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# JAST (Journal of Animal Science and Technology) TITLE PAGE

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ARTICLE INFORMATION	Fill in information in each box below
Article Type	Research article
Article Title (within 20 words without abbreviations)	Changes in ruminoreticular temperature and body activity in pregnant Hanwoo cows ( <i>Bos taurus coreanae</i> ) after lumpy skin disease vaccination
Running Title (within 10 words)	Ruminoreticular temperature after lumpy skin disease vaccination in pregnant cows
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Availability of data and material	The data analyzed or generated during this study are available upon request from the corresponding author.
Authors' contributions Please specify the authors' role using this form.	<a href="#">Conceptualization: Ha J, Yu D, Kim J, Han J, Kim M, Kim G, Jeong J, Gim GM, Kiim D</a> <a href="#">Data curation: Yu D, Kim J, Han J, Gim GM, Kiim D</a> <a href="#">Formal analysis: Ha J, Kiim D</a> <a href="#">Methodology: Ha J, Yu D, Kim M, Kim G, Gim GM, Kiim D</a> <a href="#">Software: Gim GM, Kiim D</a> <a href="#">Validation: Ha J, Kim G, Jeong J, Kiim D</a> <a href="#">Investigation: Ha J, Yu D, Kim J, Han J, Kim M, Kim G, Jeong J, Kiim D</a> <a href="#">Writing - original draft: Ha J, Yu D, Gim GM, Kiim D</a> <a href="#">Writing - review &amp; editing: Ha J, Yu D, Kim J, Han J, Kim M, Kim G, Jeong J, Gim GM, Kiim D</a>
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13 **Abstract**

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15 The first outbreak of lumpy skin disease (LSD) occurred in South Korea in October 2023, and  
16 cattle are being vaccinated countrywide to prevent its spread. However, studies regarding the  
17 changes in body temperature and activity after LSD vaccination during pregnancy are lacking.  
18 Therefore, this study aimed to compare the ruminoreticular temperature and body activity of 18  
19 pregnant and 28 non-pregnant cows using a bolus sensor after LSD vaccination. Two days after  
20 LSD vaccination, the ruminoreticular temperature of all the experimental groups increased and  
21 that of the pregnant cows remained very high 3 to 5 days after vaccination compared with that in  
22 the non-pregnant cows. The rate of maintaining  $\geq 40$  °C was 12.8% in non-pregnant cows and up  
23 to 20.8% in pregnant cows. Body activity also temporarily increased in pregnant cows compared  
24 with that in the non-pregnant cows on the 1st and 4th days after vaccination. The results of this  
25 study may be applied to prevent the rise in ruminoreticular temperature and used as raw data by  
26 veterinarians when LSD vaccine is administered during pregnancy.

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28 **Keywords:** Lumpy skin disease, lumpy skin disease vaccination, ruminoreticular temperature,  
29 body activity, pregnant cows, Hanwoo

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## Introduction

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Lumpy skin disease (LSD) in cattle and buffalo is caused by the LSD virus belonging to the family *Poxviridae* and genus *Capripoxvirus* (1-3). Mosquitoes, especially *Aedes aegypti*, can transmit the LSD virus for at least 6 days without significant loss of titer (3, 4). The incubation period for LSD is approximately 7 days. The main symptom is sporadic swelling of the skin, with the appearance of nodules having diameter 0.5–5 cm (3, 5, 6). Other symptoms include high fever of  $>40$  °C, rapid reduction in milk production, loss of appetite, nasal discharge, salivation, swollen lymph nodes, weight loss, miscarriage, and infertility (1-3, 5, 7-11).

LSD was first reported in 1929 in Zambia, from where it spread to numerous places, including South Africa, North Africa, the Middle East, Europe, and Asia (1, 3, 12-14). According to a recent report by the World Animal Health Information System and the Ministry of Agriculture, Food and Rural Affairs (MAFRA), Republic of Korea, LSD first broke out in South Korea in Seosan city on October 20, 2023.

