Disease burden of the most commonly detected respiratory viruses in hospitalized patients calculated using the disability adjusted life year (DALY) model

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ABSTRACT

Background: The most common acute infections occur in the respiratory tract. Recent discoveries of several novel viruses have markedly increased the repertoire of agents understood to cause presentations of acute respiratory disease.
Objectives: Further understanding is needed of the relative importance of newly discovered pathogens in the clinical setting to provide clinicians with an indication of appropriate diagnostic and therapeutic targets. To address this, quantification of the disease burden of respiratory viruses in hospitalized patients was undertaken.
Study design: Disease burden caused by respiratory viruses in hospitalized patients was quantified using the World Health Organization endorsed DALY model. Diagnostic testing results from samples collected over three years for adenovirus (AdV), influenzas A and B, parainfluenza viruses 1, 2 and 3 (PIV-1, -2 and -3), respiratory syncytial virus (HRSV), and previously published retrospective screening for human metapneumovirus, rhinoviruses, and four respiratory coronaviruses were applied to the DALY model. Disability weights were calculated per 1000 hospitalized patients in age banded groups.
Results: Strikingly different disease burden profiles were observed in children and adults. Adenoviruses were among the leading cause of respiratory presentations in children but not adults. HRSV and influenza A were consistently one of the greatest causes of disease regardless of sampled population. Rhinoviruses and PIV-3 were significant pathogens in all groups except those aged 16–64 years. In immunocompromised patients rhinoviruses were the leading viral cause of disease.
Conclusions: These analyses provide a framework which can be used to identify where finite resources should be directed in respiratory therapeutics and vaccine development.

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1. Introduction

Infection of the respiratory tract is the most common type of acute infection globally in both adults and children. In Europe, lower respiratory tract infection (LRTI) is the eighth leading cause of disease.[1] In recent years, discovery of several novel viruses associated with high morbidity respiratory outcomes (including human metapneumovirus (HMPV),[2] human coronaviruses (HCoVs) NL63[3] and HKU1[4] and human rhinovirus species C (HRV-C)[5]) has markedly increased the repertoire of agents associated with acute respiratory tract infections (ARTI). This presents significant challenges for microbiology diagnostics.

Attempts to prevent respiratory virus infections are usually limited to vulnerable high risk groups such as the immunocompromised or the elderly, and include measures such as vaccination, cohorting to reduce exposure risk and prophylaxis. Vaccination to protect from infection with these principal agents of respiratory disease is only widely available for influenzas A and B. In respiratory seasons where HRSV is severely epidemic, HRSV positive patients may be cohorted in attempt to reduce transmission of the virus to other patients. Near patient testing (NPT) for HRSV infection is recommended for infants requiring hospital admission with acute bronchiolitis to facilitate timely cohorting.[6]

Only one study has previously attempted to quantify disease burden attributable to respiratory viruses.[7] Respiratory tract infections in community based adults over the age of 60 were prospectively diagnosed, and disease burden associated with a panel of respiratory viruses was quantified using the method of

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the US Institute of Medicine to prioritise new vaccine development strategies for diseases of importance in the United States.

2. Objectives

The importance of clinical outcomes of infections with recently discovered and lower prevalence viruses have not been systematically quantified. The Disability Adjusted Life Year (DALY) model was endorsed by the World Health Organization in 1996 as a methodology for prioritizing interventions in the health sector based on their potential to reduce burden of disease. DALYs combine time lost due to morbidity and mortality to attribute a single numerical value as a measure of the associated burden caused by a disease outcome. This model has been employed to quantify the disease burden of the most commonly detected patients in an age stratified hospitalized patient cohort to provide an indication of where resources might be most effectively used for diagnostics, patient management and treatment.

3. Study design

Samples. Between 1 July 2006 and 30 June 2009, results of routine diagnostic screen tests of samples collected from hospitalized patients were available for adenovirus (AdV), influenza A (seasonal) influenza B, parainfluenza viruses 1–3 (PIV-1 to -3) and HRSV. Additionally, virus testing results were available for a subset of samples for HMPV,[8] human rhinoviruses species A, B and C together (456 samples)[9] and coronaviruses HCoV-229E, HCoV-HKU1, HCoV-NL63 and HCoV-OC43[10] from previous studies. For the purpose of this study, additional screening for rhinoviruses was undertaken on 579 samples collected between September 2008 and February 2009 using the previously described PCR protocol[9] for pan-rhinovirus detection. Ethical approval for this work was granted by the Lothian Regional Ethics Committee (08/S11/02/2). All samples in the respiratory archive were subject to anonymisation and so the institutional review board specifically waived the need for patient consent. Retained patient information included age, sex, place of sample collection (e.g., accident and emergency, specified hospital ward) and clinical information recorded on referral forms.

