Effects of Temperature and Position Change on Neonatal Brain Regional Oxygen Saturation in Tub Bathing: A Prospective Study

Tomohiko Nishino · Naoki Ito · Shinya Tomori · Satoshi Shimada · Misaki Kodera · Kiyoko Morita · Kazuhiro Takahashi · Masakazu Mimaki

ABSTRACT

Introduction: A major goal in neonatal medicine is to reduce stress as much as possible in routine care. Bathing is one of the important routine cares for neonates, but it makes a big environmental change for them. We aimed to examine whether water temperature, room temperature, and position changes in tub bathing serve as noxious stimuli to neonates.

Methods: This prospective trial was performed in full-term and non-low-birth-weight neonates admitted to the hospital between July 2020 and March 2021. Those with underlying diseases, fetal distress, infection, and other medical conditions were excluded. Measurements were taken during the neonates’ first tub bath since birth, which was performed by a trained nurse.

Results: In total, 17 neonates were analyzed. No changes in rSO₂ due to head washing or position change were observed; however, rSO₂ significantly decreased upon entering (78.5 ± 4.1% vs. 75.7 ± 4.1%, p < 0.001) and exiting the bath (75.8 ± 5.7% vs. 74.4 ± 5.4%, p < 0.04). The rate change in rSO₂ upon entering the bath showed a significant inverse correlation with water temperature (r = −0.53, p < 0.03), and there were no significant correlations between rSO₂ and water or room temperature upon exiting the bath. There was no change in body skin temperature before and during bathing.

Changes in regional oxygen saturation (rSO₂), determined using near-infrared spectroscopy, and water and room temperature, were examined at five different time points: upon entering the bath, head washing, position change, exiting the bath, and during the 3 min after bathing.

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after bathing, but rSO₂ gradually decreased during the 3 min after bathing.

Conclusions: Neonates may perceive certain temperatures during bathing as noxious stimuli. Therefore, methods to minimize stress associated with bathing should be implemented to reduce the difference between water temperature and room temperature during bathing.

Trial registration: This trial has been registered at UMIN repository with the trial number UMIN000041045 (https://upload.umin.ac.jp/cgi-open-bin/ctr_e/ctr_view.cgi?recptno=R000046500). The date of the final dataset was April 01, 2021.

Keywords: Bathing; Near-infrared spectroscopy; Neonatal intensive care unit; Noxious stimulus; Pediatric; Regional oxygen saturation; Room temperature; Routine care; Water temperature

Key Summary Points

Why carry out this study?
Noxious stimuli, such as pain in the neonatal period, are associated with poor neurodevelopmental prognosis.

Whether stimulation during bathing, a routine care procedure performed not only in medical settings but also at home, is also a noxious stimulus is unclear.

This trial examined whether stimuli received from water temperature, room temperature, and position changes might be perceived as noxious.

What was learned from the study?
The regional oxygen saturation decreased significantly upon entering and exiting the bath, along with change in environmental temperature, as well as for during the 3 min after bathing when no change in the body temperature was observed.

Reducing stress during bathing may be possible by bringing the water and room temperatures closer to the body temperature, with the goal of achieving an ideal bathing experience for the neonate.

