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Clinical and physiological features of postinfectious chronic cough associated with H1N1 infection

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KEYWORDS
Postinfectious chronic cough;
H1N1 influenza infection;
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Summary
Background: Post infectious chronic cough is a disabling illness. In 2009 an influenza pandemic occurred due to a novel strain of H1N1 influenza. Prolonged symptoms such as chronic cough remaining after the infection has cleared have not been examined. This study sought to investigate the prevalence, characteristics and mechanism of chronic cough following laboratory-confirmed H1N1 2009 influenza.
Methods: Out of 836 eligible patients who had been tested by PCR assay for H1N1, 136 responders participated. Nineteen underwent detailed clinical investigation of cough, and airway function using symptom questionnaires, hypertonic saline challenge, and cough monitoring.
Results: Post H1N1 chronic cough was reported by 43%, and chronic cough after non-H1N1 infection was present in 36% of participants. In the participants who progressed to testing objectively measured cough frequency was 3 times greater; there was a 9-fold increase in cough reflex sensitivity and greater quality of life impairment in the participants with postinfectious chronic cough following H1N1 infection than for the participants with no cough

Abbreviations: ACE-I, Angiotensin–converting enzyme Inhibitor; ARI, Acute Respiratory Illness; CRS, Cough Reflex Sensitivity; DRS, Dose Response Slope; EAHR-DRS, Extrathoracic Airway Hyperresponsiveness-Dose Response Slope; GORD, Gastro Oesophageal Reflux Disease; IQR, InterQuartile Range; LCQ, Leicester Cough Questionnaire; LDQ, Laryngeal Dysfunction Questionnaire; $UTCPD_{CUM15Coughs}$, Urge to Cough at provocation dose causing cumulative 15 coughs; Snot-20, 20-item sino-nasal outcome test; VAS, Visual Analogue Scale.

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following H1N1 infection and for the healthy controls.
Conclusions: This study reports the first evaluation of chronic cough following H1N1 infection. Patients that develop chronic cough after H1N1 infection display increased cough reflex sensitivity up to 220 days after confirmed infection. There is an absence of associated risk factors and less impairment in quality of life compared to those patients normally seen in a specialist cough clinic. The associated mechanism was found to be cough reflex hypersensitivity.
Trial Registration: This clinical trial has been registered with the Australian New Zealand Clinical Trials Register, ACTRN12610000540011.

Introduction

Post infectious chronic cough is a disabling illness that can occur following respiratory tract infection[1] due to laryngeal dysfunction[2,3] or bronchial hyperresponsiveness.[4] An influenza pandemic occurred in 2009 due to a novel strain of H1N1 influenza.[5–7] The influence of pandemic 2009H1N1 on chronic cough is not described. Pandemic 2009 H1N1 virus derives six genes from triple-reassortant North American swine virus lineages and two genes (encoding neuraminidase and matrix proteins) from Eurasian swine virus lineages.[8] In experimentally infected animals, the level of pulmonary replication of the 2009 H1N1 virus has been higher than that of seasonal influenza A (H1N1) viruses[9–11] but the virus generally lacks mutations that are associated with increased pathogenicity in other influenza viruses. Initial cases of H1N1 infection were identified in March 2009, and by March 2010 almost all countries had reported H1N1 cases. An estimated 59 million illnesses, 265 000 hospitalisations, and 12 000 deaths had been caused by the 2009 H1N1 virus as of mid-February 2010. In contrast, in a normal influenza season with an ordinary strain of influenza, there are 200 000 cases and 36 000 deaths in only a few months in the United States alone each year.[6]

Most illnesses caused by H1N1 infection during the pandemic have been reported as acute and self-limited, with a higher prevalence amongst children and young adults. The typical presentation is with fever and acute cough that may be accompanied by sore throat and rhinorrhea. Gastrointestinal symptoms including nausea, vomiting, and diarrhoea have been reported to be more common than in seasonal influenza.[5,7] To date, prolonged symptoms such as chronic cough remaining after the infection has cleared have not been examined. This study sought to investigate the characteristics and mechanism of chronic cough following laboratory-confirmed H1N1 2009 influenza compared to participants without cough after H1N1 2009 influenza and with healthy controls. We hypothesised that the characteristics of postinfectious chronic cough following H1N1 infection would be similar to other postinfectious chronic cough and be associated with cough reflex hypersensitivity.