Samples testing positive for more than one of the viruses included in the study were considered separately as mixed infections and excluded from subsequent analyses. Due to the differences in sampling strategy from different patient populations, it was appropriate to undertake analyses in an age-stratified way and to consider immunocompromised patients separately.

Clinical data. The DALY model quantifies morbidity associated with different clinical outcomes by assigning disability weights on a scale between 0 and 1, whereby 0 is no morbidity and 1 is death. Clinical data relating to some samples could be assigned to more than one category, and so categories were treated in a hierarchical fashion with precedent given in the order:

1. *Immunocompromised.* Included samples collected from patients with neoplasia, a known immunosuppressive disorder and transplant patients.
2. *Chronic.* Samples collected from patients with chronic respiratory conditions such as asthma and cystic fibrosis.
3. *Lower respiratory tract infection (LRTI).* Samples taken from patients with pneumonia, bronchiolitis, bronchitis, influenza-like illness (ILI), tested for HRSV using an NPT,[11] chest infection, shortness of breath, atypical pneumonia, acute respiratory distress syndrome (ARDS), difficulty breathing, community acquired pneumonia, cough, wheeze and respiratory failure if

a sample number from the same patient preceding this clinical report was associated with URTI or LRTI.
4. *Upper respiratory tract infection (URTI).* Tonsillitis, coryza, rhinorrhoea and sore throat were classified as URTI.
5. *Other.* Clinical evidence for respiratory tract infection was indeterminate e.g., apnoea, encephalitis, meningitis, ventilated, pyrexia, sepsis, headache, conjunctivitis.
6. *None.* Information/symptoms were not associated with respiratory tract infection e.g., urinary tract infection.
7. *No data.* No information about the clinical presentations of the patient from whom the sample was collected was recorded for 6225 (48.2%) of samples.

URTI and LRTI only were identified as the only outcomes for which a causal association between infection and presentation could be assumed and so only these categories were analysed using the DALY model. Where clinical data indicated no LRTI/URTI, associated samples were excluded from disease burden calculations (including samples for which no clinical data was available). Samples collected from 'chronic' patients were excluded. These included a very small proportion of all samples and current understanding of disease exacerbation precludes estimations of the proportion of these patients who would have respiratory presentations, how severe these would be and how significantly an infecting virus might contribute to these.

Literature review identified death and infection as a cause of asthma as significant outcomes of respiratory virus infection (refer to S3; supplementary material). The rates of these outcomes due to infection with each respiratory virus were estimated from the literature (Tables S1–S3; supplementary material) and incorporated into the model.

A consequence of the hierarchical categorization of clinical data was that for immunocompromised patients no information about respiratory presentations was available. There are two approaches to estimating the rate of URTI and LRTI in immunocompromised patients. The first is from the literature. The second is using the rates observed in otherwise healthy hospitalized individuals in Edinburgh. The latter approach has been chosen: the Edinburgh archive may not reflect other sampled populations in terms of the criteria for sample testing, the viruses present and clinical definitions.

The rates of death, LRTI and URTI due to respiratory virus infection were not estimated in immunocompromised patients in whom a respiratory virus infection was not identified due to the paucity of published studies of this patient group.

Disability weighting. The Global Burden of Disease study[12–14] (the 'gold standard' of studies quantifying disease burden using DALYs) weight assigned to LRTI is 0.279. In the study population, LRTI was assumed to be the cause of hospitalization, and so a higher disability weight was deemed appropriate. The range in disability weights considered to reflect the possible implications of hospitalization due to LRTI fell between 0.279 and 0.4. A weight of 0.4 was considered appropriate for an individual admitted to an intensive therapy unit (ITU). The disability weight attributed to LRTI was assigned on a virus-dependent basis in the range of 0.279–0.4 proportional to the rate of ITU admission on diagnosis of LRTI. For example, 29.4% of patients with LRTI testing singly positive for AdV were admitted to ITU, and so a disability weight of $((0.294 \times (0.4 - 0.279)) + 0.279=) 0.315$ (3 s.f.) was assigned.

URTI was not assumed to be the cause of hospitalization, but was assumed to be caused by the infecting respiratory virus. The Global Burden of Disease study disability weight of 0 was deemed inappropriate as a clinician had felt the presentations warranted documentation and so the value for pharyngitis of 0.07 was applied.[14]

Disability weights for LRTI calculated in otherwise healthy patients were applied to the immunocompromised population.

ITU admission of an immunocompromised patient could not be assumed to be due to respiratory infection and so this method was preferred to a distinct disability weight calculation in this group.

For infection as a cause of asthma, the Global Burden of Disease study disability weight of 0.043 was applied.

Other model parameters. The duration of disability associated with URTI and LRTI was estimated from the literature on a virus specific basis (Tables S1–S3; supplementary material).

