

Apple Bruise Detection by Electrical Impedance Measurement

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Abstract. Electrical impedance was used to determine the extent of tissue damage that occurred as a result of bruising of apple fruit (*Malus × domestica* Borkh, cvs. Granny Smith and Splendour). Impedance measurements were made before and after bruising. Plots of reactance against resistance at 36 spot frequencies between 50 Hz and 1 MHz traced a semicircular arc, which contracted in magnitude after bruising. A number of characteristics of these curves were then related to bruise weight. The change in resistance that occurred as a result of fruit impact ($\Delta R_{50\text{Hz}}$) was the best predictor of bruise weight, with r^2 values up to 0.71. Before bruising, resistance of fruit was higher in 'Splendour' than in 'Granny Smith' ($P < 0.001$), and at 0 °C than at 20 °C ($P < 0.001$), but was not influenced by fruit weight. The influence of apple cultivar and temperature on electrical impedance may cause difficulties when implementing these measurements in a commercial situation. However, further development of electrical impedance spectroscopy methodologies may result in convenient research techniques for assessing bruise weight without having to wait for browning of the flesh.

Electrical impedance spectroscopy (EIS) has been used to assess ripening of fruit (e.g., Furmanski and Buescher, 1979; Harker and Dunlop, 1994; Harker and Forbes, 1997; Harker and Maindonald, 1994; Varlan and Sansen, 1996). Furthermore, EIS has been used to study ripening of chill-injured nectarines [*Prunus persica* (L.) Batsch] and persimmons (*Diospyros kaki* L.) (Harker and Forbes, 1997; Harker and Maindonald, 1994). In both of these fruits, EIS can be used to detect development of chilling injury during cold storage. Unfortunately, one cannot predict when the cells will become dysfunctional. Thus, once a change in EIS is detected the fruit will then inevitably develop the visual symptoms of chilling-injury upon ripening. Freezing injury, also, is easily detected by EIS (Harker and Forbes, 1997; Harker and Maindonald, 1994). In freeze-thawed tissues, cell membranes and associated compartmentation are disrupted. This leads to a dramatic change in electrical impedance characteristics; most of the reactance component is lost, and the low frequency resistance declines dramatically (Harker and Forbes, 1997; Harker and Maindonald, 1994).

Clearly EIS has the capability to detect severe tissue damage in fruit, as occurs in freeze-thawing. Thus, EIS may also provide a technique that can detect damage that has occurred within fruit flesh as a result of physical injury and/or development of physiologi-

cal disorders. An advantage of using EIS as a measure for assessing bruises is that the extent of tissue damage can be immediately assessed without having to utilize the conventional protocol of waiting 1 day for tissue browning to develop (Klein, 1987; Schoorl and Holt, 1977). In other fruits, such as cherry (*Prunus avium* L.) and kiwifruit [*Actinidia deliciosa* (A.Chev.) C.S. Liang & A.R. Ferguson], browning is obscured by the pigments in the flesh or does not occur. Thus, the identification of alternative methods to quantify bruising may be an important future research tool.

Earlier studies (Cox et al., 1993; Greenham, 1966) have demonstrated that electrical im-

pedance measurements made using closely spaced electrodes (inter-electrode distances 1.3 and 3.5 mm, respectively) can identify bruised and nonbruised apple tissue. Indeed, Cox et al. (1993) were able to map bruising by making 100 measurements across the fruit surface. They obtained a bimodal distribution of measurements (impedance moduli), with the lower values representing measurements made within bruised regions and the higher values representing measurements of undamaged tissue. Numbers of measurements with a value lower than the average value for undamaged tissue were then used to calculate a bruise index (Cox et al., 1993). Essentially, this bruise index represented the proportion of the apple surface area that had an underlying bruise, i.e., one that would be seen after peeling. The index may also indicate that a number of bruises have occurred across the fruit surface.

The focus of the present study was to determine if EIS measurements made by placing electrodes on opposite sides of the bruise, but outside the bruise region, could provide an indication the dimensions and/or weight of a single bruise. The advantage of such measurements is that they may provide a 3-dimensional estimate of bruise dimensions/weight, rather than the 2-dimensional measures of bruise area that were made with the closely spaced electrodes used in earlier studies. Furthermore, such measurements could be made immediately after fruit impact and before any discoloration of the flesh occurred.

