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Impact of Fruit Maturity on Internal Disorders in Vapor Heat Treated Mango Cv. 'B74'

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Abstract: UN Sustainable Development Goal 12 (SDG 12) aims to reduce food losses in production and postharvest stages within supply chains. Identifying and addressing contributors to such losses is crucial to their reduction and to overall supply chain sustainability. Internal disorders (IDs) often contribute to postharvest losses and waste of highly perishable fruits like mangoes. Understanding and addressing influencers of susceptibility is limited but essential. Factors potentially associated with the expression of IDs in 'B74' mango commercial supply chains were investigated. Over three fruiting seasons (2020/21, 2021/22, and 2022/23), 43 export supply chains in Australia were monitored from two major production regions, the Northern Territory and North Queensland. Prior to export, the mangoes were subject to a mandatory phytosanitary vapor heat treatment (VHT) in which they were heated with saturated water vapor to a core temperature 46 °C maintained for 15 min and were then assessed for IDs at the end of their shelf life. The predominant IDs observed in the 'B74' fruit were flesh cavity with white patches (FCWP) and flesh browning (FB). VHT-induced FCWP, but not FB. Harvest maturity was identified as a predisposing factor. FB was generally positively correlated and FCWP was typically negatively correlated with fruit maturity at harvest. Relatively more-mature fruit was prone to FB irrespective of VHT, and relatively less-mature fruit was susceptible to FCWP post-VHT. Therefore, selective harvesting and/or sorting for optimum maturity after harvest can be practiced minimizing the incidence and severity of these two IDs in 'B74' fruit. Thus, dry matter (DM) sorting can contribute to postharvest loss reduction and the general sustainability of mango supply chains.

Keywords: disorders; export; maturity; postharvest quality; VHT

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1. Introduction

UN Sustainable Development Goal 12 (SDG 12) was introduced in 2015 to focus on sustainable consumption and production practices [1]. By 2030, it aims to fulfill 11 targets. Among these, Target 3 concentrates on global food waste reduction. It aims to cut per capita food waste at retail and consumer levels by 50% and minimize losses in production and postharvest stages and supply chains overall [1]. Inadequate knowledge of contributing factors can hamper efforts to reduce losses. Hence, understanding such factors is crucial for minimizing waste and achieving targets. Research-based recommendations are required to assist in the production of more tolerant fruit and detect susceptible fruit during postharvest handling so that they can be selectively marketed [2].

Internal disorders (IDs) that occur after harvest represent a challenge to reducing food loss and waste for fruit such as mangoes [3–7]. Jelly seed, soft nose, and spongy tissue are common IDs of mangoes, potentially sharing underlying causes [3,5,6]. Variability and poor quality in mangoes caused by IDs are major obstacles to the value, competitiveness, and consumer confidence in Australian fresh mango supply chains [8,9]. In previous years, the presence of postharvest issues in 'B74' supply chains such as FB and FCWP have led to fruit being rejected from the market with negative economic consequences [10,11].

Understanding fruit susceptibility to IDs is somewhat limited. Influences include growing location, mineral imbalance, fruit maturity at harvest, and postharvest treatment and handling [5]. Joyce and Shorter [12] reported that fruit growing in hotter locations was relatively less sensitive to postharvest heat treatment-induced IDs because it may have acclimatized. Conversely, rainfall close to or at harvest can decrease the fruit's tolerance to heat injury [13]. For example, 'Kensington Pride' mango harvested following prolonged heavy rainfall and subjected to VHT at 46.5 °C fruit core temperature maintained for 10 min exhibit internal starchy layers [14–17]. Several studies suggested that calcium (Ca) insufficiency contributes to IDs in fruits, including mangoes [4,5,7,18,19]. The severity of such IDs tends to increase as fruit mature. Examples in mangoes include watery pulp breakdown [5], stem end cavity [20], and jelly seed [20–22]. Fruit response to vapor heat treatment (VHT) is also affected by maturity [23], with less-mature fruit being more prone to VHT-induced IDs [13,23].

VHT is a mandatory market-access treatment for Australian mangoes to enter China and Japan [24,25]. VHT involves heating the fruit core to a minimum of 47 °C for 15 min at saturated vapor pressure [24]. The potential risk associated with exporting mangoes post-VHT is high [26] because such market-access treatments can have an additionally adverse impact on fruit quality [2]. Several studies have indicated that VHT induces various IDs in mangoes, such as impaired starch degradation, internal cavities, and failure to color [6,27,28]. Fruit that underwent VHT showed 23–70% internal breakdown compared to 0–7% in untreated control fruit [27]. Esgurrea and Lizda [23] also reported that 18–58% of the fruit was adversely affected by VHT compared with lower incidence (0–11%) in control fruit. Mitcham and McDonald [29] reported that fruit that underwent VHT showed 75–100% cavity- and internal breakdown compared with 25% in untreated control fruit. Cavity- and internal breakdown symptoms in the control fruit, that were similar in appearance to VHT-induced damage, may be due to exposure to high preharvest temperatures [30,31].

Not all above-mentioned pre- and postharvest variables contribute markedly to fruit sensitivity to IDs. In this context, it is important to determine and address specific factors that pertain to certain supply chains. 'B74' (Calypso™) is a popular mango cultivar developed in Australia. It accounts for 25% of fresh domestic production and ranks second among the economically important desert mango varieties [32]. Introduced in 1999 [33], 'B74' is an in-demand variety because of its unique combination of desirable skin color and flesh flavor along with freedom from flesh fibers and long shelf life [34]. However, this otherwise robust variety can experience IDs like FCWP and FB, leading to substantial losses [35].

As understanding of factors predisposing 'B74' fruit to IDs was lacking, this investigation was undertaken to discern factors mitigating expression of disorders to minimize loss and waste and underpin supply chain efficacy and sustainability.

2. Materials and Methods

2.1. Supply Chain Monitoring

Supply chain monitoring was conducted over three harvest seasons: 2020/21, 2021/22, and 2022/23. 'B74' fruits were sourced from two major mango production regions in Australia: Northern Territory (NT) and North Queensland (NQ).

The green-mature fruits were grown, harvested, de-sapped, fungicide treated, graded, and packed according to commercial practice [33]. Data on soil characteristics and fertilizer management was collected from two major growers involved in export of 'B74' fruit. Harvested fruits were transported from farms by refrigerated trucks (~16 °C) to a commercial VHT facility at Rocklea, Brisbane (Latitude: −27°32′2.4″ S, Longitude: 153°0′2.8″ E). Distances from the pack shed to market were approximately 3424 km and approximately 1720 km from NT (Latitude: −12°27′54.61″ S; Longitude: 132°15′48.49″ E) and NQ (Latitude: −17°08′33.60″ S; Longitude: 145°06′22.80″ E), respectively. Transit times and temperatures from the packing shed to VHT were monitored in the 2021/22 harvest season. A total of 9, 27, and 7 supply chains were monitored in the 2020/21, 2021/22, and 2022/23 harvest seasons from orchard blocks B1–3 from NT and B4–7 from NQ (Table 1). According to industry collaborators, their exported 'B74' fruit had a history of IDs and so were appropriate for this study.

Table 1. Production season, region, orchard block, and supply chains assessed.

Year	Region	Orchard Block Code	Supply Chain Number
2020/21	NT	B1	1–3
		B2	4
		B3	5
	NQ	B4	6–7
		B5	8–9
2021/22	NT	B3	1–5
		B2	6–13
		B1	14
	NQ	B4	15–25
		B6	26–27
2022/23	NT	B3	1
		B2	2
	NQ	B4	3–5
		B6	6
		B7	7

2.2. Fruit Sampling and Postharvest Treatment

An appropriate number of samples in experiments lies in achieving a balance between statistical power and resource efficiency [36]. If more samples are used than necessary, costs and time investment increase without meaningful gains in precision. Conversely, if insufficient replications are employed, then an experiment may lack the statistical power needed to detect meaningful differences between treatments, thereby leading to inconclusive or inaccurate conclusions. Towards determining appropriate fruit sample numbers, random sampling for allocation to treatments was undertaken at the VHT facility in Brisbane (Latitude: −27°32′2.4″ S; Longitude: 153°0′2.8″ E). The appropriate sample size was then calculated by: $n = \frac{z^2 \times p(1-p)}{\varepsilon^2}$ [37]; where, n = sample size, z = 1.96 for 95% confidence level, p = assumed disorder population proportion of 50%, and Ɛ = margin of error. By this method, sample size for a 10% margin of error was calculated to be 97. For uniform representation across treatments and replicates, 102 fruit per supply chain were randomly sampled. Of these, 51 were allocated as untreated controls (i.e., no VHT), and the remainder were allotted for VHT. Each treatment was replicated three-fold, with 17 fruit per replication.

VHT was conducted in a commercial VHT facility at Rocklea, Brisbane (Latitude: −27°32′2.4″ S, Longitude: 153°0′2.8″ E). Fruit were tipped into bins and placed in the VHT chamber. A pulp-temperature probe was inserted into the center of a randomly selected fruit in the center of each bin to monitor fruit core temperature. VHT was applied to the binned fruit as per commercial protocol. Fruit core temperature was raised and maintained at 47 °C for 15 min at >90% relative humidity (RH) [33]. Fruit temperature was gradually increased over ~4 h from ~18 °C to 47 °C. RH was maintained in the range of ~88% to 93%. Thereafter, fruit core temperature was maintained at 47 °C for 15 min for disinfestation. Fruit were removed from the treatment chamber immediately after the VHT treatment. Non-VHT control fruits were held in a room adjacent to VHT chamber at ~20 °C for the duration of treatment. VHT is a mandatory treatment for mangoes exported from Australia to China and Japan [24,25].

