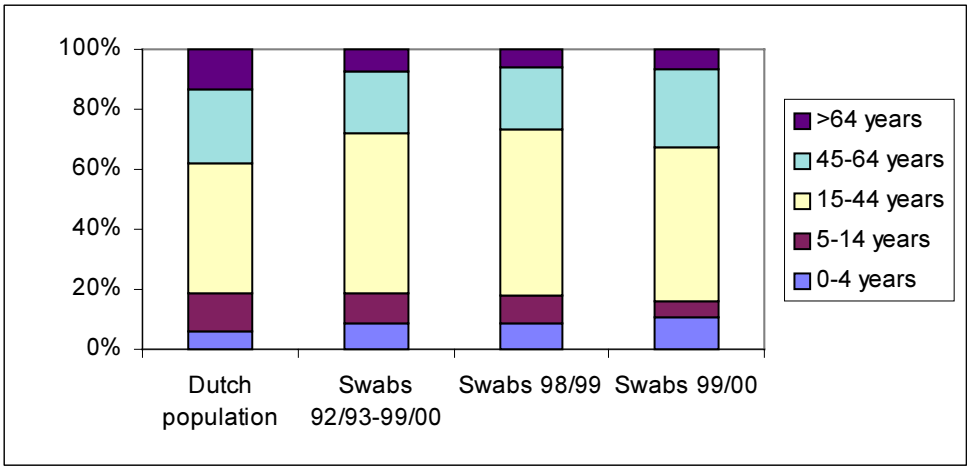

Figure 22 *Age distribution of the Dutch general population (at 1 January 1999; Source: Central Bureau for Statistics) and of patients in the NIVEL/RIVM respiratory surveillance*

3.3.2 Multiple infections

Multiple infections have been detected in every winter since 1992/1993 except in winter 1993/1994. In that winter PCR was not used, only virus culture. In winter 1995/1996 PCR was not used as well, but two multiple infections were detected. Combining the eight winters, 109 multiple infections have been detected, including seven triple infections. In 98% (107 of 109) of the multiple infections at least one of the pathogens involved was detected using PCR. The patients concerned were 59 men, 44 women and 6 people of unknown sex. Sixty-six percent of the swabs in which a multiple infection was detected was obtained from patients registered with ILI. This agrees with the fact that 65% (2198 of 3383) of all swabs is obtained from ILI patients. Relatively most multiple infections were found in children aged 0 to 4 years of age, namely in 10% of all swabs obtained in this age category (table 13). The percentage of all swabs found positive is higher in this age category than in any other age category, too (combining data from winters 1998/1999 and 1999/2000 this percentage is 85% versus an overall percentage of 55% in all age groups combined, see figures 6 and 14).

Table 13 *Number of multiple infections detected in swabs from GP patients with ARI (including ILI), winter 1992/1993 – 1999/2000*

Age (years)	Number of multiple infections	Number of swabs	Percentage multiple infection (%)
0-4	30	297	10
5-14	17	336	5
15-44	42	1 790	2
45-64	16	677	2
> 64	2	250	1
Unknown	2	33	6
All ages	109	3 383	3

Rhinovirus was part of 63% of the multiple infections (69 of 109). Influenza virus and RSV were also found often, namely in 44% and 29% of the multiple infections. Of all influenza viruses mainly type A(H3N2) was present (34% of all multiple infections). Furthermore also coronavirus type OC43 (n=22, 20%), enterovirus (n=21, 19%) and *Mycoplasma pneumoniae* (n=13, 12%) were detected often in multiple infections.
Rhinovirus / influenza virus and rhinovirus / RSV were the most common combinations in multiple infections, occurring 19 and 18 times, respectively. The combination of RSV and influenza A(H3N2) was detected in 6 swabs (6%). In six of the seven triple infections rhinovirus was detected.

3.4 Estimates of the incidence of influenza virus infection

3.4.1 Influenza incidence by age category

As described in both paragraphs 3.1.3 and 3.2.3 (figures 7 and 15), the registration of ILI (from week 40 of year *n* to week 20 of year *n+1*) and influenza virus isolation were reasonably in accordance with each other, for both winters. The reported incidence of ILI was 251 per 10 000 persons in winter 1998/1999 and 202 per 10 000 persons in winter 1999/2000. The average in the winters since 1992/1993 combined was 234 per 10 000 persons (table 14).