INTRODUCTION

Reducing noxious stimuli in neonates during routine care remains a major goal in neonatal medicine. Medical advances have focused attention on neurodevelopment in addition to lifesaving measures. Bathing is a one of the routine cares and an important component of neonatal care because, in addition to maintaining hygiene, it contributes to the formation of parental attachment [1, 2]. Neonates experience stress, even from diaper changes [3]. In preterm neonates, repeated noxious stimuli have been shown to reduce brain volume, adversely affect development, and decrease the thresholds of warmth and pain perception [3–6]. However, even full-term neonates present with susceptibility to neurodevelopmental abnormalities that persist beyond infancy due to repeated pain experiences [7]. Therefore, neonatal intensive care units (NICU) are managed with great care in terms of lighting, noise levels, and temperature [8, 9]. However, even though infants vary in their stress response depending on the bathing procedure [10], the bathing procedure is not standardized; hence, there is no consensus regarding the water temperature or procedures to be performed during bathing. Therefore, the stress experienced by neonates associated with bathing should be assessed. Most of the neonatal stress-assessment tools that have been developed to date rely on subjective evaluation [11], and do not always agree with brain activity recorded by electroencephalography [12]; thus, objective assessment methods are warranted. Near-infrared spectroscopy (NIRS) can be used to measure pulsatile-independent total oxygen saturation of arterial, venous, and capillary
blood; therefore, tissue oxygen consumption can be continuously observed by monitoring regional oxygen saturation (rSO2) [13, 14]. Significant changes in NIRS were observed during heel blood sampling, but not during von Frey hair stimulation of the plantar surface of the foot in neonates [15]. In other words, changes in NIRS are caused only by noxious stimuli and not by all stimuli. The experience of pain has been shown to increase oxygen consumption, with the changes occurring in the primary somatosensory cortex on the surface of the brain [16, 17]. NIRS is thus an excellent tool for observing these changes [15, 18, 19], which can be used to enhance the quality of routine care and reduce neonatal stress [20–24]. Because routine care is repeated daily, its optimization is important to reduce stress in neonates, which should contribute to improved neurodevelopment. This trial examined whether the stimulus caused by bathing as part of routine neonatal care was affected by temperature and position change using NIRS.

METHODS

Study Population

This prospective trial on cerebral blood flow measurement during bathing in neonates (UMIN000041045) was a quasi-experimental all-case survey conducted during the first bath after birth in a population of term neonates with a birth weight of ≥ 2500 g admitted to a level III NICU in Tokyo, Japan, between July 15, 2020, and March 31, 2021. Neonates whose conditions were generally stable were included in the trial; the first bath was included in the trial, the timing of which was normal for neonates. The final decision to initiate bathing was made by the attending physician.

Measurement Method

A digital device capable of measuring room temperature, water temperature, and room humidity simultaneously was installed at a distance from the bathtub, with the terminal of the water temperature meter fixed to the lower side of the bathtub. The rSO2 was measured using an INVOS 5100 NIRS cerebral oximeter (Covidien Somanetics, Troy, MI, USA). The neonate was placed on a table before bathing. Body skin temperature was measured at the axilla, and the NIRS probe was applied to the mid-forehead to begin measuring cerebral tissue oxygenation. The neonate was allowed to rest for at least 3 min before and after bathing. At this point, non-nutritive-sucking and tucking position was facilitated with the aim of reducing the infant’s stress as needed [25], but no cuddling or other activity that would change the position of the child was performed. Bathing was started after confirming that rSO2 was stable. The probe was attached to the same site until the end of the measurement period. After bathing, the neonate’s entire body was towel-dried on the table to maintain warmth, and care was taken to avoid hypothermia.

Bathing Method

The neonates were bathed by one of six well-trained nurses, all of whom had been working in the NICU for more than 8 years. Prior to the start of the trial, the written instructions of the trial protocol were distributed to all nurses, who reviewed the instructions together to ensure technical uniformity in following the bathing method. The timing of bathing was standardized to approximately 90–120 min after oral feeding. A two-stage bathtub was filled with warm water, and the appropriate water temperature was determined by feel by the nurse in charge, who was blinded to the temperature reading. During the trial, warm water was kept flowing into the bathtub to maintain a constant water temperature. The positioning of the neonate during bathing and the order of washing were performed according to the bathing procedure implemented at our hospital (Table 1). The nurses attempted to prevent the neonates from crying. The protocol for the timing of bathing and position change was used as a rough guide, and bathing was performed in accordance with the neonate’s crying and body movement. If the neonate was crying prior to
any of the four moments used for analysis—(A) upon entering the bath, (B) head washing, (C) position change, and (D) exiting the bath—we waited until they had stopped crying before continuing.

**Exclusion Criteria**

As this trial was performed to evaluate routine care, neonates with as close to normal medical circumstances as possible were selected. The exclusion criteria were as follows: those undergoing warming or humidification in an incubator and treatment including intravenous fluid therapy or phototherapy; those with fetal distress, postoperative bleeding, or abnormal coagulation parameters; respiratory or heart failure requiring oxygen administration; gastrointestinal symptoms; malformations including congenital heart disease; and suspected chromosomal/genetic abnormalities. Neonates whose mothers were administered antipsychotics or thyroid hormones during birth were also excluded. The data of patients who started treatment after the bathing measurements were excluded from the analysis.

