



## Article

# Impact of Electrocutation on Shoot and Tuber Vitality of Yellow Nutsedge (*Cyperus esculentus*)

Jeroen Feys <sup>1,\*</sup>, Benny De Cauwer <sup>1</sup>, Dirk Reheul <sup>1</sup>, Clara Sciffer <sup>2</sup>, Shana Clercx <sup>3</sup> and Sander Palmans <sup>3</sup>

<sup>1</sup> Department of Plants and Crops, Faculty of Bioscience Engineering, Weed Science Unit, Ghent University, 9000 Gent, Belgium

<sup>2</sup> Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), 9820 Merelbeke, Belgium

<sup>3</sup> Educational Research Center (PVL), 3950 Bocholt, Belgium

\* Correspondence: jdfey.s.feys@ugent.be

**Abstract:** *Cyperus esculentus* is an invasive perennial sedge that can cause huge losses in arable crops. Current control strategies are based on combinations of cultural, mechanical, and chemical measures, repeated over years. Recent commercial releases of safe innovative electric weeders, offer promising alternative opportunities for controlling perennial weeds with high energy/high frequency electricity. To evaluate the effect of a single electrocutation application on the efficacy of *C. esculentus* control, field experiments were performed in two locations in Belgium. Two electric weeding devices were evaluated: Zasso XP300, delivering a high-frequency, phased direct current (maximum voltage of 7000 V and maximum power output of 2000 W per square meter of green biomass, driving speeds between 1.1 and 3.0 km·h<sup>-1</sup>), and Rootwave Pro, delivering high-frequency alternating current (maximum voltage of 5000 V and power output between 7.5 and 10 kVA, treatment duration of 2 s). The impact of various technical (driving speed and voltage), biotic (clone and growth stage), and abiotic parameters on electrocutation efficacy was evaluated. Plant responses to electrocutation were evaluated by examining the vitality of treated *C. esculentus* mother tubers and shoots. Both devices were ineffective at mother tuber control, regardless of their burial depth (−5 cm to −15 cm), but were highly effective against aboveground shoots with reductions of vitality of up to 91% and 100% after a single pass with Zasso XP300 and Rootwave Pro, respectively. Maximum reductions were obtained when electricity was delivered at low speed (1.1 to 1.5 km·h<sup>-1</sup>) and on 5-leaf shoots without heat or water stress. Remarkably, the lowest efficacies were found on water-stressed soils at the time of application. Voltage had no effect on the degree of *C. esculentus* control. The efficacy of electricity was not affected by clone, irrespective of electric weeding device. Electrocutation is a useful and effective control method within any integrated control strategy for controlling emerged shoots. However, as *C. esculentus* mother tubers are not affected by a single treatment, season-long repeated treatments are needed to exhaust the mother tubers.

**Keywords:** mother tuber; prevention; thermal weeding; electrophysical weeding; voltage; driving speed; leaf stage; soil moisture content



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## 1. Introduction

*Cyperus esculentus* L. (yellow nutsedge) is a perennial weed which is very prolific and hard to control in many crops. It is reported as the sixteenth worst weed in the world [1] and can lead to huge losses in arable crops (e.g., 60% in sugar beets [*Beta vulgaris* L.] and 40% in potatoes [*Solanum tuberosum* L.]) [2]. In leek [*Allium porrum* L.], onions [*Allium cepa* L.], and Brussels sprouts [*Brassica oleracea* var. *gemmifera* (de Candolle) Zenker], high infestation levels (80–100% of the field covered with *C. esculentus*) may cause losses of 86, 90, and 93%, respectively [3]. Reproduction occurs mainly via axillary buds on tubers and basal bulbs, both of which are formed underground at the tips of rhizomes [4]. In a temperate climate, one mother tuber is able to generate 700 or more daughter tubers during one growing

season [5]. Moreover, shoot formation (shoots arise from basal bulbs) is very intensive. De Cauwer et al. [6] planted mother tubers of genetically different clones in a 10 L pot (1 mother tuber per pot). At the end of the growing season, 29 to 91 shoots (depending on the clone) were counted. In Western European conditions, mother tubers start to germinate during April and initiate a process of intensive shoot formation. Formation of daughter tubers starts in May or June and continues until autumn. Tubers can survive low temperatures (up to  $-10\text{ }^{\circ}\text{C}$  on soil surface) [5] and the most persistent may remain germinative for 10 years [7]. *C. esculentus* also produces seeds, but these rarely germinate in the field [8,9].

In Belgium, over 50,000 hectares of cropland is infected with *C. esculentus* (Feys, pers. comm.). Successful control of *C. esculentus* tuber banks in cropped areas requires years of intensive control. As a consequence of the insufficient efficacy of single control measures against *C. esculentus*, integrated weed management systems combining mechanical, chemical, and cultural control measures are required [5,10,11]. However, current *C. esculentus* control strategies in conventional agriculture are still very much herbicide-based despite the European Commission's plans to halve the risk (to human health and the environment) and overall use of chemical pesticides by 2030 as stated in the EU Green Deal framework launched in 2020. One way to decrease the overreliance on herbicides is to implement alternative curative strategies. Up to now, mostly soil-disturbing mechanical methods have been implemented but these techniques require multiple passes to exhaust carbon resources stored in the mother tubers [12] and also entrain a high risk of tuber dispersal [13]. One promising alternative to chemical or mechanical weed management is management via electricity. The use of electric energy to control weeds was first mentioned at the end of the 19th century [14] and has recently gained a lot of interest from machine developers. After direct contact between aerial plant parts and a high-voltage electrode, a current flow is established that is directed via aerial parts into belowground plant parts. As a result of the electric resistance of the plant, electric energy is converted to heat [14]. This heat causes damage to cell membranes, which leads to an accelerated loss of plant moisture [15]. So far, electrocution has hardly been practiced by farmers despite (1) the commercial availability of different types of electric weeding devices, (2) the lack of soil disturbance, and (3) its applicability in zones where herbicide use and soil tillage are legally not allowed [16].

A possible reason for the lack of implementation aside from energy input, hazard concerns, and the high equipment cost [17], might be the lack of academic literature on the use, efficacy, and efficacy-influencing factors of high energy electricity for controlling weeds and *C. esculentus* in particular.

Diprose et al. [18] and Sahin [19] concluded that small electric currents (50–100 V, 1–2  $\mu\text{A}$ ) might increase plant growth, while very large currents (5000–15,000 V) result in fast weed destruction. Moreover, the longer the electrocution time (duration of the contact between plant and electrode), the higher the electrocution effectiveness. According to Sahin and Yalınkılıç [20], the electric resistance of the plant, which is positively correlated with the plant growth stage, is an important parameter to take into account.