Table 14 *Reported incidence of ILI per 10 000 persons from week 40 till week 20, by age category*

Age (years)	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	Mean
0-4	502	581	261	430	320	381	472	375	415
5-14	424	328	143	261	203	164	195	136	232
15-44	280	242	197	230	206	151	242	179	216
45-64	225	285	183	254	221	167	265	233	229
>64	194	236	122	200	195	172	211	207	192
All ages	286	280	182	248	248	172	251	202	234

Since winter 1992/1993, the reported incidence of ILI has been highest for children between 0 and 4 years of age. It was relatively low for 5-to-14-year-old children, especially in winter 1999/2000 (table 14, figure 23).

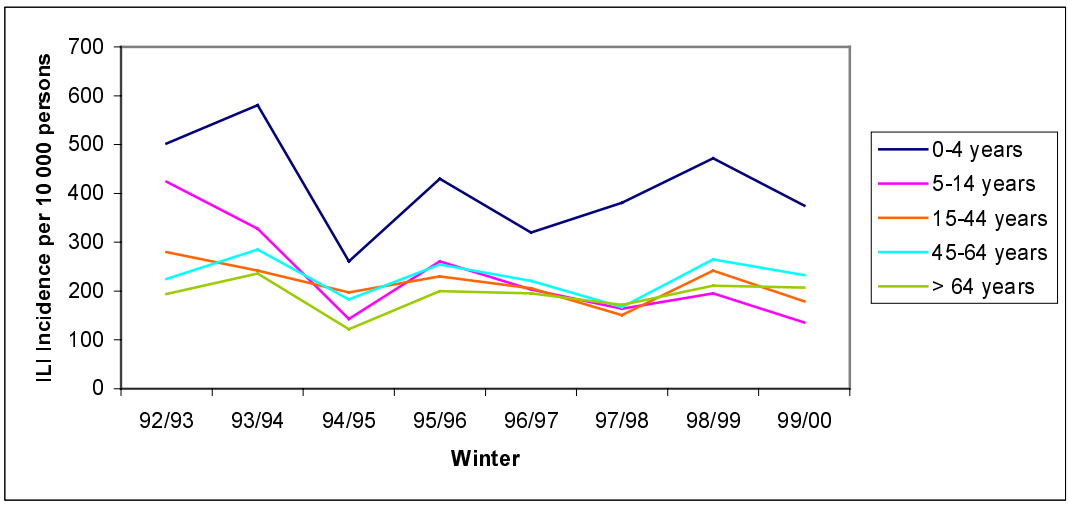

Figure 23 *Reported incidence of ILI from week 40 till week 20, by age category*

By combining the reported incidence of ILI and the number of influenza virus isolates, we can estimate the incidence of influenza virus infection in the Dutch population. With this estimation we should take the following into account:

a) not all infections with influenza virus will lead to (serious, ILI-like) symptoms;
b) not all people with a symptomatic influenza virus infection will consult their GP; it is estimated that about 33% of adults ([20-21] plus personal communication J.J. Kerssens, NIVEL) and 70% of children aged 0-4 years (personal communication J.J. Kerssens, NIVEL) consult their GP when they suffer from an ILI;
c) not all people who consult their GP for ILI are infected by influenza virus (see e.g. table 2 and 7);
d) influenza virus cannot be detected in all swabs from people who are really infected by the influenza virus;
e) GP's from the NIVEL sentinel surveillance network register ILI only five days per week;
f) GP's from the NIVEL sentinel surveillance network take swabs from a random selection of their ILI patients on four to five days per week.

The estimate of the incidence of influenza virus infection in the general population can be adjusted for factors b, c and e:

Influenza virus infection incidence per 10 000 persons = ILI * 7/5 * b * N_i/N_a
in which

ILI	=	number of ILI per 10 000 persons registered by the NIVEL GP's,
7/5	=	correction factor for the fact that the GP's register ILI five days per week,
b	=	correction factor for the number of ILI patients who do not consult their GP; 1.4 for children aged 0-4 years and 3 for people over 4 years of age,
N_i	=	number of swabs from ILI patients in which influenza virus was isolated,
N_a	=	number of swabs from ILI patients

N_i/N_a is called the 'laboratory-based correction factor'. This factor varies between age groups, with the highest value for children aged 5-14 years (table 15). All age groups and winters taken together, a swab has been taken of 8% (2 171 of 25 763) of the ILI patients registered. This percentage varied between age groups, with the highest in age group 15-44 years old (10%) and the lowest in patients over 64 years old (5%) (table 15).