**Statistical Analysis**

All statistical analyses were performed using R version 3.6.1 (R Foundation for Statistical Computing, Vienna, Austria). The measurements that were taken at five time points, at the aforementioned moments of (A), (B), (C), and (D) and a period of time (E) during the 3 min after bathing, were evaluated (Table 1). At (A), (B), (C) and (D): the rSO2 before (mean of –3–0 s before intervention) and after the intervention (mean of 6–9 s after the intervention) were compared using a paired \( t \) test. Pearson’s correlation analysis was performed to determine the correlation between the rate change in rSO2 and water or room temperature. The body skin temperatures before and after bathing were compared using a paired \( t \) test. Rate changes in rSO2 during the 3 min after bathing were calculated by setting rSO2 before (mean of –3–0 s before intervention) as 0. The \( t \) test results were expressed as mean ± standard deviation. In all analyses, \( p < 0.05 \) was considered to indicate statistical significance.

**Ethical Approval**

The trial was approved by the Teikyo University Ethical Review Board for Medical and Health Research Involving Human Subjects (protocol number 20-083). All procedures in the present trial involving human participants were performed in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. Informed consent was obtained from the day of hospitalization to the day before bathing. The parents/guardians of
The neonates were provided a written and verbal explanation of the procedures in plain language, and written consent was obtained for participation in the research and the publication of the results. Additionally, explained the right to refuse participation and withdraw consent at will were also explained.

**Trial Registration**

NSBB study has been registered at UMIN repository with the trial number UMIN000041045 (https://upload.umin.ac.jp/cgi-open-bin/ctr_e/ctr_view.cgi?recptno=R000046500). The date of the final data set was April 01, 2021.

**RESULTS**

Of the 199 neonates admitted to the NICU during the trial period, 112 met the initial inclusion criteria (Fig. 1). Among these 112 neonates, 84 met the inclusion and exclusion criteria (Table 2), and the trial sample size thus comprised 28 neonates. Measurements were performed in 18 neonates who met the inclusion criteria. One neonate was later excluded from the analysis due to the development of jaundice.

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![Flow diagram showing the study selection procedure.](https://upload.umin.ac.jp/cgi-open-bin/ctr_e/ctr_view.cgi?recptno=R000046500)

**Fig. 1** Flow diagram showing the study selection procedure. Among all neonates admitted to the neonatal intensive care unit during the trial period, 87 were born at full-term and did not have low birth weights.
decreases in rSO2 from before the intervention were observed upon entering (Fig. 3A, 78.5 ± 4.1% vs. 75.7 ± 4.1%, p < 0.001) and exiting the bath (Fig. 3B, 75.8 ± 5.7% vs. 74.4 ± 5.4%, p < 0.04). However, no variations were detected during head washing (75.6 ± 4.7% vs. 75.9 ± 5.0%, p = 0.36) or position change (74.5 ± 5.0% vs. 74.5 ± 5.1%, p = 0.94). The rate change in rSO2 upon entering the bath showed a significant inverse correlation only with water temperature (r = −0.53, p < 0.03) (Fig. 4). There were no significant correlations between rSO2 and water or room temperature upon exiting the bath. There was no significant difference in body skin temperature before and after bathing (37.0 ± 0.26 °C vs. 36.9 ± 0.31 °C, p = 0.15). During the 3 min after bathing, rSO2 showed a gradual decrease (Fig. 5). Water temperature, room temperature, and humidity were kept constant during all measurements. No adverse events occurred during the trial.

DISCUSSION

The rSO2 significantly decreased upon entering and exiting the bath in this trial. The change in rSO2 was greater with higher water temperature upon entering the bath and was less pronounced at water temperatures closer to the body skin temperature. Bathing can bring about a variety of changes such as hemodynamic in neonates; however, the lack of a uniform change in rSO2 implies that may have been