Using the assumptions described by Murray,[14] application of the same social and life expectancy parameters to immunocompromised individuals as otherwise healthy individuals was considered the most ethically viable approach for the purpose of this study (Dr. Claudia Stein, pers. Comm.).

The DALY model regards life expectancy as the maximum achievable length of life. Sex specific mean life expectancies in Scotland between 2006 and 2009 as described by the Office for National Statistics were used. DALYs were calculated in an age stratified manner with immunocompromised patients considered separately.

Calculation of disease burden. Estimates of the burden of disease associated with each respiratory virus were calculated using the DALY formula[15]:

$$-\left[\frac{DCe^{-\beta a}}{(\beta+r)^2}[e^{-(\beta+r)(L)}(1+(\beta+r)(L+a))-(1+(\beta+r)a)]\right]$$

where D is the disability weight; C and β are parameters of the age weighting function (parameters of age weighting function used were as for the World Bank study[12,14]); a is the age at disease onset (mid-range values for each age band were applied); L is the duration of disability, which in case fatalities is the time between age of onset and life expectancy; and r is the social discount rate. The social discount rate using in the Global Burden of Disease study of 0.03 was applied.[12,14]

Absolute DALY measures were standardized to a common metric (DALYs per 1000 population) by dividing the total number of DALYs calculated by the proportion of samples with clinical data available for each virus in the study. This procedure enabled a fair comparison between viruses where the number of samples taken and the number of samples with clinical data available varied. The study population of hospitalized individuals does not reflect the population of Scotland, and quantification of community based disease burden associated with respiratory viruses was outside the scope of this study.

4. Results

Quantification of the disease burden associated with each respiratory virus required calculations of sampling frequencies, breakdown of clinical data, and determination of ITU admission rates. The number of samples tested and the frequency of samples testing singly positive for each virus are summarized (Table 1). Between 1035 (rhinoviruses) and 12,883 (HRSV and influenza A) samples were tested for each virus. Detection frequencies ranged between 0.33% (PIV-2) and 18.1% (rhinoviruses). The rates of admission to ITUs (Table 1) associated with each virus are given, and fell in the range 8.5% (rhinoviruses) and 20.0% (AdV). The rates that respiratory virus infections were associated with LRTI, URTI, no respiratory outcome and immunocompromised patients are indicated (Fig. 1). HRSV was more frequently associated with LRTI than other respiratory viruses, whereas PIV-3 was frequently associated with upper respiratory presentations in comparison with other viruses. The proportion of virus positive samples collected from immunocompromised patients was highest for rhinoviruses and lowest for HRSV.

Burden of disease of the most commonly detected respiratory viruses are described (Table 2). In children under 5 years of age, HRSV was clearly the leading cause of disease, attributable with nearly 70 DALYs per 1000 hospitalized population. Adenoviruses and rhinoviruses were also significant pathogens in this age group. The parainfluenza viruses caused less disease, with PIV-1 and PIV-2 associated with the least disease of those viruses analysed.

In older children between 5 and 16 years old, HRSV was again the leading cause of disease. Different sampling strategies between age categories complicates direct comparison of different age groups, but a much lower score in this age group compared with the younger children implies less disease in older children. Rhinoviruses and adenoviruses again scored highly compared with other viruses in this age group, and parainfluenza viruses maintained a minimal role in disease.

In adults between 16 and 64 years of age, influenza A was the leading cause of disease, although the score of 2.2 DALYs per 1000 population is low in comparison with the leading causes of disease in other age groups. In this age group HMPV, HRVs and PIV-1 were not associated with any respiratory disease in the hospitalized population.

Influenza A retained its status as leading viral cause of disease in older adults, with a much higher DALY score of 107.9. Influenza B and HRSV also scored highly. While we fully acknowledge the difficulties in comparing disease burden between age groups (see above), it is interesting that the score for HRSV in this age group of 41.0 is higher than that observed in under 5 s of 67.7.

In immunocompromised subjects, HRSV and rhinoviruses scored similarly at the top of the table. Adenoviruses, influenza A, PIV-3 and HMPV had comparable DALY scores in immunocompromised patients. Adenoviruses are more frequently associated with disease outcomes outside the respiratory umbrella and so caution is required with interpretation of this finding. Coronaviruses caused least disease in the immunocompromised patients.

Between age groups, the burden of disease caused by different respiratory viruses was remarkably variable (Fig. 2). Most striking was the increasing importance of influenza viruses A and B with age, occurring inversely with the decreasing disease burden of adenoviruses. Parainfluenza- and corona- viruses were of maximal impact in the intermediate age brackets encompassing 6–64 year olds, which is largely attributable to the lack of disease caused by other viruses in these patient groups. HRSV was a substantial cause of disease regardless of age. HMPV and HRVs caused more disease in the under 16 s than in adults.