Table 15 Calculation of laboratory-based correction factor, means of winters 1992/1993 – 1999/2000 from week 40 till week 20

Age (years)	Mean patient population NIVEL	ILI reported	Swabs from ILI patients (=N_a)	% of ILI patients sampled	Positive swabs from ILI patients (=N_i)	Laboratory-based correction factor
0-4	7 997	2 656	163	6	43	0.26
5-14	16 811	3 123	215	7	102	0.47
15-44	65 497	11 319	1 184	10	314	0.27
45-64	31 675	5 800	461	8	116	0.25
>64	18 674	2 865	148	5	32	0.22
All ages	140 653	25 763	2 171	8	607	0.28

Estimates of the incidence of influenza virus infection by using the formula above, showed the highest incidence in age group 5-14 years old when combining all winters since 1992/1993 (table 16, figure 24). However, the estimated incidence of influenza virus infection shows a decreasing trend over the eight winters studied. For the other age groups, the estimated incidence of influenza virus infection remained relatively constant over time. Over the eight winters studied, the lowest incidence of influenza virus infection was estimated for people older than 64 years.

Table 16 Estimated incidence of influenza virus infection per 10 000 persons from week 40 till week 20, by age category

Age (years)	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	Mean
0-4	260	300	135	222	165	197	244	194	215
5-14	845	654	285	520	225	327	389	271	462
15-44	312	270	219	256	229	168	270	199	240
45-64	238	301	193	268	246	176	280	246	242
>64	176	214	111	182	177	156	192	188	174
All ages	336	329	214	291	225	202	295	237	274

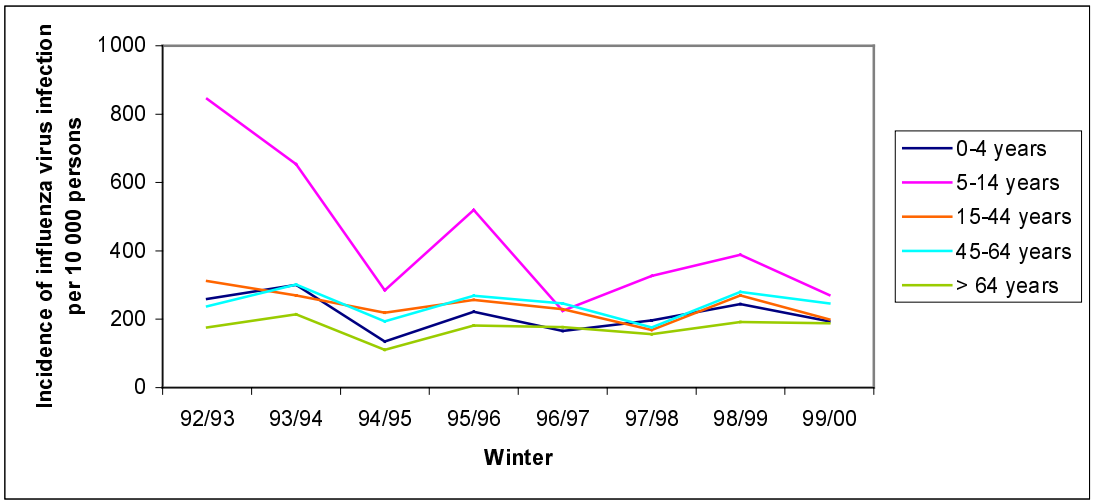

Figure 24 *Estimated incidence of influenza virus infection from week 40 till week 20, by age category*

When comparing the reported ILI incidence in GP patients with the estimated incidence of influenza virus infection in the population, it is striking that the reported ILI incidence is much higher than the estimated incidence of influenza virus infection for the age group 0-4 years. For the other age groups, the estimated incidence of influenza virus infection is higher than the reported ILI incidence, especially in the 5-to-14-year-olds (figure 25).