Table 2 Details of the patients who met the exclusion criteria

| Exclusion criteria                                      | n  |
|---------------------------------------------------------|----|
| Having already been bathed                              | 25 |
| Fetal distress                                          | 15 |
| Maternal medication                                     | 15 |
| Intrauterine infections (including suspected)           |    |
| Toxoplasmosis                                           | 3  |
| Hepatitis B                                             | 2  |
| Varicella                                               | 1  |
| Coronavirus disease 2019                                | 1  |
| Unknown                                                 | 1  |
| Neonatal infections (including suspected)               |    |
| Neonatal toxic shock-like exanthematous disease         | 1  |
| Unknown                                                 | 5  |
| Congenital malformations                                |    |
| Ventricular septal defect                               | 2  |
| Hydronephrosis                                          | 1  |
| Twin-to-twin transfusion syndrome                       | 1  |
| Esophageal atresia                                      | 1  |
| Cerebral dysplasia                                      | 1  |
| Natal tooth                                             | 1  |
| Meconium aspiration syndrome                            | 2  |
| Peripartum trauma                                       | 1  |
| Bradycardia                                             | 1  |
| Arrhythmia                                              | 1  |
| Respiratory distress syndrome                           | 1  |
| High serum creatine kinase level                        | 1  |
| Irregular antibody positivity                           | 1  |

Table 3 Patient characteristics

|                                | n = 17 |
|--------------------------------|--------|
| Sex (boy/girl)                 | 6/11   |
| Gestational age at birth (weeks)| 38.6 ± 1.4 |
| Birth weight (g)               | 3094 ± 376 |
| Apgar score at 1 min (quartile range) | 8 (8–8) |
| Apgar score at 5 min (quartile range) | 9 (9–9) |
| Disease and reason for hospitalization: |        |
| Infant of diabetic mother      | 12     |
| Transient tachypnea            | 2      |
| Transient apnea                | 2      |
| Hypoglycemia                   | 1      |
affected by temperature. Additionally, rSO₂ showed a uniform change upon exiting the bath at a room temperature of approximately 25 °C. It is noteworthy that environmental temperature would cause such a different response. Although many reports related to bathing in neonates have been published, to the best of our knowledge, this is the first trial involving the real-time measurement of cerebral oxygen saturation, and each of temperatures in neonates during bathing.

Neonates may perceive the water temperature upon entering the bath as noxious stimulation. The decrease in rSO₂ upon entering the bath observed in this trial suggests that bathing was perceived as a noxious stimulus. The change in rSO₂ was insignificant at approximately 37 °C but was substantial at approximately 40 °C and showed a significant inverse correlation with water temperature. Even if the temperature of the warm water is what a seasoned medical professional would consider appropriate for bathing, higher temperatures served as stimuli. In human somatosensory perception, temperature and pain are processed through the spinothalamic tract. Thermal stimulation at 37–40 °C, which was used in the present trial, activates mainly the C-fibers, and the magnitude of stimulation is dependent on temperature [26]. In addition, a study in rats reported that the temperature threshold for the recognition of noxious heat stimulus was low at early postnatal ages and increased over the subsequent days [27]. Therefore, even if the parent feels that the temperature is appropriate, it is important to consider the perception of water temperature by the neonate to avoid stress during bathing.