2.3. Postharvest Quality Assessments

Immediately after VHT, all fruit were transported in an air-conditioned vehicle (~20 °C) to a postharvest research laboratory at The University of Queensland, Gatton (Latitude: −27°32′21″ S Longitude: 152°16′58.7″ E; ~72 km; ~1 h). Individual fruit maturity (DM%) was measured and recorded immediately upon arrival using a portable near-infrared (NIR; F-750 Produce Quality Meter, CID Bio-Science, Inc., Camas, WA, USA) device [38]. In 2020/21, however, individual fruit DM% was not assessed using NIR for supply chains 1, 2, 4, and 5. In these cases, 20 fruit were randomly sampled, and their DM was assessed by the traditional oven-drying method [39]. All fruit were ripened at ~20 °C and ~90% RH to the eating soft stage assessed using hand firmness ratings on a 0–4 scale [34]. Time (days) to reach firmness stage 4 (eating soft) after VHT was deemed to be end of shelf life. At the eating soft stage, each fruit was sliced longitudinally on both sides of the stone (i.e., leathery endocarp and seed) and their flesh was assessed visually for incidence and severity of the IDs on a 0–3 rating scale from healthy to severe (Table S1).

2.4. Experimental Design and Statistical Analyses

To determine the effects of VHT, each supply chain was considered an independent experiment of two treatments: non-VHT (control) and VHT. Unpaired t-tests at $p < 0.05$ were applied to compare the incidence of IDs and shelf life between the two treatments. The unpaired t-test is specifically designed to compare means of two independent groups or treatments [40]. Additionally, Pearson's correlation tests were applied between individual fruit DM% and severity of IDs and between individual fruit shelf life and IDs. Pearsons's correlation considers linear relationships between variables [41]. JMP Pro 16.0.0 software was used for these analyses.

All supply chain data for each year (n = 9, 27, and 7 for years 1, 2, and 3, respectively) was pooled, and an analysis of variance (ANOVA) was performed to analyze for differences in the incidence of IDs between the supply chains. Means were separated by Tukey's HSD at $p < 0.05$. ANOVA allows for the comparison of means between multiple groups [42]. This is beneficial in dealing with categorical independent variables, such as different supply chains, and a continuous dependent variable, like incidence of IDs.

For each harvest season, fruit were sourced from the two different growing regions (NT and NQ). Each region had different orchard growing blocks, and each block provided a different number of supply chains (Table 1). To examine the effects of fruit origin on the incidence of IDs, data from all supply chains for each year were pooled. Then, variations in the incidence of IDs across seasons, regions, and blocks were evaluated by multiple regression using the linear mixed model (LMM) in the R package 'lme4'. Multiple regression offers advantages when dealing with many variables and clustered data, and can incorporate random effects, such as capturing between-group variability and handling unequal variances [43].

Time from the packing shed to VHT facility (h), ambient transit temperature (°C), postharvest time and temperature unit sums (TTUs), and individual fruit DM% were recorded for each supply chain in the 2021/22 harvest season. To examine relationships between the above-mentioned parameters and the incidence and severity of IDs, Pearson's correlations between these parameters for IDs were conducted.

3. Results

3.1. Fruit Characteristics

Monitoring trials revealed a wide range in fruit weight across different supply chains and individual harvests. For instance, in the 2020/21 monitoring trial for supply chain 1, fruit weight ranged from 405 g to 585 g, with an average weight of 481 g. In supply chain 4, weight ranged from 411 g to 594 g with an average of 489 g. In supply chains 2, 3, and 5 to 9, relatively less variation in fruit weight was observed. The least variation in fruit weight was observed in supply chains 2 and 7, where the ranges were 456 g to 504 g with an average of 477 g and 433 g to 482 g with an average of 459 g, respectively (Figure 1B). Fruit weight from different supply chains and within each harvest in the 2021/22 and 2022/23 monitoring trials also varied considerably (Figure 2).

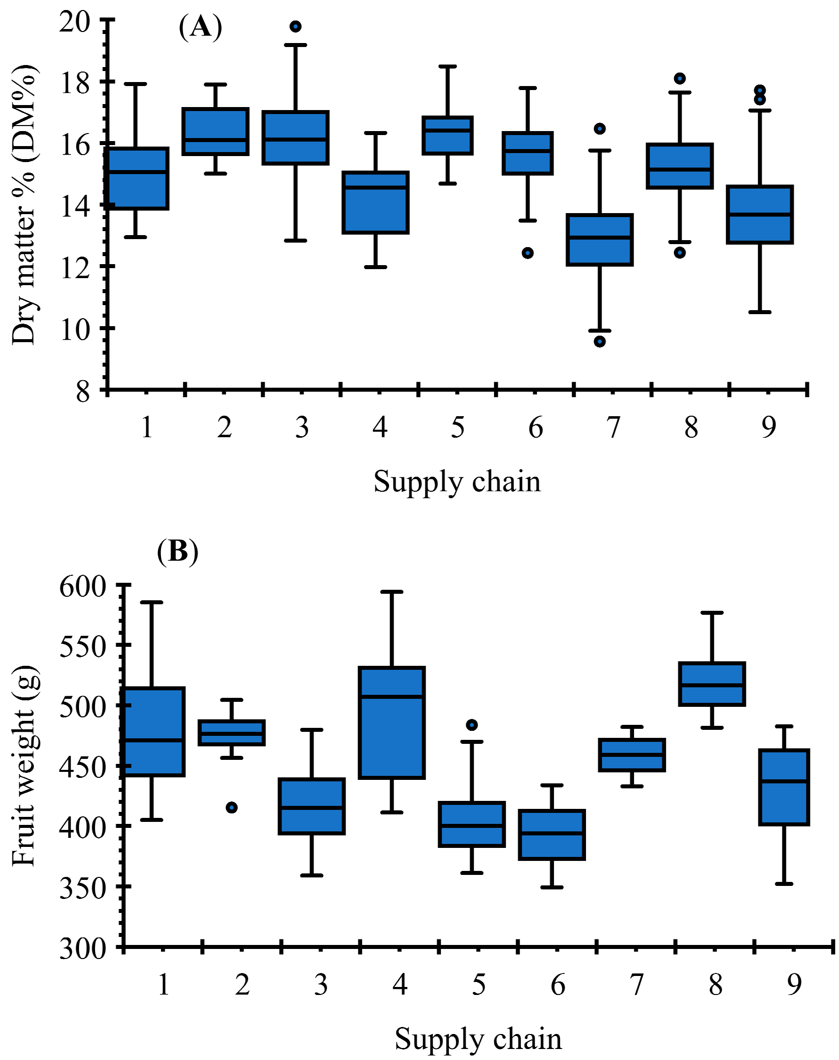

Figure 1. 'B74' mango fruit dry matter percentage (DM%) (**A**) and fruit weight (**B**) distributions in supply chains from two regions, the Northern Territory (NT) and North Queensland (NQ), in the 2020/21 harvest season. Small black dots represent outliers. Ends of lower and upper whiskers represent minimum and maximum data values, respectively. Upper and lower whiskers represent 25% of the data set. Boxes represent the intermediate 50% of the data set. The transverse line within boxes is the median value.