Schreier et al. [21] performed some field experiments in soybean (*Glycine max* (L.) Merr.) and concluded that electrocution could be part of an integrated weed management system, because it can eliminate late-season herbicide-resistant weeds. Moreover, they concluded that the effectiveness of electrocution (evaluated by visual control ratings 3 or 42 days after treatment) on some annual weed species (*Amaranthus tuberculatus* (Moq.) Sauer [waterhemp], *Xanthium strumarium* L. [cocklebur], *Ambrosia trifida* L. and *Ambrosia artemisiifolia* L. [giant and common ragweed], *Conyza canadensis* (L.) Cronquist [horseweed], *Setaria faberi* Herm. and *Setaria pumila* (Poir.) [giant and yellow foxtail], and *Echinochloa crus-galli* (L.) P. Beauv. [barnyardgrass]) is most strongly correlated to plant height and plant moisture content at the time of electrocution. The Pearson correlation coefficient between weed control (42 days after treatment) and plant height or plant moisture content was, respectively,  $-0.54$  and  $-0.26$ . Two electrocution passes led to a better control compared to one pass. This last result is confirmed in earlier research by Diprose

et al. [22] on the control of sugar beet (*Beta vulgaris* L.) bolters. According to the French Wine Institute [23], electrocution speed, which determines the contact time between electrode and plant leaves, had a significant effect on the effectiveness of electrocution for all weed species (mainly grasses) included in its study. Efficacy significantly improved by reducing speed from  $3.5 \text{ km}\cdot\text{h}^{-1}$  to  $2.5 \text{ km}\cdot\text{h}^{-1}$ . According to Vigneault and Benoît [14], soil moisture content negatively affects the effectiveness of the electrocution technique. In dry soils, more current flows through the belowground plant parts (and not through the soil volume), which leads to an increased damage to the plant root system.

To our knowledge, there are no scientific reports on the effects of a single treatment on vitality of belowground *C. esculentus* plant parts capable of vegetative reproduction (basal bulbs and mother tubers) and on efficacy-influencing (a)biotic factors. Optimization of the efficacy of a single treatment is a prerequisite to keeping the number of passes required to exhaust *C. esculentus* as low as possible.

In our study, we evaluated the effect of a single electrocution treatment on *C. esculentus* shoot and mother tuber vitality. The following hypotheses were formulated: (H1) Electrocutation effectively controls both primary *C. esculentus* shoots and corresponding mother tubers, irrespective of tuber burial depth and clone, (H2) Efficacy of electrocution is better when applied on dry soils and on small targets, and (H3) Efficacy of electrocution is enhanced by increasing the electrical dose by decreasing application speed or by increasing voltage. To address these hypotheses, the following research questions were formulated: (i) What is the impact of electrocution on the vitality of primary shoots and their corresponding mother tubers? (ii) Do growing location and the biotic factors of clone, plant growth stage, and vertical tuber position affect electrocution efficacy? (iii) What is the effect of application speed (exposure duration) and voltage on electrocution efficacy? Hereto, aboveground shoots from artificially buried mother tubers were treated with electricity delivered from two commercially available electric weeding devices applying alternating current via a handheld applicator (Rootwave) or applying phased direct current via a tractor-mounted applicator (Zasso XP300).

## 2. Materials and Methods

During the summer of 2021, two field electrocution experiments were conducted in fields artificially infested with mother tubers of *C. esculentus*. Shoots were exposed to a single electrocution treatment. Both experiments took place on two locations, Bree and Bocholt, both located in the Belgian province of Limburg. The distance between the two locations was 4.8 km. Bree has a sandy loam soil (4.0% clay, 24.5% silt, 69.5% sand) with a  $\text{pH}_{\text{KCl}}$  of 5.3, relatively rich in organic matter (1.7% organic carbon). Bocholt has a sandy soil (1.0% clay, 10.0% silt, 87.0% sand) with a  $\text{pH}_{\text{KCl}}$  of 5.1, relatively poor in organic matter (1.1% organic carbon). In both experiments, no tuber formation was observed at the time of electrocution.

### 2.1. Electrocution Equipment

In the Zasso XP300 experiment, electric weeding was performed with a Zasso XP300 Xpower, from hereon in named 'Zasso' (Figure 1). The machine (working width of 3 m, weight of 2500 kg) is developed by Zasso and commercialized by CNH [AGXTEND], Switzerland. This device consists of a tractor-mounted separate power alternator, and a front applicator unit consisting of 24 positive electrodes which make direct contact with the plant leaves (2 rows of 12 positive electrodes). The alternator, driven by the tractor (150 units of horsepower needed) via a cardan joint, produces an alternating current of 230 V that is routed to power electronic modules that convert the alternating current into a high-voltage phased direct current (maximum voltage 7000 V). Each power module has a maximum power output of 3 kW electrical power that is eventually delivered to one of the 24 single, 12.5 cm wide positive electrodes. The uniform fragmentation of the power via separate electrodes ensures a homogeneous power supply over the entire working width. As the tractor is moving forward and weeds are touched by the positive electrodes,

a current flows via aerial parts into belowground plant parts and the soil volume. After passing the soil volume, the current is captured via a set of negative electrodes touching the soil (and closing the electrical circuit). The power requirements may depend on the plant species, plant morphology, plant growth stage, weed density, and soil properties [24].



**Figure 1.** Picture of the Zasso XP300 Xpower (left) and Rootwave Pro (right) electric weeding devices.

In the Rootwave experiment, electrocution was performed with a Rootwave Pro (Rootwave Pro-001-2, Ubiquitek<sup>®</sup>, Kineton, United Kingdom), from hereon in named ‘Rootwave’ (Figure 1). The machine consists of a power module and alternator, a hand-operated treatment lance (1.5 m long) and a treatment return. The generator produces an alternating current of 230 V (power output between 7.5 kVA and 10 kVA, 50–60 Hz), which is transformed by the power module into high voltage electricity. This current is then transported to the hand-operated treatment lance. The voltage delivered by this treatment lance is adjustable: 3000, 4000, or 5000 V. The alternating current (1.5 A) flows through the plant and soil to the treatment return (grounding electrode) which is inserted into the ground at a distance of 10 m from the treated plants. The recommended voltage and treatment duration (varies from 1 to 10 s) depends on the weed stem diameter and the weed height (before treatment, trimming might be useful in very dense weed areas for safety reasons). In order to operate safely, dielectric insulating knee boots are needed [25,26].

## 2.2. Experimental Set-up

### 2.2.1. Zasso Experiment

The Zasso experiment was a split-split plot design in 4 replicates with all combinations of 5 application speeds (main factor, inclusive the untreated control), 2 genetically different clones (split-plot factor) and 4 *C. esculentus* plant types (split-split-plot factor). To account for possible interclonal differences in sensitivity to electrocution, plant responses were tested for two clones namely ‘Meulebeke’, a clone producing heavy tubers (mean fresh tuber weight of  $456.4 \pm 25.4$  mg) and ‘Ardooie’, a clone producing light tubers (mean fresh tuber weight of  $217.1 \pm 20.2$  mg). These clones were named after the place in Belgium where they were originally sampled and are genetically (based on unpublished AFLP analysis) and morphologically distinct [6]. To evaluate the effect of plant type on the efficacy of electrocution, mother tubers were planted at three times (13, 21, and 29 days before electrocution) and at two depths (7 and 15 cm below surface) according to the planting scheme provided in Table 1. The factor plant type refers to a particular combination of mother tuber depth and plant age, as indicated in Table 1. To assess the effect of application rate, and hence exposure duration, electrocution was performed at 4 speeds 1.1, 1.5, 2.2, and  $3.0 \text{ km} \cdot \text{h}^{-1}$  apart from an untreated control. The maximum power was set to 2000 W per square meter of green surface. The experimental unit was a plot ( $1.5 \text{ m} \times 2.5 \text{ m}$ ) with 25 pre-germinated mother tubers of a particular clone planted at a particular planting depth and time. The tubers were produced in 2020 and kept in the fridge ( $5 \text{ }^\circ\text{C}$ ) from harvest until planting. In order to pre-germinate, tubers were laid on a Copenhagen germination table (regime of 16 h light [ $24 \text{ }^\circ\text{C}$ ] and 8 h dark [ $18 \text{ }^\circ\text{C}$ ]) for 3 days. Tubers were planted in two longitudinal parallel rows at an inter- and intra-row spacing of 50 and 11 cm, respectively. Only medium sized tubers were used: these tubers have a fresh weight falling between

80% and 120% of the clone-specific mean fresh tuber weight. Plots were kept weed-free until the day of electrocution to maximize the contact between *C. esculentus* shoots and the positive electrodes.

