Acta Med. Okayama, 2011
Vol. 65, No. 2, pp. 97–103

Acta Medica Okayama
http://escholarship.lib.okayama-u.ac.jp/amo/

Original Article

Environmental Factors and Seasonal Influenza Onset in Okayama City, Japan: Case-Crossover Study

Yuuki Tsuchihashi[a]*, Takashi Yorifuji[a], Soshi Takao[a], Etsuji Suzuki[a], Shigeru Mori[b], Hiroyuki Doi[a], and Toshihide Tsuda[c]

[a]Department of Epidemiology, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama 700–8558, Japan, [b]Department of Pediatrics, Momotaro Clinic, Okayama 701–0205, Japan, and [c]Department of Environmental Epidemiology, Okayama University Graduate School of Environmental Science, Okayama 700–8530, Japan

Seasonal influenza infection is a major challenge in public health. The term "seasonal influenza" refers to the typical increase in the number of influenza patients in the winter season in temperature zones. However, it is not clear how environmental factors within a single flu season affect influenza infection in a human population. Therefore, we evaluated the effects of temperature and humidity in the 2006–7 flu season on the onset of seasonal influenza using a case-crossover study. We targeted patients who attended one pediatric clinic in Okayama city, Japan and who were diagnosed as being infected with the seasonal influenza virus. Using 2 references (time-stratified and symmetric bidirectional design), we estimated the effects of average temperature and relative humidity from the onset day (lag 0) to 10 days before (lag 10). The total number of subjects was 419, and their onset days ranged from 26 December 2006 to 30 April 2007. While the onset was significantly associated with lower temperature, relative humidity was not related. In particular, temperatures before the 3-day incubation period had higher-magnitude odds ratios. For example, the odds ratio and 95% confidence interval for average temperature at time lag 8 was 1.12 (1.08–1.17) per 1.0℃ decrease. Low environmental temperature significantly increased the risk of seasonal influenza onset within the 2006–7 winter season.

Key words: seasonal influenza in humans, temperature, humidity, case-crossover study

In recent years, awareness of the epidemiology and prevention of infectious diseases has grown due to the appearance of new pathogens, such as the pandemic (H1N1) 2009 virus, avian influenza virus and Severe Acute Respiratory Syndrome (SARS) coronavirus. Therefore, research findings that elucidate the mechanisms of transmission and can help forecast infections such as influenza are eagerly sought [1]. A study of environmental factors affecting influenza infections may contribute to the clarification of transmission mechanisms. It is not clear how environmental factors such as temperature and humidity affect influenza infections in human populations.

Several studies have examined the association between environmental factors and the influenza virus in a laboratory environment and in humans. In the laboratory, survival of the influenza virus was longer and virus transmission was promoted at low-temperature and low-humidity conditions [2, 3]. In human populations, although weekly or monthly data has indicated that, in the temperature zone, the number of influenza patients increases in the winter season [4–6], there has been no clear evidence that virus transmission is promoted at relatively low-tempera-

Received September 29, 2010; accepted December 8, 2010.
*Corresponding author. Phone:+81–86–223–7151; Fax:+81–86–235–7178
E-mail:yuuki.tsuchihashi@gmail.com (Y. Tsuchihashi)

ture and low-humidity conditions within a single season. Therefore, we do not know what environmental factors might contribute to epidemics of influenza in the temperate zone [3, 7] or how environmental factors affect the pattern of influenza infection in a human population within a single winter season. On the other hand, in the tropical and subtropical zones, which do not have a winter season, it has been reported that the seasonality of influenza was either unclear or tended to increase in the rainy season [8–10]. Therefore, it is worthwhile to investigate the association between environmental factors and influenza infections within the same season. Previous researchers have used weekly or monthly data to investigate the associations between seasons and influenza infections [4–6]. In the present study, we used daily data and a case-crossover design to evaluate whether environment factors such as low temperature and low humidity increased the risk of influenza infections among humans within a single winter season.

