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Short communication

Electrostatic measurement of dischargeable electricity and bioelectric potentials produced by muscular movements in flies

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A B S T R A C T

A simple electrostatic apparatus was devised to measure dischargeable electricity and bioelectric potentials produced by flies. The apparatus involved two insulated electrodes, ICW(−) and ICW(+), oppositely charged with equal voltages supplied by two voltage-generators. In the electric field, the flies became net positive by instantaneously discharging their electricity and were attracted to negative surface charges on ICW(−). The tail-lifting movement by the attracted insect was an action creating electric potentials that could cause discharge of ICW(−). The discharge transiently appeared in response to individual movements and was larger when the tail was lifted at higher angles.

1. Introduction

The bioelectric characteristics of insects are a major focus in developing an electrostatic pest trapping technique for crop protection [1]. Of particular interest is to demonstrate their ability to generate an electric field to dispel the electrostatic attraction force of a trap. The cuticle layer covering the insect body is interesting because of its highly conductive nature [2–6]. Under the influence of an electric field, conductors are electrified as a result of uneven distribution of electricity (free electrons). This implies that insects experience an electrification of their surface cuticle layer in an electric field and that they would be forced to generate an electric potential inside their bodies to oppose the external electric potential of an electric field [7]. Our original idea was to devise a new method to evaluate the potential electric production by insects.

The main focus of this study is to compare insects to a biological voltage generator and to specify a power source for electric power generation. Some insects generate bioelectricity through muscular movement [4,5,8–10] and/or neural excitation [11]. Also in our previous work [12], we reported electric current-associated muscular movements by vinegar flies in an electric field; insect movements were clear enough to track the electric power generation linked to individual movements.

The electrostatic attraction force is safe for insects, and therefore is available for holding test insects on a probe of an electric current detector without causing any harm. Stable holding of an insect is essential to consecutively analyse a series of muscular movements and their corresponding current flows. We attempted to deprive test insects of the electricity in their cuticle layer in an electric field because electrification of the surface layer is thought to be harmless to the insect. This method is easier and safer than conventional microsurgical operations by which microelectrodes are inserted into muscular or nervous tissues [4,13]. Electrified insects became net positive and could be attracted to the cathodic pole used to form the electric field [1]. The necessary equipment for this experiment is simple. Basically, only three components (insulated wires for electrodes, voltage generators and current detectors) are needed.

Using this method, electrostatic measurements are implemented at pre- and post-attraction stages. The pre-attraction stage determines the amount of dischargeable electricity from an insect, when the insect electricity instantaneously discharged by the

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mechanically charged voltages (external voltages) is measured. The subsequent post-attraction stage determines the electric potentials generated by insect muscular movements, when the electrode-accumulated electricity mobilised by biologically generated voltages (internal voltages) is measured. Dynamic analysis of the video-recorded data of the attracted insects visualises electric current generation associated with individual muscular movements. We used three kinds of flies belonging to different families to comparatively analyse muscular movement-mediated electric power generation.

2. Materials and methods

2.1. Experimental equipment

Electrodes were constructed using two insulated conductor wires (ICWs; Fig. 1A). An iron wire (2 mm diameter, 2 cm length) was passed through a vinyl sleeve (1 mm thick; bulk resistivity, $1 \times 10^9\ \Omega$) to make an ICW. Two ICWs were paralleled at a 5-mm interval and linked to negative and positive direct current (DC) voltage generators (Max Electronics, Tokyo, Japan). The opposite ends of the ICWs were closed by inserting an insulator polypropylene rod (2 mm diameter, 5 mm length) into the sleeve. Both generators were linked to grounded lines, and a galvanometer PC7000 (Sanwa Electric Instrument, Tokyo, Japan) was integrated into each line. The generators were operated with 12 V storage batteries to supply equal negative and positive voltages to ICWs; negatively and positively charged ICWs are represented as ICW(−) and ICW(+), respectively. Cover sleeves were dielectrically polarised positively on the surface of the iron wire side and negatively on the outer surface of the insulator sleeve in ICW(−) and vice versa in ICW(+) (Fig. 1B) [14]. Opposite surface charges on the ICWs act as dipoles that form an electric field between them.