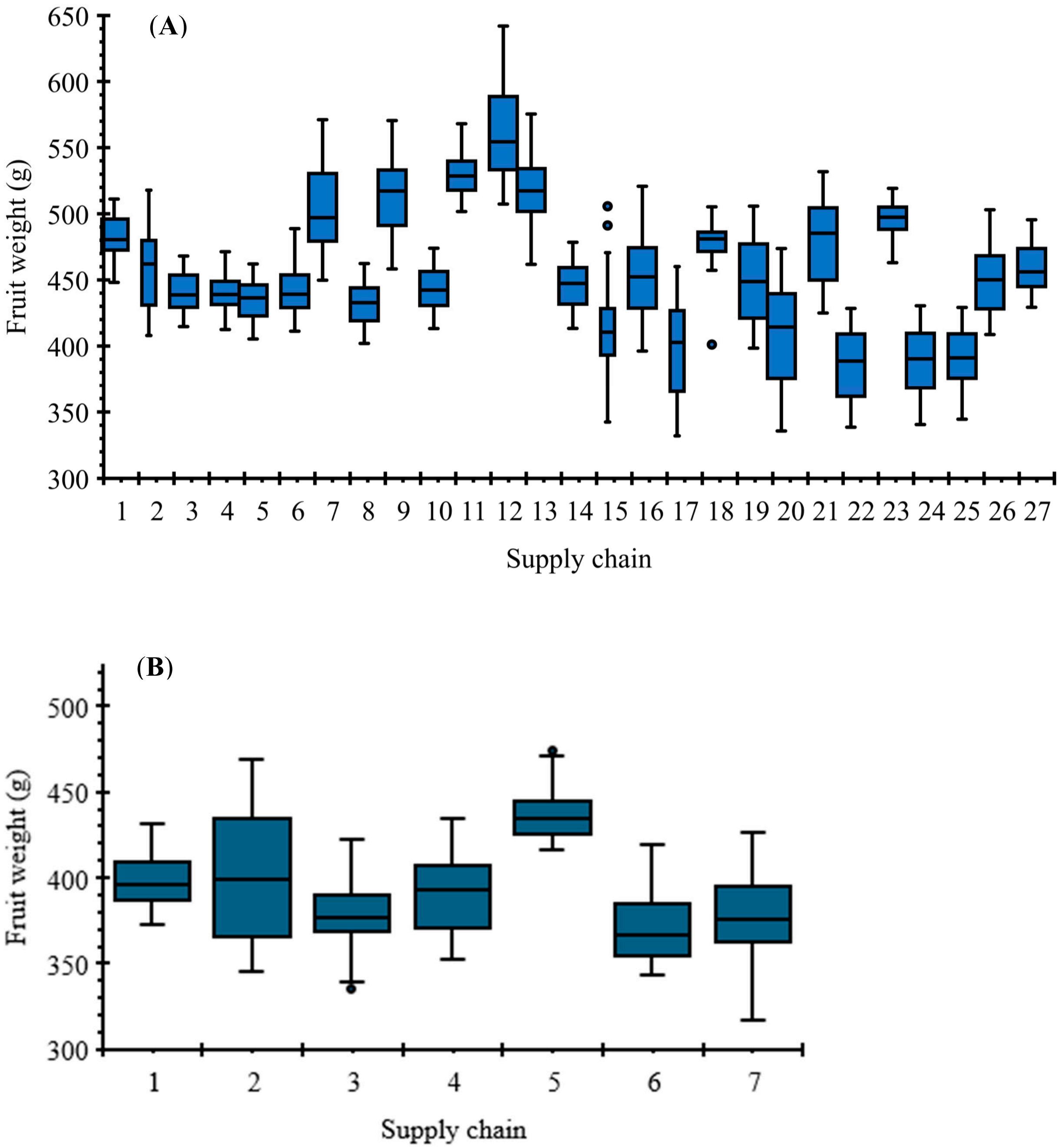

Figure 2. 'B74' mango fruit weight distribution in supply chains from two regions, Northern Territory (NT) and North Queensland (NQ), in 2021/22 (**A**) and 2022/23 (**B**) harvest seasons. Small black dots represent outliers. Ends of lower and upper whiskers represent minimum and maximum data values, respectively. Upper and lower whiskers represent 25% of the data set. Boxes represent the intermediate 50% of the data set. Transverse line within the boxes is the median value.

DM% distribution in fruit from season 2020/21 varied markedly across different supply chains. More than 50% of fruit in supply chains 1, 4, 7, 8, and 9 had $\leq$ 15% DM (Figure 1A). In supply chains 2, 3, 5, and 6, DM% was mostly >15% (Figure 1A). Fruit DM% from different supply chains in the 2021/22 and 2022/23 monitoring trials also varied considerably (Figure 3).

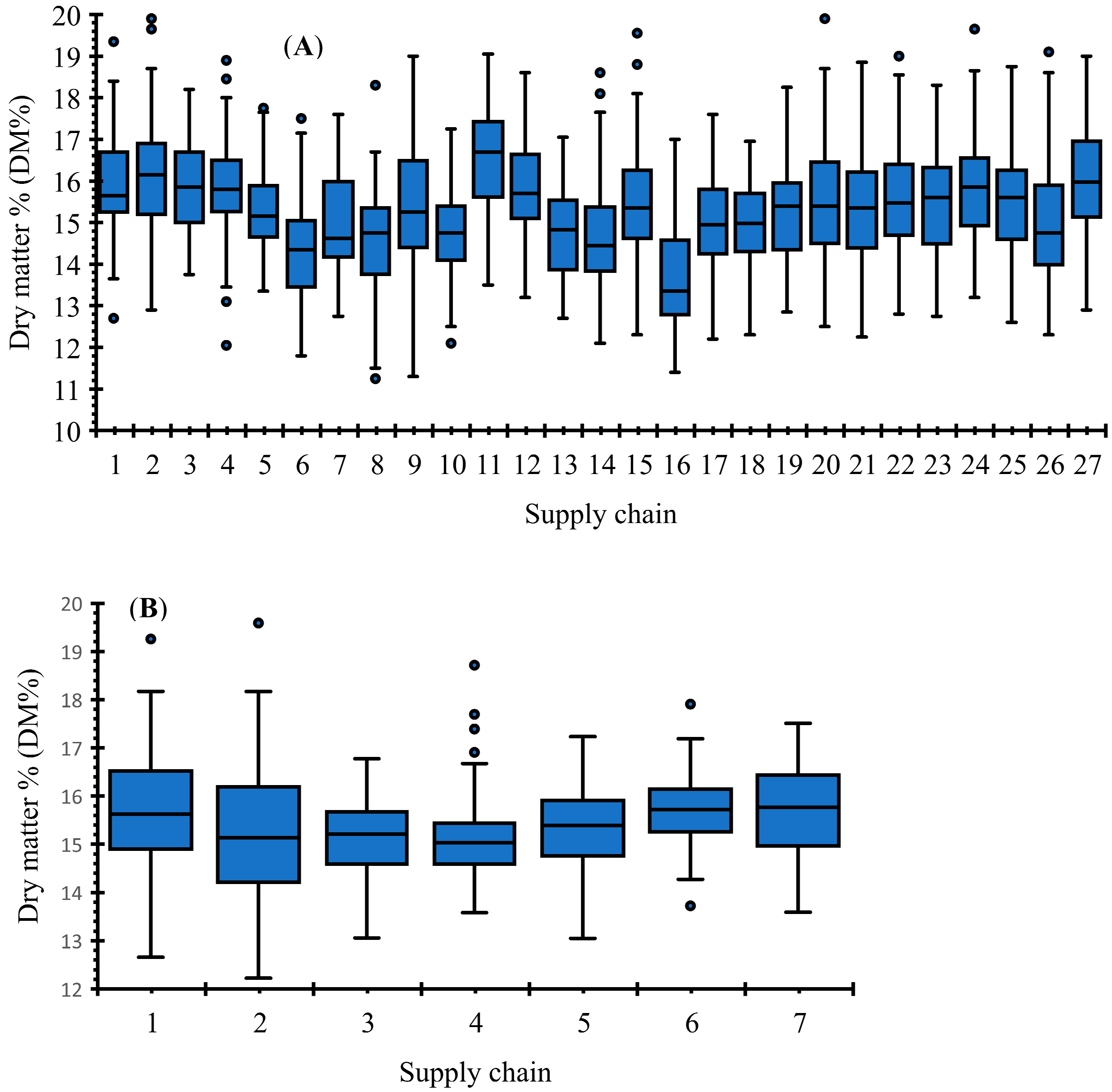

Figure 3. 'B74' mango fruit dry matter percentage (DM%) distribution in supply chains covering two regions of Australia, the Northern Territory (NT), and North Queensland (NQ), in 2021/22 (**A**) and 2022/23 (**B**) harvest seasons. Dots represent outliers. Ends of lower and upper whiskers represent minimum and maximum data values, respectively. Upper and lower whiskers represent 25% of the data sets. Boxes represent the intermediate 50% data set. Transverse line within the boxes is the median.

No significant effects ($p > 0.05$) of VHT on 'B74' mango shelf life were observed in the 2020/21 season in any supply chain (Table S2). However, and except for supply chains 2 and 6, non-VHT fruit exhibited a relatively longer shelf life than did VHT fruit (Table S2). This tendency was also observed in season 2022/23 supply chains, except for supply chain 4, in which there was no difference ($p > 0.05$) in shelf life between the two treatments (Table S3). In 2021/22 supply chains 1, 2, 3, 4, 5, 6, 7, 11, 17, 20, and 23, however, non-VHT fruit had a significantly ($p < 0.05$) longer shelf-lives than did VHT fruit (Table 2).

Table 2. Effect of treatment (+/− vapor heat treatment (VHT)) on shelf life (mean ± SE) of 'B74' mango in 27 case study supply chains during the 2021/22 harvest season. Shelf life was calculated from the day of VHT to the day the fruit reached a hand firmness rating scale of 4. No data appear for supply chain 16 because only fruit that underwent VHT were available during monitoring.

Supply Chain	Shelf Life (Days)		
	+VHT	−VHT	Sig.
1	13.46 ± 0.83 [b]	18.27 ± 0.13 [a]	*
2	13.75 ± 0.25 [b]	16.40 ± 0.82 [a]	*
3	14.55 ± 1.41 [b]	19.43 ± 0.97 [a]	*
4	12.00 ± 1.73 [b]	18.33 ± 0.33 [a]	*
5	18.50 ± 0.32 [b]	21.67 ± 0.43 [a]	**
6	15.80 ± 0.71 [b]	19.47 ± 0.61 [a]	**
7	13.87 ± 0.99 [b]	19.16 ± 0.73 [a]	**
8	16.47 ± 0.92	16.94 ± 3.38	NS
9	15.64 ± 1.52	17.22 ± 2.42	NS
10	19.90 ± 0.45	21.13 ± 0.34	NS
11	21.98 ± 0.51 [a]	20.61 ± 0.63 [b]	*
12	19.75 ± 0.11	20.92 ± 0.92	NS
13	22.39 ± 0.84	23.3 ± 0.49	NS
14	16.35 ± 16.35	14.25 ± 0.67	NS
15	22.65 ± 0.07	22.88 ± 0.06	NS
16	-	-	-
17	18.08 ± 0.13 [b]	19.47 ± 0.35 [a]	*
18	20.16 ± 0.83	22.25 ± 0.62	NS
19	20.12 ± 1.09	21.44 ± 0.61	NS
20	20.51 ± 0.91 [b]	23.88 ± 0.67 [a]	*
21	20.87 ± 1.82	23.10 ± 0.56	NS
22	21.16 ± 0.42	22.18 ± 0.66	NS
23	18.7 ± 0.21 [b]	24.08 ± 0.50 [a]	**
24	22.33 ± 0.90	23.9 ± 1.21	NS
25	20.12 ± 0.89	21.79 ± 1.35	NS
26	21.76 ± 1.10	21.80 ± 0.53	NS
27	17.1 ± 0.99	23.02 ± 2.60	NS

VHT: Vapor heat treated; −VHT: Not vapor heat treated. Means followed by the same letter in a row within incidence and severity are not statistically different. Significance levels: *: 0.05, **: 0.01; and, NS: not significant.