Materials and Methods

Case identification. The investigation was conducted from 26 December 2006 to 2 May 2007. We targeted patients who attended one pediatric clinic in Okayama city, Japan and who were diagnosed as being infected with the influenza virus. The clinic serves an average of approximately 150 outpatients daily and is regarded as one of the largest pediatric clinics in Okayama city. The diagnosis of influenza infection was made by the Point-of-Care Testing (POCT) product of influenza Capillia FluA + B®. This is a test which involves immunochromatography and uses a monoclonal antibody having a high specificity to the influenza virus. According to the test documentation, the sensitivity and specificity of the monoclonal antibody to the type A influenza virus are 96.4% and 94.3%, respectively, and the sensitivity and specificity to the type B influenza virus are 83.8% and 100%, respectively. All patients whom doctors suspected of influenza infection and whose parents agreed to their being tested were examined. In total, we identified 422 subjects who had a positive reaction to POCT. Details regarding the first day of fever were available for all subjects. Since most influenza patients develop fever within 24 h of onset, we defined the first day of fever as the onset day. All subjects developed fever.

Meteorological data. We obtained daily data of average temperature (Celsius, ℃) and relative humidity (%) in Okayama city (34° 39'6" N, 133° 55'0" E) from the Japan Meteorological Agency (<http://www.data.jma. go.jp/obd/stats/etrn/index.php> accessed May, 2007). We defined the daily average temperature and relative humidity as exposure variables. The distance between the temperature and humidity measurement point in Okayama city and the pediatric clinic was about 6 kilometers.

Study design. We conducted a case-crossover study, which is a variation of a case-control study [11]. The major difference between case-crossover design and matching case-control studies is that each case serves as its own control in the case-crossover design [11]. The association between exposure and disease is assessed by comparing the exposure status of a case when manifesting symptoms with his or her exposure status at earlier or later periods when symptoms were not present. The case-crossover design has been used in studies of temperature and stroke [12, 13] and in another study of air pollution and respiratory morbidity [14]. This design is also used in the field of infectious disease [15–17]. We examined 2 ways of selecting references to overcome the time trend of exposure, which is a limitation of the case-crossover design: time-stratified design and symmetric bidirectional design.

We used time-stratified design, which can control for the season and the day of the week [18]. Furthermore, this design is considered to lead to unbiased conditional logistic regression estimates [18]. Reference days were the same days of the week as the onset day in other weeks of the same month; for example, when the onset day was 7 Feb 2007 (Wed), we considered 14 (Wed), 21 (Wed), and 28 (Wed) Feb 2007 as reference days. In addition, we compared the exposure before the onset day with the exposure before the reference days, shifting each exposure from the onset day or reference days (lag 0) to 10 days before (lag 10) in the same manner [19]. As a result, the design had a 3 : 1 or 4 : 1 (reference: case) matched case-crossover design. We also used symmetric bidirectional design, although this design is considered to lead to bias, in comparison with the time-stratified design, during conditional logistic regression analysis [18]. Here, the same day

of the week as the onset day in the week before and after onset were selected as the reference days; for example, when the onset day was 7 Feb 2007 (Wed), we considered 31 Jan (Wed) and 14 Feb (Wed) 2007 as reference days. The design had a 2 : 1 matched case-crossover design [12, 13].

The study procedures were approved by the Ethics Committee on Epidemiologic Research of Okayama University Graduate School of Medicine, Dentistry, and Pharmaceutical Sciences (No. 171).

Statistical analysis. In both time-stratified and symmetric bidirectional designs, we used average temperature and relative humidity as covariates in the conditional logistic regression model. These meteorological data were treated as continuous variables. In all data analysis, we adjusted average temperature and relative humidity with each other; i.*e.*, when one of them was treated as the exposure variable, the other was considered as a confounder. We then estimated the odds ratios (ORs) and 95% confidence intervals (CIs) from lag0 to lag10 exposure period, respectively. In addition, we categorized meteorological data into quartiles in the time-stratified design, which is known to be a better design than the symmetric bidirectional design, and also assessed the dose-response relationships. We defined the highest average temperature or relative humidity category as the reference groups.

Further, as a supplementary analysis, we also conducted a stratified analysis by influenza types using the time-stratified design. To minimize the possibility of residual confounding due to time-dependent factors (*e.g.*, socializing around New Year's days), we also conducted a supplementary analysis by restricting the subjects to those who had an onset in March, 2007 by using the time-stratified design.

We used SPSS11.0 for Windows to analyze the data. A p value less than 0.05 (two-sided) was considered statistically significant.

