Oxygen Uptake during Aerobic Cycling Exercise Simultaneously Combined with Neuromuscular Electrical Stimulation of Antagonists

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Abstract

The purpose of this study was to evaluate oxygen uptake ($\dot{VO}_2$) during aerobic exercise with a Hybrid Training System (HTS) at a moderate intensity. Recently it is said that the combined application of electrical stimulation (ES) and Volitional Contractions (VC) is effective. A Hybrid Training System (HTS) has been developed as a resistance exercise method combining ES with VC, and using electrically stimulated eccentric antagonist muscle contractions as a resistance to voluntary agonist muscle contractions. The benefits of combining aerobic exercise and resistance exercise have also been reported. Therefore, we devised an exercise method that combines the resistance exercise of HTS with aerobic cycling exercise. However, the influence of HTS on aerobic exercise has not been confirmed. Outcome measurements of expired gas were compared during conventional ergometer exercise with and without HTS. 11 healthy young men exercised on a cycle ergometer starting at 20 Watts and increasing by 20 Watts every 3 minutes to 100 Watts, with volitional contractions alone (VER) and with HTS (HER). During each VER and HER test, $\dot{VO}_2$, carbon dioxide output ($\dot{VCO}_2$), expired ventilation ($VE$) and heart rate (HR) was measured. $HR$, $\dot{VO}_2$, $\dot{VCO}_2$, and $VE$ showed a linear relation with workload during both VER and HER. $\dot{VO}_2$ during HER was significantly higher than during VER at an average of about 21.1% (p<0.001). HER at moderate intensity seems to result in a linear relationship between $\dot{VO}_2$ and the work rate in the same way as conventional aerobic exercise. Furthermore HER resulted in stronger exercise intensity than VER with the same workload. HTS may be a novel exercise technique that could combine resistance exercise with aerobic exercise.

Keywords: Analysis of expired gas; Ergometer; Exercise intensity; Metabolic cost.

Abbreviations: ES-Electrical Stimulation; HR-Heart Rate; HTS-Hybrid Training System; NMES-Neuromuscular Electrical Stimulation; VC-Volitional Contraction; $\dot{VCO}_2$-Carbon dioxide Output; $VE$-Expired Ventilation; $\dot{VO}_2$-Oxygen Uptake; VT-Ventilatory Threshold

Introduction

Recently it is said that the combined application of Electrical Stimulation (ES) and Volitional Contractions (VC) is more effective for muscle strengthening and muscle hypertrophy than ES or VC alone [1-2]. A Hybrid Training System (HTS) that creates resistance to the motion of a voluntarily contracting agonist muscle by means of the force generated by its electrically stimulated antagonist has been developed [3] (Figure 1). HTS can train both voluntarily contracting agonist muscles and electrically stimulated antagonist muscles simultaneously, and is one of the methods that serve to make up for the limitations of Neuromuscular Electrical Stimulation (NMES) [4]. Resistance training using HTS has been shown to improve muscle hypertrophy and strength while using relatively low ES intensity compared with ES alone and/or conventional weight training [3-6]. In the upper extremity, elbow flexion isometric torque had increased about 56% and the biceps muscle had enlarged by about 14% using HTS over an 8-week period, and this was significantly greater than or similar to isotonic weight training and NMES [4]. In the lower extremity, the knee extension isokinetic torque at 30°/sec had increased about 28% using HTS over a 6-week period, and it was comparable to weight training with 15 repetition maximum loads [5]. In these studies, HTS has been used as exercise resistance in joint bending exercise as a substitute for weights. However, HTS has not been previously used in combination with other exercise to add further resistance (e.g. dumbbell exercise or cycling exercise). Theoretically, HTS can be used for exercise in every agonist-antagonist group of muscles [7]. We devised a new exercise technique that combines HTS

Figure 1: Schematic model of hybrid training system
Note that both the voluntarily activated agonist and the electrically stimulated antagonist contract during joint motion. The result is that both muscles are trained and a longitudinal compressive load is placed on the bone.
with cycling exercise based on the concept that aerobic exercise and electrically eccentric exercise are possible simultaneously. However, the influences that HTS has on aerobic exercise are not known.