3.2. Internal Disorders

In simulated export supply chains, 'B74' mangoes are afflicted by three IDs: flesh cavity (FC), flesh cavity with white patches (FCWP), and flesh browning (FB; Figure 4). In simulated export, FC (Figure 4—LHS) was not severe and did not markedly affect potential marketability of the fruit. In contrast, FCWP and FB were severe and deemed to be of profound economic importance. FCWP was characterized by cavities within the white, starchy, and tough matrix in mesocarp tissue near the seed (Figure 4—middle). FB was characterized by diffuse brown flesh discoloration, usually near the seed (Figure 4—RHS).

3.2.1. Flesh Cavity with White Patches

FCWP was observed exclusively in VHT fruit. Pronounced variation was observed in FCWP incidence across growing regions, harvest seasons, orchard blocks, and supply chains. For example, NT fruit consistently had significantly ($p < 0.05$) less FCWP than did NQ fruit; FCWP in 2022/23 was significantly ($p < 0.05$) less than in 2020/21 and 2021/22; and orchard blocks B1, B2, and B3 had significantly ($p < 0.05$) lower FCWP than did B4–7 (Table 3).

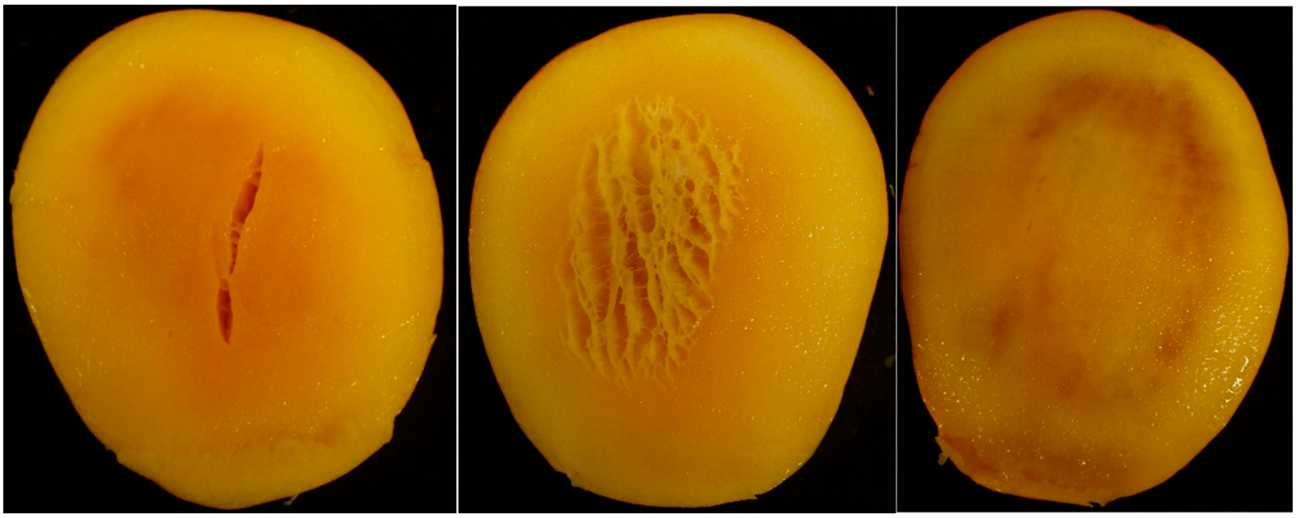

Figure 4. Internal disorders observed during simulated supply chain monitoring: flesh cavity (FC; **left-hand side**), flesh cavity with white patches (FCWP; **middle**), and flesh browning (FB; **right-hand side**).

Table 3. Effect of treatment (+/− vapor heat treatment (VHT)), region (Northern Territory (NT) and North Queensland (NQ)), harvest season (2020/21, 2021/22, and 2022/23) and harvest orchard blocks (B1–7) on Flesh Cavity with White Patches (FCWP) incidence of 'B74' mango fruit in 3-year monitoring trials. Data from all supply chains for each year were pooled for multiple regression. The variations in FCWP incidence across the season, region, and blocks were evaluated using the linear mixed model (LMM) in the R package 'lme4'.

Variable	Incidence %	Standard Error (SE) %	Adjusted SE	*z* Value	*p* Value
Intercept	8.1	5.2	5.2	1.6	0.11
Treatment VHT	39.8	2.5	2.5	15.8	0.00 ***
Region NT	−18.7	2.6	2.6	7.3	0.00 ***
Harvest season 2021/22	4.8	3.5	3.5	1.4	0.16
Harvest season 2022/23	−9.0	4.6	4.6	1.7	0.04 *
Block B1	−29.3	10.2	10.3	2.8	0.00 **
Block B2	−25.0	9.3	9.3	2.7	0.00 **
Block B3	−22.1	9.4	9.4	2.3	0.02 *
Block B4	−6.1	9.0	9.1	0.7	0.50
Block B5	−14.8	11.3	11.4	1.3	0.19
Block B6	0.7	9.8	9.9	0.1	0.94

Significance levels: '***' 0.001 '**' 0.01 '*' 0.05.

In 2020/21, significant variation ($p < 0.05$) in FCWP incidence was observed across different supply chains (Figure 5). Specifically, supply chain 6 had higher FCWP incidence (74.1%), as did supply chain 9 (66.2%). In contrast, supply chain 3 had lower ($p < 0.05$) FCWP incidence (1.96%), similar to supply chains 1, 5, and 8. Supply chains 1–3 were from the same harvest block (B1) but harvested at different times, and supply chain 3 had lower ($p < 0.05$) FCWP incidence than did supply chain 2. Supply chains 6 and 7 were from the same harvest block (B4) but harvested at different times and had different ($p < 0.05$) FCWP incidences (Figure 5). Such variation in FCWP incidence among supply chains originating from the same orchard blocks was also observed in the 2021/22 and 2022/23 monitoring trials (Figures 6 and 7).

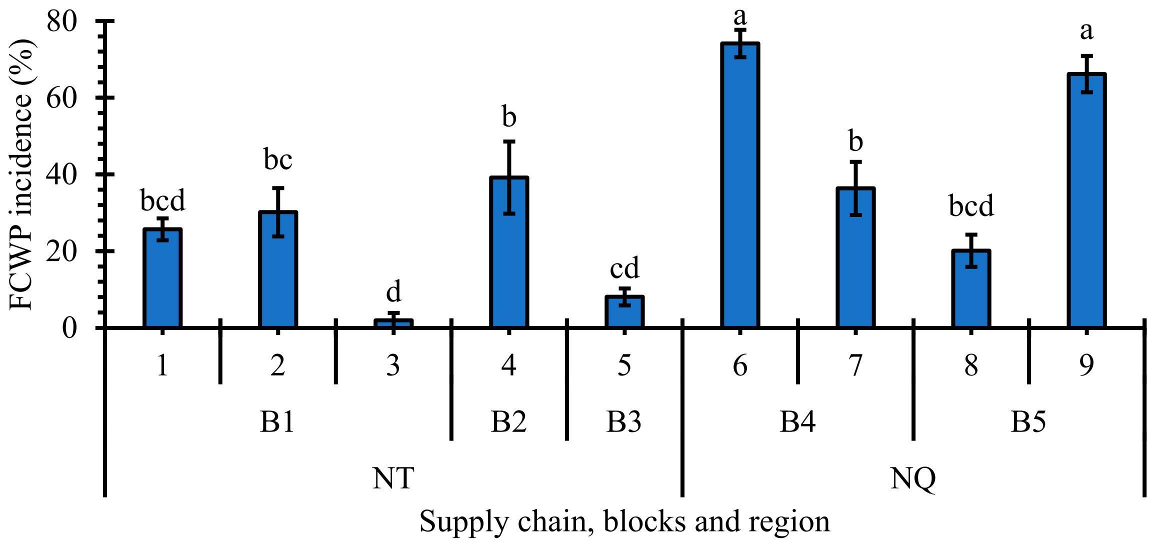

Figure 5. Incidence of flesh cavity with white patches (FCWP) in nine different supply chain case studies over the 2020/21 harvest season for vapor heat treated (VHT) fruit. Values not represented by the same letter are significantly different at $p < 0.05$.

