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Premature Fruit Drop in Mango (*Mangifera indica* L.) in Northern Vietnam

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Abbreviations

AZ	abscission zone
CK	Cytokinin
CPPU	N-(2-chloro-4-pyridyl)-N-phenylurea.
Dafb	days after full bloom
2,4-D	2,4-dichlorophenoxyacetic acid
ET_o	reference crop evapotranspiration [mm day^{-1}]
GAs	giberellins
GA_3	giberellic acid
GMA	glycol methacrylate
HAc	acetic acid
IAA	indole-3-acetic acid
Kc	crop coefficient
LSD	least significant difference
MMA	methyl methacrylate
M	mol
MPa	megapascal
NAA	1-naphtaleneacetic acid
ng	nanogram
PGR	plant growth regulator
ppm	parts per million
°R	réaumur scale
RH	relative humidity [%]
RIA	radio-Immuno-assay
VPair	air vapour pressure
VPD	vapour pressure deficit
VPsat	air saturation pressure

Summary

Mango production in Northern Vietnam is mainly organized in farmer-owned, small-scale orchard operations. However, the production is limited due to excessive fruit drop presumably caused by unfavourable climatic conditions in combination with plant stresses during the fruit set period. There is a general belief that this phenomenon is caused by different combinations of stressing factors which may vary between regions and sites. In the mountainous area of Northern Vietnam, fruit drop of two main local cultivars 'Hôi' and 'Tròn' may be caused by environmental cues occurring particulary during fruit set. Environemntal stress factors may include excessive air temperature, low relative humidity, strong prevailing winds and little rainfall. These multiple stressors are likely associated with a time dependent change of the endogenous plant hormone auxin (indole-3-acetic acid, IAA) exported from fruit and within the pedicel. Field trials revealed, that fruit shedding could be reduced by irrigation and plant growth regulator (PGR) application throughout the 3-year experiment study; however, it remains unclear how climatic conditons might induce hormonal response and thus enhance fruit shedding at different stages of fruit development.

The present research project consists of three studies. First it was to determined which single or multiple climatic cues trigger fruit shedding in mango. Second, it was attempted to alleviate fruit abscission by PGR spray application at post-bloom and early developmental stages of the fruit in comparison with regular irrigation scheduling to reduce extensive fruit abscission. Third, the morphological changes in the abscission zone of mango pedicel during fruit abscission were studied. The timely changes of plant tissue IAA concentration and its key role in the abscission process was also evaluated.

The research work was conducted in a commercial orchard near the township of Yen Chau in 2007, 2008 and 2009. The experimental design consisted of 20 randomly selected 10-year-old mango trees of each of the cultivars 'Hôi' and 'Tròn'. Half of the trees were irrigated at 3-day-intervals by mircospinkler and the remaining trees served as non-irrigated controls. For the PGR applications, 3 trees of each cultivar in 2008, and 6 trees of each cultivar in 2009 were used. In both experiments, 10 randomly selected inflorescences per tree were labelled and counted twice per until end of the fruit drop period. Fruit tissue was collected on-site from irrigated and non-irrigated trees. The fruit export of the endogenous indole-3-acetic acid was

analyzed by Radio-Immuno-Assay (RIA). Further, to clarify the morphological changes within the abscission zone (AZ) of mango pedicels, samples of irrigated and control trees were collected, fixed and embedded, using a modified dehydration and embedding technique by vacuum infiltration.

The results of the first part of this study indicate that the onset of the hot, dry prevailing winds induced the fruit drop. Whether fruit drop was reduced by irrigation seems to depend on the level of soil water deficiency, hence the reduction of plant water potentials. The results of the second part of the study showed that PGR applications reduced excessive fruit drop. Although all chemical treatments indicated significant effects, a single spray application of N-(2-chloro-4-pyridyl)-N-phenylurea (CPPU) and 1-naphtaleneacetic acid (NAA) effectively improved fruit retention. Furthermore, those spray applications led to a greater fruit set per inflorescennce than irrigation.

However, IAA-export was not clearly affected by irrigation or PGR applications. The results of the third part of the study indicate positive effect of irrigation on fruit retention in both cultivars, which might be influenced by increased pedicel thickness thus increased carbon/nutrient availability to the fruit during critical environmental periods. Moreover, shortage of carboydrate supply to the fruit may be associated with a reduction of IAA-export out of the fruit and this in turn triggers the abscission process.