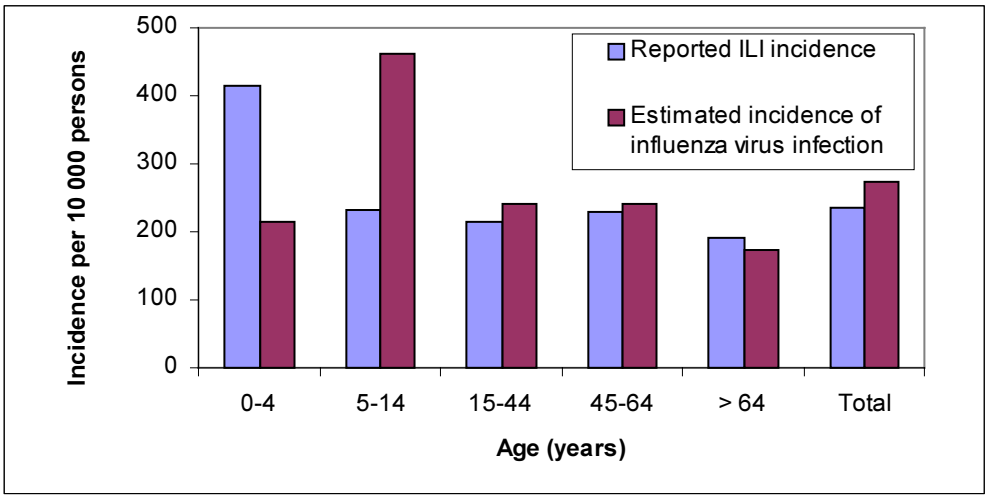

Figure 25 *Reported incidence of ILI in GP patients and estimated incidence of influenza virus infection in the general population, means of winters 1992/1993-1999/2000, from week 40 till week 20*

Both observations can be explained in different ways. First, the laboratory-based correction factor is relatively high for the age group of 5 to 14 years (0.47, table 15). In other words, the percentage of swabs found positive for influenza virus was 47% of all swabs from ILI patients in this age category, versus approximately 26% in the other age categories. An explanation for this could be that young children with influenza-like-symptoms visit their GP

in an earlier stage than older people, leading to a higher chance of detecting a pathogen if it is present. However, then we would expect the laboratory-based correction factor for children in the youngest age group to be in the same range as the one for children aged 5 to 14 years old. However, we did not observe this (laboratory-based correction factor=0.26, table 15). Furthermore, in winter 1998/1999 and 1999/2000, no differences between age categories were observed for duration of symptoms at the time of swab-taking.
Second, the correction factor for the number of patients with ILI not consulting their GP also attributes to the calculation of the estimated incidence of influenza virus infection. We used 1.4 as value for this correction factor for children aged 0-4 years and 3 for persons older than 4 years based on literature ([20-21] plus personal communication J.J. Kerssens, NIVEL). In a recent study, two GP's estimated that only 15% of people older than 4 years with ILI symptoms consult their GP and 33% of the young children[22]. This would result in a correction factor of 3 for 0-to-4-year-olds and 6.7 for people older than 4 years. With 3 and 6.7 as correction factors, estimated incidences of influenza virus infection in the general population would become significantly higher (model 2 in table 17). Thus using different values for this correction factor alters the estimated incidence significantly. Ideally, a new survey in the Dutch population should investigate which values are correct.

Table 17 *Estimated incidence of influenza virus infection in the general population, by different estimate of GP consulting rate, means of winters1992/1993-1999/2000, week 40 till week 20*

Age (years)	Reported incidence of ILI per 10 000 persons	Estimated incidence of influenza virus infection per 10 000 persons	
		Model 1[a]	Model 2[b]
0-4	415	215	460
5-14	232	462	1 031
15-44	216	240	537
45-64	229	242	541
>64	192	174	390

a) correction factor for the number of patients with ILI not consulting their GP is 1.4 for children aged 0-4 years, and 3 for persons older than 4 years

b) correction factor for the number of patients with ILI not consulting their GP is 3 for children aged 0-4 years, and 6.7 for persons older than 4 years

Even if all corrections made (item b, c and e on page 37) are correct, estimating the incidence of influenza virus infection as described here underestimates the real incidence.
The chance of detecting a pathogen if it is there, depends for instance on the time between the onset of symptoms and the moment the swab is being analysed in the laboratory (figure 21 in section 3.3.1). This would contribute to item d on page 37.
There is also a limit to the sensitivity of the detection methods used (also part of item d on page 37). For rhinovirus, RSV and enterovirus e.g. PCR has been shown to have a high added value compared with virus culture (tables 1 and 6).

The incidence of influenza virus infection in the general population was also estimated by influenza subtype (table 18). Influenza virus A(H3N2) clearly had the highest impact in all age categories. In children aged 5 to 14 years, influenza B also had a high incidence. Influenza A(H1N1) did not play a significant role in influenza virus incidence in these eight winters.