**Table 1.** Characteristics (planting date, treatment date, plant age, and planting depth) of the four plant types treated in the Zasso experiment in 2021.

| Plant Type | Planting Date | Planting Depth of Mother Tuber (cm) | Treatment Date | Plant Age at Treatment (Days after Planting) |
|------------|---------------|-------------------------------------|----------------|--|
| 1          | 28 May        | 7                                   | 10 June        | 13   |
| 2          | 20 May        | 7                                   | 10 June        | 21   |
| 3          | 20 May        | 15                                  | 10 June        | 21   |
| 4          | 12 May        | 15                                  | 10 June        | 29   |

At time of electrocution, soil moisture content at a depth of 10 and 20 cm was 14.0 and 16.5% in Bree, and 5.5 and 7.6% in Bocholt, respectively. Each location contained 80 experimental units (2 clones  $\times$  4 plant types  $\times$  5 application speeds  $\times$  4 replicates).

The climatic conditions during the pre-treatment period (12 May–10 June) and electrocution (10 June between 9 AM and 1 PM) are provided in Table 2. Table 3 provides the leaf number of the treated plants as a function of the clone, location, and plant type. At Bree, 40 plants outside the experiment were extracted to assess the moisture content in the shoots. Gravimetric shoot moisture content after 16 h drying at 75 °C varied between 74.3 and 86.3%, irrespective of the plant growth stage.

**Table 2.** Climatic and pedohydrological parameters (in 2021 and 2022) before, during, and after the Zasso and Rootwave electrocution treatments in Bree and Bocholt. T. = temperature (°C), R.H. = relative humidity (%). The meteorological data were obtained via the KMI (Royal Meteorological Institute of Belgium).

| Parameter                         | Zasso   |  | Rootwave  |  |
|-----------------------------------|---|--|---|--|
|                                   | Pre-Electrocution Period (12 May–10 June)                                   |  | Pre-Electrocution Period (25 June–16 July 2022) |  |
|                                   | Bree/Bocholt  |  | Bree (25 June–22 July)                          |  |
| Precipitation (mm)                | 39.7  |  | 151.8   |  |
| Average T. (°C)                   | 14.9  |  | 18.4  |  |
| Average R.H. (%)                  | 71.4  |  | 78.9  |  |
|                                   | Day of treatment (10 June)  |  | Day of treatment (16 July 2022)                 |  |
|                                   | Bree  |  | Bocholt   |  |
| Soil moisture content 0–20 cm (%) | 15.3  |  | 6.6   |  |
| Treatment hour                    | 9 AM–11 AM  |  | 11 AM–1 PM                                      |  |
| Average T. during treatment (°C)  | 22.1  |  | 24.4  |  |
| Average R.H. during treatment (%) | 57.0  |  | 50.5  |  |
| Weather type                      | clear-partially cloudy  |  | partially cloudy                                |  |
|                                   | Post-electrocution period (June 2021–May 2022, vitality screening at Melle) |  |   |  |
|                                   | 10 June–31 August   |  | 1 September–30 November                         |  |
| Average T. (°C)                   | 17.7  |  | 11.8  |  |
| Average R.H. (%)                  | 78.9  |  | 84.8  |  |
|                                   |   |  | 1 December–28 February                          |  |
|                                   |   |  | 1 March–31 May                                  |  |
|                                   |   |  | 5.6   |  |
|                                   |   |  | 89.2  |  |
|                                   |   |  | 11.3  |  |
|                                   |   |  | 66.7  |  |

**Table 3.** Characteristics of the plants on the day of electrocution with the Zasso or the Rootwave electric weeding device.

| Plant Type          | Location | Clone     | Depth of Mother Tuber (cm) | Plant Age (Days after Planting) | Mean Leaf Number of the Primary Shoot <sup>1</sup> |
|---------------------|----------|-----------|----------------------------|---------------------------------|--|
| Zasso experiment    |          |           |                            |                                 |  |
| 1                   | Bree     | Ardooie   | 7                          | 13                              | 5.2 ± 0.63   |
|                     |          | Meulebeke | 7                          | 13                              | 4.1 ± 0.64   |
|                     | Bochholt | Ardooie   | 7                          | 13                              | 4.5 ± 0.17   |
|                     |          | Meulebeke | 7                          | 13                              | 4.7 ± 0.25   |
| 2                   | Bree     | Ardooie   | 7                          | 21                              | 5.8 ± 0.25   |
|                     |          | Meulebeke | 7                          | 21                              | 4.3 ± 0.17   |
|                     | Bochholt | Ardooie   | 7                          | 21                              | 6.7 ± 0.45   |
|                     |          | Meulebeke | 7                          | 21                              | 5 ± 0.36   |
| 3                   | Bree     | Ardooie   | 15                         | 21                              | 4.4 ± 0.31   |
|                     |          | Meulebeke | 15                         | 21                              | 2.9 ± 0.43   |
|                     | Bochholt | Ardooie   | 15                         | 21                              | 4.5 ± 0.13   |
|                     |          | Meulebeke | 15                         | 21                              | 4 ± 0.36   |
| 4                   | Bree     | Ardooie   | 15                         | 29                              | 4.1 ± 0.10   |
|                     |          | Meulebeke | 15                         | 29                              | 3.7 ± 0.51   |
|                     | Bochholt | Ardooie   | 15                         | 29                              | 4.7 ± 0.35   |
|                     |          | Meulebeke | 15                         | 29                              | 6.5 ± 0.21   |
| Rootwave Experiment |          |           |                            |                                 |  |
| /                   | Bochholt | Ardooie   | 7                          | 22                              | 6.0 ± 0.42   |
| /                   | Bochholt | Meulebeke | 7                          | 22                              | 4.4 ± 0.35   |
| /                   | Bree     | Ardooie   | 7                          | 28                              | 7.1 ± 0.48   |
| /                   | Bree     | Meulebeke | 7                          | 28                              | 5.6 ± 0.21   |

<sup>1</sup> Some plants developed secondary and tertiary shoots. The primary shoot was defined as the shoot with the highest number of leaves.

### 2.2.2. Rootwave Experiment

The Rootwave experiment was a randomized split-plot design in 4 replicates with all combinations of 4 voltages (main factor) and 2 clones (split-plot factor). To account for interclonal differences in sensitivity to electric current, the same clones ('Ardooie', 'Meulebeke') as in the Zasso experiment were used. Medium-sized mother tubers were planted at a depth of 7 cm on 25 June. To define the optimal voltage, shoots were for 2 s exposed to 4 voltages: 0 (untreated control), 3000 V, 4000 V, and 5000 V. The exposure time was kept constant at 2 s. The grounding electrode was inserted at 10 m from the treated plants.