Cycling exercise is widely used as an aerobic exercise method to improve exercise capacity or physical fitness [8-11]. In general, its effect on muscle strengthening has been not demonstrated [12,13], although there are some reports that cycling exercise improved muscle strength of the lower extremity [14-16]. On the other hand, eccentric muscle contractions combined with cycling exercise have been studied [17-19]. Because eccentric muscle contractions generate 30% to 50% more muscle force than concentric muscle contractions, eccentric exercise is more effective for muscle strengthening than concentric exercise [20]. Furthermore, in eccentric exercise, a large force is produced with very little energy demand [21,22]. Lastayo et al. reported that 8 weeks of eccentric cycling exercise produced a 36% isometric leg strength improvement and 52% fiber area increase while training at exercise intensities that did not promote strength or size increases concentrically in healthy young subjects [18]. Although this eccentric cycling exercise is passive exercise, HTS has not only passive (electrically stimulated) eccentric exercise but also active voluntary concentric exercise. Electrically stimulated eccentric antagonist contractions can increase muscle strength and activity of not only the antagonist but also the agonist [7]. Therefore, it is inferred that metabolic cost can increase by combining electrically stimulated eccentric contractions with voluntary concentric contractions.

For exercise prescription, the percentage of maximal oxygen uptake (VO2) is commonly used as one of the targets of exercise intensity. Therefore, in order to calculate an appropriate exercise prescription it is necessary to know VO2 during the exercise with HTS. We evaluated VO2 during HTS with a moderate cycle ergometer (HER) by comparing it with VO2 during a volitional moderate cycle ergometer alone (VER).

Methods
Setting and Participants
The Ethics Committee of Kurume University and the Japan Aerospace Exploration Agency approved the clinical design of this study protocol. The study was designed in accordance with the ethical standards of the Helsinki Declaration of 1975 and received the approval of the Ethics Committee of Kurume University and the Japan Aerospace Exploration Agency. All procedures were fully explained to the participants who gave their written informed consent to participate. 11 healthy young men [mean (standard deviation (SD)) age 21 (1.3) yrs; height 173.3 (6.5) cm; and weight 67.7 (8.1) kg] agreed to participate. The participants were examined after giving consent, by an orthopedic specialist who was not involved in this study. They were excluded if they clearly deviated from the inclusion criteria. This included the requirements that they had no adverse medical history, and an examination for normal physical fitness, strength, sensation [23], and range of motion according to the criteria of the Japanese Orthopedic Association [24]. They also had not participated in any regular or on-going sports activities although occasional sports were allowed. Randomization was performed using a computer-generated randomized sequence of exercise order created before the beginning of this investigation. Each participant was tested during either VER or HER (randomly selected), and was tested for the other on same day an hour later.

Intervention
All participants were measured for height and body weight, and performed the ramp exercise test to determination their peak oxygen consumption (VO2peak) and two cycle ergometer exercise tests. The first exercise test consisted of a ramp protocol performed on an electronically braked cycle ergometer until exhaustion levels were reached. On the second exercise test, which was carried out on another day, participants were measured for gas exchange during cycle ergometer exercise according to the following protocol. They pedaled with their feet strapped into the pedals during all exercise tests.

Ramp exercise test protocol
After a two minute rest sitting on the cycle ergometer (STB-2400, Nihon Kohden, Tokyo, Japan), the test started at 20 W and the workload was increased by 20 or 30 W/min according to the physiological profile of each subject [25]. Pedaling cadence was kept constant at 60 to 80 rev/min using a pedal frequency meter depending on the subject’s preference. The ramp rate was determined so that the test duration was 8-12 min. The exercise test was terminated when the pedal cadence could not be maintained at 60 rev/min, and VO2peak was determined. Verbal encouragement was given during the ramp exercise test. The ventilatory threshold (VT) was determined as power output (W) corresponding to the first breakpoint in carbon dioxide production (VCO2) with respect to VO2, (the so called V-slope method) [26].