Methods

Subjects

Eligible participants were asymptomatic with no cough symptoms prior to infection and had received H1N1 influenza testing by the Hunter Area Pathology Service at least 8 weeks prior to completion of the symptoms questionnaire, were aged above 18 years, and gave their written and free consent to participate in the study. Ineligible participants were pregnant or breast feeding women, current smokers and those who had a current active respiratory disease such as COPD, asthma or bronchiectasis and/or were unable to attend study visits.

Study design

H1N1 2009 influenza testing by multiplexed tandem polymerase chain reaction (MT-PCR) was available to residents of the Hunter region of NSW, Australia with symptoms of an acute respiratory tract infection suspected as influenza, from 20th June 2009 onwards. Over this period test requests came from both outpatients and inpatients with influenza-like illness. The testing was performed by the Hunter Area Pathology Service (HAPS) located at John Hunter Hospital in Newcastle, New South Wales (NSW) Australia. During October 2009, the HAPS database was used to identify people who had been tested for H1N1 2009 influenza by MT-PCR assay at least 8 weeks previously. These people were then sent a letter of invitation and a study information and consent sheet requesting that they identify current symptoms including cough, breathing difficulties and voice changes, and current cough severity which was scored on a visual analogue scale (VAS) 100 mm in length with 0 representing "no cough" to 100 representing "worst cough". Eight hundred and thirty six letters were sent and 136 responded by completing the current symptoms form. Nineteen of these progressed to clinical testing at the hospital.

Clinical investigations

Nineteen participants completed a range of investigations to assess cough and airway function. These consisted of the Cough VAS[12,13] for cough severity, the Leicester cough questionnaire (LCQ),[14] Cough Reflex Sensitivity (CRS) using 4.5% hypertonic saline challenge[15,16] and cough frequency measured during the clinic visit by a cough recording monitor.[17,18] Upper airway dysfunction was assessed by the Laryngeal dysfunction questionnaire (LDQ)[19,20] and hypertonic saline challenge for extrathoracic airway hyperresponsiveness (EAHR).[21,22] Bronchial hyperresponsiveness (BHR) was assessed using spirometry and hypertonic saline challenge.[22] To investigate prevalence of risk factors

Table 1 H1N1 laboratory tested participant characteristics.

	Pos H1N1	Neg H1N1	*P* value
N (%)	58	78	
Gender, M/F	16/42	38/40	0.013
Age, yr Mean ± SD	38.4 ± 18.1	53.7 ± 23.3	0.0001
Time, days (swab to questionnaire letter) Median (IQR)	103.5 (34.0)	100.5 (24.0)	0.13
Cough (%)	43.1	35.9	0.39
Breath (%)	25.9	30.8	0.53
Voice (%)	27.6	16.7	0.12
Cough Severity, Mean ± SD	43.0 ± 23.1	49.0 ± 25.0	0.38

associated with persistent cough and upper airway dysfunction, the following questionnaires were also completed: Rhinosinusitis (SNOT-20),[23] Gastroesophageal reflux disease (GORD),[24] ACE-I use, and Obstructive Sleep Apnoea (Berlin Questionnaire).[25]

All subjects provided written informed consent for this study, which was approved by the University of Newcastle's Human Research Ethics Committee and the Hunter New England Human Research Ethics Committee.

Clinical methods

Multiplexed tandem polymerase chain reaction Method

Influenza MT-PCR testing on pooled nose and throat swabs was performed by the MT-PCR method targeting influenza A and B nucleoprotein and influenza A hemagglutinin gene sequences. The assay has a synthetic internal control sequence to act as an amplification control.[26] Control influenza RNA for influenza A strains, including pandemic strain 2009 H1N1 was run in parallel. The results distinguished influenza A (seasonal strains), influenza 2009 H1N1 and influenza B.