In conclusion, this research proved that prevailing environmental conditons, particulary hot, dry winds, induce premature fruit drop in mango in Northern Vietnam. The identification of the physiological basis of premature fruit drop allowed the development of effective crop management strategies (e.g. PGR applications, irrigation) to overcome unfavourable environmental conditions and to reduce or even inhibit plant responses associated with premature fruit drop. The presented results suggest, that setting up an irrigation system increased fruit retention, which; however, is a great investment for farmers. It was shown that the ease of PGR application and the efficiency of the treatment is a promising alternative to irrigation in oder to prevent excessive fruit drop in mango. The development of effective, fruit drop reducing crop management strategies may also optimized crop loads and enhanced financial returns to growers.

Zusammenfassung

Die Mangoproduktion in Nordvietnam ist überwiegend in bäuerlichen Kleinbetrieben organisiert. Der Anbau ist jedoch stark eingeschränkt durch einen übermäßigen Fruchtfall, vermutlich verursacht durch örtlich vorherrschende ungünstige klimatische Bedigungen. Es wird allgemein angenommen, dass dieses Fruchtfall-Phänomen durch eine Kombinationen verschiedener Stressfaktoren verursacht wird, die je nach Region und Lage variieren. In der Bergregion von Nordvietnam, genauer der Son La Provinz, kann der Fruchtabfall von ungünstigen Umwelteinflüssen verursacht werden, die zumeist während des Fruchtansatzes auftreten. Zu diesen negativen Umweltfaktoren zählen sowohl extrem hohe Lufttemperaturen, eine niedrige relative Luftfeuchtigkeit, aber auch die vorherrschenden heißen Winde mit wenigen Niederschlägen. Ein Zusammenwirken dieser multiplen Stressoren kann vermutlich eine Reduzierung des Auxins Indol-3-Essigsäure (IAA) Exports durch den Fruchtstiel bewirken. Feldversuche zeigten, dass das Muster und die Intensität des Fruchtfalls bei Mango während der gesamten 3-jährigen Versuchsperiode durch Bewässerung sowie die Anwendung von Pflanzenwachtumsregulatoren verändert, respektive vermindert werden konnten. Es bleibt jedoch unklar, wie die Klimabedingungen eine hormonelle Reaktion hervorrufen könnten, und folglich den Fruchtfall in verschiedenen Stadien der Fruchtentwicklung verstärken.

Das vorliegende Forschungsprojekt besteht aus drei Studien. Das Ziel der ersten Studie war es, die einzelnen bzw. kombinierten klimatischen Faktoren, die den vorzeitigen Fruchtfall bei Mango induzieren, zu bestimmen, bzw. durch den Einsatz von regelmäßiger Bewässerung diesen Stressoren entgegenzuwirken und somit den Fruchtfall zu reduzieren. Ziel der zweiten Studie befasste sich mit den möglichen Auswirkungen der Behandlung mit Pflanzenwachtumsregulatoren und deren Potential den vorzeitigen Fruchtfall kurz nach der Blüte und während des frühen Entwicklungsstadiums der Mangofrüchte zu vermindern, bzw. deren Effekte mit den Ergebnissen des Bewässerungsexperiments zu vergleichen. Im dritten Teil des vorliegenden Forschungsprojekts wurden die morphologischen Veränderungen in der Abszissionszone von Mangostielen während des Fruchtfalls untersucht. Die Untersuchungen des Auxins-Exports der zweiten und dritten Studie dienten hauptsächlich dem genaueren Verständnis über den Auxinfluß aus der sich entwickelnden Frucht und somit der endogenen Hormonkonzent-

ration im Trenngewebe über die kritische Zeit des Fruchtfalls und deren Schlüsselrolle im Abszissionsprocess, besonders in Abhängigkeit von der Bewässerung.