VER test protocol
The VER test was performed to obtain an analysis of expired gas. After a 2 minutes rest sitting on the cycle ergometer, the test started at 20 W and the workload was increased by 20 W to 100 W every 3 minutes. Pedaling cadence was kept constant at 60 rev/min with the aid of a pedal frequency meter and a metronome, which is not thought to affect blood pressure or heart rate [27]. To prevent muscle fatigue, exercise test time was limited to 15 minutes.

HER protocol
The HER test was performed to obtain an analysis of expired gas. After a 2 minute rest sitting on the cycle ergometer, the test started at 20 W and the workload was increased by 20 W to 100 W every 3 minutes. For the HER test, HTS was performed simultaneously during a volitional moderate cycle ergometer with the participant’s hamstrings electrically stimulated as he volitionally extended his knee, and his quadriceps electrically stimulated as he volitionally flexed his knee to provide motion resistance (Figure 2). The pedaling cadence was kept.

Figure 2: Protocol of electrical stimulation during cycling exercise with hybrid training system.

The position of the left pedal was the highest at 0 degrees. The position of the saddle was inclined backward to 10 degrees from vertical. Subject sat on a cycle ergometer, with his hamstrings electrically stimulated as he volitionally extended his knee and his quadriceps electrically stimulated as he volitionally flexed his knee. The timing of the electrical stimulation was controlled by a joint motion sensor attached to the knee.
constant at 60 rev/min. During HER both lower extremities were stimulated in synchronization to the bending motions of the knee by using HTS. The joint range of motion was set at nearly 90° arc that extended from 20° to 110° (0° indicating full knee extension) by adjusting the height of the saddle.

**Electrical stimulation protocol**

The ES device has been described previously [3,4] and consists of a custom designed waveform generator capable of delivering stimulating signals with unique frequencies and waveforms to as many as 8 pairs of electrodes and a joint motion sensor (Mutoh Engineering Inc., Tokyo, Japan) that triggers stimulation of the antagonist once it senses the initiation of an agonist's VC [28]. Pairs of 3×6-cm low impedance gel-coated silver fiber electrodes (Nihon Medix Co, 315-1, Mukai-machi, Minami-hanashima, Matsudo-shi, Chiba-ken, Japan.) were placed over each motor point on the quadriceps and the hamstrings and a detector was also attached (Top Co, 19-10, Senju-nakai-machi, Adachi-ku, Tokyo, Japan).

**Stimulation Parameters**

The stimulation waveform used in this study is similar in some ways to that of “Russian stimulation” [29] and consists of a 5,000 Hz carrier frequency modulated at 40 Hz (2.4 ms on, 22.6 ms off) to deliver a rectangular biphasic pulse [3]. The electrical stimulator gives constant voltage stimulus to the human body (regulated voltage). It has a stimulus pattern with interlock and a limiter for safety. Therefore, the effective current is interlocked at 20 mA, and the peak voltage and current is limited to under 72V and 90 mA. Stimulation intensities were determined one week before the evaluation session began. We regulated stimulation intensity so that the exercise intensities were adjusted to 80% of the maximum comfortable intensity in order to successfully improve muscle strength and mass without causing pain [4,6]. The mean stimulating voltages were 32.4 (SD=9.7) V and 30.6 (SD=7.8) V for the quadriceps femoris and hamstring muscles, respectively. At these ES intensities, all subjects were able to pedal during HER.

**Measuring Methods**

Environmental conditions were similar for all exercise tests (21 to 24 degrees centigrade, 45 to 55% relative humidity). During the exercise tests, gas exchange data was collected continuously using an automated breath by breath system (AE-300S; Minato Medical Science Co. Ltd., Osaka, Japan) implementing the standard technique. The AE-300S consists of a microcomputer, a hot wire flowmeter, and a gas analyzer, which contains a sampling tube, filter, suction pump, O₂ analyzer made by a paramagnetic oxygen transducer and an infrared CO₂ analyzer. Ventilatory parameters were measured using a hot-wire flow meter, and the flow meter was calibrated with a syringe of known volume (2.0 l). A zirconium sensor and an infrared absorption analyzer, respectively, measured O₂ and CO₂ concentrations. The gas analyzer was calibrated to known standard gas levels (O₂ 15.16%, CO₂ 0.023%) before each test. Also heart rates (HR, beats/min) were continuously monitored by electrocardiogram during the tests. VO₂, VCO₂, and pulmonary ventilation (VE) were calculated and recorded during the cycle ergometer exercise tests. The values of VO₂ (ml/kg/min), VCO₂ (L/min), VE (L/min) and HR were averaged during the last 1 minute of the steady rate portion of each workload (3 min, 6 min, 9 min, 12 min, and 15 min after exercise start) and used for data analysis.