Pulmonary function test with inspiratory flow (FIF50%)

Inspiratory-expiratory Flow Volume Loops were measured using a KoKo K323200 Spirometer (Technipro, North Parramatta, Australia). Forced expiratory and inspiratory times were held constant to ensure consistency and accuracy.

Hypertonic saline challenge for bronchial hyperresponsiveness, extrathoracic airway hyperresponsiveness and cough reflex sensitivity

Hypertonic saline challenge for BHR and EAHR has been described previously.[27] Bronchial hyperresponsiveness, was calculated as the dose response slope (DRS), according to the percent fall in FEV_1 divided by the cumulative dose of hypertonic saline (mL) delivered. Extrathoracic Airway Hyperresponsiveness DRS was calculated by dividing the percent fall in $FIF_{50\%,}$ by the cumulative dose of hypertonic saline (mL) delivered. Cough Reflex Sensitivity was also calculated as DRS: cumulative number of coughs divided by the cumulative dose of hypertonic saline (mL) delivered. The participant's perceived urge to cough[28] at cumulative 15 coughs was also recorded. Cough frequency was defined as the number of coughs recorded during the visit on the cough monitor and expressed as coughs/hr.

Statistical analysis

Data analysis was conducted using STATA statistical software (Statacorp, Texas, USA). Dichotomous categorical variables were tested by Pearson's chi2 with Fishers' exact test. Continuous non-parametric data analyses were tested using the Kruskal–Wallis2 Test for comparison of more than 2 groups. Continuous parametric data were tested using ANOVA with Bonferroni correction. A p- value result of less than 0.05 was considered statistically significant.

Results

Current symptoms questionnaire (letter) responding participant characteristics

From the 136 participants who returned the completed symptoms questionnaire, 58 (43%) tested positive for H1N1 and 78 (57%) tested negative for H1N1. H1N1 infected participants were on average 15 years younger than the H1N1 negative group ($p = 0.0001$, Table 1) and included a greater proportion of females ($p = 0.013$, Table 1). There was a high prevalence of ongoing symptoms present at a median of 3 months after the acute respiratory illness. 43% of positive for H1N1 participants reported chronic cough with moderate severity, 26% reported ongoing dyspnoea and 28% reported ongoing voice symptoms. Similarly, 36% of negative for H1N1 participants reported chronic cough with moderate severity, 31% reported dyspnoea, and 17% reported ongoing voice symptoms. The youngest age group (participants less than 40 years of age, median age 22 years) had the highest H1N1 prevalence of 60%, and 50% of those had persisting cough compared to 23% without a confirmed H1N1 respiratory infection, $p = 0.12$. The oldest age group (age >60 years, median age of 72 years) had the lowest prevalence of H1N1 at 17.8%.

Not all patients with H1N1 infection developed a chronic cough however, for those that do cough reflex hypersensitivity was found to be still prevalent 220 days after confirmed infection (Table 2).

Clinical testing

19 participants attended for clinical testing, of whom 6 (32%) were positive for H1N1 and chronic cough, 6 (32%) were positive for H1N1 with no cough, and 7 (37%) were negative for H1N1 with no cough (healthy controls). These participants were tested a median (IQR) 221(40) days after laboratory PCR testing for H1N1 influenza, and 113(46) days after their questionnaire (letter) responses.

The characteristics and associated mechanisms of post-infectious chronic cough was examined in the 6 participants with laboratory-confirmed H1N1 influenza and CC and compared to those with no cough after laboratory-confirmed H1N1 infection and to the healthy control group. Participants with postinfectious chronic cough had heightened cough reflex sensitivity compared to participants without cough ($p = 0.020$) and compared with the control group (Table 2, Fig. 1). They had an increased urge to cough

Table 2 Characteristics and associated risk factors of postinfectious chronic cough following H1N1 influenza infection compared with no cough following H1N1 influenza infection and healthy controls.

