**Supplemental Material**

**Sampling method**

*Ambient air*

The ambient air in and outside of patients rooms were collected from all participants by an air sampler (AirPort MD8®, Sartorius Stedim Biotech, Germany) with a gelatin filter (Type 175, pore size 3 µm, Sartorius Stedim Biotech); 1,000 L of ambient air was collected over approximately 20 min. To collect inside room air, an air sampler was placed on a 1.0 to 1.5 m high platform located approximately 1.5 m from the right pillar on the head side of the patient beds. To collect outside room air, an air sampler was placed on a 1.0 m high platform located approximately 1.5 m from the doorknob of the room on the corridor side, at the same time the indoor air was collected. The gelatin filter was melted by 10 mL of RNA-free distilled water warmed to 70 °C.

*Pharyngeal and vesicular swabs*

Pharyngeal and vesicular swabs were collected with the patient’s permission using an eSwab 480C (Copan, Italy) or Opti-swab (Puritan, Maine), both of which contained 1 mL of the preservation solution. Although the latter is not suitable for virus transport, cryopreservation was performed on the day of specimen collection to ensure preservation of viral DNA. Five pharyngeal and vesicular swabs were collected by Opti-swab from 3 and 2 patients with disseminated herpes zoster (DZ) and localized herpes zoster (HZ), respectively.

All vesicular swabs were PCR positive, and two of five pharyngeal swabs were positive (40%), which was not significantly different from the positivity rates of the specimens collected by eSwab (91% and 54%, respectively).

**PCR method**

Viral DNA was extracted from 200 µL of samples using the spin protocol by Maxwell® RSC Pathogen TNA Kit (Promega, Wisconsin). Viral DNA was extracted from 200 μL samples using Maxwell® RSC 48 (Promega). VZV was detected by real-time PCR for ORF62 region1 using cobas® z480 (Roche Diagnostics, Switzerland). Primers, 5’-CCTCCGTATCGGGACTTCAA and 5’-TGACCGTCCTCGCATACGTA, and a probe, 5’-FAM-TTGGCGAAGAGCTAAC-MGB, were used for a quantitative PCR assay for the VZV ORF62 gene. The assay thermal conditions were 50 °C for 2 min and 95 °C for 10 min, with 45 cycles at 95 °C for 30 s and 60 °C for 30 s. For the PCR assay, 5 µL of template was used (0.005 swabs on blister contents and pharyngeal secretions and 0.5 L min of air). Synthetic nucleic acid plasmids were used as positive controls in the target region of ORF62, adjusted for copy number. A calibration curve was created at six concentrations (102, 103, 104, 105, 106, and 108 copies/μL). We re-tested samples with an error of extrapolated concentration in the calibration curve due to extreme high or low viral load with calibration curves on the other concentrations (103, 104, 105, 106, 107, and 108 copies/μL) after 100 to 1,000-fold dilution, or on the others (9.8, 2.0×10, 3.9×10, 1.6×102, 6.3×102, 2.5×103, and 104 copies/μL), respectively. The viral load of VZV was calculated using calibration curves with crossing point (Cp) values. However, Cp values calculated with no clearly observable waveform were considered invalid. If VZV DNA was detected in one or more of the three replicates without error, the viral load was calculated as the average of the three replicates. If re-tests were performed, only the re-test results were included.

**Information collection**

The humidity and temperature inside and outside the room were also measured during air sampling. The following clinical information was collected: age, sex, type of hospital room (negative-pressure room or not), underlying diseases, immunosuppressants, chemotherapy for cancer, initial symptoms, all symptoms until sampling, onset date, neutrophil and lymphocyte counts, history of varicella zoster vaccination, antiviral agents, number of days of medication, and type of VZV infection (HZ, DZ, or varicella [VA]).

**Environment results**  
The median temperature inside the rooms was 25.9 °C (25.0–26.5 °C) and the relative humidity was 45% (25–59%), while the temperature outside the rooms (corridor) was 25.7 °C (25.4–26.0 °C) and the relative humidity was 40% (25–57%). Temperature and humidity were not significantly different between VA, DZ, and HZ.

Although the room pressure was not measured, the median value of the average atmospheric pressure on the day of collection was 1010.8 hPa (1008.5-1014.9 hPa). The differential pressure between the room and the corridor was about -0.2 to +0.2 hPa. Therefore, the indoor and outdoor pressures were approximately equal to atmospheric pressure.

**References**

1. Inoue N, Matsushita M, Fukui Y, et al. Identification of a varicella-zoster virus replication inhibitor that blocks capsid assembly by interacting with the floor domain of the major capsid protein. *J Virol* 2012;86:12198–12207.