**Data Analysis**

All variables are presented as means (SD). The linear relationship between each workload (20W, 40W, 60W, 80W, 100W) and VO₂ (ml/kg/min), VCO₂ (L/min), (VE) (L/min) and HR (beats) was checked graphically. For each of the comparisons which satisfied linearity, the linear relationship equation was estimated by the mixed effect model. We judged whether a slope for VER and one for HER were the same by interaction (group×workload) terms. If p for the interaction term was larger than 0.25, we judged their slopes were common and conducted the mixed effect model without the interaction term again.

All the statistical analyses were performed using SAS Version 9.3 statistical software (SAS Institute Inc., Cary, NC, USA) and p values ≤ 0.05 were considered to be statistically significant.

**Results**

Both height and weight of participants were average for Japanese height 173.3 (6.5) cm; and weight 67.7 (8.1) kg. The national average for 20-24 yr males Japanese is height 172.2 (5.7) cm; and weight 65.5 (8.7) kg [23]. All participants completed all of the exercise tests without any problems such as not being able to continue pedaling the ergometer from pain or fatigue, although six of eleven participants complained of slight post exercise delayed-onset muscle soreness on the day after the tests.

VT and VO₂peak were 20.8 (2.9) ml·kg⁻¹·min⁻¹ and 41.0 (4.2) ml·min⁻¹, respectively. These values were standard for Japanese adults [23]. The ratio for VO₂peak of VT was 51.0 (8.0)%. At 100W during VER, 3 participants were higher than VT, also 5 participants at 100W during HER were higher than VT. All participants pedaling at a workload between 20W and 80W during VER and HER were pedaling below their individual VT.

The graph of VO₂ during both VER and HER showed the linear relationship to the workload (Figure 3A, 3B, and Figure 4A). Furthermore, for VCO₂ VE and HR, a linear relationship to the workload was observed similar to VO₂ (Figure 4B, 4C, 4D). The interaction terms (group×workload) in the mixed effect model in VO₂, VCO₂, VE and HR were larger than 0.25. Therefore, we estimated the linear relationship equations by the mixed effect model without the interaction term. The results were summarized in Table 1. The common slope of VO₂ was 0.14 (0.02). Each intercept of VER was 21.1 (0.17) (Figure 5A). VO₂ During HER was significantly higher by 2.1 (0.17) ml/kg/min than VER at moderate intensities (20W-100W) (p<0.001) (Figure 5A). This increase was about 20% of VER. The common slopes of VCO₂, VE and HR were 9.1 (1.12), 0.23 (0.03), and 0.39 (0.05), respectively. Each intercept of VER of VCO₂, VE, and HR was 260.10 (44.25), 10.31 (1.35), 85.8 (2.84) (Figures 5B, 5C, 5D). VCO₂, VE, and HR during HER was significantly higher than VER 93.33 (12.16) ml/min, 2.97 (0.41) l/min, and 5.07 (1.06) beats (all, p<0.001) (Figure 5B, 5C, 5D).

**Discussion**

This is the first report of HTS combined with aerobic exercise. The findings of this study showed that HTS, utilizing electrically stimulated eccentric antagonist muscle contractions, increased VO₂ about 2.1 (0.17) ml/kg/min from aerobic cycle ergometer exercise at moderate-intensity. Furthermore, similar to conventional aerobic exercise, VO₂, VCO₂, VE and HR during HER had a linear relationship to workload. Therefore HER resulted in stronger exercise intensity than VER at the same workload.