	Control	Pos H1N1+Neg CC	Pos H1N1+Pos CC	*p*-value
N	7	6	6	
Age, Mean ± SD	54.7 ± 22.5	46.7 ± 11.6	35.5 ± 12.8	0.154
Gender M/F, n	4/3	1/5	2/4	0.427
Time, days (swab to clinic testing)	236 (56)	203 (91)	223 (33)	0.116
ACE-I use, %	14.3	16.7	0	1.000
Berlin risk for sleep apnoea, %	28.6	50.0	50.0	0.722
GORD Score, Median (IQR)	11 (16)	14 (6)	13 (12)	0.874
SNOT-20 Score, Median (IQR)	0.3 (0.2)	1.13 (1.60)	1.45 (1.00)	0.186
FEV1 %pred, Mean ± SD	99.4 ± 13.9	103.0 ± 11.3	86.3 ± 20.3	0.175
FVC %pred, Mean ± SD	100.2 ± 14.4	102.8 ± 13.8	85.6 ± 16.9	0.132
FEV1/FVC%, Mean ± SD	81.9 ± 4.8	83.1 ± 3.1	84.1 ± 7.5	0.781
FIF50 %pred, Mean ± SD	117.2 ± 22.1	129.4 ± 15.5	105.6 ± 39.2	0.343
Cough Frequency during hypertonic saline challenge, (calculated as coughs/hr), Mean ± SD	42.3 ± 33.7	56.2 ± 62.6	162.8 ± 168.5	0.108
Leicester Cough Questionnaire, GMean (95% CI)	20.16 (19.26, 21.11)	20.27 (19.39, 21.19)	18.27 (16.32, 20.46)[b]	0.036
Laryngeal Dysfunction Questionnaire, Median (IQR)	3 (3)	2 (4)	6.5 (2)	0.056
BHR-DRS, Median (IQR)	2.29 (3.17)	0.294 (0.359)	1.09 (2.97)	0.157
EAHR-DRS, Median (IQR)	1.53 (11.01)	1.58 (0.868)	5.01 (12.3)	0.246
CRS-DRS, GMean (95% CI)	3.33 (1.41, 7.84)	1.16 (0.24, 5.64)	11.07 (2.79, 43.92)[a]	0.022
Urge to Cough at PDCUM15Coughs, Median (IQR)	3 (1)	2.5 (2)	4.5 (1)[b]	0.050
Cough Severity, Median (IQR)	0	0	17 (11)[a]	0.003

ACE-I = Angiotensin conversion enzyme inhibitor, GORD = Gastrooesaphageal reflux disease, SNOT-20 = Sino Nasal Outcome Test, FIF50% = mid inspiratory flow, DRS = Dose Response Slope, EAHR = Extrathoracic Airway Hyperresponsiveness.
[a] v Pos H1N1+ Neg CC, $p \leq 0.020$.
[b] v Pos H1N1+Neg CC, $p < 0.05$.

($p < 0.05$ v participants without postinfectious H1N1 cough) and increased cough severity, $p = 0.003$ (Table 2). Objectively measured cough frequency during testing was 3 times greater in the participants with postinfectious H1N1 chronic cough compared to the participants without postinfectious H1N1 cough and 4 times greater than the healthy control participants (Fig. 3) however this was not significant. The postinfectious H1N1 chronic cough participants also displayed decreased quality of life compared to the participants without postinfectious H1N1 cough, and compared to the healthy control participants, $p = 0.036$ (Table 2,Fig. 2). Participants with postinfectious H1N1 chronic cough tended to have more laryngeal symptoms and increased extrathoracic airway hyperresponsiveness than the participants without postinfectious H1N1 CC and healthy control participants (Table 2) but this was not significant.

There was no difference in associated risk factors between the three groups and spirometry was unremarkable (Table 2).