| Patient number | Age (days) | Room temperature (°C) | Humidity (%) | Water temperature (°C) | BT before bathing (°C) | BT after bathing (°C) |
|----------------|-----------|-----------------------|--------------|------------------------|------------------------|----------------------|
| 1              | 2         | 26.4                  | 52           | 39.1                   | 36.8                   | 36.3                 |
| 2              | 3         | 25.8                  | 42           | 37.5                   | 37.2                   | 37.0                 |
| 3              | 4         | 26.0                  | 42           | 37.5                   | 37.0                   | 37.1                 |
| 4              | 3         | 26.8                  | 39           | 37.2                   | 37.3                   | 37.1                 |
| 5              | 3         | 25.9                  | 35           | 39.1                   | 36.9                   | 37.1                 |
| 6              | 4         | 24.8                  | 31           | 36.4                   | 37.1                   | 36.9                 |
| 7              | 2         | 25.6                  | 33           | 37.6                   | 37.5                   | 36.9                 |
| 8              | 4         | 25.4                  | 37           | 38.9                   | 37.3                   | 36.7                 |
| 9              | 2         | 25.1                  | 42           | 38.9                   | 36.9                   | 37.1                 |
| 10             | 3         | 24.8                  | 44           | 39.9                   | 36.4                   | 36.9                 |
| 11             | 3         | 24.8                  | 34           | 39.0                   | 37.0                   | 37.1                 |
| 12             | 3         | 25.1                  | 34           | 39.0                   | 36.8                   | 36.4                 |
| 13             | 4         | 25.4                  | 39           | 38.2                   | 36.6                   | 37.0                 |
| 14             | 3         | 25.5                  | 45           | 39.5                   | 37.1                   | 37.7                 |
| 15             | 3         | 25.1                  | 37           | 39.2                   | 36.9                   | 37.0                 |
| 16             | 3         | 26.2                  | 50           | 37.9                   | 37.0                   | 36.7                 |
| 17             | 3         | 26.0                  | 36           | 37.5                   | 37.0                   | 36.7                 |
The room temperature at the time of exiting the bath may be stressful for neonates. The decrease in rSO\(_2\) observed in this trial upon exiting the bath was uniform even at the recommended controlled room temperature of 25–27 °C. The World Health Organization recommends a warm room with a temperature not less than 25 °C with no draft as the environment for neonatal care [8]. However, this view is a recommendation for the general environment and does not take into account the room temperature when exiting the bath. The activation of transient receptor potential melastatin 8, which is mainly involved in perception of temperatures of approximately 25 °C, is associated with insensible temperatures due to its analgesic properties [28]. However, as sensory processes continue to develop after birth [29], the temperature response in the neonatal period remains immature. Transient receptor potential melastatin 8 expression continues from the fetal stage to several weeks after birth [30]. This suggests that the neonates perceived room temperature as a noxious stimulus due to their immature transient receptor potential channel expression. However, it is arguable that only room temperature is responsible for the change in rSO\(_2\), as temperature perception is a complex response involving many channels [26], and the neonates included in this trial had been housed at constant room temperatures prior to bathing. The temperature set point at

![Fig. 2 Trend of rate changes in regional oxygen saturation (rSO\(_2\)) from 3 s before to 9 s after each moment of intervention. In each figure, the vertical axis represents the rate change in rSO\(_2\) and the horizontal axis represents the time from 3 s before to 9 s after the intervention. The rate change in rSO\(_2\) was set to (each rSO\(_2\))/rSO\(_2\) before intervention; mean of −3 to 0 s before treatment), and the trend of rate change in rSO\(_2\) from each 3 s before to 9 s after intervention is shown. A On entering the bath; B Head washing; C Position change; D Exiting the bath. The dotted lines show the values for each patient, and the thick solid lines show the average for all patients.](image-url)
which the channel activity of thermoreceptors is induced is affected by various substances [31, 32], and some factors during the neonatal period may have caused an effect. Consideration should be given to the room temperature at the time of exiting the bath, but further investigation of this issue is required.

Temperature maintenance for neonates after bathing must be performed more carefully rather than merely evaluating the body skin temperature. In this trial, rSO₂ gradually decreased during the 3 min after bathing. In contrast, no significant change in body skin temperature was observed. These results indicate that body skin temperature may be compensated, which indicates that the stress experienced by neonates cannot be assessed by observing body skin temperature alone. Previous reports have shown a decrease in body temperature and an increase in peripheral oxygen consumption before and after bathing in 19 neonates based on NIRS [33]. The difference in temperature between the bath water and room air contributes to heat dissipation and decreases the body skin temperature, leading to compensatory heat production by pheochromocytes [34]. Various bathing methods involving temperature maintenance have been attempted by evaluating body temperature as an indicator [35, 36]; however, it should be noted that stress may be applied even if no change in body temperature is observed. In addition, to the best of our knowledge, previous reports have not examined room temperature after exiting the bath. Oxygen consumption in neonates is highly dependent on environmental temperature [33]. Therefore, reducing stress in neonates is possible by maintaining a higher room temperature to eliminate temperature differences upon exiting the bath.