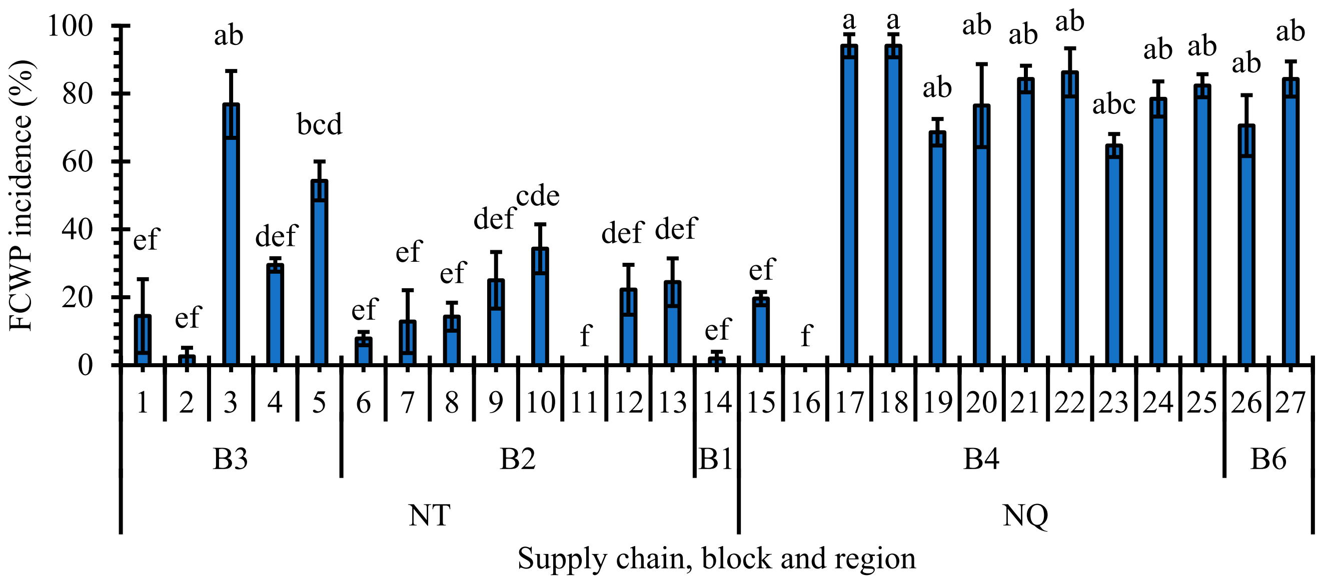

Figure 6. Incidence of flesh cavity with white patches (FCWP) in 27 different supply chain case studies over the 2021/22 harvest season for vapor heat treated (VHT) fruit. Values not represented by the same letter are significantly different at $p < 0.05$.

3.2.2. Flesh Browning

FB was evident in both VHT and non-VHT fruit. VHT fruit had lower ($p < 0.05$) FB incidence than non-VHT fruit (Table 4). Considerable variation in FB incidence was evident across different orchard blocks, harvest seasons, and supply chains. FB in the 2022/23 harvest season was higher ($p < 0.05$) than in the 2020/21 and 2021/22 seasons. Source blocks were also different ($p < 0.05$) in FB incidence (Table 4). In the 2020/21 season, significant variation ($p < 0.05$) in FB incidence was observed among different supply chains (Figure 8). Supply chains originating in NT, except for 1 and 5, typically had significantly higher ($p < 0.05$) FB incidence than did those originating from NQ. FB incidence in supply chains 1 and 5 from NT was akin to all the supply chains from NQ (Figure 8). Variation in FB incidence among the different supply chains originating from the same orchard blocks was observed in the 2021/22 and 2022/23 monitoring trials (Figures 9 and 10).

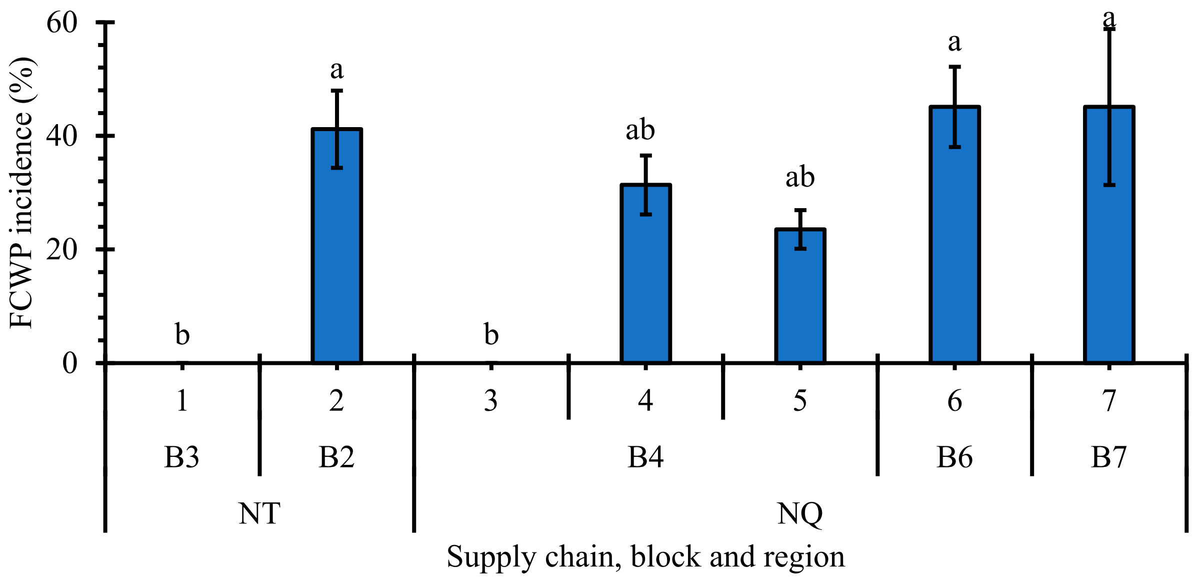

Figure 7. Incidence of flesh cavity with white patches (FCWP) in seven different supply chain case studies over the 2022/23 harvest season for vapor heat treated (VHT) fruit. Values not represented by the same letter are significantly different at $p < 0.05$.

Table 4. Effect of treatment (+/− vapor heat treatment (VHT)), region (Northern Territory (NT) and North Queensland (NQ)), harvest season (2020/21, 2021/22, and 2022/23), and harvest orchard blocks (B1–7) on Flesh Browning (FB) incidence of 'B74' mango fruit in 3 years of monitoring trials. Data from all supply chains for each year were pooled for multiple regression. The variations in FB incidence across the season, region, and blocks were evaluated using the linear mixed model (LMM) in the R package 'lme4'.

Variable	Incidence %	Standard Error (SE) %	Adjusted SE	*z* Value	*p* Value
Intercept	64.8	11.4	11.5	5.6	0.00 ***
Treatment VHT	−13.5	2.7	2.7	4.9	0.00 ***
Region NT	−5.1	3.1	3.1	1.6	0.11
Harvest season 2021/22	8.3	4.4	4.4	1.9	0.06
Harvest season 2022/23	16.6	5.6	5.6	3.0	0.00 **
Block B1	−41.1	11.7	11.8	3.5	0.00 ***
Block B2	−56.8	10.3	10.3	5.5	0.00 ***
Block B3	−26.2	10.4	10.4	2.5	0.01 *
Block B4	−38.3	10.0	10.0	3.8	0.00 ***
Block B5	−55.5	13.0	13.1	4.3	0.00 ***
Block B6	−32.8	10.8	10.8	3.0	0.00 **

Significance levels: '***' 0.001 '**' 0.01 '*' 0.05.

3.3. Relationship between DM%, FCWP, FB, and Shelf Life

In the 2020/21 season, fruit from supply chains with higher average DM% generally exhibited lower incidence of FCWP than did supply chains with lower average DM%, and vice versa (Figure 11). For example, supply chains 3 and 5 had lower incidences of FCWP and higher fruit DM%. Conversely, supply chains 4, 7, and 9 had higher FCWP and relatively lower average DM% (<14.2%). Supply chains 1 and 2 were from the same block harvested at different times earlier than chain 3. Fruit harvested earlier had relatively lower DM%, which coincided with higher FCWP incidence as compared to late-harvested fruit of higher DM% (Figure 11). During 2021/22, most supply chains reflected negative correlations between FCWP severity and fruit DM%, especially supply chains 17, 18, 21, 22, 24, 25, and 27 (Table 5). This suggests that fruit maturity is an important factor governing fruit susceptibility to VHT-induced FCWP.

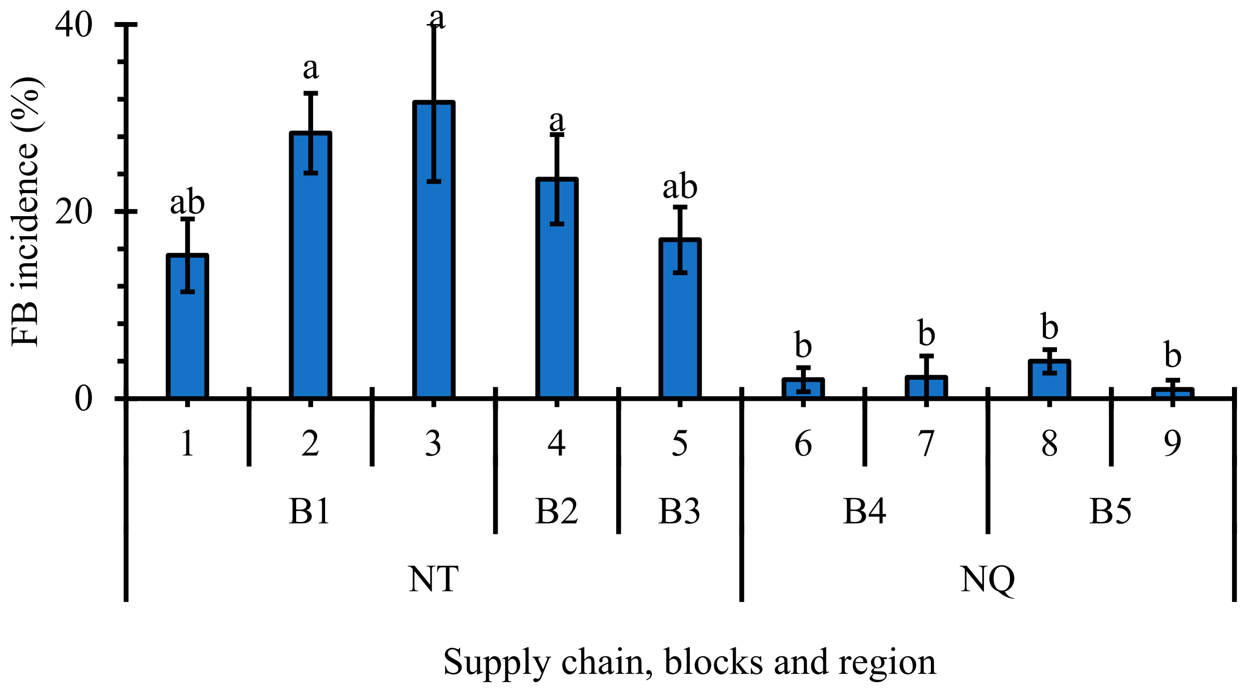

Figure 8. Flesh browning (FB) incidence in nine separate case study supply chains during the 2020/21 harvest season. Levels not represented by the same letter are significantly different at $p < 0.05$.