Materials and Methods

Plant material. Apple fruit ('Granny Smith' and 'Splendour') with an average weight of 186 g were obtained from commercial growers and stored at 0 °C until required. Individual fruit were weighed before being bruised at flesh temperatures of 20 or 0 °C (10 fruit were

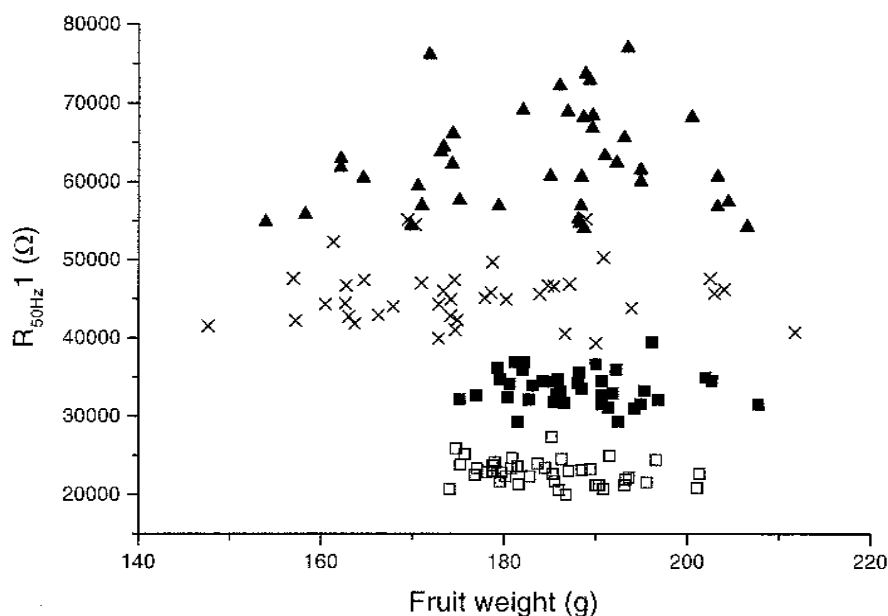


Fig. 1. The influence of fruit weight, cultivar and temperature on initial fruit resistance ($R_{50\text{Hz}}$). Points represent individual 'Granny Smith' apples at 0 °C (■) and 20 °C (□), and 'Splendour' apples at 0 °C (▲) and 20 °C (×).

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bruised for each cultivar \times temperature \times drop height combination). Electrical impedance was measured before and after bruising.

Bruising treatments. To induce bruising, fruit were suspended in a sling and swung in a pendulum through a known arc into a vertical impact surface (as described by Mohsenin, 1970). Fruit were carefully positioned within the sling to ensure that position of the initial impedance measurement was identical with the position that was subsequently bruised. The fruit were dropped from 10-, 20-, 30-, and 50-cm heights and the rebound height for each impact was recorded using a video camera.

About 24 h after impact, the fruit were peeled around the point of impact and bruise diameter (length and width) was measured. The apple was then cut in half and the depth of the bruise measured, followed by removal of all bruised tissue for weighing.

Impedance measurements. Electrical impedance was measured using methods described earlier (Harker and Forbes, 1997; Harker and Maindonald, 1994). Two Ag/AgCl electrodes (2 mm diameter \times 5 mm length, model EP2; World Precision Instruments, Sarasota, Fla.) were impaled in the fruit 35 mm apart, to an approximate depth of 3 mm. The resistance and reactance components of electrical impedance were measured at frequencies between 50 Hz and 1 MHz using a Hewlett-Packard precision LCR meter (model HP4284A; Hewlett-Packard, Hyogo, Japan). Electrodes were removed and the fruit were dropped as previously described. Immediately after impact, the electrodes were reinserted into the holes made during initial measurements, and impedance reassessed as described above. The 35-mm inter-electrode distance used in this study was sufficient to ensure that bruises up to 31 mm in diameter were located between the two electrodes.