In this trial, no changes in rSO₂ associated with position change or head washing during bathing were observed, which suggests stability during tub bathing. In a previous study, the cerebral tissue oxygenation index obtained using NIRS was significantly altered by a 15° head-up tilt in 17 healthy, full-term neonates 2–4 weeks after birth [37]. Thus, rSO₂

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**Fig. 3** Comparison of regional oxygen saturation (rSO₂) before and after the intervention on A entering the bath and B exiting the bath. In each panel, the vertical axis shows rSO₂ values and the horizontal axis shows the rSO₂ values before (mean of –3–0 s before intervention) and after (mean of 6–9 s after intervention) the intervention. A paired t test was used for analyses.
monitoring by NIRS can also be performed to evaluate the effects of rapid repositioning on neonates. Traditional sponge bathing is known to affect hemodynamics and respiratory status and cause stress in neonates [38]. Tub bathing has recently attracted attention due to preventing the lowering of body temperature, the decrease in dryness and redness of skin after bathing, as well as improved stability compared to a sponge bath during bathing [38, 39]. The stability and safety of the procedure are important because the bathing of neonates is generally performed by families who may be unfamiliar with neonatal care.

This trial has several limitations. First, NIRS can only measure the pain response and not the pain stimulus itself. Because a time lag occurs before oxygen consumption, the pain response occurs several seconds after the stimulus is applied. The timing of this trial was chosen to

Fig. 4 Correlations between rate change in regional oxygen saturation (rSO2) and water temperature and room temperature. In each figure, the vertical axis represents the rate change in rSO2 and the horizontal axis represents the temperature. Rate change in rSO2 was set to (rSO2 after intervention: mean of 6–9 s after intervention) / (rSO2 before intervention: mean of 3–0 s before intervention). Differences between A on entering the bath and water temperature; B on entering the bath and room temperature; C exiting the bath and water temperature; D exiting the bath and room temperature.
be similar to a previous study [40], and it is considered to have a certain reliability. In addition, the change in rSO2 is known to vary with blood flow [14], which may be affected by factors such as water pressure or buoyancy. Interestingly, however, only a small change in rSO2 was observed upon entering the bathtub when the water temperature was close to the body temperature. This may have occurred due to the lesser influence of the aforementioned factors. As this trial was performed to evaluate routine care, the conditions were maintained as similar as possible to normal bathing. Therefore, heart rate, saturation of percutaneous oxygen, and blood pressure, all of which require the use of a measurement device, were not measured during bathing. These factors, along with respiratory rate and crying, might have affected rSO2 and should be examined in future studies. Second, the sample size was small due to the small number of full-term births at a single institution. We aimed to have a normally distributed sample of 30 patients, but this was not achieved because many families did not consent to participate in the trial. One of the most common reasons for this was because parents wished to bathe their child for the first time themselves rather than allowing it to be performed by a nurse. The results of all parameters in this trial showed normality and were suitable for statistical analysis, with maintenance of a certain level of reliability. Moreover, by limiting the number of patients to a single institution, it was possible to ensure a uniform bathing method by skilled medical personnel that kept neonates in a stable condition. The stress experienced by infants during bathing due to the technique of the caregiver has been well described [41]. This problem was addressed in the present trial by standardizing the routine

![Fig. 5](image-url)
care performed. During the trial, no cases of aborted measurements due to electrode dislodgement were recorded, indicating the stability of the technique. However, further studies including a larger study population and implementing strict temperature controls should be conducted.

CONCLUSIONS

We suggest the implementation of a water temperature close to the body temperature of neonates upon entering the bath and decreasing the temperature changes upon exiting the bath with utmost care as temperature changes can be perceived by neonates as noxious stimuli. Because bathing is necessary for hygiene and fostering parental attachment, determining the best method for minimizing stress experienced by neonates during bathing is essential.

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Compliance with Ethics Guidelines. The trial was approved by the Teikyo University Ethical Review Board for Medical and Health Research Involving Human Subjects (Protocol Number 20-083). All procedures in studies involving human participants were performed in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. Informed consent was obtained from the day of hospitalization to the day before bathing. The parents/guardians of the neonates were provided a written and verbal explanation of the procedures in plain language, and written consent was obtained for participation in the research and the publication of the results. Additionally, explained the right to refuse participation and withdraw consent at will were also explained.

Data Availability. The datasets generated during the present study are available from the UMIN repository: https://upload.umin.ac.jp/cgi-bin/ctr_e/ctr_view.cgi?recptno=R000046500. The date of the final dataset is April 1, 2021.

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