The ratio of disease burden attributable to virus infections compared to that of respiratory disease with non-viral or unidentified causes diminished with increasing age (Table 2), likely reflecting the increasing incidence bacterial infections in older age groups. A secondary contributor to this may be reduced disease burden of respiratory virus infections with increasing age.

Disease burden profiles were compared between males and females (Fig. 3). While greater scores for disease burden were usually observed in males than females for each virus, the proportion of disease attributable to each virus was not different between males and females.

5. Discussion

In the hospitalized population, HRSV was the leading cause of disease across children and immunocompromised patients and was a significant cause of disease in adults. The closely related HMPV caused disease in all hospitalized age cohorts (although this was less than HRSV) consistent with previous reports of its association with severe disease and mortality in older individuals.[16–20]

Table 1
Detection of the most common respiratory viruses.

Virus	No. samples tested in each age range (yrs)					No. single infections[a]	No. positive samples[a]	Detection frequency (%)	ITU admission rate (regardless of clinical presentation)
	All	<5	6–15	16–64	>65				
AdV	12,486	7025	1507	3059	895	464	610	4.89	20.0
Coronavirus	11,529	6339	1378	2989	823	164	183	1.59	10.9
HMPV	7078	4202	893	1560	423	87	103	1.46	17.5
HRSV	12,883	7096	1528	3333	926	990	1273	9.88	15.6
HRVs (all)	1035	612	110	225	88	158[b]	187[b]	18.1[b]	8.5
-HRV-A							82	97	9.37	
-HRV-B							14	14	1.35	
-HRV-C							56	63	6.09	
Influenza A	12,883	7096	1528	3333	926	305	391	3.04	16.1
Influenza B	12,558	6901	1497	3266	894	110	133	1.06	12.8
PIV-1	12,830	7068	1520	3317	925	57	72	0.56	11.1
PIV-2	11,989	6553	1447	3130	859	30	39	0.33	15.4
PIV-3	12,831	7068	1521	3317	925	257	333	2.60	11.7
Mixed infection	3081	1845	420	658	158	134	157	5.09	13.0

[a] Represents the number of samples in which virus was detected as the sole pathogen (excepting samples analysed for mixed infections).
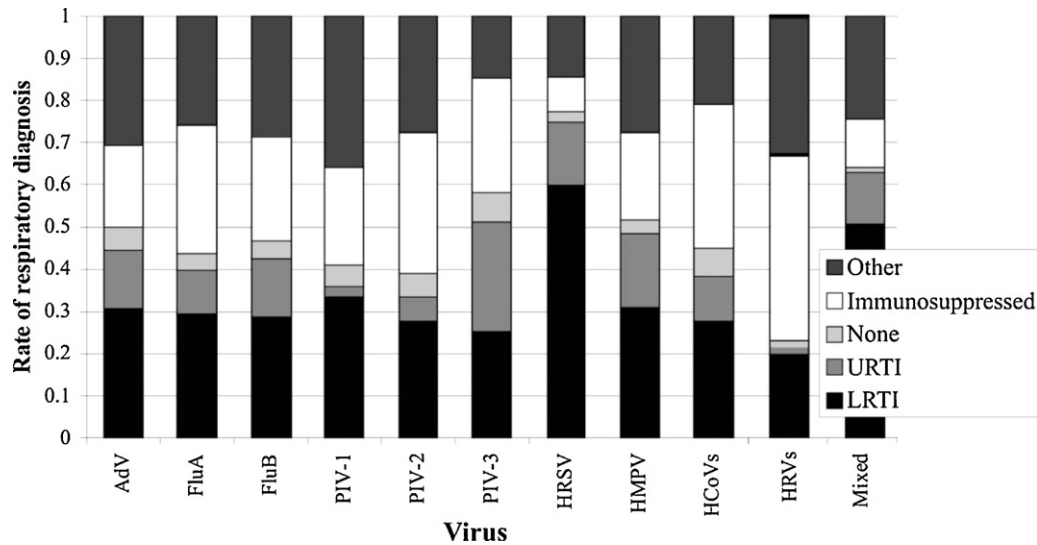[b] Thirteen HRV positive samples were untyped.

Fig. 1. Clinical presentations associated with the most commonly detected respiratory viruses. AdV, adenovirus; Flu, influenza; PIV, parainfluenza virus; HRSV, human respiratory syncytial virus; HCoVs, human coronaviruses; HRV, human rhinovirus.

Table 2
Disease burden attributable to the most commonly respiratory viruses in hospitalized patients calculated per 1000 population.