Resistance measurements made at 50 Hz before and after bruising are represented by the abbreviations $R_{50\text{Hz}1}$ and $R_{50\text{Hz}2}$, respectively. The change in resistance ($R_{50\text{Hz}1} - R_{50\text{Hz}2}$) was represented by $\Delta R_{50\text{Hz}}$. The resistance measurement made at 1 MHz after bruising was represented by $R_{1\text{MHz}2}$. Two other calculations were used to characterize the electrical properties of damaged apple tissue: $100 \times R_{50\text{Hz}2}/R_{1\text{MHz}2}$ (Greenham and Daday, 1957) and P_y , the volume fraction of intact cells ($100 \times R_{\text{extracellular}}/R_{\text{extracellular}} + R_{\text{intracellular}}$; Pliquet et al., 1995). P_y was estimated by the calculation $100 \times R_{50\text{Hz}2}/R_{50\text{Hz}2} + R_{1\text{MHz}2}$, since $R_{50\text{Hz}}$ and $R_{1\text{MHz}}$ are good estimates of $R_{\text{extracellular}}$ and $R_{\text{intracellular}}$, respectively (Harker and Maindonald, 1994). Statistical analysis was by analysis of variance, and means of the main effects are presented in the tables and text.

Results and Discussion

The initial measurement of resistance ($R_{50\text{Hz}1}$) was not influenced by fruit weight (Fig. 1). This indicated that the volume of tissue encompassed within the electric field generated between the two electrodes was much smaller than the volume of the whole fruit. However, cultivar and temperature in-

fluenced $R_{50\text{Hz}1}$. At 20 °C, 'Granny Smith' apples had a lower $R_{50\text{Hz}1}$ than did 'Splendour' fruit (22,813 Ω c.f. 45,590 Ω ; $P < 0.001$; Fig. 1). Furthermore, $R_{50\text{Hz}1}$ was substantially lower at 20 °C than at 0 °C (22,813 Ω c.f. 33,573 Ω , and 45,590 Ω c.f. 62,570 Ω for 'Granny Smith' and 'Splendour', respectively; Fig. 1). Temperature is known to influence the conductivity of electrolyte solutions (Weast et al., 1984), and thus our observations can be explained in terms of the physical chemistry of electrolytes. Differences in $R_{50\text{Hz}1}$ that exist between

the two cultivars may have reflected differences in tissue structure, such as the arrangement and volume of air spaces, which could alter the resistance of the apples.

The experimental approach required that electrical properties of the fruit did not change as a result of the removal and reinsertion of the electrodes. This was confirmed by examining changes in fruit impedance that occurred after electrodes were removed, the fruit left on the bench for 5 min and the electrodes reinserted. Paired t tests indicated that there was no sig-

Table 1. Influence of removal and reinsertion of electrodes on fruit resistance. Values represent means \pm SE for six fruit, P was determined using a paired t test.

Frequency	Resistance		P
	First value	Second value	
50 Hz	21,710 \pm 591	21,677 \pm 641	0.556
1 MHz	1,380 \pm 42	1,367 \pm 48	0.816