Results

In total, 422 patients consulted the pediatric clinic from 26 December 2006 to 2 May 2007 and were diagnosed as having influenza by POCT. We excluded three cases because the date of fever onset was unknown. Therefore, we analyzed 419 cases. The dates of fever onset ranged from 26 December 2006 to 30 April 2007. Demographic characteristics of the subjects are shown in Table 1. Mean values for meteorological data by month are shown in Table 2. The average temperature and relative humidity ranged from 2.6℃ to 18.6℃ and from 37% to 90%, respectively. The number of subjects who had fever onset on each day is plotted in Fig. 1, along with daily meteorological data. The peak of the influenza outbreak during the 2006/07 season was observed in the middle of March.

Table 3 shows ORs and their 95% CIs between average temperature/relative humidity and onset of influenza from lag0 to lag10 exposure period using the time-stratified design. Regarding average temperature, the crude and adjusted point estimates of ORs were 1.05–1.12 and 1.05–1.12 per 1.0℃ decrease, respectively. In comparison with temperatures during the lag0 to lag3 exposure period, which is the incubation period of the influenza virus [20], temperatures during lag4 or before this period had a higher magnitude of ORs. With regard to relative humidity, although some ORs were significant in the crude data, the associations were almost non-significant after adjustment.

Table 4 shows ORs and their 95% CIs between

Table 1 Demographic characteristics of the study subjects

Characteristic	Subjects (n = 419)
Sex	
Male	219 (52.3)
Female	200 (47.7)
Age, y	5.7 ± 3.6
Influenza type	
A	240 (57.3)
B	179 (42.7)

Data are no. (%) of subjects or mean ± standard deviation.

Table 2 Meteorological data by month; Okayama city, Japan 2006–2007

Month	Average temperature (℃)	Relative humidity (%)
December	8.0 ± 1.8	69 ± 10
January	6.2 ± 1.1	64 ± 8
February	7.9 ± 2.3	62 ± 10
March	9.4 ± 3.5	57 ± 9
April	14.1 ± 2.7	57 ± 11

Data are mean ± standard deviation.

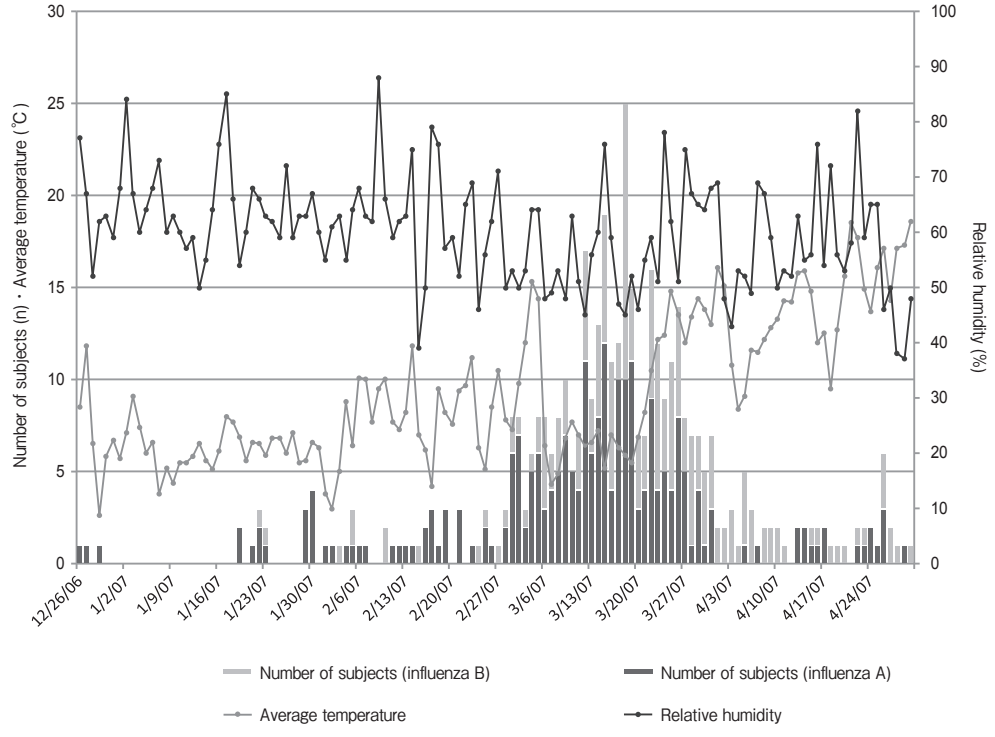

Fig. 1 Number of subjects at onset date and the meteorological data in Okayama city for 2006–2007.