To prevent the spread of LSD, the immediate slaughter of all cattle that have come in contact with infected cattle and elimination of the initial source of infection are recommended (3, 12, 15). However, if the disease has already spread widely, vaccination is recommended in most countries because this is the only method of prevention (6, 15). According to a report by MAFRA, 3 types of LSD vaccines were used in Republic of Korea. As of 14:00 on November 5, 2023, the status of LSD vaccination in Korea was 90.9% (3 766 000/4 076 000 cattle). To complete the nationwide vaccination by November 10, 2023, cows in areas at risk of LSD are being vaccinated by city/county vaccination groups (2065 people from 931 classes nationwide) and farm owners (self-vaccination).

Studies have shown that cows demonstrate fever (83.9%), decreased feed intake (85.9%), and reduced milk production (94.6%) when the LSD vaccine is administered (16). The analysis of changes in rectal temperature according to the LSD vaccination showed that compared with that of the control animals, the rectal temperature increased, and high-dose vaccinations resulted in rise of temperatures to  $\geq 40$  °C (11). Other studies showed that the highest rectal temperature was recorded 8 days after LSD vaccination, and milk production decreased by up to 16% (17).

While vaccination is required to prevent LSD, studies comparing body temperature and activity in pregnant and non-pregnant cows have not been conducted to date. Therefore, this study aims to analyze the patterns of changes in ruminoreticular temperature and body activity measured using a bolus sensor after LSD vaccination in pregnant and non-pregnant Hanwoo cows.

## Materials and Methods

### Animals and Management

The cows used in this study were bred at the Gyeongsangbuk-do Livestock Technology Research Institute, fed according to the Korean Feeding Standard for Hanwoo, and housed in pens (rearing space=300 m<sup>2</sup>/15 cows) equipped with stanchions. Before beginning the experiment, cows with no abnormalities in the ovaries and uterus were selected by ultrasound examination. Finally, 46 cows (18 pregnant cows, 28 non-pregnant cows) were chosen for the study.

All the experiments were approved by the Animal Ethics Committee of the Gyeongsangbuk-do Livestock Research Institute (approval number: protocol code GAEC/140, approval date: December 14, 2021). Table 1 shows the age, parity, and pregnancy day of the cows used in the experiment.

### Ruminoreticular Temperature and Body Activity Measurement

Six months before beginning the experiment, a bolus sensor (smaXtec Co., Inc. New Zealand) was orally administered and placed in the cow's rumen or reticulum; the adaptation period was 6 months. Information regarding the sensor used in the experiment and the method of measuring temperature and activity in the rumen every 10 min have been described in detail previously (18).

### LSD Vaccination

LSD vaccine (Lumpyvax®, Republic of South Africa; each 1 mL [1 dose] of the vaccine contains 10<sup>4</sup> TCID<sub>50</sub> of freeze-dried, live, attenuated virus) was administered after disinfecting the vaccination site using 70% alcohol. The powder was dissolved in the dilution solution and subcutaneously injected (1 mL/cow) into the neck of the cow using a disposable syringe.

### Pregnancy Test

Two weeks before beginning the experiment, a pregnancy test was conducted using rectal ultrasound equipment (DRAMINSKI iScan mini, Dramiński S.A., Gietrzwałd, Poland).

### Statistical Analysis

Changes in ruminoreticular temperature and body activity in pregnant and non-pregnant cows were statistically analyzed by two-way ANOVA using GraphPad Prism (version 8.0.1; GraphPad Software, Inc., La Jolla, CA, USA). p-value ≤0.05 was considered significant.

## Results

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99 The ruminoreticular temperatures of pregnant and non-pregnant cows were measured at 10-  
100 minute intervals before and after LSD vaccination using the sensor. The average temperature for  
101 4 hours is shown in Figure 1. Two days before LSD vaccination, the average ruminoreticular  
102 temperatures of the pregnant and non-pregnant cows were  $38.89 \pm 0.01$  °C and  $38.74 \pm 0.01$  °C,  
103 respectively (Figure 1). Two days after LSD vaccination, the ruminoreticular temperatures in  
104 both the groups gradually increased; this continued until 6 days after vaccination ( $p < 0.001$ ). The  
105 rise in ruminoreticular temperature was greater in pregnant cows than it was in non-pregnant  
106 cows 3–5 days after vaccination ( $p < 0.001$ ).