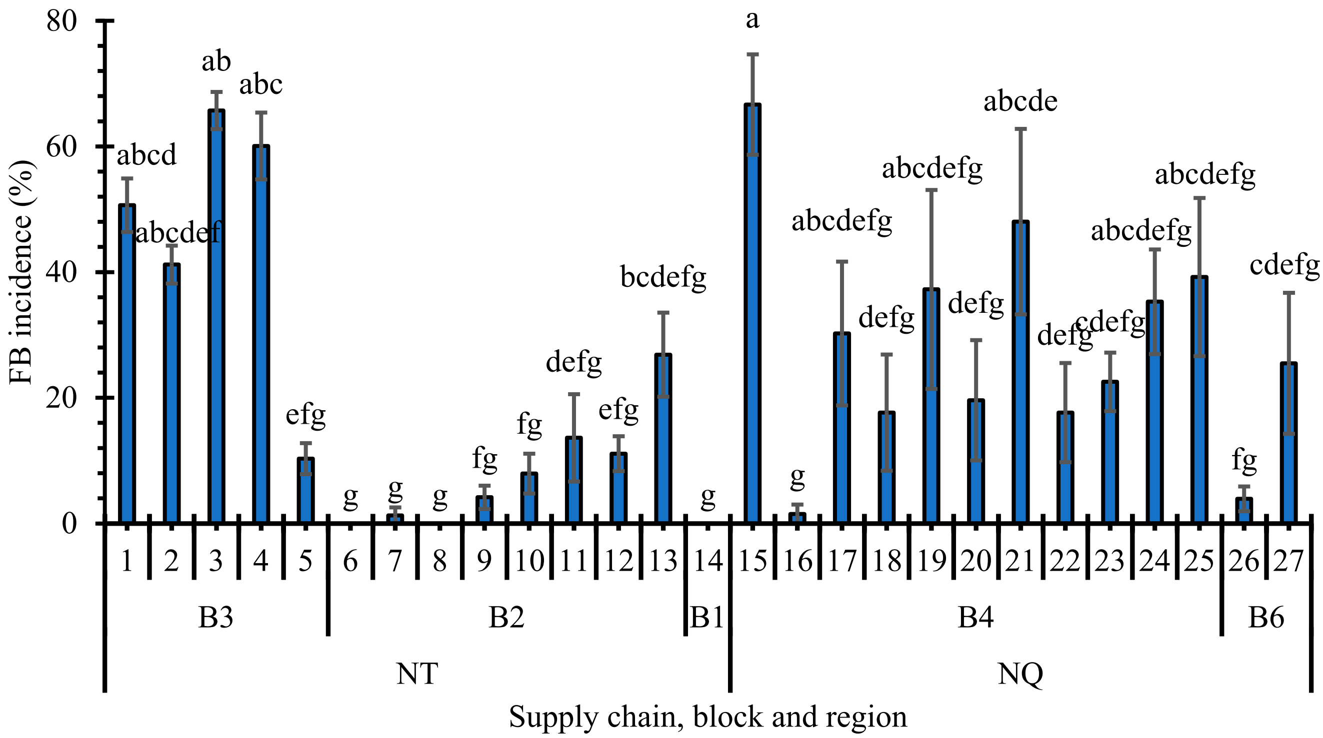

Figure 9. Flesh browning (FB) incidence in 27 separate case study supply chains during the 2021/22 harvest season. Levels not connected by the same letter are significantly different at $p < 0.05$.

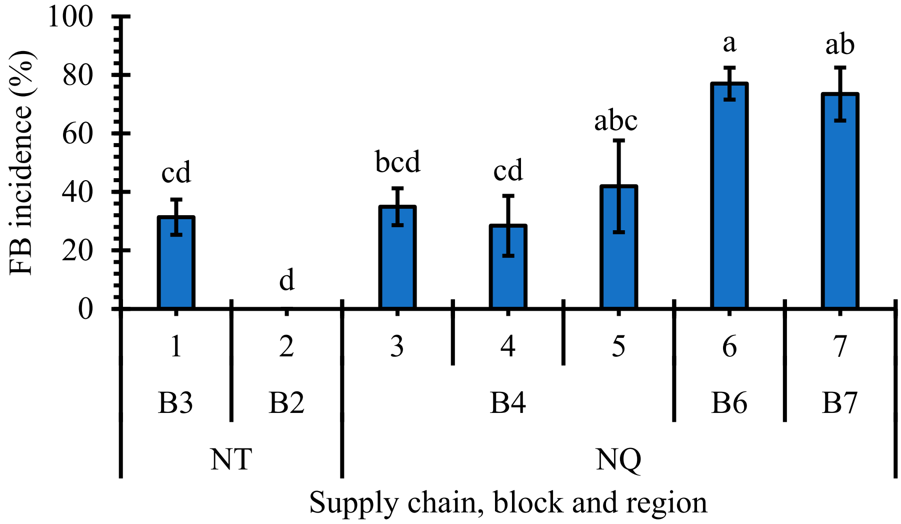

Figure 10. Flesh browning (FB) incidence in seven separate case study supply chains during the 2022/23 harvest season. Levels not connected by the same letter are significantly different at $p < 0.05$.

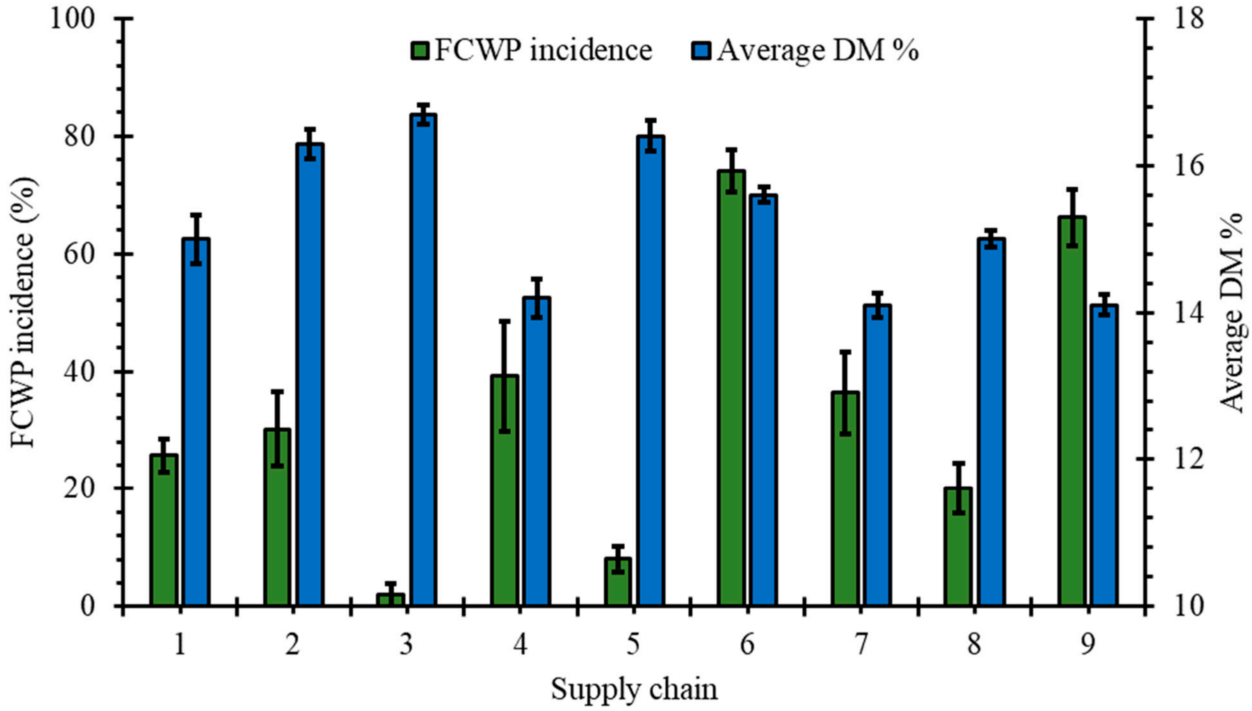

Figure 11. Average dry matter (DM; n = 102) and flesh cavity with white patches (FCWP) incidence (%) in nine case study supply chains for 'B74' mango fruit subjected to vapor heat treatment (VHT). FCWP incidence % presented is the sum of incidences for three severities: viz., slight, moderate, and severe. Vertical bars show ± standard error of the mean.

In 2020/21, fruit from supply chains with a relatively higher average DM% generally exhibited higher FB incidence (Figure 12). For instance, supply chains 2, 3, and 5 had higher FB incidences, corresponding to relatively higher average fruit DM%. Reciprocally, fruit from supply chains having lower average DM (<15%) generally exhibited lower FB incidence. Supply chains 1, 2, and 3 were from the same block harvested sequentially at different times. Earlier harvested fruit (supply chain 1) had relatively lower DM% with lower FB incidence compared to late-harvested fruit with higher DM% (Figure 12). In the 2021/22 season, FB severity was positively correlated with individual fruit DM% in all other supply chains (Table 5).