Table 18 *Estimated incidence of influenza virus infection per 10 000 persons, by subtype, means of winters 1992/1993 - 1999/2000 from week 40 till week 20*

Age (years)	A(H3N2)	A(H1N1)	B	Total
0-4	170	20	25	215
5-14	286	14	163	462
15-44	143	6	90	240
45-64	184	4	52	239
>64	136	0	38	174
All ages	180	8	86	274

3.4.2 Influenza incidence by degree of urbanisation and by region

Reported incidence of ILI was lowest in towns, and higher in villages and cities (table 19). By using the laboratory-based correction factor (table 20), the incidences of influenza virus infection in the general population by urbanisation were estimated (table 21). These estimates showed the highest incidence in villages. We expected a higher incidence of influenza virus infection in cities, since when people live closer together, the virus can spread more easily. Only in winter 1992/1993 and 1999/2000, the highest incidence was estimated for cities. In the other winters the highest incidence was estimated for villages. This could be explained by the higher laboratory-based correction factor in villages. From all patients with ILI reported in the eight winters combined, GP's from cities took more swabs (16%) than GP's in villages and towns (5% and 6% respectively). However, the percentage of the swabs found positive was highest in villages (34%) (table 20). Perhaps the higher estimated incidence of influenza virus infection in villages is due to differences in the contacts with other people or in the general condition of people according to degree of urbanisation.

Table 19 *Reported incidence of ILI per 10 000 persons from week 40 till week 20, by degree of urbanisation*

Urbanisation	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	Mean
Village	299	310	236	328	279	264	354	180	281
Town	235	240	153	219	184	140	218	194	198
City	400	357	226	283	281	227	306	254	292

Table 20 *Calculation of laboratory-based correction factor, means of winters 1992/1993 – 1999/2000 week 40 till week 20, by degree of urbanisation*

Urbanisation	Mean patient population NIVEL	ILI reported	Swabs from ILI patients (=N_a)	% of ILI patients sampled	Positive swabs from ILI patients (=N_i)	Laboratory-based correction factor
Village	21 042	4 731	232	5	80	0.34
Town	92 141	14 457	932	6	240	0.26
City	27 454	6 435	1 017	16	294	0.29

Table 21 *Estimated incidence of influenza virus infection per 10 000 persons in the general population from week 40 till week 20, by degree of urbanisation*

Urbanisation	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	Mean
Village	433	449	342	475	404	382	513	261	407
Town	254	260	165	237	199	151	236	210	214
City	486	433	274	344	341	276	372	308	354

For this surveillance, the Netherlands is divided into four regions: north (the provinces Groningen, Friesland and Drente), east (Overijssel, Flevoland and Gelderland), west (Noord-Holland, Zuid-Holland and Utrecht) and south (Zeeland, Noord-Brabant and Limburg). Despite the fact that the Netherlands is a small country, regional differences in ILI activity were found. Over the eight winters combined, the mean ILI incidence reported was highest in the east of the Netherlands: 271 per 10 000 persons. However, the last three winters the highest ILI incidence was found in the western region. Continuously, in the north the lowest incidence was found (table 22). GP's in the west took relatively more swabs (10%) than GP's in the remainder of the country (table 23). By using the laboratory-based correction factor, the incidences of influenza virus infection in the general population showed the same ranking order as the reported ILI incidence in GP patients; highest overall incidence in the east with the incidence in the western region highest in the last three winters (table 24). There is no obvious reason for these regional differences.

Table 22 Reported incidence of ILI per 10 000 persons from week 40 till week 20, by region

Region	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	Mean
North	201	192	91	176	115	133	146	168	153
East	382	363	237	322	259	178	229	201	271
West	271	292	151	226	230	204	327	224	241
South	295	242	244	265	215	151	251	191	232

Table 23 Calculation of laboratory-based correction factor, means of winters 1992/1993 – 1999/2000 week 40 till week 20, by region

Region	Mean patient population NIVEL	ILI reported	Swabs from ILI patients ($=N_a$)	% of ILI patients sampled	Positive swabs from ILI patients ($=N_i$)	Laboratory-based correction factor
North	27 342	2 821	166	6	43	0.26
East	32 299	6 860	435	6	138	0.32
West	52 120	9 866	1 026	10	306	0.30
South	32 808	6 068	554	9	127	0.23