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| Supplemental Table S1. Patient characteristics associated with the presence of varicella zoster virus in indoor air | | | |
| (a) All cases |  |  |  |
|  | Inside air negative | Inside air positive | p value |
| N | 40 | 6 |  |
| Varicella, n (%) | 4 (10.0) | 3 (50.0) |  |
| Disseminated zoster, n (%) | 16 (40.0) | 2 (33.3) |  |
| Localized herpes zoster, n (%) | 20 (50.0) | 1 (16.7) |  |
| Age, years, median, interquartile range (IQR) | 64.5 (47.0-74.0) | 58.0 (36.8-77.0) | >0.1 |
| Sex (male), n (%) | 19 (47.5) | 2 (33.3) | >0.1 |
| Underlying diseases, n (%) | 23 (57.5) | 2 (33.3) | >0.1 |
| Solid cancer (active), n  Solid cancer (inactive), n  Hematological malignancy, n  Bone marrow transplantation, n  Solid organ transplantation, n  Collagen diseases, n  Diabetes mellitus, n  Others, n | 3  1  1  2  1  8  6  7 | 1  0  0  0  0  1  0  0 |  |
| Immunosuppressive agents, n (%)  (including low-dosage oral corticosteroids and methotrexate) | 14 (35.0) | 2 (33.3) | >0.1 |
| WBC (/µL), median (IQR) | 5870.0 (4947.5-7645.0) | 3735.0 (3550.0-4617.5) | 0.006 |
| ANC (/µL), median (IQR) | 3772.9 (2932.8- 5666.4) | 2507.5 (2057.4- 2850.9) | 0.026 |
| ALC (/µL), median (IQR) | 1332.9 (924.1- 1632.3) | 809.7 (704.7- 1092.6) | 0.029 |
| Prior antiviral agents, n (%) | 37 (92.5) | 3 (50.0) | 0.026 |
| Intravenous, n (%) | 36 (90.0) | 2 (33.3) | 0.005 |
| Oral, n (%) | 6 (15.0) | 2 (33.3) | >0.1 |
| Median time between onset of rash and sampling, days, median (IQR) | 4.0 (3.0-5.2) | 3.5 (2.2-4.8) | >0.1 |
| Median time between initiating antiviral agents and sampling, days, median (IQR) | 3.0 (2.0-4.0) | 1.5 (1.0-2.0] | 0.034 |
| Vesicle swabs: median viral load, log10 copies/test (IQR) | 7.6 (5.8-7.9) | 7.1 (7.0-7.7) | >0.1 |
| Pharyngeal swabs: median viral load, log10 copies/test (IQR) | 1.0 (0-2.6)† | 4.4 (3.9-5.0) | 0.004 |
|  |  |  |  |
| (b) Except varicella cases |  |  |  |
|  | Inside air negative | Inside air positive | p value |
| N | 36 | 3 |  |
| Disseminated zoster, n (%) | 16 (44.4) | 2 (66.7) |  |
| Age, years, median, interquartile range (IQR) | 67.5 (52.5-74.5) | 78.0 (76.0-85.5) | 0.048 |
| Sex (male), n (%) | 16 (44.4) | 1 (33.3) | >0.1 |
| Underlying diseases, n (%) | 22 (61.1) | 2 (66.7) | >0.1 |
| Solid cancer (active), n  Solid cancer (inactive), n  Hematological malignancy, n  Bone marrow transplantation, n  Solid organ transplantation, n  Collagen diseases, n  Diabetes mellitus, n  Others, n | 3  1  1  2  1  8  6  6 | 1  0  0  0  0  1  0  0 |  |
| Immunosuppressive agents, n (%)  (including low-dosage oral corticosteroids and methotrexate) | 13 (36.1) | 2 (66.7) | >0.1 |
| WBC (/µL), median (IQR) | 6130.0 (4950.0- 7815.0) | 4860.0 (4375.0- 5180.0) | >0.1 |
| ANC (/µL), median (IQR) | 4161.7 (2975.8- 5755.2) | 2964.6 (2734.9- 3660.3) | >0.1 |
| ALC (/µL), median (IQR) | 1332.9 (931.2- 1632.3) | 871.4 (809.7- 1018.9) | >0.1 |
| Prior antiviral agents, n (%) | 35 (97.2) | 2 (66.7) | >0.1 |
| Intravenous, n (%) | 34 (94.4) | 2 (66.7) | >0.1 |
| Oral, n (%) | 6 (16.7) | 1 (33.3) | >0.1 |
| Median time between onset of rash and sampling, days, median (IQR) | 4.0 (3.0-6.0) | 5.0 (3.5-6.0) | >0.1 |
| Median time between initiating antiviral agents and sampling, days, median (IQR) | 3.0 (2.0-4.0) | 2.0 (1.5-3.0) | >0.1 |
| Vesicle swabs: median viral load, log10 copies/test (IQR) | 7.6 (5.8-7.9) | 7.1 (7.0-7.1) | >0.1 |
| Pharyngeal swabs: median viral load, log10 copies/test (IQR) | 0 (0-2.3)† | 4.1 (3.9-4.3) | 0.056 |
| †”log0” is a value that does not exist, but for convenience, the number of copies less than 1 copies/test was replaced by “0”. | | | |

Figure S1. Study flow diagram



VZV, Varicella zoster virus

Figure S2. Viral load of VZV in vesicle swab, pharyngeal swab, or inside ambient air after onset of rash of each VZV infection type



The p-values for the Jonckheere-Terpstra test (significance level 0.05) were 0.40, 0.039, and 0.35 for (a), (b), and (c), respectively, with only (b) showing a decreasing trend.

Figure S3. Viral load of VZV in vesicle swab, pharyngeal swab, or inside ambient air after initiating antiviral agents in each VZV infection type



The p-values for the Jonckheere-Terpstra test (significance level 0.05) were 0.31, 0.041, and 0.11 for (a), (b), and (c), respectively, with only (b) showing a decreasing trend.

\*Day 1 is defined as the day on which antivirals were initiated, with no antivirals in all patients included on day 1.