The experimental unit was a plot (2 m × 1.5 m) with 12 pre-germinated mother tubers of a particular clone planted in two longitudinal rows at an inter- and intra-row spacing of 50 and 11 cm, respectively. Plots were manually kept free of other weeds to assure optimal contact with the treatment lance. There were 32 experimental units (2 clones × 4 voltages × 4 replicates) on each location, so the total treated area was 96 m<sup>2</sup> (3 m<sup>2</sup> per plot).

At time of electrocution, the soil moisture content in the soil layers 0–10 cm and 10–20 cm was, respectively, 14.3% and 14.9% in Bochholt and, respectively, 18.6% and 19.1% in Bree.

The climatic conditions during the pre-treatment period and during electrocution are provided in Table 2. Due to the extreme wet soil conditions in Bree, the date of electrocution was postponed to a later date (22 July). Table 3 provides the leaf number of the treated plants as a function of the clone and location.

### 2.3. Efficacy Measurements

In both experiments, the efficacy of electrocution on emerged shoots and their mother tubers was assessed through evaluation of shoot vitality and tuber resprouting capacity during the 12-month period following electrocution (June 2021–June 2022). The long

screening period was chosen to give putative dormant tubers or basal bubs the opportunity to resprout. The treated shoots with their mother tuber were carefully exhumed 1 to 3 h after electrocution and stored in the dark at 5 °C. The next day, mother tubers and shoots were disconnected (by cutting the vertical rhizome connecting them) and individually planted 2 cm deep in 150-cell trays (mother tubers) or 96-cell trays (shoots). The substrate in the trays was a 1:1 mixture of potting soil and steamed sandy loam soil containing 2.6% organic matter, 46.7% silt (2–50 µm), 43.4% sand (>50 µm), and 10.0% clay (>2 µm) with a pH-K<sub>Cl</sub> of 5.5. After planting, the trays were placed under a rain shelter greenhouse and optimally irrigated by overhead sprinklers at a rate of 2.5 to 3.8 mm·day<sup>-1</sup> depending on daily water evapotranspiration.

During the 12-month screening period all tray cells were bi- to triweekly monitored for resprouting or regrowth. Regrowing shoots and resprouting tubers were counted and removed from the trays. For each experimental unit, the vitality of mother tubers and shoots was calculated by dividing the number of resprouting/regrowing mother tubers/shoots by the total number of exhumed mother tubers/shoots planted in the trays (between 23 and 25). During frosty days the greenhouse was kept free of frost (min. temperature of 2 °C). Outdoor climatic conditions during this post-electrocution period are given in Table 2 (both experiments).

#### 2.4. Statistical Analysis

All data were analyzed in R version 4.1.3 [27]. All data were analyzed using parametric or non-parametric tests run at the 5% significance level. ANOVA tests (4-way ANOVA and 3-way ANOVA for the Zasso and Rootwave experiments, respectively) were performed to detect significant main and interaction effects of the factors clone origin, plant type, location, and application speed (Zasso experiment) or to detect significant main and interaction effects of the factors voltage, clone origin, and location (Rootwave experiment). Then, reduced models were constructed by retaining the significant highest order (interaction) terms. The model assumptions homoscedasticity and normality were checked with the Levene test and a QQ-plot, respectively. As these assumptions were always met, the Tukey HSD test was used to search for significant differences between factor levels (for all significant main and interaction factors).

### 3. Results

#### 3.1. Tuber Vitality

##### 3.1.1. Zasso Experiment

The four-way ANOVA test indicated that only the main factor plant type had a significant effect on the *C. esculentus* tuber vitality (Table 4). The tuber vitality for plant types 1, 2, 3, and 4 was 94.2% ± 0.96%, 96.8% ± 0.47%, 98.0% ± 1.41%, and 93.5% ± 4.66%, respectively. Tuber vitality within plant type 3 differed significantly from tuber vitality within plant types 1 and 4. Generally, tuber vitalities of electrocuted plants varied between 84.0% and 100.0% and were not significantly different from tuber vitalities (88.3%–100.0%) of untreated plants.

**Table 4.** Zasso experiment. Effects of the parameters speed, location, clone, plant type, and their interactions on vitality of mother tubers from *C. esculentus* plants electrocuted with the Zasso device in 2021.

| Experimental Factor | Df | F-Value | p-Value |
|---------------------|----|---------|---------|
| Speed               | 4  | 0.195   | 0.903   |
| Location            | 1  | 2.078   | 0.124   |
| Clone               | 1  | 0.644   | 0.470   |
| Plant type          | 3  | 4.996   | 0.002 * |
| Speed:location      | 4  | 0.629   | 0.684   |

**Table 4.** *Cont.*

| Experimental Factor             | Df | F-Value | p-Value |
|---------------------------------|----|---------|---------|
| Speed:clone                     | 4  | 1.101   | 0.371   |
| Location:clone                  | 1  | 0.081   | 0.706   |
| Speed:plant type                | 12 | 0.925   | 0.523   |
| Location: plant type            | 3  | 1.024   | 0.411   |
| Clone: plant type               | 3  | 1.224   | 0.355   |
| Speed:location:clone            | 4  | 1.288   | 0.280   |
| Speed:location:plant type       | 12 | 1.229   | 0.295   |
| Speed:clone:plant type          | 12 | 1.567   | 0.058   |
| Location:clone:plant type       | 3  | 2.027   | 0.129   |
| Speed:location:clone:plant type | 12 | 0.751   | 0.713   |

(\*) Significant parameters ( $p < 0.05$ ) are indicated with the asterisk symbol.

### 3.1.2. Rootwave Experiment

The three-way ANOVA test indicated that none of the factors location, clone, and voltage had a significant effect on tuber vitality (Table 5). Tuber vitality of all factorial combinations of location, clone, and voltage (inclusive the control) varied between 72.9% and 93.2%, irrespective of the location, clone, or voltage. Tuber vitality averaged over locations and clones was  $85.0 \pm 3.34\%$  for electrocuted plants (3000, 4000, and 5000 V) and  $88.7 \pm 5.33\%$  for untreated plants (0 V).

**Table 5.** Rootwave experiment. Effects of the parameters voltage, location, clone, and their interactions on vitality of mother tubers from *C. esculentus* plants electrocuted with the Rootwave device in 2021.

| Factor                 | Df | F-Value | p-Value |
|------------------------|----|---------|---------|
| Voltage                | 3  | 0.446   | 0.721   |
| Location               | 1  | 0.753   | 0.391   |
| Clone                  | 1  | 0.716   | 0.403   |
| Voltage:location       | 3  | 0.773   | 0.516   |
| Voltage:clone          | 3  | 1.734   | 0.176   |
| Location:clone         | 1  | 0.000   | 0.995   |
| Voltage:location:clone | 2  | 0.002   | 0.998   |

## 3.2. Shoot Vitality

### 3.2.1. Zasso Experiment

The four-way ANOVA test indicated significant two-factor interactions between speed and location, speed and plant type, and location and plant type (Table 6). Therefore, a separate three-way ANOVA test was performed between the two locations, Bree and Bocholt (Table 6).