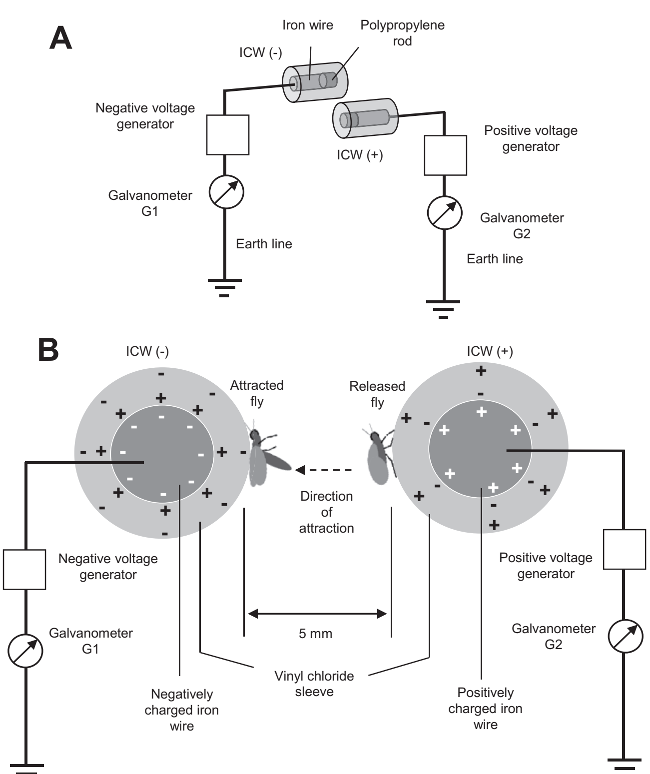

Fig. 1. Diagram (A) and cross section (B) of an electrostatic apparatus to measure dischargeable electricity and bioelectric potentials produced by muscular movements in flies. Two insulated iron conductor wires (ICWs) were oppositely charged with two DC voltage generators, and the direction and electric current magnitude were measured with galvanometers integrated into the grounded lines of the voltage generators.

2.2. Test flies

Three flies from different genera, humpbacked fly (*Megaselia spiracularis*, Schmitz: Phoridae), vinegar fly (*Drosophila melanogaster*, Meigen: Drosophilidae) and, greenhouse shore fly (*Scatella stagnalis*, Fallen: Ephydridae) were used as test insects. Pupae of test flies were purchased from Sumika Technoservice (Hyogo, Japan) and incubated for eclosion in a growth chamber (25.0 ± 0.5 °C, 12-h photoperiod of 4000 lux). The flies were reared following the method of Matsuda et al. [1], and newly emerged adults, 15–24 h after eclosion, were used as active flies for experiments. To collect insects, we constructed an insect aspirator consisting of a polypropylene tube (10 mm diameter) with a pointed tip (1 mm tip diameter). The opposite open end of the tube was linked to an aspirator (aspiration pressure 1.2 kg/cm^2). The insect was attracted to the pointed tip and released at a particular site on the ICW(+) (Fig. 1B) by stopping aspiration. All collected flies walked and flew normally and appeared to be unhurt by the collection. Body sizes of flies (length from head to wing edge) were measured using 30 adult test insects collected randomly: 4.02 ± 0.16, 3.68 ± 0.15 and 3.53 ± 0.26 mm for humpbacked fly, vinegar fly, and greenhouse shore fly, respectively.

2.3. Measurement of electric currents

2.3.1. Mechanical discharge

Both ICWs were oppositely charged with 1.0–9.0 kV to determine the range of voltages that cause mechanical discharge (constant transfer of electricity between both electrodes). In this electricity transfer, the direction and magnitude of electric current were measured with G1 and G2 galvanometers.

2.3.2. Insect discharge

Both ICWs were symmetrically charged, and adult flies were singly released at a particular site on ICW(+) to measure insect discharge (instantaneous transfer of electricity from an insect to ground) before the insect was attracted to the ICW(−). The current direction and magnitudes were detectable with a G2 galvanometer. Twenty adults were used per voltage and insect species.

2.3.3. Electric potential produced by insect movements

Both ICWs were charged with voltages causing no mechanical discharge. Electric currents linked to muscular movements by the attracted insect on ICW(−) were measured with G1 and G2. The profiles of the electric currents were recorded with a current detector (detection limit, 0.01 μA) integrated into the galvanometer (G2). The electric potential (voltage; muscular movement-derived electric power to mobilise electricity) was estimated based on a voltage–current calibration measure presented in this study. Movements by the attracted flies were video-recorded with a digital EOS camera (Canon, Tokyo, Japan) equipped with a dissecting microscope while applying voltages. Elevation angles of the tip lifted by the attracted insect were measured from video pictures, and the current magnitudes corresponding to individual tail-lifting movements were recorded. Movements were observed continuously (for 1 min) until the flies were removed from the ICW(−) after voltage to the ICW was stopped. All experiments were conducted at 25 ± 2 °C and 60 ± 3% relative humidity.