Table 24 Estimated incidence of influenza virus infection per 10 000 persons in the general population from week 40 till week 20, by region

Region	1992/93	1993/94	1994/95	1995/96	1996/97	1997/98	1998/99	1999/00	Mean
North	219	209	99	191	125	145	159	183	166
East	509	484	316	429	345	237	305	268	362
West	339	366	189	283	288	256	410	281	301
South	284	233	235	255	207	145	242	184	223

4. Concluding remarks and recommendations

Acute respiratory infections are very common in the general population; yearly, upper airway complaints are the reason for 3.2 million contacts to general practitioners[23]. For effective prevention and control of ARI, it is essential to know the relative role of the various micro-organisms. For example, in only one third of the nose/throat swabs from patients registered with ILI over the last eight winters, influenza virus was detected. In one third another micro-organism was detected, being a potential cause of the symptoms. Unjust claims of influenza vaccine failure can be refuted with these findings.
Further, the finding that in more than half of the swabs from ARI (including ILI) patients a virus is detected should support general practitioners in restrictive prescription of antibiotics for ARI. This is a positive development with regard to the growing prevalence of resistance to antibiotics[24]. Another example is related to development of vaccines against infection with RSV and parainfluenza virus. When these vaccines become available it is essential to know of whom the target groups for vaccination exist. The same holds for the new antivirals that have been developed against infections with influenza virus (neuraminidase inhibitors)[25-26] and that are being developed against infections with rhinovirus[27]: e.g. since rhinoviruses are often detected in ARI patients, more effort could be put into developing suitable antivirals against these viruses. For the neuraminidase inhibitors to be effective it is essential that they are taken within 48 hours after onset of symptoms. Ross et al.[28] found that only 23% of the ILI patients in the United Kingdom consulted their GP within 48 hours after onset of symptoms. In our surveillance this percentage was 36% for winters 1998/1999 and 1999/2000 combined. If prescription of neuraminidase inhibitors would be encouraged, patients should be made clear to consult their GP in an earlier stage of disease. Registration of prescripts of neuraminidase inhibitors by the GP's in the sentinel network takes place since January 2000. These drugs were prescribed to less than 1% of the registered ILI patients between January and May 2000.

The NIVEL/RIVM respiratory surveillance is a unique system for various reasons. Most systems survey hospital patients whereas this surveillance yields information on GP patients. Surveillance of GP patients next to hospital patients has added value. GP patients are likely to better reflect the general population. For instance it is shown that RSV infections do not occur exclusively in babies[29]. Influenza virus types isolated from GP patients sometimes differ from those isolated from hospital patients[30]. Also unique in the NIVEL/RIVM respiratory surveillance is that it contains information on ILI since 1970 and on the aetiology of ARI including ILI since winter 1992/1993.

So far, the NIVEL/RIVM respiratory surveillance consisted of analysing swabs from patients with ARI, including ILI, but only ILI patients have been registered. Therefore, estimates of the incidence of the various micro-organisms can only be based on swabs from ILI patients

and not on all swabs. If registration of all ARI patients would be added to the surveillance, the data could be used more completely. Adding ARI registration to ILI registration also enables comparison with the reports of fourteen European countries, participating in the European Influenza Surveillance Scheme (EISS)[31]. In some countries registration of ARI takes place, in other countries the number of ILI patients is registered, and in a few both.

For rhinovirus, enterovirus and RSV both virus culture and PCR were used in this surveillance. PCR proved to be more sensitive than viral culture for these m.o.'s. Using PCR to detect other m.o.'s as well could lower the diagnostic deficit. However, there is still little known about how long viral RNA can be detected after an ARI[6]. At least theoretically a positive PCR result can be obtained after a non-recent infection. Because a positive PCR result means that a part of the genomic material of the m.o. was present that could serve as a template for PCR. This does not necessarily indicate a viable m.o., i.e. a recent infection. Comparing PCR results from swabs obtained from people with and without ARI signs and symptoms could shed more light on this matter, as well as analysing follow-up samples taken regularly and frequently from symptomatic patients with a positive PCR result.