Table 3 Onset of influenza and 24-hour average temperature or relative humidity by Time-stratified design

Lag Days*	Average Temperature		Relative Humidity	
	Crude	Adjusted**	Crude	Adjusted**
Lag0	1.05 (1.02–1.09)	1.05 (1.02–1.09)	1.01 (1.00–1.02)	1.00 (0.99–1.01)
Lag1	1.05 (1.02–1.08)	1.05 (1.01–1.08)	1.01 (1.00–1.02)	1.00 (0.99–1.02)
Lag2	1.06 (1.02–1.10)	1.08 (1.04–1.12)	1.00 (0.99–1.01)	0.99 (0.98–1.00)
Lag3	1.08 (1.04–1.11)	1.09 (1.05–1.13)	1.00 (0.99–1.01)	0.99 (0.98–1.00)
Lag4	1.09 (1.06–1.13)	1.11 (1.06–1.15)	1.01 (0.99–1.02)	0.99 (0.98–1.00)
Lag5	1.11 (1.07–1.15)	1.09 (1.05–1.13)	1.03 (1.01–1.04)	1.01 (1.00–1.02)
Lag6	1.12 (1.08–1.16)	1.09 (1.05–1.14)	1.03 (1.02–1.04)	1.02 (1.00–1.03)
Lag7	1.10 (1.07–1.14)	1.12 (1.08–1.16)	1.01 (1.00–1.02)	0.99 (0.98–1.00)
Lag8	1.12 (1.08–1.16)	1.12 (1.08–1.17)	1.02 (1.01–1.03)	1.00 (0.99–1.01)
Lag9	1.12 (1.08–1.16)	1.10 (1.05–1.15)	1.03 (1.02–1.04)	1.01 (1.00–1.03)
Lag10	1.10 (1.06–1.14)	1.09 (1.05–1.13)	1.02 (1.00–1.03)	1.00 (0.99–1.02)

Data are odds ratio (95% confidence interval), which are calculated per 1.0℃ temperature decrease and per 1.0% relative humidity decrease.

*For example, lag0 means the onset day, lag1 means a time lag of one day before the onset day and lag2 means a time lag of 2 days before the onset day.

**Simultaneously, adjusted for average temperature and relative humidity.

average temperature/relative humidity and onset of influenza from the lag0 to lag10 exposure period using symmetric bidirectional design. The results using this design showed the same trend as the results obtained from the time-stratified design, although the point estimates were closer to the null.

Table 4 Onset of influenza and 24-hour average temperature or relative humidity by symmetric bidirectional design

Lag Days*	Average Temperature		Relative Humidity	
	Crude	Adjusted**	Crude	Adjusted**
Lag0	1.04 (1.00–1.08)	1.03 (0.99–1.07)	1.01 (1.00–1.03)	1.01 (1.00–1.02)
Lag1	1.02 (0.98–1.06)	1.01 (0.97–1.05)	1.01 (1.00–1.02)	1.01 (0.99–1.02)
Lag2	1.02 (0.98–1.06)	1.04 (1.00–1.08)	0.99 (0.98–1.00)	0.99 (0.97–1.00)
Lag3	1.03 (0.99–1.07)	1.04 (1.00–1.08)	1.00 (0.99–1.01)	0.99 (0.98–1.01)
Lag4	1.04 (1.00–1.08)	1.05 (1.01–1.10)	1.00 (0.99–1.01)	0.99 (0.98–1.00)
Lag5	1.05 (1.01–1.09)	1.04 (1.00–1.09)	1.01 (1.00–1.03)	1.01 (0.99–1.02)
Lag6	1.07 (1.02–1.11)	1.05 (1.00–1.09)	1.02 (1.01–1.03)	1.01 (1.00–1.03)
Lag7	1.05 (1.01–1.09)	1.07 (1.03–1.12)	1.00 (0.98–1.01)	0.99 (0.97–1.00)
Lag8	1.05 (1.01–1.10)	1.06 (1.02–1.11)	1.00 (0.99–1.02)	1.00 (0.98–1.01)
Lag9	1.04 (1.00–1.08)	1.02 (0.98–1.07)	1.02 (1.00–1.03)	1.01 (1.00–1.03)
Lag10	1.02 (0.98–1.07)	1.02 (0.97–1.06)	1.01 (0.99–1.02)	1.00 (0.99–1.02)

Data are odds ratio (95% confidence interval), which are calculated per 1.0℃ temperature decrease and per 1.0% relative humidity decrease.