Die entsprechenden Forschungsarbeiten wurden in einer Plantage in der Nähe der Stadt Yen Chau in den Jahren 2007, 2008 und 2009 durchgeführt. Der Versuchsaufbau beinhaltete jeweils 20 zufällig ausgewählte 10-jährige Mangobäume der Sorten 'Hôi' und 'Trôn'. Für das Beregnungsexperiment wurden jeweils die Hälfte der Bäume in 3-tägigen Abständen mittels Mikrosprinklern bewässert, wobei die verbliebenden Bäume als nicht-bewässerte Kontrolle dienten. Zur Evaluiering der Sprühapplikation der Pflanzenwachstumsregulatoren wurden separat 3 Bäume in 2008 bzw. 6 Bäume im Versuchsjahr 2009 ausgewählt. In beiden Experimenten wurden 10 Infloreszenzen zufällig über den Baum verteilt etikettiert und zweimal pro Woche über die Fruchtfallperiode hinweg ausgezählt. Für die hormonellen Untersuchungen wurden Früchte ab stecknadelgröße von bewässerten und nicht-bewässerten Bäumen eingesammelt und auf Diffusionspuffer gesetzt. Die Bestimmung der diffusiblen IAA-Konzentration erfolgte mittels radioimmunologischer Methoden. Ferner, um die morphologischen Veränderungen innerhalb der Abszissionszone der Mangofruchtstiele zu verfolgen, wurde entsprechendes Probenmaterial von bewässerten sowie Kontrollbäumen gesammelt, in einer Formalin-Essigsäure-Alkohol Lösung fixiert und unter Anwendnung einer modifizierten Dehydrierungs- und Einbettungstechnik mittels Vakuuminfiltration in Glykolmethacrylat-Methylmethacrylat eingebettet.

Die Ergebnisse der ersten Studie zeigten, dass der Beginn des vorzeitigen Fruchtfalls zeitgleich mit dem Aufkommen der heißen, trockenen, Winde ausgelöst wurde. Die Auswirkung der Bewässerung auf den Fruchtfall scheint von der vorhandenen Bodenfeuchtigkeit, bzw. dem verfügbaren Bodenwasserpotential abzuhängen.

In der zweiten Studie konnte nachgewiesen werden, dass durch Anwendung von Pflanzenwachstumsregulatoren der vorzeitige Fruchtfall in Mango reduziert werden konnte. Zwar wurden druch alle chemischen Behandlungen der Fruchtfall signifikant reduziert, die vorliegenden Ergebnisse zeigten jedoch deutlich, dass eine einmalige Applikation von N-(2-chloro-4-pyridyl)-N-phenylurea (CPPU) sowie 1-Naphtylessigsäure (NAA) den vorzeitigen Fruchtabfall am effektivsten verringerte und den Fruchtbehang bis unmittelbar vor dem Erntezeitpunkt am positivsten beeinflusste. Darüber hinaus zeigten die einmaligen chemischen Applikationen einen im Vergleich zum Bewässerungsexperiment gesteigerten durchschnittlichen

Fruchtbehang pro Infloreszenz. Dennoch konnten die über die Fruchtfallperiode hinweg festgestellten Veränderungen des IAA-Exports die positive Wirkung der Bewässerung auf die Fruchtretention nicht erklären.

Die mikroskopischen Ergbnisse der dritten Studie lassen darauf schliessen, dass dickere Fruchtstiele die bedarfsgerechte Kohlenhydratversorgung der Früchte während umweltbedingter kritischer Temperatur- und Wasserdampfdefizitbedingungen positiv beeinflussen. Darüberhinaus dürfte eine Kohlenhydratunterversorgung der Frucht möglicherweise in Kombination mit einem reduzierten IAA-Export aus der Frucht als Startsignal für den pflanzenphysiologischen Prozess der Ausbildung eines Abszissions-gewebes dienen.