The NIVEL/RIVM respiratory surveillance is limited to people who develop signs and symptoms of an ARI **and** who consult their GP for these symptoms. By combining the reported ILI incidence with the number of influenza viruses isolated, we estimated the incidence of influenza virus infection in the general population. Essential in this estimate is the correction factor for people experiencing ILI signs and symptoms, but **not** visiting their GP. We assumed that 70% of patients 0 to 4 years old and 33% of patients over 4 years old with ILI symptoms consult their GP. However, these assumptions are based on American data[20], Dutch data from one general practice in winter 1990/91[21] and a relatively small Dutch survey from 1994 (personal communication J.J. Kerssens, NIVEL). In a recent study estimates by two GP's varied between 15% and 33%[22]. Using different values for this correction factor has a great impact on the estimated incidence of influenza virus infection (see table 17). Ideally, a new survey in the Dutch population should investigate which values are correct.

In summary:

- this unique surveillance is a valuable system for monitoring the incidence of ILI and of associated pathogens in the Netherlands, and should therefore be continued,
- the microbiological data collected in this surveillance could be used more completely if patients consulting their GP for signs and symptoms of ARI other than ILI would be registered, next to the current registration of ILI patients,
- the clinical value of the PCR could be investigated further,
- it would be worth-while to re-investigate the percentage of people in the general population with signs and symptoms of ILI (and preferably also other ARI) that consult their GP.

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Appendix I Mailing list

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Appendix II Form accompanying nose/throat swabs

LIS/VIR-F117

Inzendformulier monsters NIVEL/RIVM virologische surveillance van IAZ en andere acute respiratoire infecties, seizoen 1999/2000

Naam arts: ..... Code peilstation: .....

Naam patiënt: ..... Geslacht patiënt: M/V

Geboortedatum patiënt: ……. / ..…. / ..…… (dag/maand/jaar) Ziekteduur: …… dagen

Datum afname materiaal: …… / …… / ….…. (dag/maand/jaar) □ neuswat □ keelwat

Symptomen:
- □ Acuut begin
- □ Hoesten
- □ Rhinorrhoe
- □ Keelpijn
- □ Rode keel
- □ Dyspnoe
- □ Koorts, …… °C
- □ Malaise
- □ Spierpijn
- □ Hoofdpijn
- □ Buikpijn
- □ Misselijk
- □ Braken
- □ Diarree
- □ Exantheem

Diagnose:
- □ Sinusitis
- □ Otitis
- □ Conjunctivitis
- □ Pharyngitis
- □ Pseudocroup
- □ Tonsillitis
- □ Laryngitis
- □ Bronchitis
- □ Bronchiolitis
- □ Pneumonie
- □ Tracheïtis

Heeft u de patiënt aangemeld als IAZ?	□ ja	□ nee
Zijn er soortgelijke zieken in de omgeving van de patiënt?	□ ja	□ nee
Heeft de patiënt een influenza-vaccinatie voor dit seizoen?	□ ja	□ nee
Lijdt de patiënt aan een bewezen respiratoire allergie?	□ ja	□ nee
Heeft de patiënt regelmatig contact met kinderen < 5 jaar ?	□ ja	□ nee
Lijdt de patiënt aan immunosuppressie?	□ ja	□ nee
Rookt de patiënt?	□ ja	□ nee
Zo nee, heeft de patiënt ooit gerookt?	□ ja	□ nee

- **Materiaal afnemen bij patiënten met een IAZ of andere acute respiratoire infectie.**
- **Materiaal afnemen tot uiterlijk 5 dagen na het begin van de ziekte, omdat in later afgenomen materiaal te weinig virus aanwezig is.**
- **Materiaal in transportvloeistof direct versturen. Kan dit niet, dan bewaren bij 4°C (niet invriezen).**
- **De onbeënte transportvloeistof kan bij kamertemperatuur, in het donker (d.w.z. in ongeopende verzendzak), worden bewaard. De vloeistof blijft op deze wijze 2 jaar bruikbaar.**
- **Materiaal op maandag tot en met donderdag naar het RIVM sturen.**
- **Maximaal 2 monsters per week insturen.**
- **Voor logistieke vragen kunt u contact opnemen met Cees Verweij, tel. 030-274 2392 of 030-274 2247.**
- **Als u inhoudelijke vragen heeft, bijvoorbeeld over de uitslag, kunt u terecht bij Wendelien Dorigo (arts-microbioloog), tel. 030-274 3705 of 030-274 2889 (secretariaat).**

Niet invullen door inzender

Datum ontvangst materiaal: …... / …... / ..…… (dag/maand/jaar)

Uitslag: ..... Datum: ..…. / ..…. / ….…. (dag/maand/jaar)