Table 5. Pearson's correlation coefficients between individual fruit dry matter (DM)% from each supply chain with the flesh cavity with white patches (FCWP) and flesh browning (FB) severity in 27 different case study supply chains monitoring during 2021/22 harvest season. FCWP and FB severity for individual fruit were rated on a scale of 0 to 3.

Supply Chain		FCWP	FB	Supply Chain		FB
DM%	1	−0.29	0.35 **	Shelf life	1	0.21
	2	0.17	0.24 *		2	0.26 *
	3	−0.16	0.02		3	0.21 *
	4	0.2	0.26 *		4	0.12
	5	0.21	0.29 **		5	0.15
	6	−0.04	0.00		6	0.00
	7	0.49 ***	0.00		7	0.18
	8	0.05	0.00		8	0.00
	9	0.18	0.18		9	0.08
	10	−0.12	0.00		10	0.27 *
	11	0.00	0.07		11	0.41 ***
	12	0.08	−0.09		12	0.09
	13	−0.12	0.23		13	0.33 ***
	14	−0.04	0.00		14	0.00
	15	−0.21	0.31 **		15	0.16
	16	0.00	0.16		16	-
	17	−0.44 **	0.36 ***		17	0.27 ***
	18	−0.38 **	0.32 **		18	0.27 ***
	19	−0.03	−0.02		19	0.43 ***
	20	−0.11	0.45 ***		20	0.40 ***
	21	−0.44 **	0.34 ***		21	0.16
	22	−0.34 *	0.23 *		22	0.33 ***
	23	−0.24	0.33 *		23	0.33 ***
	24	−0.28 *	−0.01		24	0.12
	25	−0.40 *	0.32 ***		25	0.17
	26	−0.11	0.05		26	0.21 *
	27	−0.43 **	0.20 *		27	0.48 ***

Significance levels *: 0.05, **: 0.01; ***: 0.001.

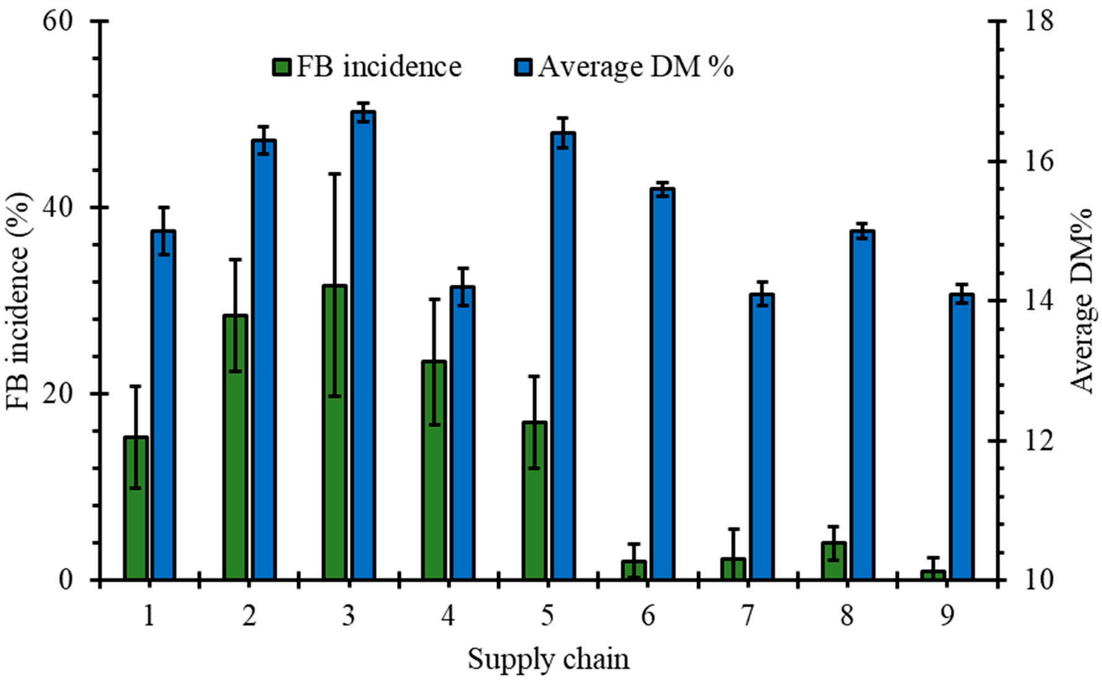

Figure 12. Average dry matter (DM; n = 102) and flesh browning (FB) incidence (%) in nine different case study supply chains for 'B74' mango fruit. The FB incidence % presented is the sum of incidences over the three severity classes: slight, moderate, and severe. Vertical bars are the ± standard error of the mean.

During the three harvest seasons, fruit shelf life and FB severity were correlated across the majority of monitored supply chains. In the 2021/22 harvest season, positive correlations were observed between FB severity and the shelf life of all monitored supply chains (Table 5).

3.4. Relationships between DM%, Postharvest Time-Temperature Units, and Time from Packing Shed to VHT with Incidence of FCWP and FB

Correlations in the 2021/22 trial between DM%, postharvest TTUs, time from packing shed to VHT, and incidence of FCWP and FB are presented in Table 6. No significant correlation was observed between FCWP incidence and time from packing shed to VHT, postharvest TTUs before VHT, or fruit DM%. In contrast, FB incidence was significantly positively correlated with time from packing shed to VHT (r = 0.49), postharvest TTUs (r = 0.39), and DM% (r = 0.61) (Table 6).

Table 6. Pearson's correlation coefficients between time from packing to vapor heat treatment (VHT) facility, postharvest time (h), and temperature unit sums (TTUs) from packing to VHT, and the individual fruit DM% with flesh cavity with white patches (FCWP) and with flesh browning (FB) incidence in 27 different case study supply chains monitored during the 2021/22 harvest season. Postharvest TTUs are the product of average daily temperature with time.

	Disorder	Correlation (r)	Probability Value
Time (h)	FCWP incidence	0.05	0.79
	FB incidence	0.49 **	0.00
Postharvest TTUs	FCWP incidence	−0.04	0.83
	FB incidence	0.39 *	0.05
DM%	FCWP incidence	0.15	0.45
	FB incidence	0.61 ***	0.00

Significance levels: *: 0.05, **: 0.01; ***: 0.001.

4. Discussion

4.1. Flesh Cavity with White Patches

FCWP was directly attributable to VHT as it was exclusively found in fruit subject to this phytosanitary treatment. Nevertheless, not all VHT fruit displayed FCWP, as evidenced by the different supply chains (Figures 5–7). Thus, susceptibility to FCWP is influenced, if not determined, by factors pre-VHT.

Early-harvested fruit with relatively low DM% had higher FCWP incidence than did late-harvested fruit with higher DM% (Figure 11). FCWP incidence tended to be negatively related to DM%, the highest correlation being r = −0.44, $p < 0.001$ (Table 5). While not necessarily causation, this correlation suggests that maturity-related attributes contribute to expression of FCWP. Furthermore, the relatively low correlation suggests that factors beyond maturity may influence fruit sensitivity to VHT.

A significant difference in the incidence of FCWP was evident between fruit from the NT versus from NQ (Table 3). Across the three seasons of monitoring, fruit from NT had a consistently lower incidence of FCWP than fruit from NQ (Table 3). Joyce and Shorter [12] reported that growing locations influence sensitivity to heat treatment. They postulated that fruit growing in warmer locations may be less sensitive to heat due to acclimation [7].

However, the present study did not discern specific and/or consistent influences for regional growing conditions on the incidence of FCWP. Notably, fruit from the same harvest block or growing conditions showed significantly ($p < 0.05$) different FCWP incidence. For instance, in the 2020/21 monitoring trial, supply chains 1, 2, and 3, originating from the same block (B1), exhibited varied FCWP incidence (Figure 5). Likewise, so did supply chains 6 and 7, and 8 and 9, originating from the same blocks, namely B4 and B5, respectively (Figure 5). Similarly, in 2021/22, despite supply chains 1–5, 6–13, and 15–25 originating from the same blocks, significant variation in FCWP incidence was observed among these supply chains (Figure 6).

The disparity in FCWP incidence between the two regions may also or alternatively be associated with differences in the transit time from harvest to VHT. Fruit from the NT endured comparatively longer handling and transit times of 5–7 d from harvest to VHT as compared to the shorter times of 3–4 d for fruit from NQ. Holmes [10] found that time from packing to VHT was a major contributing factor to damage caused by VHT.

The role of the physiological stage or age of fruit in rendering them more tolerant to disinfestation heat treatment is discussed by Jacobi et al. [44]. They observed that fruit treated immediately after harvest had relatively higher starch and lower sucrose levels in association with more severe starch layer disorder expression post-heat treatment. It is established that solutes, including sugars, can have a significant role in protecting plant tissues, including fruit, against various stresses, such as high CO_2, desiccation, cold, and heat [45].

Furthermore, VHT applied to fruit of different physiological stages within a single batch may be reflected in differential effects. If ripe fruits were used as the fruit to temperature probe during VHT, then treatment time was extended by as much as 30 min [10]. The relative temperature lag in riper fruit may be associated with compositional changes (e.g., solutes) and/or density (e.g., intercellular air space) variations. Carefully controlled experiments with the fruit of the same maturity at harvest subsequently held to obtain different physiological stages at the time of VHT could help elucidate this phenomenon.

Fruit size in single VHT batches varied (Figure 1B). Sivakumar et al. [46] reported that relatively smaller fruits were more susceptible to heat damage than were larger fruit. This observation was affirmed by Jacobi and Giles [17] on 'Kensington Pride' mango. Smaller fruits typically heat faster and are thereby exposed to treatment temperature for a longer duration, making them more susceptible to heat damage [17].