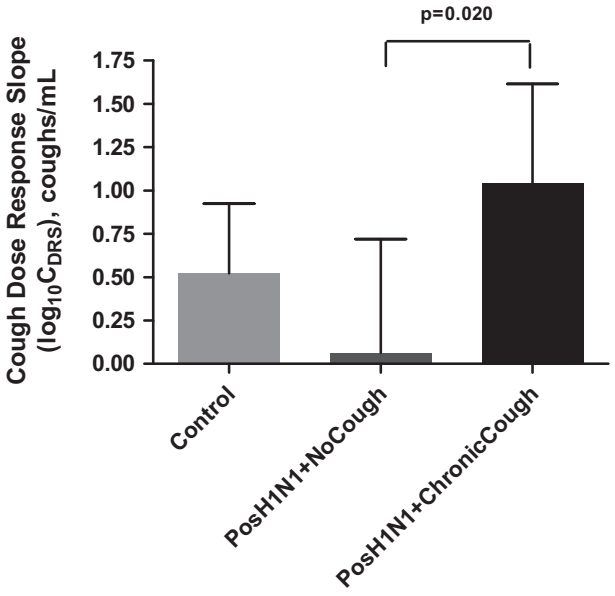

Figure 1 A comparison of cough reflex sensitivity across the three study groups Control (no H1N1 or cough) v positive H1N1 infection without cough v positive H1N1 infection with chronic cough.

Discussion

This study reports the first evaluation of chronic cough following the pandemic H1N1 influenza outbreak in 2009. We found that participants experience moderate chronic cough severity after infection but this tended to improve

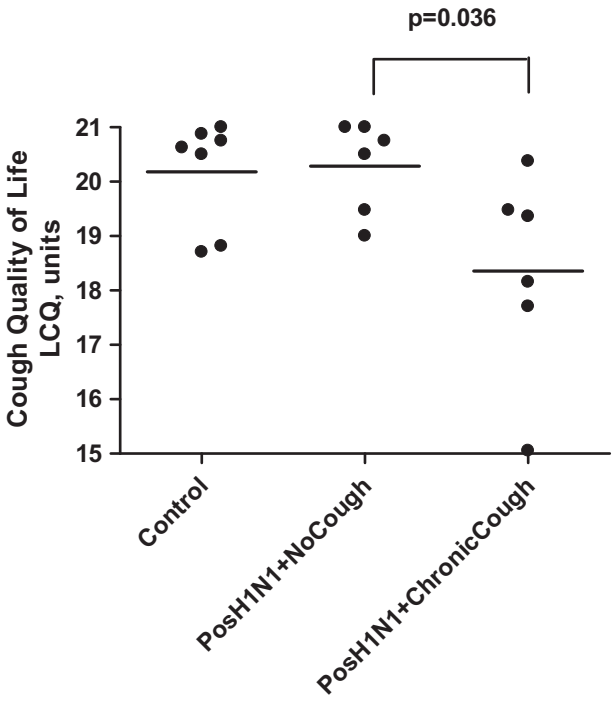

Figure 2 A comparison of cough quality of life, LCQ across the three study groups Control (no H1N1 or cough) v positive H1N1 infection without cough v positive H1N1 infection with chronic cough.

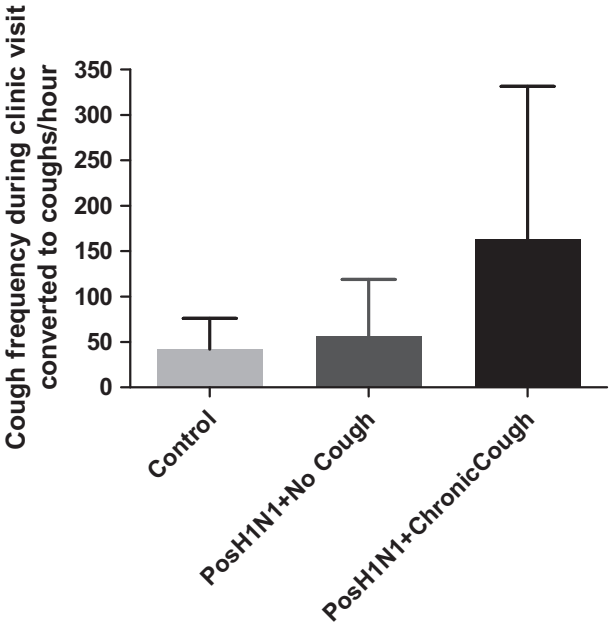

Figure 3 A comparison of cough frequency during clinic testing across the three study groups Control (no H1N1 or cough) v positive H1N1 infection without cough v positive H1N1 infection with chronic cough, coughs per hour, Mean ± SD.

spontaneously over time. The characteristics of postH1N1 chronic cough were similar to other postinfectious chronic cough[29] and associated with cough reflex hypersensitivity.