**Table 6.** Zasso experiment. Results of the four-way ANOVA test performed in order to evaluate the main and interaction effects of the factors speed, location, clone, and plant type on *C. esculentus* shoot vitality. As the interaction term speed:location was significant, a separate three-way ANOVA test was performed for each location to assess the main and interaction effects of the factors speed, clone and plant type on *C. esculentus* shoot vitality.

| Factor                 | Df | F-Value | p-Value                  |
|------------------------|----|---------|--------------------------|
| General four-way ANOVA |    |         |                          |
| Speed                  | 4  | 166.025 | $<2 \times 10^{-16} *$   |
| Location               | 1  | 84.847  | $<2 \times 10^{-16} *$   |
| Clone                  | 1  | 6.218   | 0.013 *                  |
| Plant type             | 3  | 10.392  | $<2 \times 10^{-16} *$   |
| Speed:location         | 4  | 14.509  | $1.53 \times 10^{-10} *$ |



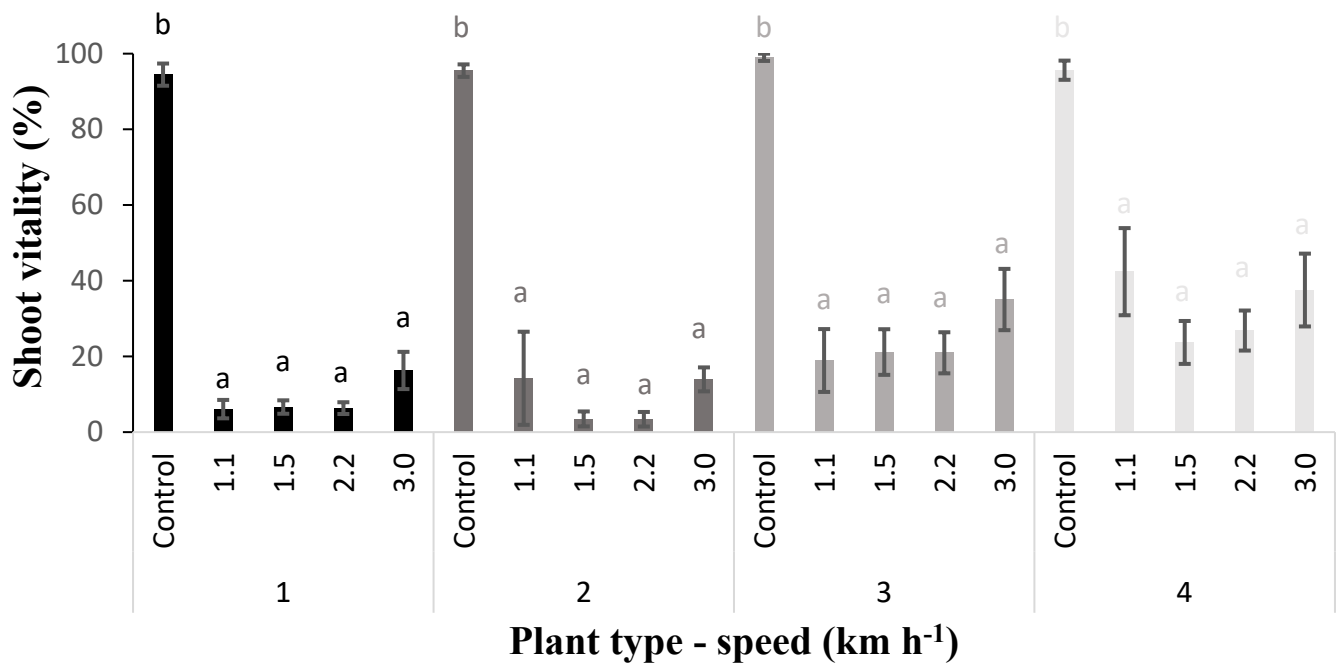
Table 6. Cont.

| Factor                          | Df | F-Value | p-Value                |
|---------------------------------|----|---------|------------------------|
| Speed:clone                     | 4  | 0.989   | 0.414                  |
| Location:clone                  | 1  | 0.324   | 0.570                  |
| Speed:plant type                | 12 | 2.290   | 0.009 *                |
| Location:plant type             | 3  | 3.008   | 0.028 *                |
| Clone:plant type                | 3  | 0.357   | 0.784                  |
| Speed:location:clone            | 4  | 0.799   | 0.527                  |
| Speed:location:plant type       | 12 | 1.239   | 0.258                  |
| Speed:clone:plant type          | 12 | 1.156   | 0.316                  |
| Location:clone:plant type       | 3  | 0.953   | 0.416                  |
| Speed:location:clone:plant type | 12 | 1.697   | 0.069                  |
| Three-way ANOVA for Bree        |    |         |                        |
| Speed                           | 4  | 141.394 | $<2 \times 10^{-16}$ * |
| Clone                           | 1  | 1.877   | 0.173                  |
| Plant type                      | 3  | 13.203  | $<2 \times 10^{-16}$ * |
| Speed:clone                     | 4  | 0.378   | 0.824                  |
| Speed:plant type                | 12 | 1.229   | 0.271                  |
| Clone:plant type                | 3  | 1.390   | 0.249                  |
| Speed:clone:plant type          | 12 | 0.914   | 0.536                  |
| Three-way ANOVA for Bocholt     |    |         |                        |
| Speed                           | 4  | 50.818  | $<2 \times 10^{-16}$ * |
| Clone                           | 1  | 3.604   | 0.060                  |
| Plant type                      | 3  | 1.216   | 0.308                  |
| Speed:clone                     | 4  | 1.319   | 0.268                  |
| Speed:plant type                | 12 | 2.195   | 0.017 *                |
| Clone:plant type                | 3  | 0.117   | 0.950                  |
| Speed:clone:plant type          | 12 | 1.769   | 0.063                  |