Using smaller fruit for the temperature probe during treatment in a batch of heterogeneous fruit may lead to inadequate heat treatment of larger fruit in the batch. Conversely, the use of larger fruit for the probe could expose smaller fruit to longer high treatment temperature exposure than required or safe. Therefore, it is recommended to use fruit of similar size during treatment to reduce the negative effects of VHT and ensure proper disinfestation treatment.

Based on the fruit sampling across supply chains, it is evidently desirable to conduct trials to identify, compare, and contrast predisposing factors additional to maturity that are associated with FCWP in 'B74' supply chains.

4.2. Flesh Browning

FB was observed both in VHT and non-VHT fruit (Table S4). However, the VHT effect was not significant in 2020/21. Moreover, in 2021/22, for 10 of the 27 supply chains monitored, non-VHT fruit exhibited a significantly higher ($p < 0.05$) FB incidence than did VHT fruit (Table S5).

As contrasting results were obtained across supply chains (Tables S4–S6), it was not definitively affirmed that VHT is the primary cause of FB disorder in 'B74' export supply chains. For instance, FB was observed in fruit regardless of whether they had undergone VHT. Moreover, non-VHT fruit had higher FB than VHT-treated fruit (Table 4). Similarly, Brecht et al. [47] discerned no definitive role of hot water quarantine treatment in the development of 'corte negro', a mango fruit disorder like FB. However, Esguerra et al. [27], Esguerrra and Lizada [23], and Mitcham and McDonald [29] reported VHT damages mango fruit due to internal O_2 deficiency leading to undesirable fermentation products and internal breakdown like FB symptoms.

Fruit with a higher DM% exhibited greater susceptibility to FB compared to those with a lower DM% (Figure 12). Likewise, early harvested fruit with relatively lower DM% exhibited lower FB incidence than did late harvested fruit with higher DM% (Figure 12). Results of the three-year monitoring trial indicated that nearly all supply chains exhibited a positive correlation, the highest being $r = 0.45$, $p < 0.001$ (Table 5) between FB and DM%. This suggests that fruit maturity only contributes in part to FB susceptibility.

Significant positive correlations (r = 0.39, $p < 0.05$) between FB incidence and postharvest TTUs and between FB incidence and time from packing to the VHT facility (r = 0.49, $p < 0.01$) were discerned in the 2021/22 season (Table 6). This putative association further suggests that fruit experiencing supply chain distribution practices are predisposed to the expression of IDs. Thus, 'aging' associated with postharvest handling operations can potentially have a relatively greater influence than preharvest conditions on the fruit's expression of FB.

The monitoring trial in 2021/2022 also revealed a positive correlation between shelf life and FB incidence (Table 5), with fruit taking longer to ripen having higher incidence and severity of FB. Overall, but not exclusively, non-VHT fruit had relatively higher FB than did VHT fruit (Tables S4–S6). Several previous studies have discerned that FB is a function of storage time [34,47–49]. Brecht et al. [50] and Brecht et al. [51] considered that internal flesh discoloration/browning in mangoes developed because of chilling injury.

4.3. Plant Nutrition

Significant variation in FCWP and FB incidence in the present study across different supply chains originating from the same block was observed over the 3 years of monitoring (Tables 3 and 4). Fruit from the same block may be presumed to have similar nutrient levels. While not analyzed for minerals, they 'on average' are grown in the same environment with the same management practices. In this purported scenario, the main differences would be postharvest handling practices and fruit maturity.

Putative roles of N and Ca in fruit predisposition to IDs are debated in the literature [4,5,7,18,19]. It is generally considered that too much N and / or too little Ca available during fruit growth and development increase the risk of fruit disorders. In the present study, however, soil analyses commissioned by the 'B74' mango growers (Tables S7 and S8) suggested balance to provide adequate Ca during early fruit development and growth phases. Yet, the incidence of IDs was relatively high (Figures 11 and 12).

In blocks with higher and lower soil Ca levels, such as block B1with soil Ca at 2106 ppm versus block B3 with soil Ca at 912 ppm (Table S8), IDs did not differ significantly (Figures 5 and 8). This may imply that postharvest handling operations are more influential in rendering fruit either more or less susceptible to express IDs than is nutritional balance.

Nevertheless, it cannot be discounted that fruit mineral nutrition plays a role in fruit robustness and susceptibility to IDs. Thus, further work on VHT-associated IDs in 'B74' is likely warranted to explore the putative role of fruit mineral nutrient compositions in FCWP and FB. Further studies might employ a broad range of Ca and N application rates and examine the harvesting of fruit at various maturities.

5. Conclusions

FCWP and FB in 'B74' supply chains were identified as being of economic importance. This study over three serial seasons identified harvest maturity and postharvest TTUs as factors that contribute to the expression of IDs in 'B74' mango fruit. Fruit maturity was identified as the more important factor, with FCWP being negatively correlated and FB being positively correlated, with these defects (Table 5). FCWP was clearly associated with mandatory VHT market-access treatment (Table 3). By contrast, FB was evidently independent of ±VHT (Tables S4–S6). Thus, the study did not affirm the working hypothesis that VHT is a primary cause of FB disorder in 'B74' export supply chains.

Results overall suggest that the two specific disorders are intransigent in the context of interacting predisposing factors versus discrete cause and effect. Nonetheless, it was clearly established that harvesting optimally mature fruit and segregating them on relative maturity after harvest along with limiting transit time TTUs offers reduced FCWP and FB in 'B74' mango supply chains.

6. Practical Implication of the Research Outcomes

A range in maturity levels (DM%) within a single harvest is a common problematic phenomenon. To address DM% variation in a single harvest, maturity mapping of the orchard and harvesting fruit based on maturity zones could ensure a relatively homogenous desirable maturity range within harvest batches [20]. In-field maturity mapping allows the identification of different rates of fruit maturation and provides guidance for scheduling the order of harvest across an orchard.

Upon in-field maturity mapping, a reduced degree of mixed fruit maturity would exist within a harvest batch [52]. To address this, non-destructive fruit sorting using in-line NIR might be employed to sort fruit into more discrete maturity batches [2]. In practice, rigorous sorting should minimize IDs in the market.

7. Research Limitations and Future Research

'B74' fruit susceptibility to IDs varies with region, block, and season, rendering it challenging to identify specific causes in any single export supply chain. Follow-on research should focus on narrowing the 'B74' fruit dry matter in any one harvest batch. It might also explore predictive modeling of the relationships between preharvest and postharvest variables and fruit predisposition to express IDs post-VHT. Also, relatively simple decision support tools, such as decision trees, could be devised to support workers on farms and subsequent supply chain stakeholders in reducing FCWP and FB disorders and lessening fruit losses in the context of assuring consumer satisfaction. Future work could entail more comprehensive fruit maturity comparisons in conjunction with postharvest TTU experiments over harvest. Through better understanding, including measuring, modeling, and managing pre- and postharvest interactions governing fruit robustness or resilience, the profitability of 'B74' mango supply chains could be maintained in concert with reduced food loss at wholesale, retail, and in the home.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/su16135472/s1, Table S1. Rating scale for Flesh Cavity with White Patches (FCWP) and flesh browning (FB) based on severity. Images on left panel show the rating scale for FCWP, and the right panel shows the rating scale for FB. Both disorders were rated on a 0–3 scale; 0: healthy, 1: slight, 2: moderate, and 3: severe; Table S2. Effect of +/− vapor heat treatment (VHT) on shelf-life (mean ± SE) of 'B74' mango in nine case study supply chains during the 2020/21 harvest season. Shelf-life was calculated from the day of VHT to the fruit reaching hand firmness rating scale of 4. No data in supply chain seven as only VHT fruit was available during monitoring. In each supply chain, there were 102, evenly divided between 51 +VHT treatment and 51 −VHT. Table S3. Effect of +/− vapor heat treatment (VHT) on shelf-life (mean ± SE) of 'B74' mango in seven case study supply chains during the 2022/23 harvest season. Shelf-life was calculated from the day of VHT to the fruit reaching hand firmness rating scale of 4. Table S4. Incidence and severity of Flesh Browning (FB) (mean ± SE) in nine separate case study supply chains during the 2020/21 harvest season with +/− vapor heat treatment (VHT). The data were collected from two regions in Australia, Northern Territory (NT; supply chains 1–5) and North Queensland (NQ; supply chains 6–9). Table S5. Incidence and severity of Flesh Browning (FB) (mean ± SE) in twenty-seven separate supply chains during 2021/22 harvest season with +/− vapor heat treatment (VHT). The data were collected from two regions in Australia, Northern Territory (NT; supply chains 1–14) and North Queensland (NQ; supply chains 15–27). Table S6. Incidence and severity of Flesh Browning (FB) (mean ± SE) in seven separate case study supply chains during the 2022/23 harvest season with +/− vapor heat treatment (VHT). The data were collected from two regions in Australia, Northern Territory (NT, supply chains 1and 2) and North Queensland (NQ, supply chains 3–7). Table S7. Nutrient management plan adopted by a 'B74' mango grower who exports 'B74' mangoes. Export of 'B74' mangoes is from only two major growers in Northern Territory (NT) and in North Queensland (NQ), respectively. The 'baseline' data presented were collected in 2022 and reflect nutrient management practices over 3 years. Table S8. Soil test parameters of 'B74' growing blocks registered for export. Export of 'B74' mangoes is from two major growers in Northern Territory (NT) and in North Queensland (NQ), respectively. Only data from three registered blocks from each site are tabulated. They were collected in 2022.

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