<5 yrs		6–15 yrs		16–64 yrs[b]		>65 yrs[b]		Immunocompromised	
Virus	DALY score	Virus	DALY score	Virus	DALY score	Virus	DALY score	Virus	DALY score
No virus	89.4	No virus	43.8	No virus	70.4	No virus	1166		
Total viruses[a]	247.9	Total viruses	45.0	Total viruses	6.4	Total viruses	231.4		
HRSV	67.7	HRSV	22.9	Flu A	2.2	Flu A	107.9	HRSV	6.8
AdV	49.7	HRVs	8.8	HRSV	1.0	Flu B	70.0	HRVs	6.5
HRVs	34.4	AdV	3.0	PIV-3	0.7	HRSV	41.0	AdV	3.5
Flu A	6.8	Flu A	1.5	Flu B	0.6	HRVs	8.1	Flu A	3.5
PIV-3	4.4	HCoVs	0.9	AdV	0.4	HMPV	2.1	PIV-3	3.1
HMPV	4.0	HMPV	0.6	PIV-2	0.4	PIV-3	1.2	HMPV	2.8
HCoVs	1.7	Flu B	0.5	HCoVs	0.3	HCoVs	0.5	Flu B	1.4
Flu B	1.7	PIV-3	0.4			PIV-1	0.4	PIV-2	0.7
PIV-1	0.7	PIV-1	0.4			AdV	0.3	PIV-1	0.5
PIV-2	0.3	PIV-2	0.3					HCoVs	0.3

[a] Total for all viruses includes disease burden of mixed infections, which are not shown as only a proportion of samples were tested for mixed infections, but are included as this gives a closer estimate of the total disease burden of respiratory viruses.
[b] In 16–64 year olds, no infections with PIV-1, HMPV or HRVs were associated with respiratory presentations, and in over 65 year olds, no single infections with PIV-2 were identified.
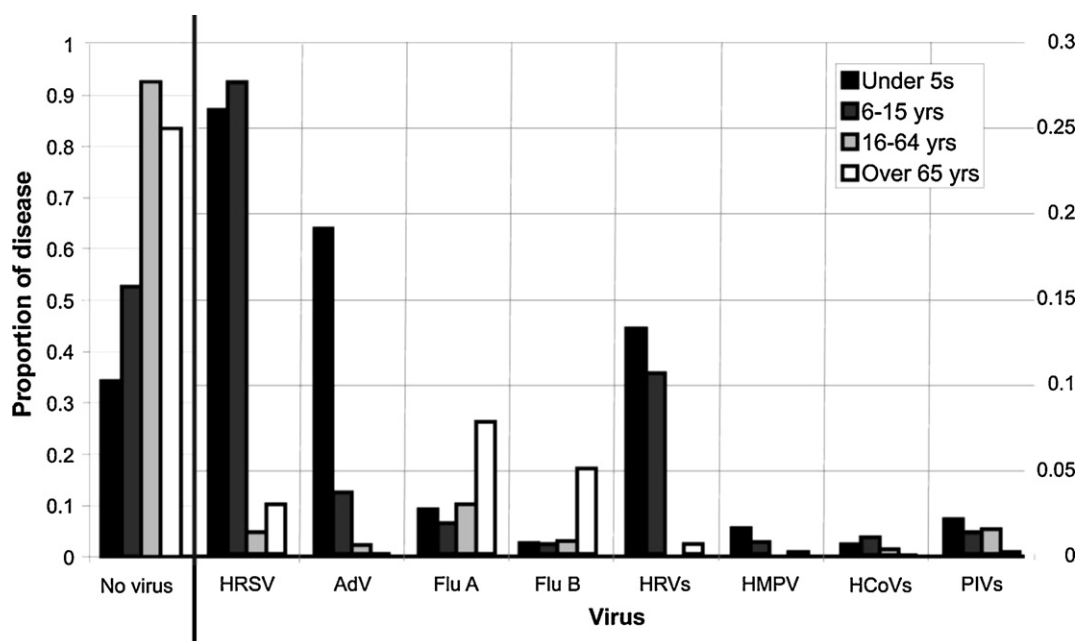
Fig. 2. Relative proportion of disease burden caused by the most commonly detected respiratory viruses by age group. AdV, adenovirus; Flu, influenza; PIV, parainfluenza virus; HRSV, human respiratory syncytial virus; HCoVs, human coronaviruses; HRV, human rhinovirus.

Rhinoviruses were a significant cause of respiratory illness, particularly in young children (Fig. 2). There are three known species of rhinoviruses (HRV-A, HRV-B and HRV-C), comprising well over a hundred types[21–24]; around a quarter to a third of all rhinovirus detections in the Edinburgh study population are of species C.[25] There is increasing evidence to suggest that HRV-C types are of similar clinical relevance as rhinoviruses assigned to other species.[5,26,27] The finding that rhinoviruses are a leading cause of acute viral respiratory illness in immunocompromised patients presents them as important opportunistic pathogens. These data together highlight the importance of including rhinoviruses in diagnostic screening protocols.