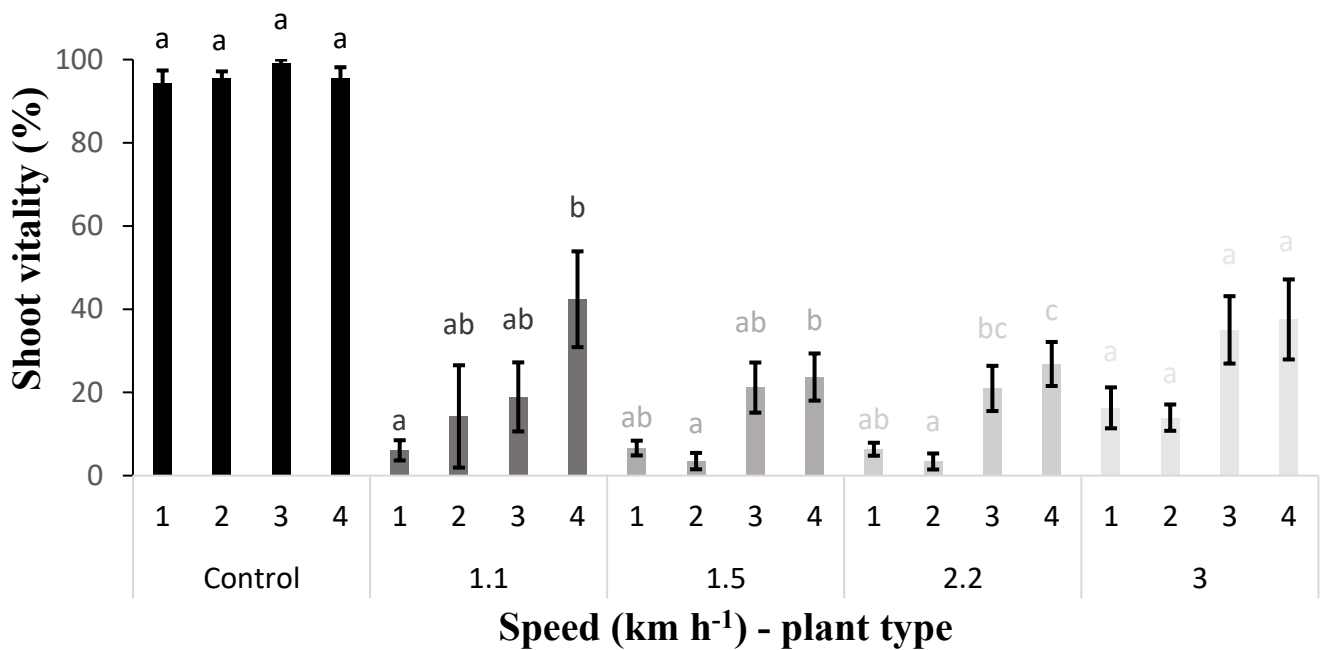
(\*) Significant parameters ( $p < 0.05$ ) are indicated with the asterisk symbol.

At Bree, a significant effect of the factors speed and plant type was observed. For the speed levels 0 (untreated control), 1.1, 1.5, 2.2, and 3.0 km·h<sup>-1</sup>, the *C. esculentus* shoot vitality over all plant types was 96.2% ± 1.07%, 20.4% ± 5.11%, 13.7% ± 2.60%, 14.4% ± 2.59%, and 25.3% ± 3.73%, respectively. The shoot vitality of the untreated plants (= 'speed 0') differed significantly from the shoot vitality of the treated plants. For the plant types 1, 2, 3, and 4, the *C. esculentus* shoot vitality over all speed levels was 26.0% ± 5.66%, 26.1% ± 6.13%, 39.0% ± 5.58%, and 45.4% ± 5.34%, respectively. Shoot vitality of plant type 3 and 4 was significantly higher compared to plant type 1 and 2.

To obtain more information, additional one-way ANOVA tests (with associated Tukey post hoc tests) were performed in order to evaluate the effect of speed level within each plant type and vice versa. The results are given in Figures 2 and 3. Within all plant types, the shoot vitality in the control group differed significantly from the shoot vitality in the other treatment groups (speed of 1.1, 1.5, 2.1, or 3.0 km·h<sup>-1</sup>) (Figure 2). No significant differences were observed between the treatment groups 1.1, 1.5, 2.1, and 3.0 km·h<sup>-1</sup>. Figure 3 shows a significant difference in shoot vitality between plant types 1 and 4 (36.3% higher vitality for plant type 4 compared to type 1) if a speed of 1.1 km·h<sup>-1</sup> was maintained. If a speed of 1.5 km·h<sup>-1</sup> was maintained, plant type 4 showed significantly higher shoot vitality than plant type 2 (20.2% higher). At a speed of 2.2 km·h<sup>-1</sup>, shoot vitality was significantly higher for plant type 4 compared to plant types 1 and 2 (20.5 to 23.4% higher), and for plant type 3 compared to plant type 2 (17.6% higher). Within the speed levels of 0 (control) and 3 km·h<sup>-1</sup>, no significant differences in shoot vitality between plant types were observed.



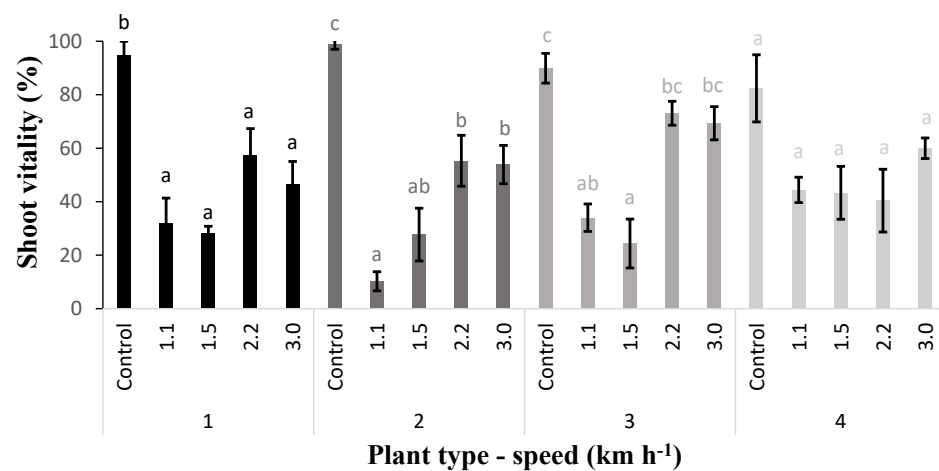
**Figure 2.** Shoot vitality (%) of electrocuted *C. esculentus* plants for each combination of plant type and speed (km·h<sup>-1</sup>) in the Zasso experiment at Bree in 2021. Means sharing the same letter are not significantly different ( $p < 0.05$ ); comparison within plant type only.



**Figure 3.** Shoot vitality (%) of electrocuted *C. esculentus* plants for each combination of speed (km·h<sup>-1</sup>) and plant type in the Zasso experiment at Bree in 2021. Means sharing the same letter are not significantly different ( $p < 0.05$ ); comparison within speed only.

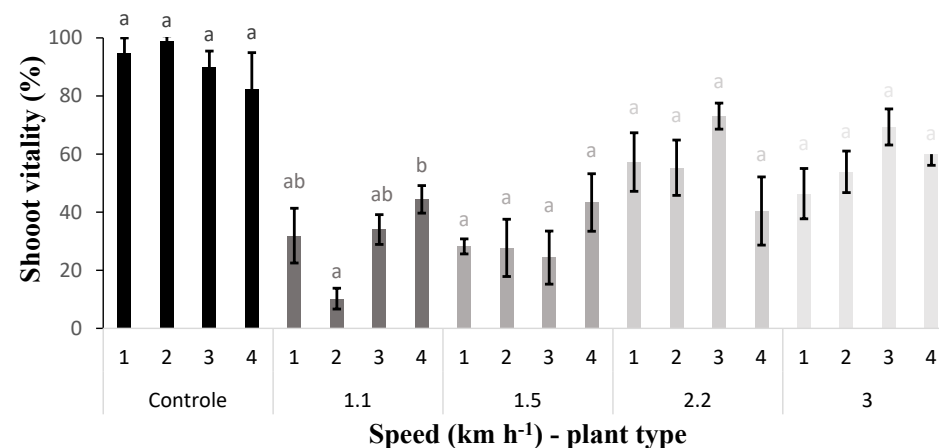
At Bocholt, a significant interaction between speed and plant type was observed (Table 6). Additional one-way ANOVA tests (with associated Tukey post hoc tests) were performed in order to evaluate the effect of speed level within each plant type and vice versa. Within plant type 1, electrocuted shoots showed significantly lower shoot vitality compared to the untreated shoots, irrespective of speed rate with reductions of 63.0, 66.7,