107 The body activity of pregnant and non-pregnant cows were measured at 10-minute intervals  
108 before and after LSD vaccination using the sensor. The average body activity for 4 hours is  
109 shown in Figure 2. Two days before LSD vaccination, the mean body activity of the pregnant  
110 and non-pregnant cows were  $2.41 \pm 0.06$  V and  $2.81 \pm 0.12$  V, respectively (Figure 2). No  
111 significant difference in the body activity of pregnant and non-pregnant cows was observed  
112 before and after LSD vaccination. However, the body activity of pregnant cows temporarily  
113 increased 1 and 4 days after vaccination compared with that in non-pregnant cows ( $p < 0.001$ ).

114 Additionally, the rate of rise in temperature of  $>40$  °C was measured for 4 hours at 10-minute  
115 intervals for 9 days after LSD vaccination and analyzed by group. The rate at which a  
116 temperature of  $40$  °C was maintained was higher in pregnant cows than it was in non-pregnant  
117 cows. A maximum of 12.8% non-pregnant cows and 20.8% pregnant cows demonstrated  
118 temperatures  $>40$  °C between 5 and 6 days after vaccination (Figure 3).

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## Discussion

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123 There are research reports comparing changes in ruminoreticular temperatures and body activity  
124 according to estrus(19), pregnancy(20), parturition(21), and FMD vaccination(22), which are  
125 behavioral characteristics of cows using bolus sensors. Because the sensors are located in rumen  
126 or reticulum, ruminoreticular temperatures temporarily decreases rapidly due to the effect on  
127 water consumption after ingestion of the feed, and body activity increases than usual because  
128 they are mixed with the feed due to feed intake(19, 20, 23-25). This characteristic is the result of  
129 normal feed intake, so it is also the basis for accurately determining whether cows consume feed.  
130 Governments are encouraging vaccinations to prevent the outbreak of infectious diseases, such  
131 as LSD, Akabane disease, and foot-and-mouth disease (FMD) among animals (6, 15, 22, 26, 27),  
132 considering this is the most efficient way to prevent infection. (6, 15, 26). However, the side  
133 effects of vaccination must be studied, and vaccination methods to minimize these side effects  
134 should be developed (11, 16, 17, 22, 23, 26).

135 Recently, Abutarbush et al. (16) reported that LSD vaccination causes fever, decreased feed  
136 intake, and reduced milk production in dairy cows. Furthermore, Bamouh et al. (11) found that  
137 the rectal temperature increased significantly in the vaccinated group than it did in the non-  
138 vaccinated experimental group. Additionally, the body temperature was found to gradually  
139 increase up to 6 days after LSD vaccination (11), which conforms to the results of this study.  
140 Bamouh et al. (11) also showed that a high vaccine dose caused a rise in temperature to  $\geq 40$   
141 degrees; this finding was similar to that of our study. Nevertheless, analyzing the changes in  
142 temperature depending on the dose of LSD vaccination in cows cannot help elucidate the  
143 changes in body temperature according to pregnancy status.

144 Katsoulos et al. (17) measured rectal temperature using digital thermometers after LSD  
145 vaccination and found that the highest rectal temperature was recorded 8 days after vaccination,  
146 and milk production decreased by up to 16%. However, analyzing milk production status after  
147 parturition also cannot help clarify the changes in body temperature after LSD vaccination  
148 according to pregnancy status.

149 Body temperature is very closely related to physiological mechanisms, and technologies have  
150 been developed to monitor body temperature using non-invasive methods, such as by using bolus  
151 sensors (22, 26). Our research team previously conducted a study to investigate changes in  
152 ruminoreticular temperature and body activity depending on estrus status (19), gestation period  
153 (20), and parturition (21). A study has also been conducted to compare and analyze the

154 ruminoreticular temperature after administering FMD vaccine to cows in early- and late-  
155 pregnancy stages (22).