Zusammenfassend lässt sich feststellen, dass die vorherrschenden Umweltbedingungen im Norden von Vietnam, insbesondere die heißen, trockenen Winde während der frühen Fruchtentwicklungsphase einen vorzeitigen Fruchtfall bei Mango verursachen. Die hier beschriebenen fruchtphysiologischen Arbeiten zum vorzeitigen Fruchtfall bei Mango führten zu der Entwicklung wirksamer Fruchtbehandlungsstrategien wie dem Aufbau eines Bewässerungssystems und/oder dem Einsatz von Pflanzenwachstumsregulatoren, um ungünstige Umweltbedingungen zu überwinden, und um die Reaktionen der Planzen zu reduzieren oder sogar zu unterbinden, die mit dem verfrühtem Fruchtfall in Verbindung stehen. Die vorgelegten Ergebnisse legen nahe, dass der erntenahe Fruchtbehang durch die Errichtung eines Bewässerungssystems gesteigert werden kann; wobei solche Systeme für Bauern in den Anbaugebieten eine große Investition darstellen. Eine Behandlung mit Pflanzenwachtumsregulatoren, bedingt durch die Einfachheit der Applikation in Kombination mit der chemischen Wirksamkeit, stellt eine vielversprechende Alternative dar, um den exzessiven vorzeitigen Fruchtbfall beim Mango zu verhindern. Diese könnte einen Beitrag dazu leisten, einen ökonomisch rentableren Anbau von Mango in dieser Region zu ermöglichen.

1. General Introduction

1.1 Mango

1.1.1 Mango in the World

Mango (*Mangifera indica* L.) belongs to the order Sapindales in the family of *Anacardiacea* which is a family of mainly tropical species, with a few representatives in temperate regions. Apart from mango, cashew, spondias and pistachio are other well-know crops of the family. The classification of *Mangifera* species comprise of 69 species according to Kostermans and Bompard (1993) which at least 26 of this species bear edible fruits primarily found in south-east Asia. Nowadays, mangos grow in over 100 countries from the equator to close to 36° latitudes, indicating both the great adaptability to different environments and cultural management techniques in the different producing countries (Sauco, 1997). Producing areas can be grouped in 6 main regions; (1) USA (Florida), (2) Mexico, (3) West Indies (Caribbean Islands), (4) South America, (5) Africa/Arabian Peninsula, and (6) Indian subcontinent and Indochina. However, by continents, Asia is the main producer with approximately 77% of the total world production, followed by America with nearly 14% and Africa with 9%, respectively (Sauco, 2004). According to FAO (2010) India, China, Thailand, Mexico, Pakistan, Brazil, Philippines, Indonesia, Nigeria and Vietnam are the leading producing countries. In fact, these ten countries cover 85% of the entire world production (Gunjate, 2009) but India accounts for 55% of the total mangoes produced worldwide (Naidu and Naidu, 2009). However, new mango plantings occurred mainly in China, Brazil, the Philippines and Vietnam (Sauco, 2004). Mango is one of the most important tropical fruits worldwide in terms of production and consumer acceptance (FAO, 2010). The fruit, morphologically classified as drupe, is the 5th most important fruit crop in the world (after citrus, grapes, bananas and apples). Demand for mangoes is presently rising in developed countries; however, the vast majority of mango is sold in local markets and consumed domestically (Santoso, 2000). The mango tree is not solely used for fruit production; it also yields wood, forage, and has several medicinal uses in eastern Asia (Kostermans and Bompard, 1993). The fruit has a high nutritional value and represents an important source for Vitamins A and C as well as for potassium and ß-carotene (Ribeiro *et al.*, 2008). Due to its availability at affordable price, the fruit is important part of the human diet

even economically weaker part of the society. This indicates its importance as carotene-rich nutritive source of Vitamin A in most developing tropical and subtropical countries (English and Badcock, 1998).

1.1.2 Mango in Vietnam

In Vietnam mangos are grown mainly for national consumption. From 1998 to 2008, Vietnam increased mango production from 18.0500 t to 370.000 t with an average yield of about 7 t/ha in 2008 (FAO, 2010). In the period 2001 to 2006, due to increasing demand, planted fruit area grew averagely 5% per annum, reaching more than 774,000 ha in 2006 (IFPRI, 2001). Fruit trees were mostly planted in the Mekong Delta, the Northeast and the Southeast region. In the year 2010, the target is to achieve one million hectares of orchards (Khoi and Tri, 2003). Mango is one of the most important subtropical fruit crops of Vietnam after longan, litchi, rambutan, citrus which account of 73% of the area planted with fruit crops in Vietnam (IFPRI, 2001). In Vietnam mangoes have been traditionally cultivated in the central and southern parts of Vietnam. Main production areas are DongNai (22.000 ha), BaRia–VT (8300 ha), Dong Thap (8.000 ha) and Tien Giang (5.000 ha) (FAO, 2004). Until the mid 1990s the Mekong Delta with its 13 provinces, accounted for over half of the total fruit area in the country. However, since 1995 the fruit area continues to expand less rapidly than in other regions, so its share of the national total has fallen to 38% (IFPRI, 2001). In contrast, fruit production areas in the Northern part have grown about 23 %, which reflects the growing local demand for fruit in Hanoi and bordering Chinese provinces in the North. According to AGROINFO (2008) the average retail price of fresh mango was more than 18,500 VND/kg in 2008, thus increased by 21% compared to 2006.