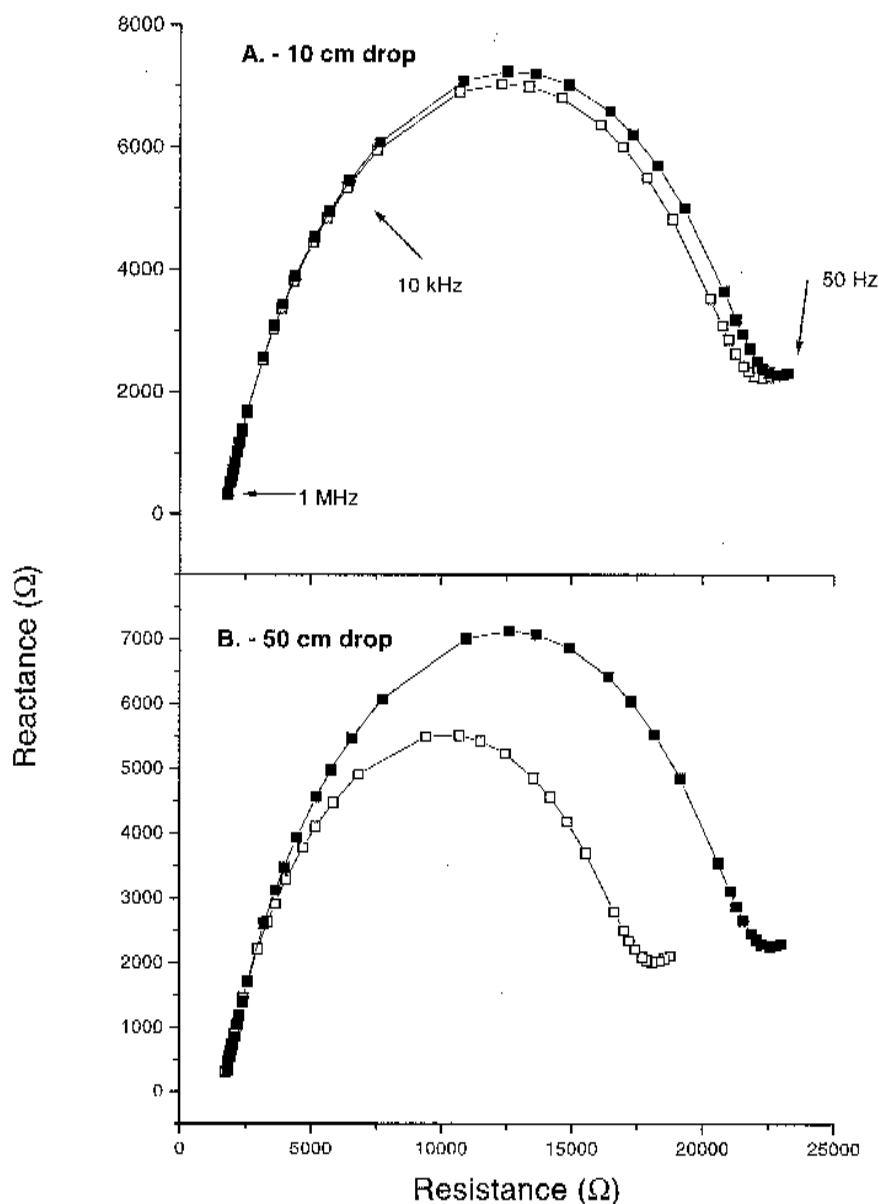


Fig. 2. Plots of reactance against resistance for 'Granny Smith' apples before (■) and after (□) bruising. Fruit were dropped from 10 cm (A) and 50 cm (B). Points represent measurements at 36 spot frequencies between 50 Hz and 1 MHz at 20 °C.

nificant difference between $R_{50\text{Hz}1}$ and $R_{50\text{Hz}2}$ (Table 1).

Impedance is generally separated into resistance and reactance components. At lower frequencies, resistance of the apoplast is measured, while at higher frequencies the resistance of the entire tissue, including the symplast, is measured (Cole, 1972; Harker and Maindonald, 1994; Stout, 1988). In biological systems, reactance is largely a measure of the capacitance of biological membranes (Harker and Maindonald, 1994). Plots of reactance against resistance over the 50 Hz to 1 MHz frequency range traced out a semicircular arc (Fig. 2). The arc contracted in magnitude as a result of bruising, which indicated that both resistance and reactance of the tissue were reduced. These changes were most apparent at frequencies below 10 kHz (Fig. 2). These results indicate that the volume of apple tissue accessible to low-frequency current increased as a result of bruising.

Bruise diameter, depth, and weight increased with drop height (Table 2). The relationships between electrical measurements, $R_{50\text{Hz}2}$ and $\Delta R_{50\text{Hz}}$, and bruise diameter, depth, and weight were examined on an individual fruit basis. Correlations between $\Delta R_{50\text{Hz}}$ and bruise weight (Fig. 3) were higher than all other correlations between other electrical and bruise characteristics (data not shown). The correlation between bruise weight and $\Delta R_{50\text{Hz}}$ was consistent for both 'Granny Smith' and 'Splendour' apples at 0 °C and at 20 °C (Fig. 3). However, $\Delta R_{50\text{Hz}}$ for individual fruit with small and large bruises often overlapped. From a practical perspective, there remained an element of uncertainty when converting a $\Delta R_{50\text{Hz}}$ value into a bruise weight. When the data for both cultivars and both temperatures were combined, there were significant increases in $\Delta R_{50\text{Hz}}$ as drop height was increased from 10 to 20, 30 and 50 cm (mean $\Delta R_{50\text{Hz}}$ for all apples increased from 2083 to 3525, 6005 and 9895 Ω , respectively; $P < 0.001$). Mean $\Delta R_{50\text{Hz}}$ (combined drop heights) was much smaller for 'Granny Smith' (3173 Ω) than for 'Splendour' apples (7581 Ω) ($P < 0.001$). This difference in $\Delta R_{50\text{Hz}}$ between the cultivars may in part have been influenced by differences in magnitude of the initial resistance ($R_{50\text{Hz}1}$), since $R_{50\text{Hz}1}$ was 42% lower in 'Granny Smith' than in 'Splendour' (Fig. 1). When $\Delta R_{50\text{Hz}}$ was recalculated as a percentage of $R_{50\text{Hz}1}$ ($\% \Delta R_{50\text{Hz}}$), cultivar variability was reduced at 20 °C (Table 3). However, at 0 °C, $\% \Delta R_{50\text{Hz}}$ was still higher in 'Splendour' (16%) than in 'Granny Smith' (12%).