156 However, to the best of our knowledge, changes in ruminoreticular temperature and activity after  
157 administering LSD vaccine during pregnancy have not been analyzed to date. Therefore, the  
158 contribution of our study, which shows the relative rise in ruminoreticular temperature of  
159 pregnant cows after LSD vaccination when compared with that of non-pregnant cows, is  
160 significant.

161 The current study shows that the rate at which a ruminoreticular temperature of  $>40$  °C was  
162 maintained was higher in pregnant cows than it was in non-pregnant cows after LSD vaccination.

163 Hence, prescribing antipyretic drugs and close monitoring are necessary to prevent miscarriage.

164 While no miscarriage or stillbirth occurred while conducting this experiment, additional large-  
165 scale studies are required to investigate adverse reactions of LSD vaccination.

166 In conclusion, the results of this study can be used as raw data to understand the physiological  
167 changes in ruminoreticular temperature and body activity depending on pregnancy status after  
168 LSD vaccination in Hanwoo. In addition, based on the results of this study, we plan to conduct a  
169 study to investigate cases of miscarriage, premature birth, and stillbirth following LSD  
170 vaccination in the future and develop ways to prevent them.

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179 "Development of techniques to improve the reproductive performance in Korean native cows for  
180 the domestic FMD vaccination."

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289 **Tables and Figures**

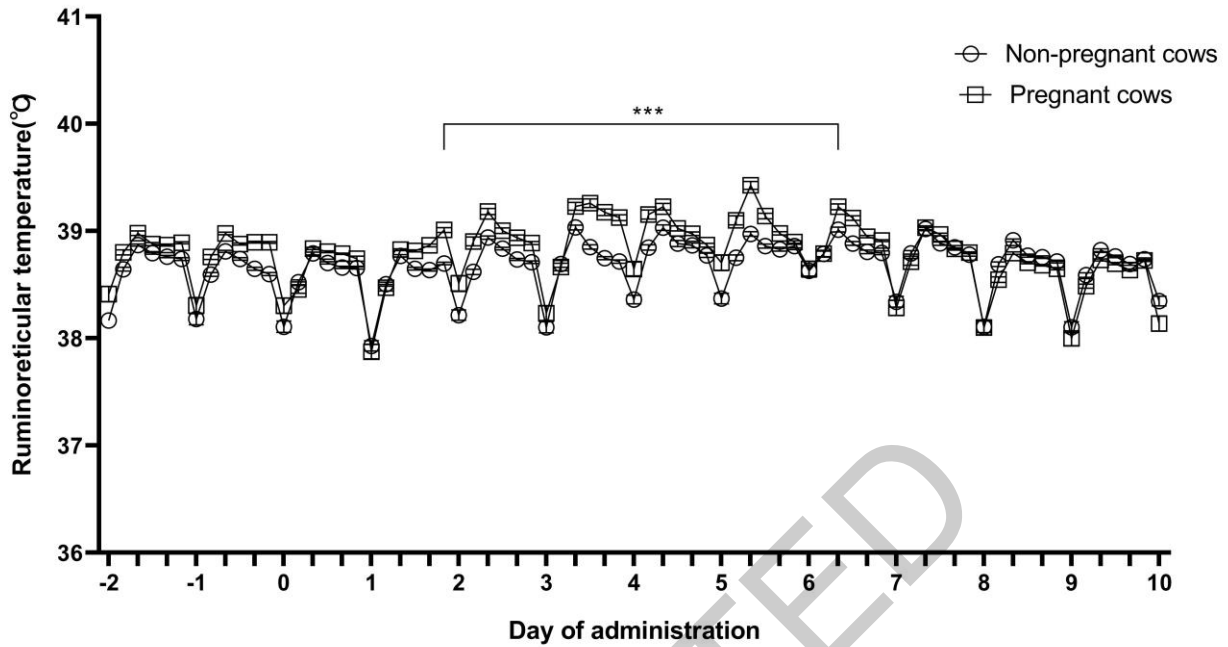
290 Table 1. Information regarding experiment group (n=46)

Group	Number of cows	Age of months	Parity	Days of pregnancy
Non-pregnant cows	28	51.2±4.0	1.8±0.1	
Pregnant cows	18	47.0±3.5	1.6±0.2	173.6±3.7
Total	46	49.7±2.9	1.7±0.1	