‘Xoai’ is the Vietnamese term for mango. It is customary to use in front of mango cultivars in Vietnam. The major cultivars predominately grown in the South are ‘Xoai Cat Hoa Loc’, ‘Xoai Cat Chu’, ‘Xoai Hon’ and ‘Xoai Buoi’ (FAO, 2004). The most widespread mango varieties are the ‘Cat Hoa Loc’ and the ‘Buoi’ variety with around 30 % and 27 % of producers, respectively (IFPRI, 2001).

Mango is typically not commercially grown in Northern Vietnam and, due to traditional extensive cultivation and lack of experience in modern orchard management practices, tree

productivity is generally low. However, with increased security of land tenure and grower training seminars by governmental institutions (Huong, 2007), production areas have increasingly expanded to previously less commercially developed fields of the provinces such as Ha Giang, Lai Chau and Son La. In the later province mango production occurs in particular in the districts Mai Son, Phu Yen, Song Ma and Yen Chau district (Fig. 1).

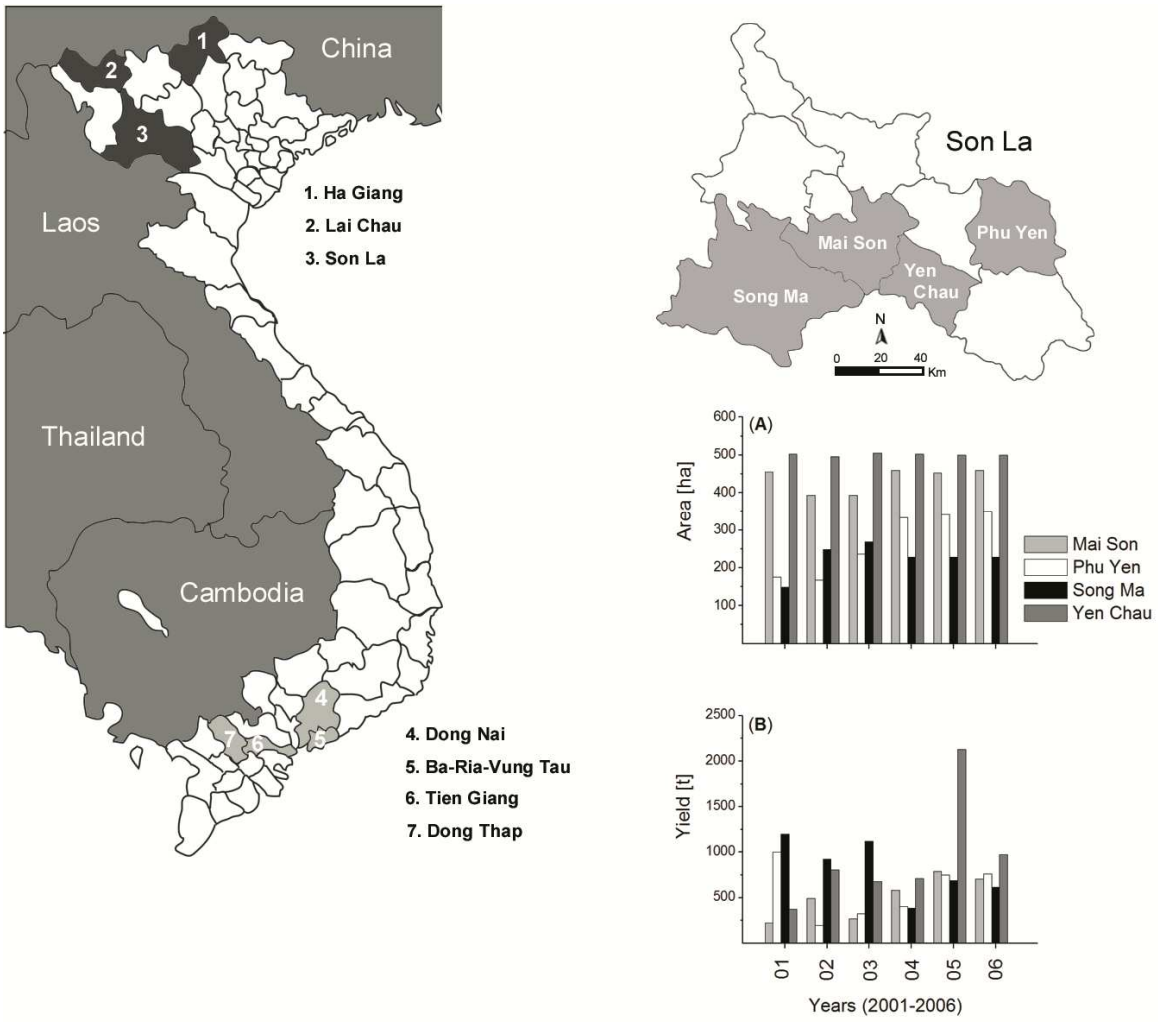

Figure 1: Mango production areas by provinces in South and North Vietnam. In (A) area and (B) yield of mango plantations in Son La are shown by provinces and districts.

Mango has a long history of cultivation in Son La province and ranks behind plum and longan at 3rd position in terms of planted area with 200% increased yield performance between 1993 and 2006 (Yen Chau, 2008). The local cultivars 'Hôi' and 'Tròn' are widely planted by farm-

ers of different ethnic minorities in Yen Chau. With more than 500 ha, Yen Chau district supplies annually approximately 800 to 1000 t of mango to local markets (Huong, 2010). Thus, there is a rapid transition from local to national market-orientated production, due to the good taste and aroma of the fruit.

Nevertheless, mango growers are afflicted with severe yield loss due to premature fruit drop following fruitlet abscission as natural process which has been described for various mango cultivars (Guzman-Estrada, 1996). Premature fruit drop can be caused by several factors, like for example lack of pollination or embryonic degeneration (Beyschlag *et al.