*For example, lag0 means the onset day, lag1 means a time lag of one day before the onset day and lag2 means a time lag of 2 days before the onset day.

**Simultaneously, adjusted for average temperature and relative humidity.

Table 5 Dose-response relationship between meteorological exposures and onset of influenza at time Lag8

	Average Temperature		Relative Humidity	
	Crude	Adjusted*	Crude	Adjusted*
1st**	1 (Reference)	1 (Reference)	1 (Reference)	1 (Reference)
2nd	1.63 (1.13–2.33)	1.68 (1.15–2.45)	1.48 (1.08–2.01)	1.17 (0.84–1.64)
3rd	2.84 (1.99–4.04)	2.77 (1.91–4.02)	1.42 (1.03–1.95)	1.07 (0.76–1.50)
4th	2.54 (1.79–3.61)	2.54 (1.70–3.79)	1.82 (1.32–2.50)	1.10 (0.75–1.61)

Data are odds ratio (95% confidence interval).

*Simultaneously, adjusted for average temperature and relative humidity.

**Categorized into quartiles as follows: 1st (12.5–18.5℃ or 65–88%), 2nd (8.2–12.4℃ or 59–64%), 3rd (6.5–7.8℃ or 52–58%), and 4th (3.0–6.4℃ or 39–51%).

Finally, we assessed the association between average temperature/relative humidity and onset of influenza using categorized exposure data from the lag0 to lag10 exposure period. Clear dose-response relationships were observed between average temperature and onset of influenza in almost all lag periods. For example, as shown in Table 5, the result of lag8 exposure, which had one of the strongest associations (Table 3), showed that subjects in the lowest temperature category (3.0–6.4℃) were about 2.5 times more likely to be infected compared to those in the highest temperature category (12.5–18.5℃) (results of other lags are not shown). With regard to the adjusted relative humidities, no obvious associations were observed in almost all lag periods.

When we conducted a stratified analysis by influenza type, we found no substantial differences between influenza A and B; their point estimates of adjusted ORs in average temperature were 1.05–1.14 and 1.03–1.11 per 1.0℃ decrease, and were 0.99–1.02 and 0.98–1.01 per 1.0% decrease, respectively. Moreover, when we restricted the subjects to those who had onset in March, we observed slightly higher point estimates of ORs in average temperature, whereas we found no clear patterns in relative humidity; the adjusted point estimates of ORs in average temperature and relative humidity were 1.06–1.16 per 1.0℃ decrease and 0.98–1.03 per 1.0% decrease, respectively.

Discussion

In the present study, we demonstrated that the risk of influenza onset within a single temperate-zone winter season was associated with lower temperatures and especially with lower mean temperature before the 3-day incubation period. Relative humidity showed no relation to the risk of influenza. This study is the first to quantify the effects of environmental factors (average temperature and relative humidity) on influenza infections within a single season in a human population.

Our results agree with previous studies carried out in laboratories which have shown that low temperatures increased the survival time and accelerated the transmission of the influenza virus [2, 3]. In the present study, we showed that low temperatures for all lags were associated with the onset of influenza, which would reflect that there are many mechanisms between exposure and onset (*e.g.*, the survival time of the virus, the immune response of the host, the distribution of protease/protease inhibitor in the host, and the clearance ability of the virus in the host [6, 21]). We considered these mechanisms as intermediate variables. Furthermore, among all lags, lower temperatures before the incubation period had higher-magnitude ORs. This implies that temperatures before influenza viruses invade the human body are a more important factor than temperatures after the incubation period has begun. So, low temperatures in the environment would promote the entrance of the virus into the human body. However, we did observe an association between low temperature at lag 0 and influenza onset, which is not compatible with a biological explanation. This effect at lag 0 could well be due to the residual temperature correlation between "day 0" and the etiologically relevant days.