Generally, aerobic exercise improves exercise capacity and/or physical fitness. On the other hand, resistance exercise mainly improves muscle strength and/or mass. Therefore, in conformity with the training purpose or individual ability, an exercise prescription...
In elderly women, Ferketich et al. reported that to add -Oxygen consumption; On the other hand, in HER (eccentric Carbon dioxide production; and HR. at moderate exercise intensity. Therefore, HER seems to be by -Oxygen consumption.

were very similar [33]. during HER showed a linear relationship -Expired muscle strength and endurance as well as exercise capacity (peak the combined aerobic and resistance training were effective to improve benefits of combining aerobic exercise and resistance exercise have been reported [30-32]. For heart failure, Mandic et al. reported that eccentric cycling exercise was to consider higher exercise HR to reach a comparable level of VO₂peak at moderate exercise intensity. Therefore, HER seems to be an exercise technique which can add electrical resistance exercise to aerobic cycling exercise simultaneously. Mackenzie et al. reported that electrically stimulated eccentric antagonist contractions could increase muscle strength and activity of agonist muscles, and provided resistance of motion for the agonist [7]. In this study, the resistance newly added by HTS to VER seemed to increase VO₂.

Furthermore, because VO₂ during HER showed a linear relationship with workload, an adjustment of the exercise intensity in conformity with the training target would be possible by adjusting the workload in the same way as conventional aerobic exercise. Moreover, it was similar in HR too. The metabolic cost of HER may be able to be predicted easily from HR similar to conventional exercise. In eccentric cycling exercise, the relationships between HR and VO₂ were very similar [33]. Therefore eccentric cycling training is currently based on exercise HR [18,19, 34]. However, HR was higher during eccentric cycling exercise than concentric cycling exercise even though the metabolic demand was similar [35]. Dufour et al. showed that an important application for eccentric cycling exercise was to consider higher exercise HR to reach a comparable level of VO₂ [17]. On the other hand, in HER (eccentric cycling combined with VC) the relationships not only between VO₂ and workload but also between HR and workload were almost parallel unlike eccentric cycling exercise (without VC). Therefore HER training based on exercise HR seems to be possible. NMES activation is considered nonselective with regard to the

is chosen from aerobic exercise or resistance exercise. Recently the benefits of combining aerobic exercise and resistance exercise have been reported [30-32]. For heart failure, Mandic et al. reported that the combined aerobic and resistance training were effective to improve muscle strength and endurance as well as exercise capacity (peak

### Table 1: Estimated slope and intercept of \( VO_2, VCO_2, VE \) and HR.

<table>
<thead>
<tr>
<th></th>
<th>Estimate</th>
<th>Standard error</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>( VO_2 ) (ml/kg/min) ( \beta_1 = 0.14 )</td>
<td>0.02</td>
<td>&lt;.0001</td>
<td></td>
</tr>
<tr>
<td>( VO_2 ) (ml/kg/min) ( \beta_2 = 2.09 )</td>
<td>0.17</td>
<td>&lt;.0001</td>
<td></td>
</tr>
<tr>
<td>Intercept for VER ( \beta_0 = 4.56 )</td>
<td>0.50</td>
<td>&lt;.0001</td>
<td></td>
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<tr>
<td>( VCO_2 ) (L/min) ( \beta_1 = 9.09 )</td>
<td>1.12</td>
<td>&lt;.0001</td>
<td></td>
</tr>
<tr>
<td>( VCO_2 ) (L/min) ( \beta_2 = 93.33 )</td>
<td>12.16</td>
<td>&lt;.0001</td>
<td></td>
</tr>
<tr>
<td>Intercept for VER ( \beta_0 = 280.10 )</td>
<td>44.25</td>
<td>&lt;.0001</td>
<td></td>
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<tr>
<td>( VE ) (L/min) ( \beta_1 = 0.23 )</td>
<td>0.03</td>
<td>&lt;.0001</td>
<td></td>
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<tr>
<td>( VE ) (L/min) ( \beta_2 = 2.97 )</td>
<td>0.41</td>
<td>&lt;.0001</td>
<td></td>
</tr>
<tr>
<td>Intercept for VER ( \beta_0 = 10.31 )</td>
<td>1.35</td>
<td>&lt;.0001</td>
<td></td>
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<tr>
<td>HR (beats) ( \beta_1 = 0.38 )</td>
<td>0.05</td>
<td>&lt;.0001</td>
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<tr>
<td>HR (beats) ( \beta_2 = 5.07 )</td>
<td>1.06</td>
<td>&lt;.0001</td>
<td></td>
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<tr>
<td>Intercept for VER ( \beta_0 = 85.82 )</td>
<td>2.84</td>
<td>&lt;.0001</td>
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</table>