291

292

ACCEPTED

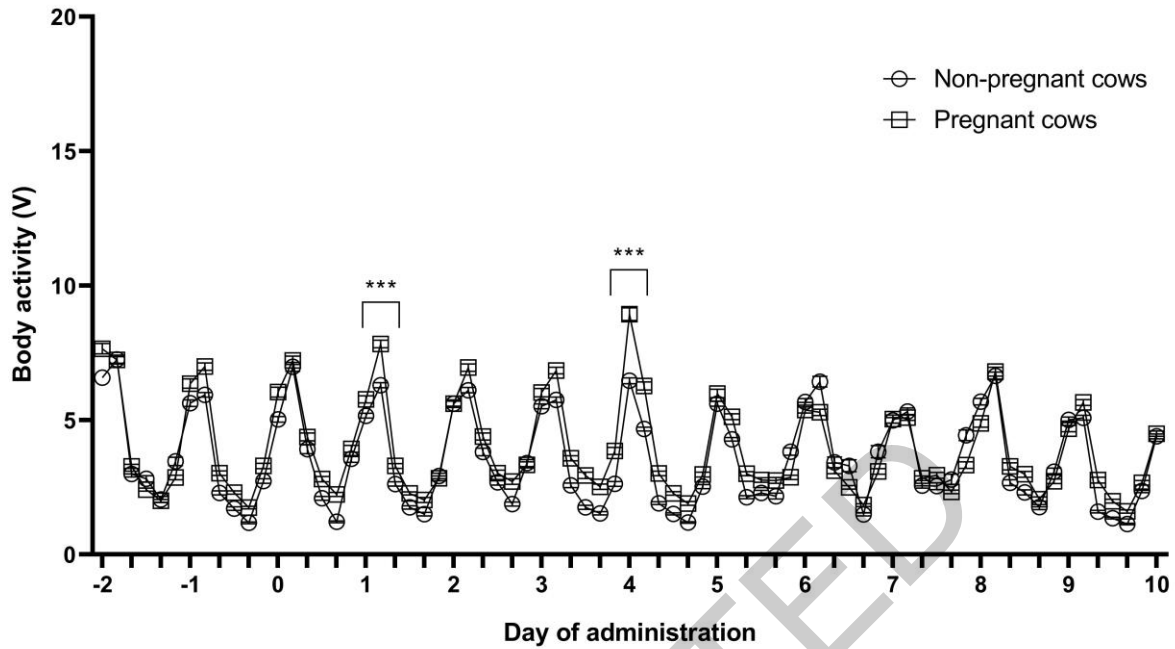


294

295 Figure 1. Changes in ruminoreticular temperature of pregnant and non-pregnant cows depending  
 296 on days after lumpy skin disease vaccination (n=46).  $\square$  represents the mean values for the  
 297 pregnant group and  $\circ$ , the mean values for the non-pregnant group. The day of vaccine  
 298 administration is 0 day, and the error bar is presented as standard error of the mean (SEM).