*, 1992; Lovatt, 1997; Nunez-Elisea and Davenport, 1983; Singh, 1997; Singh, 1961; Treharne *et al.*, 1985; Yuan and Huang, 1988). Moreover, mango crop load in the previous season (Mukherjee, 1953; Pandey, 1989), pest and disease pressure (Arauz, 2000; Estrada *et al.*, 2000; Pena *et al.*, 1998; Verghese *et al.*, 2005) and carbohydrate deficiency for fruit growth (Burondkar *et al.*, 2000; Chacko *et al.*, 1995) have also been linked to premature fruit abscission. In addition, unfavourable weather conditions combined with or intensified by genetic predisposition or insufficient fertilization may also be crucial factors in premature fruit drop in mango (Chacko *et al.*, 1970; Lakshminarayana and Aguilar, 1975; Sukhvibul *et al.*, 2005).

Mango production in Northern Vietnam is typically characterised by adequate air temperature, low humidity (FAO, 2004) and well-defined periods of drought at pre-flowering time (Phan, 2005). Low temperatures during anthesis have been shown to reduce the number of hermaphrodite flowers in mango (Sukhvibul *et al.*, 1999a; Sukhvibul *et al.*, 1999b). This was suggested earlier by Issarakraisila and Considine (1994), demonstrating that air temperatures below a critical threshold of 10°C during anthesis or at an early stage of fruit development (Whiley *et al.*, 1988) might prevent fertilization or ovule development. Additionally, according to Nunez Elisea and Davenport (1983), higher temperatures throughout anthesis and early fruit set leads to embryo abortion. This is confirmed by observations of reduced pollen viability, thus low fruit set, within periods of high temperature during the critical period of flowering (Issarakraisila and Considine, 1994). Heat associated with hot, dry winds also has detrimental effects on pollination and fruit set in avocado (Wolstenholme, 2002). Moreover hot dry winds in Western Australia negatively affect fruit set in mango (Johnson and Parr, 1999), which might be accelerated by periods of water deficiency. Although several studies indicate that irrigation increases yields in subtropical evergreen fruit trees (Koo, 1979; Lahav and