The possibility that impedance measurements made after impact could be used to assess bruising was investigated. Measurements of resistance at 50 Hz and 1 MHz ($R_{50\text{Hz}2}$ and $R_{1\text{MHz}2}$) were used as well as estimates of tissue damage, including $100 \times R_{50\text{Hz}2}/R_{1\text{MHz}2}$ (Greenham and Daday, 1957) and P_y ($100 \times R_{\text{extracellular}}/R_{\text{extracellular}} + R_{\text{intracellular}}$; Pliquet et al., 1995). The calculation $100 \times R_{50\text{Hz}2}/R_{1\text{MHz}2}$ provides a measure of the resistances of the apoplast relative to the resistance of the entire tissue (apoplast and symplast), and often provides the best estimate of cell damage

Table 2. Influence of drop height on bruise dimensions and weight in two apple cultivars. Values represent means of 20 fruit.

Drop ht (cm)	Cultivar					
	Granny Smith			Splendour		
	Diam (mm)	Depth (mm)	Wt (g)	Diam (mm)	Depth (mm)	Wt (g)
10	16.2 a ²	4.7 a	0.77 a	18.6 a	5.0 a	0.90 a
20	21.2 b	6.5 b	1.57 b	24.0 b	6.7 b	1.87 b
30	25.0 c	8.5 c	2.66 c	26.6 c	8.4 c	2.73 c
50	29.4 d	10.8 d	4.00 d	30.8 d	10.9 d	4.45 d
LSD _{0.05}	0.9	0.6	0.23	0.9	0.6	0.23

²Mean separation within columns by LSD, $P < 0.05$.

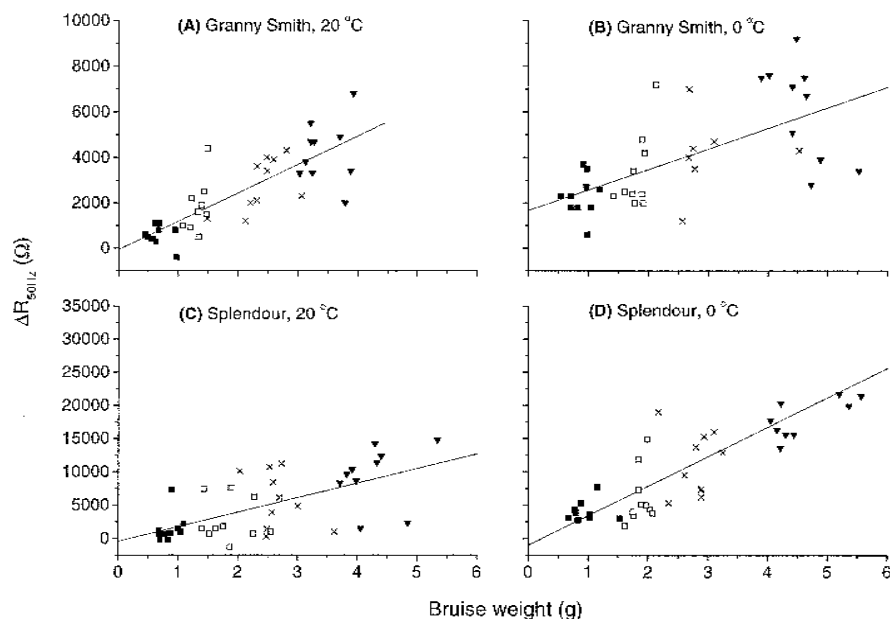


Fig. 3. Relationships between fruit resistance ($\Delta R_{50\text{Hz}}$) and bruise weight for 'Granny Smith' apples at 20 °C (A) and 0 °C (B), and 'Splendour' apples at 20 °C (C) and 0 °C (D). Points represent individual fruit dropped from 10 cm (■), 20 cm (□), 30 cm (×), and 50 cm (▼). R^2 values were 0.71, 0.37, 0.36, and 0.65 for data in A, B, C, and D, respectively. Note that scales for the cultivars differ.