299 \*\*\*p&lt;0.001

300

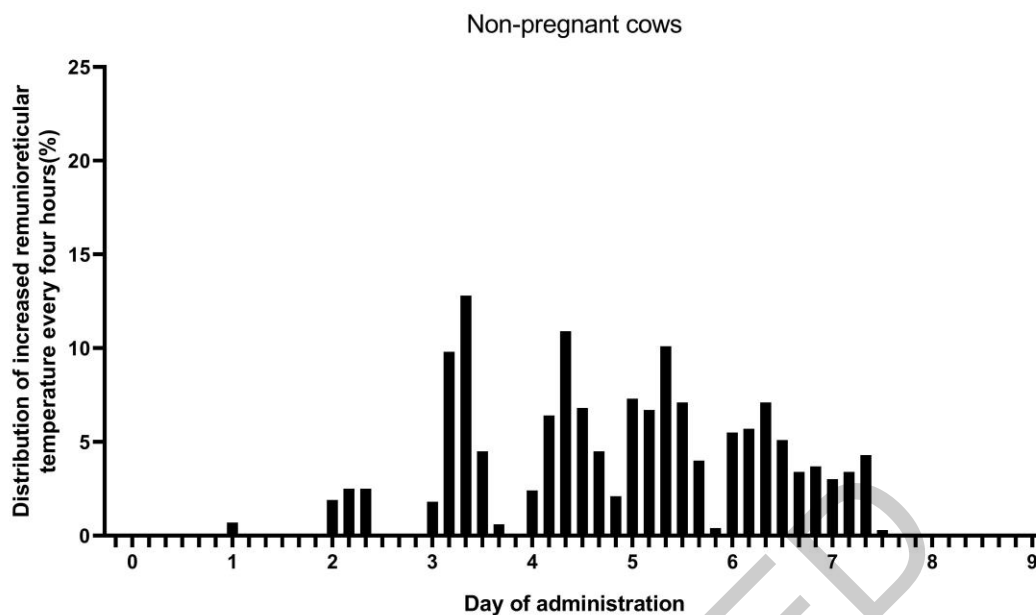
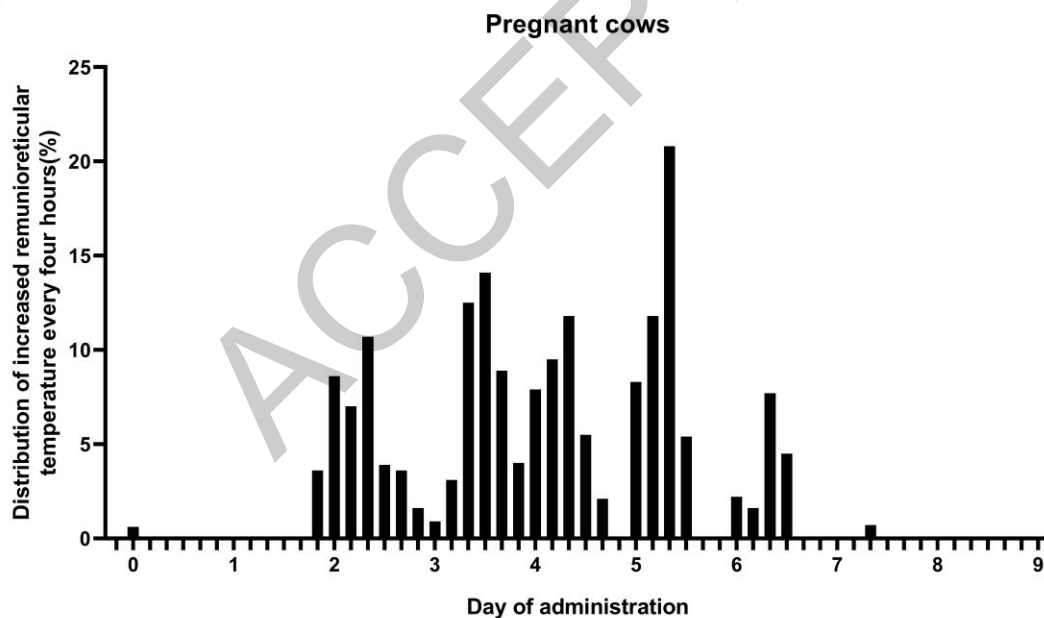


302

303 Figure 2. Changes in body activity in pregnant and non-pregnant cows depending on days after  
 304 lumpy skin disease vaccination (n=46).  $\square$  represents the mean values for the pregnant group and  
 305  $\circ$ , the non-pregnant group. The day of vaccine administration is 0 day, and the error bar is  
 306 presented as standard error of the mean (SEM). \*\*\*p<0.001.

307



**A****B**

308  
309 Figure 3. Distribution of increased ruminoreticular temperature of  $>40^{\circ}\text{C}$  every 4 hours in (A)  
310 non-pregnant and (B) pregnant cows after lumpy skin disease vaccination (n=46). The black bar  
311 represents the percentage of increased ruminoreticular temperature of  $>40^{\circ}\text{C}$  every 4 hours.