Cough reflex sensitivity, the urge to cough and cough severity were all enhanced in the participants with post-infectious H1N1 chronic cough compared to the participants without postinfectious H1N1 cough and compared to the healthy control participants. The objectively measured cough frequency during clinical testing in this current study was similar to what we have previously observed in the cough clinic.[30] Although the quality of life impairment in post infectious H1N1 chronic cough was significant, it was not as severe as seen in people with chronic cough presenting to specialist cough centres.[18,20,31] Similarly, the prevalence of specific cough—related co morbidities of reflux, rhinitis, and vocal disturbance was also much lower than in cough referral clinics. This indicates that although postinfectious cough is common, it usually causes less distress than cough from other causes. Our data support the hypothesis that seeking medical attention for chronic cough requires additional factors such as a related co-morbidity, for example GORD, rhinitis, asthma or a voice disorder. This would also explain why therapy targeting these conditions, as in the anatomic-diagnostic protocol, is so successful in treating chronic cough in the cough clinic setting.[1] While these statistical results are significant, they should be interpreted as merely descriptive due to the small sample size, a recognised limitation of this study.

The pathogenesis of postinfectious cough is believed to be related to inflammation and epithelial damage to the upper and lower airways. Several potential mechanisms for post-infectious cough have been proposed. These include postinfectious bronchial hyperresponsiveness[4]; transient vocal cord dysfunction[3]; cough reflex hypersensitivity[32]; nasal and sinus inflammation[33]; and GORD.[34] We examined several of these mechanisms in this study and our results support increased cough reflex sensitivity as the dominant mechanism for postinfectious H1N1chronic cough since there was a 3—9-fold increase in cough reflex sensitivity and no significant increase in bronchial or extrathoracic hyper-responsiveness in the postinfectious H1N1 chronic cough participants compared with the healthy participants and post H1N1 without cough participants. The other mechanisms may be relevant in individual patients, but do not emerge as a general mechanism for postinfectious chronic cough.

Relevant treatment strategies for postH1N1 infectious chronic cough include components of a diagnostic assessment approach.[35] Vaccination appears to be effective at reducing post pertussis chronic cough.[36] An inactivated, split-virus 2009 H1N1 vaccine is available and is both immunogenic and safe in healthy adults.[37] The efficacy of this vaccine in reducing cough prevalence needs further study.

Limitations of this study include the low response rate to the questionnaire (current symptoms letter). An over representation of participants with cough in the response group is noted as a potential bias. To some extent this was accounted for in the clinical testing arm of the study where an even representation of cough, no cough and healthy controls were tested. It is recommended however that future studies aim for a more representative group. A second limitation of this study is that participants were only clinically tested on one occasion; a preferred scenario would be to conduct longitudinal testing enhancing characterisation of this group. A third recognised limitation of this study was that we did not conduct treatment on the participants therefore we cannot prove causes of the persistent cough.

In conclusion, this is the first study to evaluate chronic cough following the pandemic H1N1 influenza outbreak of 2009. H1N1 infected participants were younger, primarily female and tended to have a higher prevalence of chronic cough and voice symptoms than those without H1N1. Clinical testing showed that those participants with H1N1 postinfectious chronic cough were still symptomatic with increased cough reflex sensitivity some 220 days after infection although cough severity was found to be significantly improving by this stage. The characteristics of postH1N1 chronic cough are similar to other postinfectious cough and associated with cough reflex hypersensitivity.

Author's contributions

NR, JF, PW and PG planned the study. JF collated and co-ordinated MT-PCR data and authored MT-PCR Method. NR recruited the participants, performed the objective cough and respiratory tests and questionnaires, collected and collated data, interpreted outcomes, drafted and edited manuscript. PG interpreted data, co-drafted and edited manuscript. AV edited manuscript and all authors read and approved the final manuscript submission.

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Conflict of interest

The authors wish to declare that there is no conflict of financial or personal interest related to this manuscript.

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