37.6, and 48.5% for 1.1, 1.5, 2.2, and 3 km·h<sup>-1</sup> relative to the untreated control (Figure 4). No significant differences were observed among speed rates within the range of 1.1 to 3 km·h<sup>-1</sup>. However, shoots electrocuted at a speed of 1.1 km·h<sup>-1</sup> showed significantly lower vitality compared to shoots electrocuted at higher speeds (2.2 and 3.0 km·h<sup>-1</sup>) within plant types 2 and 3. Within plant type 2, shoot vitality was significantly 43.7 to 45.1% higher for plants electrocuted at a speed of 2.2 or 3.0 km·h<sup>-1</sup> compared to a speed of 1.1 km·h<sup>-1</sup>. Moreover, shoot vitality of the untreated plants differed significantly from the shoot vitality of the treated plants (range from 43.5 to 88.6%), irrespective of the speed. Within plant type 3, shoot vitality was significantly 44.9 to 65.5% lower for plants electrocuted at a speed of 1.5 km·h<sup>-1</sup> compared to the control plants or plants electrocuted at a speed of 2.2 or 3.0 km·h<sup>-1</sup>. Finally, within plant type 4, shoot vitality of electrocuted plants was not significantly affected by speed. Shoot vitality of treated plants did not differ significantly from shoot vitality of untreated plants.



**Figure 4.** Shoot vitality (%) of electrocuted *C. esculentus* plants for each combination of plant type and speed (km·h<sup>-1</sup>) in the Zasso experiment at Bocholt in 2021. Means sharing the same letter are not significantly different ( $p < 0.05$ ); comparison within plant type only.

Shoot vitalities of plant types within speeds were only significantly different when plants were electrocuted at the lowest speed (1.1 km·h<sup>-1</sup>) (Figure 5). At this speed shoot vitality of plant type 2 was significantly 34.2% lower than shoot vitality of plant type 4.



**Figure 5.** Shoot vitality (%) of electrocuted *C. esculentus* plants for each combination of speed (km·h<sup>-1</sup>) and plant type in the Zasso experiment at Bocholt in 2021. Means sharing the same letter are not significantly different ( $p < 0.05$ ); comparison within speed only.

### 3.2.2. Rootwave Experiment

The three-way ANOVA indicated that only the factor voltage had a significant effect on *C. esculentus* shoot vitality (Table 7). For the voltages 0 (control), 3000, 4000, and 5000 V, shoot vitality averaged over locations and clones was  $98.5\% \pm 1.03\%$ ,  $15.3\% \pm 4.84\%$ ,  $8.6\% \pm 3.24\%$ , and  $11.1\% \pm 3.55\%$ , respectively. Compared to untreated plants (0 V), electrocuted plants had significantly 63.9 to 100.0% lower shoot vitality. However, no significant difference in shoot vitality was found among voltages.

**Table 7.** Rootwave experiment. Results of the three-way ANOVA test performed in order to evaluate the main and interaction effects of the factors voltage (V), clone, and location on *C. esculentus* shoot vitality.

| Factor                 | Df | F-Value | p-Value               |
|------------------------|----|---------|-----------------------|
| Voltage                | 3  | 170.854 | $<2 \times 10^{-6}$ * |
| Clone                  | 1  | 1.163   | 0.288                 |
| Location               | 1  | 1.443   | 0.237                 |
| Voltage:clone          | 3  | 0.677   | 0.572                 |
| Voltage:location       | 3  | 1.556   | 0.216                 |
| Clone:location         | 1  | 0.541   | 0.467                 |
| Voltage:clone:location | 2  | 1.790   | 0.180                 |

(\*) Significant parameters ( $p < 0.05$ ) are indicated with the asterisk symbol.

## 4. Discussion

Hypothesis 1 was partly supported. In both the Zasso and the Rootwave experiments, a single electrocution treatment did not affect the vitality of the mother tubers. In the Zasso experiment, tuber vitalities obtained for electrocuted plants (84.0 to 100.0%) were comparable to the ones obtained for untreated control plants (88.3 to 100.0%). Additionally, in the Rootwave experiment, tuber vitalities obtained for plants electrocuted at a voltage of 3000 to 5000 V (72.9 to 93.2%) were comparable to the ones obtained for untreated control plants (77.2 to 100.0%). Seemingly, the electric current was not able to lethally harm mother tubers even when planted at a superficial depth of 5 cm. Most likely the vertical rhizome connecting the mother tuber with the basal bulb of the primary shoot constitutes a poor conductor of electricity in comparison with the conductivity of the surrounding soil substrate as a result of its thinness (1–2 mm), low water content (<50%), and fibrousness. Indeed, the inner cortical cells of rhizomes are often thick-walled and lignified [28]. In both the Zasso and the Rootwave experiment, both shoot and mother tuber vitality of treated plants was not affected by clone. Hence, the two genetically different *C. esculentus* clones showed similar sensitivity to electricity, irrespective of the weeding device.

Hypothesis 2 was also partly supported. Mother tuber vitality of electrocuted plants was high (72.9 to 100%), irrespective of location, plant growth stage, and electric weeding device, and did not differ significantly from the mother tuber vitality of the untreated control plants (77.2 to 100%). On the other hand, shoot vitality of electrocuted plants differed among the plant growth stage and the location.

In the Zasso experiment, electrocution efficacy clearly depended on the plant type. At Bree, effects of plant type on efficacy of electrocution were more pronounced at lower application speeds (1.1, 1.5, and 2.2 km·h<sup>-1</sup>) than at the highest speed (3.0 km·h<sup>-1</sup>), at which no significant differences in shoot vitality were observed among plant types. At a speed of 1.1 km·h<sup>-1</sup>, shoot vitality was significantly higher within plant type 4 compared to plant type 1 (difference of 36.3%). Additionally, shoot vitality was significantly higher within plant type 4 compared to plant type 2 if a speed of 1.5 km·h<sup>-1</sup> was applied (difference of 20.5%). At a speed of 2.2 km·h<sup>-1</sup>, differences were even more apparent. Shoot vitality was significantly higher within plant type 4 compared to plants type 1 and 2 (difference of, respectively 20.5% and 23.4%). Moreover, shoot vitality was significantly higher within plant type 3 compared to plant type 2 (difference of 17.6%). Similar but smaller effects of plant type were observed at Bocholt. Differential sensitivity of plant types to electricity

reflects differential leaf number of treated plant types. At time of electrocution, plants belonging to plant types 1 and 2 had on average 0.7 leaves more than plants belonging to plant type 3 and 4. Additionally, in the experiment of Schreier et al. [21], there was a trend towards greater weed control when electrocution was performed on larger weed targets.