Kalmar, 1977, 1983), there are conflicting reports about the need for irrigation in mango. However, the degree of fruit drop and final fruit retention are mainly dependent on variety, although water and nutritional stress generally increases fruit drop. The intensity of abscission of premature mango fruit varies considerably with the developmental stage of the fruit. Several studies indicated that, depending on cultivar, most of the fruit drop occurred at around 7 (Lam *et al.*, 1985), 28 (Nunez-Elisea and Davenport, 1983) or 25 to 50 days after full bloom (Guzman-Estrada, 1996), respectively. Fruit retention seems to depend on plant signals sent from the fruit to the tree and thereby suppressing the activation of the abscission zone and the ability of the fruit to compete for carbohydrates (Agusti *et al.*, 2002). The present study was carried out to investigate premature fruit drop of mango in Northern Vietnam. Environmental cues such as unfavourable weather conditions during early fruit set, are suggested to be the key factors for premature abscission.

1.2 Fruit abscission in Mango

1.2.1 Pollination and Fertilisation

The understanding of mango flowering in the tropics and subtropics is essential to efficiently utilize crop management systems which extend both the flowering and crop production season (Chacko, 1991; Whiley *et al.*, 1991). Flowering and fruit set are the most critical events after establishing a tree crop. In nature, mango trees produce large numbers of flowers of which only a small proportion set fruit. There are two different kinds of flowers; male, which have one or more stamens and staminodes and a completely abortive or reduced pistil, and hermaphrodite flowers with one or more fertile stamens and functional female organs (Kostermans and Bompard, 1993). Only perfect (hermaphrodite) flowers are able to set fruit, however, the number of perfect flowers per inflorescence varies between cultivars or is variable from year to year, depending on the location of the inflorescence in the tree (Singh and Arora, 1965; Singh, 1954; Singh, 2005). Mango flowers are small, 5-10 mm in diameter, and according to Mukherjee (1953) both male and hermaphrodite flowers offer nectar and pollen. Depending on the cultivar, fertilization occurs within 48 to 72 h after pollination (Davenport and Nunez-Elisea, 1997; Singh, 1997). However, a successful fruit set is dependent on the use of selective pollinizers (Ram *et al.*, 1976) and pollen viability has been considered to be a major

factor limiting yields in mango (Davenport and Nunez-Elisea, 1997). Cross-pollination increased fruit set and retention in most cases (Singh, 2005) because self-incompatibility has been reported for several mango cultivars, however, cross-incompatibility between certain cultivars was also described (Ram *et al.,* 1976). According to Singh (2005) mango flowers are generally pollinated by insects, predominantly by diptera (Anderson *et al.,* 1982; Singh, 1997). The efficiency of pollination and fertilization defines fruit set in mango; however, environmental cues, particularly air temperature, strongly determines timing and intensity of flowering and fruit set, and later on influences fruitlet abscission.

1.2.2 Air temperature

Mango trees are adapted to a wide range of subtropical and tropical climatic and edaphic conditions (Schaffer *et al.,* 1994); however, favourable weather conditions are crucial for flowering and fruit retention (Chacko *et al.,* 1970; Lakshminarayana and Aguilar, 1975; Sukhvibul *et al.,* 2005). In particular, temperature extremes and strong winds during pollination and fruit set, are known to negatively affect the crop. Low temperatures during anthesis have been shown to reduce particularly the number of hermaphrodite flowers in mango (Sukhvibul *et al.,* 1999a; Sukhvibul *et al.,* 1999b) which was suggested earlier by Issarakraisila and Considine (1994). It was demonstrated that air temperatures below a critical threshold of 10°C during anthesis or at an early stage of fruit development might prevent fertilization or ovule development (Whiley *et al.,* 1988). Additionally, according to Nunez Elisea and Davenport (1983), high temperatures throughout anthesis and early fruit set leads to embryo abortion. This was confirmed by observations of reduced pollen viability, within periods of higher temperature during the critical period of flowering (Issarakraisila and Considine, 1994). Fruit that set during periods of high temperature did not develop to maturity compared to those set during lower temperature periods (Chacko, 1984). For avocado it was shown that heat associated with dry winds has detrimental effects on pollination and fruit set (Wolstenholme, 2002). For mango grown in Western Australia hot, dry winds negatively affect fruit set (Johnson and Parr, 1999), which might be accelerated by periods of water deficiency. Temperatures above 30°C increases vegetative growth in some tropical and subtropical trees including mango (Higuchi *et al.,* 1999; Menzel and Simpson, 1986; Whiley *et al.,* 1989). Optimum temperature range for the growth of mango trees has been reported to be between 24°C and 30°C