Table 3. Effects of cultivar and temperature on the change in fruit resistance after bruising ($\% \Delta R_{50\text{Hz}}$). Values represent the means for 80 fruit.²

Cultivar	Fruit temp (°C)	
	0	20
Granny Smith	11.71	10.42
Splendour	16.22	10.73

² $P = 0.049$; LSD = 2.402.

Table 4. Influence of drop height on bruise weight and measurements of electrical impedance after impact. Values represent the means for 40 fruit (20 of 'Granny Smith', 20 of 'Splendour').

Drop ht (cm)	Bruise wt (g)	Impedance measurements			
		$R_{50\text{Hz}2}$ (ohms)	$R_{1\text{MHz}2}$ (ohms)	$R_{50\text{Hz}2}/R_{1\text{MHz}2} (\times 100)$	P_y
10	0.84 a ²	38,530 a	2837 a	1343 a	93.0 a
20	1.72 b	37,013 ab	2823 a	1292 a	92.7 a
30	2.69 c	36,585 b	2717 ab	1331 a	92.9 a
50	4.94 d	31,828 c	2686 b	1181 b	92.1 b
LSD _{0.05}	0.16	1893	127	61	0.3
P	<0.001	<0.001	0.046	<0.001	<0.001

²Mean separation within columns by LSD, $P < 0.05$.

(Greenham and Daday, 1957). The calculation $\Delta R_{50\text{Hz}}$ is used to give an estimate of the volume fraction of intact cells, and has been used to characterize meat quality (Pliquett et al., 1995). In the present study $\Delta R_{50\text{Hz}}$ was estimated as $R_{50\text{Hz}}/R_{50\text{Hz}} + R_{1\text{MHz}}$. All measurements ($R_{50\text{Hz}}$ and $R_{1\text{MHz}}$) and calculations ($100 \times R_{50\text{Hz}}/R_{1\text{MHz}}$ and $\Delta R_{50\text{Hz}}$) were significantly affected by cultivar ($P < 0.001$). However, while measurements of $R_{50\text{Hz}}$ and $R_{1\text{MHz}}$ were significantly lower at 20 °C compared to 0 °C ($P < 0.001$), there was no significant effect of temperature on calculations of $100 \times R_{50\text{Hz}}/R_{1\text{MHz}}$ or $\Delta R_{50\text{Hz}}$ (temperature data not presented).

The effect of drop height on bruise weight was significant at all drop heights (Table 4). $R_{50\text{Hz}}$ measurements made at 10-, 30-, and 50-cm drop heights were significantly different. However, only measurements made at 50 cm differed from those made at other drop heights when $R_{1\text{MHz}}$, $100 \times R_{50\text{Hz}}/R_{1\text{MHz}}$, and $\Delta R_{50\text{Hz}}$ were measured (Table 4). Clearly, $100 \times R_{50\text{Hz}}/R_{1\text{MHz}}$ and $\Delta R_{50\text{Hz}}$ were not sufficiently sensitive to evaluate the effect of drop height on bruising. While $R_{50\text{Hz}}$ was the best of these four predictors of bruising, none of the measurements ($R_{50\text{Hz}}$, $R_{1\text{MHz}}$, $100 \times R_{50\text{Hz}}/R_{1\text{MHz}}$, or $\Delta R_{50\text{Hz}}$) were as closely related to bruise weight as was $\Delta R_{50\text{Hz}}$.

In conclusion, this study has confirmed that electrical impedance measurements, particularly $\Delta R_{50\text{Hz}}$, can be correlated with physi-

cal bruise damage. However, $\Delta R_{50\text{Hz}}$ is not horticulturally meaningful, since it is unlikely that original resistance ($R_{50\text{Hz}}$) will be known in practice. Despite this, EIS has shown considerable potential as a method for assessing fruit bruises, both in this and earlier studies. With further development, the methodology may provide convenient and rapid estimates of bruise volume/weight immediately after impact.

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