In the Zasso experiment, shoot vitality of treated plants was lower at Bree (a relatively wet soil) than at Bocholt (a relatively dry soil). The shoot vitality of treated plants varied from 3.4 to 42.4% at Bree, and from 10.2 to 73.1% at Bocholt. This contradicts the expectation that the control efficacy of electrocution is higher when soil moisture content is low [14]. Under dry soil conditions, more current flows through the plant and not through the soil as a result of the high electrical resistivity of the soil that non-linearly increases with decreasing soil moisture content [29,30]. However, apart from soil electrical conductivity, conductivity or resistivity of the plant itself is also an efficacy-determining factor. Indeed, Nadler et al. [31] performed some experiments with trees (mango [*Mangifera indica* L.], banana [*Musa* spp.], date [*Phoenix dactylifera* L.], and olive [*Olea europaea* L.]) and observed a positive correlation between stem water content and stem electrical conductivity. The differential efficacy of electrocution between locations may possibly be explained by a differential electrical conductivity of the plants. In contrast with plants growing on the wetter and heavier soil of Bree, plants growing on the drought-sensitive, light-textured soil of Bocholt most likely experienced (more) heat and water stress resulting in a thicker cuticle and/or lower plant moisture content hampering the entry or transport of the electric current into the plant. Indeed, during the experimental period (12 May–10 June), weather conditions were relatively dry (20–30 mm less rain compared to average years). Moreover, the first ten days of June were very warm for Belgian standards (around 4 °C warmer compared to average years). The higher electric resistivity of the plants at Bocholt might also be explained by the time of electrocution. The electrocution treatments at Bree and Bocholt took place from 9 AM till 11 AM, and from 11 AM till 1 PM, respectively. As a result, the treatment at Bocholt took place at warmer and drier conditions (difference of 2.3 °C and 6.5% relative humidity).

Contrary to the Zasso experiment, no significant differences in shoot vitality between locations were observed in the Rootwave experiment, despite the slightly lower shoot vitalities at Bree. Shoot vitality of treated plants varied from 7.1 to 33.3% at Bocholt, and from 0 to 13.4% at Bree. The differential significance of the location factor between both experiments could be explained by differential weather conditions and soil water status around the time of electrocution and their impact on the plant's sensitivity to electricity. In the Rootwave experiment, *C. esculentus* shoots growing at Bree and Bocholt did not encounter any heat or water stress. On both locations, the experimental period (25 June–16 July 2022) was characterized by very wet weather ( $\pm 150$  mm of rainfall) and average daily maximum temperatures (around 23 °C). Moreover, soil water content (soil layer 0–20 cm) around the time of electrocution was high with only small differences between locations (18.9 and 14.6% for Bree and Bocholt, respectively). Hence, heat and water stress levels were very low and similar at both locations, which most probably led to a similar electric resistivity of the plants at both locations. This was clearly not the case in the Zasso experiment, where a difference of electric resistivity between locations was very likely.

Hypothesis 3 was partly supported. In the Zasso experiment at Bree, shoot vitality of electrocuted plants varied from 3.4 to 42.4%, but no significant differences were observed between applied speeds. This is contradictory with the results of the French Wine Institute [23], where weeds (mainly grasses) were better controlled when applying a speed of 2.5 km·h<sup>-1</sup> compared to 3.5 km·h<sup>-1</sup>. However, at Bocholt, plants belonging to plant types 2 or 3 were significantly better controlled when treated at 1.1 or 1.5 km·h<sup>-1</sup> (shoot vitality from 10.2 to 34.0%) than at 2.2 or 3.0 km·h<sup>-1</sup> (shoot vitality from 53.9 to 73.1%). Within plant types 1 and 4, differences were still present but smaller and not significant. A possible explanation for the differential effect of speed between locations could be that the control levels obtained at Bocholt were much lower (Bocholt: shoot vitality from 10.2 to 73.1%,

Bree: shoot vitality from 3.4 to 42.4%). As a result, the potential effect of the factor speed became more apparent at Bocholt.

The effect of voltage on electrocution efficacy was evaluated in the Rootwave experiment. The applied voltage clearly had no effect on both mother tuber and shoot vitality. After electrocution, mother tuber vitality varied from 72.9 to 93.2%, irrespective of the applied voltage. This result is not surprising because it was already clear that electrocution is not able to lethally heat mother tubers, presumably as a result of the low electric conductivity of the vertical rhizome connecting the basal bulb with the mother tuber. Shoot vitality of treated plants varied from 0 to 31.6%, irrespective of the voltage. An applied voltage of 3000 V already led to relatively low shoot vitalities; increasing the voltage with 1000 or 2000 units had no effect. Increasing voltage may further reduce shoot vitality as a higher voltage leads to an increased amount of transferred energy [14], but on the other hand it may increase shoot vitality when the amount of energy may become so high that it may cause leaf tissue breakage, thus hampering energy transport to the basal bulb.

To conclude, electrocution had no effect on mother tuber vitality but exhibited a strong effect on the vitality of exposed shoots, regardless of the type of electric current. This implies that several electrocution passes are required to exhaust energy reserves of the mother tuber by killing successive flushes of newly initiated or regrowing shoots. According to Matthiesen [32], a mother tuber is able to resprout up to six times depending on tuber size and clone. The first, second, and third resprouting consumes about 60, 10, and 10% of energy reserves of the mother tuber, respectively [33].

In absence of a crop, electrocution should preferably be repeated on regrowing shoots which have around five leaves, as the electrocution efficacy was highest at this growth stage. Applying electricity at this particular stage also implies maximum depletion of energy reserves stored in belowground tissues. Indeed, at the five-leaf stage the plant has reached its compensation point (i.e., the minimum level of belowground reserves) as indicated by Schröder et al. [34]. In naturally infested fields, primary shoots do not emerge simultaneously as tubers may exhibit differential degrees of tuber dormancy or emerge from differential depths. Generally, 80 to 85% of the tubers are located in the upper 15 cm soil layer, and more than 95% are located in the upper 45 cm soil layer [35,36]. Possibly, tuber dormancy may be released by an electric current flowing in the soil matrix in accordance with the findings of Kocaçalışkan et al. [37] showing that dormant seed tubers of potato (*Solanum tuberosum* L.) resprouted after treatments with a low voltage current of 50 and 100 V. Hence, successful *C. esculentus* control with electricity requires season-long repetitive electrocution passes targeting shoots from newly sprouting tubers or resprouting tubers. Repeated passes at the two–five-leaf stage have also been advised for mechanical methods [38]. However, in contrast with mechanical methods based on tillage, electrocution does not disturb the soil, thus avoiding tuber dispersal across the whole field [10], nor does it stimulate soil mineralization. Löbmann et al. [39] found no evidence that electrocution treatments might negatively impact soil organisms. In contrast with chemical *C. esculentus* control, electrocution leaves no potentially harmful chemicals in food or the environment, and its control efficacy is not affected by clone origin. Herbicide efficacy indeed widely varies across *C. esculentus* clones as shown by De Cauwer et al. [6]. However, there are still some challenges. Intensive weed control with electrocution requires season-long black fallow, which may increase the risk of soil erosion [40]. Moreover, the technique is still quite expensive and requires stringent safety procedures.

## 5. Conclusions

Efficacy of *C. esculentus* control with electrocution was maximized when applied at low speeds (1.1 to 1.5 km·h<sup>-1</sup>, and on five-leaf shoots without heat or water stress. The parameters voltage and genetic clone had no effect on the degree of *C. esculentus* control. Electrocution is a useful tool in any conventional or organic integrated weed management system targeting *C. esculentus* but should be considered as an exhaustion tool and therefore be repeated over time or complemented with other methods to achieve sufficient season-

long control. We recommend future research to investigate optimal treatment interval and appropriate sequences of mechanical and electrophysical passes.

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