\( \text{VER; } y=\beta_0+\beta_1\cdot \text{workload}+\beta_2\cdot \text{workload}+\beta_0 \cdot \text{Intercept for VER.} \)

\( \text{HER; } y=\beta_0+\beta_1\cdot \text{workload}+\beta_2\cdot \text{workload}+\beta_0 \cdot \text{Intercept for VER.} \)

\( VO_2 \) -Oxygen consumption; \( VCO_2 \) -Carbon dioxide production; \( VE \) -Expired ventilation; HR-Heart rate; VER-Cycle ergometer exercise; HER-Cycle ergometer exercise combined with HTS.
type of motor unit and synchrony, and it preferentially activates Type II fibers as compared with voluntary muscle contractions [1,36]. Therefore, NMES is not commonly used for endurance training, but is used for resistance training. HTS is a type of NMES, and is resistance exercise using electrically stimulated eccentric contractions [3,4]. The presence of an eccentric component may also be beneficial in that these contractions are neuromuscularly more efficient [37], less metabolically demanding [21,34-35], and more conducive to hypertrophy than concentric contractions [18,38]. Indeed in previous studies, HTS had successfully improved muscle strength and muscle mass in healthy men when used for bending exercises of the elbow or knee [3-5]. It is known that a given level of stimulation will produce an eccentric contraction that is 20-30% stronger than its isometric counterpart [39]. Therefore, an eccentric contraction of HTS is thought to improve muscle strength and mass as effective resistance exercise [3-5]. LaStayo et al. reported that eccentric cycling training at low exercise intensity (54-65% of peak heart rate) without VC could increase the size and strength of muscles even though a classic cycle ergometer could not [18]. HER is utilizing eccentric cycling training, too. Therefore HER would have the potential of providing resistance exercise, and muscular strength and mass augmentation could be expected. HER at the same pedaling intensity (workload) as VER would increase not only the perceived exercise intensity but also the metabolic cost. Moreover increasing the metabolic rate using HER would lead to increased caloric expenditure during exercise. HER may be available where exercise opportunity and/or equipment is limited such as microgravity conditions or extended bed rest.

**Study Limitations**

This study had some limitations. There were the limitations of the small number participants and the fact that all of them were young men. A long-term training study is necessary to show that HER is an effective technique combining aerobic and resistance exercise by evaluating exercise capacity, physical fitness, muscle strength, muscle mass, and so on. This study showed the influences of HER on aerobic cycling exercise, and it is the pilot study that showed HER serves as aerobic exercise with simultaneous electrical resistance exercise. Furthermore, a future training study was enabled by discovering the exercise intensity of HER through this study.

**Conclusions**

In conclusion, the finding of this study is that the combined application of aerobic cycling exercise and resistance through an electrically stimulated antagonist (cycle ergometer with HTS) could result in increased \( \text{VO}_2 \) with a linear relationship to workload in comparison to aerobic cycling exercise alone. Furthermore HER resulted in stronger exercise intensity than VER with the same workload. HER may be a new exercise technique in which aerobic exercise and resistance exercise are combined.
Acknowledgments

Author Contributions

All authors contributed to writing. Dr. Ohmoto, Dr. Matsuse and Dr. Shiba (including development of Hybrid exercise system) provided the concept/project design. Dr. Takano measured and calculated the data, and Dr. Yamada and Dr. Ohshima provided consultation.

Financial Disclosures

The authors have declared that no competing interests exist.

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Institutional Review

The study was designed in accordance with the ethical standards of the Helsinki Declaration of 1975 and received the approval of the Ethics Committee of Kurume University and the Japan Aerospace Exploration Agency.

All procedures were fully explained to the participants who gave their written informed consent to participate.

Participant follow-up

The authors do not plan to inform the participants of the publication of this study because of a lack of contact information.

References