In this study, relative humidity did not affect the onset of influenza. This was probably due to the fact that the humidity was high with only small fluctuations in Okayama city during the study period (*i.e.*, mean relative humidity in the 06/07 winter season was 57–69%). It has been demonstrated that the relative humidity most favorable for virus transmission in guinea pigs is 20–35% [3], much lower than that found in Okayama city during the study period. In a relatively humid environment like Okayama city, temperature would be a more important factor than relative humidity for the onset of influenza. Future studies in other climate zones are warranted to examine the comparative effects of temperature and relative humidity to humans.

The advantages of our study design are as follows: First, unknown factors, especially time-independent factors, were adjusted using the case-crossover design. For example, matching with natural immune ability, immune level by vaccination, individual environmental factors such as heating equipment, and behaviors could be achieved. Although information about vaccination of influenza was not available, we believe that subjects who sought it were vaccinated by the beginning of this study because influenza vaccination is available from around October annually. Secondly, by using the time-stratified design, we could adjust confounding due to time trends of exposure, seasonality, and weeks as much as possible. Third, we examined 2 ways to select references. The point estimates of ORs obtained from the symmetric bidirectional design were lower than those obtained from the time-stratified design. However, the trends of the results were similar [22]. Fourthly, as exposures (average temperature and relative humidity) were exogenous, the design could be considered a natural experiment [23]. Therefore, we did not adjust for the following factors: type of feeding, attendance or not at day-care and exposure to second-hand smoke. Fifth, we used individual and daily data, not aggregated and weekly data as used out in previous studies. As a result, we could conduct more detailed analyses within the same season. Finally, dropouts amounted to only 3 influenza-positive patients.

The disadvantages of this study are as follows: First, because we used meteorological data obtained from a measurement point in Okayama city which was a distance from the pediatric clinic, exposure misclassification may have been introduced. However, this non-differential bias would simply distort the present findings toward the null [24]. Second, the number of patients who refused the influenza test is unknown. Nevertheless, we think this is unimportant, as refusals were independent from exposure. Third, as we only used data from one pediatric clinic in one winter season, the generalizability of the current findings may be limited. However, our data showed a similar trend to the overall influenza data compiled for Okayama city (<http://www.city.okayama.jp/con-

tents/000076391.pdf> accessed November, 2010). Further, our data are relatively compatible with the national surveillance data in the 2006/07 season, the national agents surveillance showed that the epidemic predominantly consisted of influenza AH3 (49.1%) and B (40.3%), and each epidemic curve was almost the same pattern (<http://idsc.nih.go.jp/iasr/index-j.html> accessed on November, 2010). The peak week of the epidemic in the national diseases surveillance data was the 11th week of 2007 (12–18 in March) (<http://idsc.nih.go.jp/idwr/index.html> accessed November, 2010).

We showed that low temperatures, especially a low mean temperature before the incubation period (lag3 to lag0), significantly increased the risk of influenza onset within a single winter season. Given the relatively humid environment such as Okayama city, temperature was a more important factor for the onset of influenza. Further epidemiologic studies are necessary to examine the effect of humidity more closely and to clarify the mechanism of human influenza epidemics in temperate zones.

Acknowledgments. We appreciate Professor Eiji Yamamoto, Okayama University of Science for his statistical comments, as well as the staff at the Department of Pediatrics, Miyake Clinic for their efforts.