Influenza was the leading viral cause of disease in all hospitalized patients over the age of 16 years. The increasing morbidity of influenza resembles infection outcomes of other human viruses, such as measles, varicella zoster virus and Epstein Barr virus which show greater disease severity in adults compared to children.

No differences in the proportion of disease burden attributable to the most commonly detected viruses were found between males and females (Fig. 3). In infant males, a relatively smaller airway size compared with females of the same age increases likelihood of blockages and so exacerbated respiratory disease.[28] Age stratified serologic study has demonstrated seroconversion rates approaching 100% for all ubiquitous respiratory viruses studied,[2,29–34] when
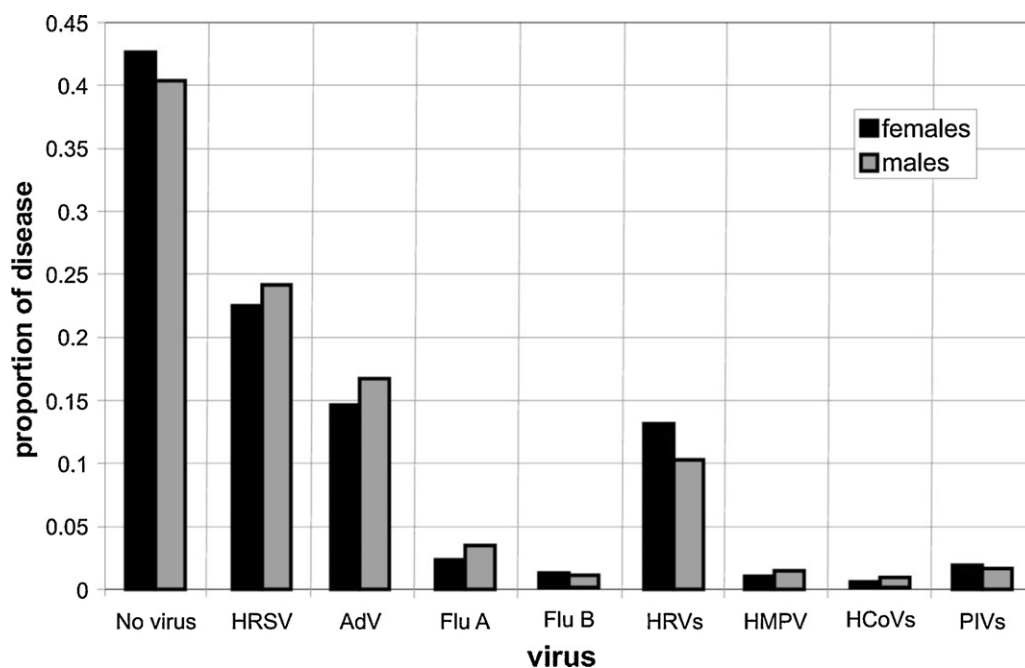

Fig. 3. Relative disease burden caused by the most commonly detected respiratory viruses in males compared with females. AdV, adenovirus; Flu, influenza; PIV, parainfluenza virus; HRSV, human respiratory syncytial virus; HCoVs, human coronaviruses; HRV, human rhinovirus.

physiological differences of the lungs between males and females are still apparent. The greater disease severity observed in young males compared with their female peers during HRSV infection[35] may therefore be more widely applicable to other respiratory virus infections.

The differences between community based and hospital based causes of disease burden are apparent by comparison of this data with the Nicholson study,[7] which determined the following virus rankings in descending order of importance in community based adults over 60 years of age:

1. Rhinoviruses (study predated the discovery of rhinovirus species C)
2. Causes of unknown aetiology
3. Coronaviruses (study prior to discovery of HCoV-HKU1 and HCoV-NL63)
4. Influenza viruses A and B
5. HRSV

The study did not quantify the disease burden of PIV-3 or HMPV, which were the fifth and sixth greatest causes of disease in adults over 65 years of age, ahead of coronaviruses. This study predated the discovery of HMPV, two of the four coronaviruses and rhinovirus species C. Nicholson found rhinoviruses were associated with greater disease burden that coronaviruses in the community, in keeping with observations in the hospitalized population. Influenzas A and B were far more significant in hospitalized than community based adults.

The DALY model is dynamic – changing prevalence, susceptible population and clinical presentations can be captured by and incorporated into the model to determine the worst causes of disease in real time. Though not investigated here, it is likely that DALY scores will fluctuate between seasons and year on year. The value of this method in allowing rapid determination of the clinical significance of recently discovered (or newly emerged) viruses is evident.