References

1. Fisman DN: Seasonality of infectious diseases. Annu Rev Public Health (2007) 28: 127–143.
2. Harper GJ: Airborne micro-organisms: survival tests with four viruses. J Hyg (Lond) (1961) 59: 479–486.
3. Lowen AC, Mubareka S, Steel J and Palese P: Influenza virus transmission is dependent on relative humidity and temperature. PLoS Pathog (2007) 3: 1470–1476.
4. Urashima M, Shindo N and Okabe N: A seasonal model to simulate influenza oscillation in Tokyo. Jpn J Infect Dis (2003) 56: 43–47.
5. Viboud C, Boelle PY, Pakdaman K, Carrat F, Valleron AJ and Flahault A: Influenza epidemics in the United States, France, and Australia, 1972–1997. Emerg Infect Dis (2004) 10: 32–39.
6. Eccles R: An explanation for the seasonality of acute upper respiratory tract viral infections. Acta Otolaryngol (2002) 122: 183–191.
7. Dowell SF and Ho MS: Seasonality of infectious diseases and severe acute respiratory syndrome-what we don't know can hurt us. Lancet Infect Dis (2004) 4: 704–708.
8. Shek LP and Lee BW: Epidemiology and seasonality of respiratory tract virus infections in the tropics. Paediatr Respir Rev (2003) 4: 105–111.
9. Shih SR, Chen GW, Yang CC, Yang WZ, Liu DP, Lin JH, Chiu SC, Chen HY, Tsao KC, Huang CG, Huang YL, Mok CK, Chen CJ, Lin TY, Wang JR, Kao CL, Lin KH, Chen LK, Eng HL, Liu YC, Chen PY, Lin JS, Wang JH, Lin CW, Chan YJ, Lu JJ, Hsiung CA, Chen PJ and Su IJ: Laboratory-based surveillance and molecular epidemiology of influenza virus in Taiwan. J Clin Microbiol (2005) 43: 1651–1661.
10. Viboud C, Alonso WJ and Simonsen L: Influenza in tropical regions. PLoS Med (2006) 3: e89.
11. Maclure M and Mittleman MA: Should we use a case-crossover design? Annu Rev Public Health (2000) 21: 193–221.
12. Hong YC, Rha JH, Lee JT, Ha EH, Kwon HJ and Kim H: Ischemic stroke associated with decrease in temperature. Epidemiology (2003) 14: 473–478.
13. Kyobutungi C, Grau A, Stieglbauer G and Becher H: Absolute temperature, temperature changes and stroke risk: a case-crossover study. Eur J Epidemiol (2005) 20: 693–698.
14. Barnett AG, Williams GM, Schwartz J, Neller AH, Best TL, Petroeschevsky AL and Simpson RW: Air pollution and child respiratory health: a case-crossover study in Australia and New Zealand. Am J Respir Crit Care Med (2005) 171: 1272–1278.
15. Dixon KE: A comparison of case-crossover and case-control designs in a study of risk factors for hemorrhagic fever with renal syndrome. Epidemiology (1997) 8: 243–246.
16. Fisman DN, Lim S, Wellenius GA, Johnson C, Britz P, Gaskins M, Maher J, Mittleman MA, Spain CV, Haas CN and Newbern C: It's not the heat, it's the humidity: wet weather increases legionellosis risk in the greater Philadelphia metropolitan area. J Infect Dis (2005) 192: 2066–2073.
17. Reintjes R, Kajueter H, Ehrhard I, van Treeck U and Ammons A: Applying a case-crossover study design to examine transient exposures in the transmission of N. meningitidis. Eur J Epidemiol (2005) 20: 629–633.
18. Janes H, Sheppard L and Lumley T: Case-crossover analyses of air pollution exposure data: referent selection strategies and their implications for bias. Epidemiology (2005) 16: 717–726.
19. D'Ippoliti D, Forastiere F, Ancona C, Agabiti N, Fusco D, Michelozzi P and Perucci CA: Air pollution and myocardial infarction in Rome: a case-crossover analysis. Epidemiology (2003) 14: 528–535.
20. American Public Health Association: Control of communicable diseases manual: an official report of the American Public Health Association. 18th Ed, American Public Health Association, Washington, D.C. (2004) 281–287.
21. Kido H, Chen Y, Yamada H and Okumura Y: [Host cellular proteases trigger the infectivity of the influenza virus in the airway and brain]. Nippon Yakurigaku Zasshi Folia Pharmacol Japonica (2003) 122: 45–53 (in Japanese).
22. Basu R, Dominici F and Samet JM: Temperature and mortality among the elderly in the United States: a comparison of epidemiologic methods. Epidemiology (2005) 16: 58–66.
23. Hulley SB: Designing clinical research, 3rd Ed, Lippincott Williams & Wilkins, Philadelphia, PA (2007) 136.
24. Szklo M and Nieto FJ: Epidemiology: beyond the basics. 2nd Ed, Jones and Bartlett Publishers, Sudbury, MA (2000) 121–125.