With the development of large-throughput screening techniques and improving clinical data management, there is increasing potential for future studies of this kind to identify the greatest causes of disease to inform diagnostic and therapeutic decision making. This study provides a framework for prospective analyses which capture clinical data for 100% of patients and follow up to determine disease duration and case fatality rates for both viral and bacterial respiratory pathogens. Only when such data is available can DALY scores be applied directly to clinical decision making. It is our hope that this is the direction clinical management will drive in the future.

Funding

Conflicts of interest

The authors declare no conflicts of interest with the design, execution or reporting of the study.

Ethical approval

Ethical approval for this work was granted by the Lothian Regional Ethics Committee (08/S11/02/2). All samples in the respiratory archive were subject to anonymisation and so the institutional review board specifically waived the need for patient consent.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.jcv.2011.07.017.

References

1. World Health Organization. World health report; 2001.
2. van den Hoogen BG, de Jong JC, Groen J, Kuiken T, de Groot R, Fouchier RAM, et al. A newly discovered human pneumovirus isolated from young children with respiratory tract disease. *Nat Med* 2001;**7**:719–24.
3. van der Hoek L, Pyrc K, Jebbink MF, Vermeulen-Oost W, Berkhout RJ, Wolthers KC, et al. Identification of a new human coronavirus. *Nat Med* 2004;**10**(4):368–73.
4. Woo PCY, Lau SKP, Chu CM, Chan KH, Tsoi HW, Huang Y, et al. Characterization and complete genome sequence of a novel coronavirus: coronavirus HKU1, from patients with pneumonia. *J Virol* 2005;**79**(2):884–95.
5. Lau SKP, Yip CCY, Tsoi HW, Lee RA, So LY, Lau YL, et al. Clinical features and complete genome characterization of a distinct human rhinovirus genetic cluster, probably representing a previously undetected HRV species, HRV-C, associated with acute respiratory illness in children. *J Clin Microbiol* 2007. p. JCM. 01254-07.
6. Scottish intercollegiate guidelines network, bronchiolitis in children: a national clinical guideline; 2006. Edinburgh.
7. Nicholson KG, Kent J, Hammersley V, Cancio E. Acute viral infections of upper respiratory tract in elderly people living in the community: comparative, prospective, population based study of disease burden. *BMJ* 1997;**315**(7115):1060–4.
8. Gaunt E, McWilliam-Leitch EC, Templeton K, Simmonds P. Incidence: molecular epidemiology and clinical presentations of human metapneumovirus; assessment of its importance as a diagnostic screening target. *J Clin Virol* 2009;**46**(4):318–24.
9. Wisdom A, Leitch EC, Gaunt E, Harvala H, Simmonds P. Screening respiratory samples for detection of human rhinoviruses (HRVs) and enteroviruses: comprehensive VP4-VP2 typing reveals high incidence and genetic diversity of HRV species C. *J Clin Microbiol* 2009;**47**(12):3958–67.
10. Gaunt ER, Hardie A, Claas ECJ, Simmonds P, Templeton KE. Epidemiology and clinical presentations of the four human coronaviruses 229E: HKU1, NL63 and OC43 detected over 3 years using a novel multiplex real-time PCR. *J Clin Microbiol* 2010;**48**(8):2940–7.
11. Mackenzie A, Hallam N, Mitchell E, Beattie T. Near patient testing for respiratory syncytial virus in paediatric accident and emergency: prospective pilot study. *BMJ* 1999;**319**(7205):289–90.
12. Murray CJL, Lopez AD. *Global health statistics: global burden of disease and injury series*, vol. II. Boston: Harvard School of Public Health; 1996.
13. The World Bank. World development report, 1993: investing in health. New York: Oxford University Press; 1993.
14. Murray CJL, Lopez AD. *The global burden of disease: global burden of disease and injury series*, vol. I. Boston: Harvard School of Public Health; 1996.
15. Murray CJ. Quantifying the burden of disease: the technical basis for disability-adjusted life years. World Health Organization; 1994.
16. Falsey AR, Erdman D, Anderson LJ, Walsh EE. Human metapneumovirus infections in young and elderly adults. *J Inf Dis* 2003;**187**(5):785–90.
17. Williams JV, Martino R, Rabella N, Otegui M, Parody R, Heck JM, et al. A prospective study comparing human metapneumovirus with other respiratory viruses in adults with hematologic malignancies and respiratory tract infections. *J Inf Dis* 2005;**192**(6):1061–5.
18. Sivaprakasam V, Collins TC, Aitken C, Carman WF. Life-threatening human metapneumovirus infections in West of Scotland. *J Clin Virol* 2007;**39**(3):234–7.
19. Hamelin ME, Côté S, Laforge J, Lampron N, Bourbeau J, Weiss K, et al. Human metapneumovirus infection in adults with community-acquired pneumo-

nia and exacerbation of chronic obstructive pulmonary disease. *Clin Inf Dis* 2005;**41**(4):498–502.

20. O'Gorman C, McHenry E, Coyle P. Human metapneumovirus in adults: a short case series. *Eur J Clin Microbiol Inf Dis* 2006;**25**(3):190–2.

21. Simmonds P, McIntyre C, Savolainen-Kopra C, Tapparel C, Mackay IM, Hovi T. Proposals for the classification of human rhinovirus species C into genotypically assigned types. *J Gen Virol* 2010;**91**(10):2409–19.

22. Savolainen C, Blomqvist S, Mulders MN, Hovi T. Genetic clustering of all 102 human rhinovirus prototype strains: serotype 87 is close to human enterovirus 70. *J Gen Virol* 2002;**83**(2):333–40.

23. Laine P, Savolainen C, Blomqvist S, Hovi T. Phylogenetic analysis of human rhinovirus capsid protein VP1 and 2A protease coding sequences confirms shared genus-like relationships with human enteroviruses. *J Gen Virol* 2005;**86**(3):697–706.

24. Ledford RM, Patel NR, Demenczuk TM, Watanyar A, Herbertz T, Collett MS, et al. VP1 sequencing of all human rhinovirus serotypes: insights into genus phylogeny and susceptibility to antiviral capsid-binding compounds. *J Virol* 2004;**78**(7):3663–74.

25. Wisdom A, Kutkowska AE, McWilliam Leitch EC, Gaunt E, Templeton K, Harvala H, et al. Genetics, recombination and clinical features of human rhinovirus species C (HRV-C) infections; interactions of HRV-C with other respiratory viruses. *PLoS One* 2009;**4**(12):e8518.

26. Jin Y, Yuan XH, Xie ZP, Gao HC, Song JR, Zhang RF, et al. Prevalence and clinical characterization of a newly identified human rhinovirus C species in children with acute respiratory tract infections. *J Clin Microbiol* 2009;**47**(9):2895–900.

27. Piotrowska Z, Vazquez M, Shapiro ED, Weibel C, Ferguson D, Landry ML, et al. Rhinoviruses are a major cause of wheezing and hospitalization in children less than 2 years of age. *Pediatr Infect Dis J* 2009;**28**(1):25–9, 10.1097/INF.0b013e3181861da0.

28. Holberg CJ, Wright AL, Martinez FD, Ray CG, Taussing LM, Lebowitz MD, Health Medical Associates Group. Risk factors for respiratory syncytial virus-associated lower respiratory illnesses in the first year of life. *Am J Epidemiol* 1991;**133**(11):1135–51.

29. Dijkman R, Jebbink MF, El Idrissi NB, Pyrc K, Muller MA, Kuijpers TW, et al. Human coronavirus NL63 and 229E seroconversion in children. *J Clin Microbiol* 2008;**46**(7):2368–73.

30. Severance EG, Bossis I, Dickerson FB, Stallings CR, Origoni AE, Sullens A, et al. Development of a nucleocapsid-based human coronavirus immunoassay and estimates of individuals exposed to coronavirus in a U.S. metropolitan population. *Clin Vaccine Immunol* 2008;**15**(12):1805–10.

31. Monto AS, Bryan ER, Rhodes LM. The Tecumseh study of respiratory illness: VII. Further observations on the occurrence of respiratory syncytial virus and mycoplasma pneumoniae infections. *Am J Epidemiol* 1974;**100**(6):458–68.

32. Kim HW, Arrobio JO, Brandt CD, Jeffries BC, Pyles G, Reid JL, et al. Epidemiology of respiratory syncytial virus infection in Washington DC: I. Importance of the virus in different respiratory tract disease syndromes and temporal distribution of infection. *Am J Epidemiol* 1973;**98**(3):216–25.

33. Kim HW, Brandt CD, Arrobio JO, Murphy B, Chanock RM, Parrott RH. Influenza A and B virus infection in infants and young children during the years 1957–1976. *Am J Epidemiol* 1979;**109**(4):464–79.

34. Kim HW, Canchola JG, Brandt CD, Pyles G, Chanock RM, Jensen K, et al. Respiratory syncytial virus disease in infants despite prior administration of antigenic inactivated vaccine. *Am J Epidemiol* 1969;**89**(4):422–34.

35. Parrott RH, Kim HW, Arrobio JO, Hodes DS, Murphy BR, Brandt CD, et al. Epidemiology of respiratory syncytial virus infection in Washington, D.C.: II. Infection and disease with respect to age, immunologic status, race and sex. *Am J Epidemiol* 1973;**98**(4):289–300.