(Mukherjee, 1953; Whiley *et al.*, 1989). The significance of tree water status for flowering, thus fruit set, and possible interactions with temperature is not known (Pongsomboon *et al.*, 1997). In tropical climates flowering is more likely problematic, where the dry period appears to be the main flowering trigger, compared to subtropics, where winter cold is the main environmental cue (Schaffer *et al.*, 1994).

1.2.3 Water relations

Typical mango environments in the tropics and subtropics indicate extreme water deficits due to prolonged periods of droughts. Generally, plants with laticifers or resin ducts are adapted to drought by modulating the water status of the plant, thus reducing the loss of water by transpiration (Downton, 1981; Kallarackal *et al.*, 1990). The differentiation, structure and distribution of resin ducts in trunks, shoots, leaves and fruit exocarp of mango have been reported to be in close association with the vascular tissue (Joel, 1980, 1981; Joel and Fahn, 1980). The mango tree can withstand considerable periods of water stress and is considered to be drought tolerant, however, during the reproductive phase a higher amount of water supports fruit set and fruit retention (Whiley and Schaffer, 1997). Although several studies indicate that irrigation increases yields in subtropical evergreen fruit trees there are conflicting reports about the need for irrigation in mango (Koo, 1979; Lahav and Kalmar, 1977, 1983; Nagle *et al.*, 2010). It was also described that a water deficit at pre-flowering time might enhance flowering intensity, however, detrimental effects at the stage of flowering, pollination and fruit set were observed (Chacko, 1984; Gonzalez *et al.*, 2004; Lu *et al.*, 2000). It was reported that fruit abscission on trees under severe drought stress was enhanced (Schaffer *et al.*, 1994). Fruit drop in mango at an early developmental stage is associated with low soil moisture and excessive loss can be prevented by adequate irrigation, particularly during flowering and the first six weeks after fruit set (Larson *et al.*, 1989; Spreer *et al.*, 2007). Irrigation also can increase individual fruit size and overall yield as described by Spreer *et al.*, (2007) and Whiley and Schaffer (1997). However, mango orchards in Northern Vietnam generally are not irrigated properly (Gunjate, 2009), especially during the critical time of flowering and early fruit development, which takes place during the hot and dry season.

1.2.4 Hormonal regulation

Hormonal regulation of fruit abscission has been observed in many fruit crops (Addicott, 1968; Roberts *et al.*, 2002) and endogenous hormones play a major role in fruit growth and fruit drop in mango (Chattha *et al.*, 1999). Generally, they function at relatively low ppm-concentrations in fruits to regulate the formation and activation of the abscission zone within the separation layer (Singh *et al.*, 2005). Like in other fruit crops, fruit abscission in mango is regulated by a complex interaction of phytohormones, which protect the abscission zones or induct the separation process. The efficiency of various phytohormones and their synthetic analogs, called plant growth regulators (PGR) for the control of fruit drop in mango have been conducted in several studies (Chen, 1983; Notodimedjo, 2000; Oosthuyse, 1995; Ram *et al.*, 1983).

1. Auxin: The major common auxin in plants is indole-3-acetic acid (IAA). IAA is one of the principal hormones that control plant growth and development. All natural auxins are found in plants either as free acids and/or in conjugated forms. IAA is synthesized from tryptophan or indole primarily in leaf primordia, young leaves and in developing seeds (Davies, 2004). In combination with other hormones, auxins are involved in the development of vascular tissue and regulation of plant growth primarily through control of cell division and elongation. (Abel and Theologies, 1996). Additionally, the hormone is involved in abscission process mainly by facilitating the separation of organs from plants (Osborne, 1979; Sexton and Roberts, 1982). IAA is transported basipetally from cell to cell mainly in the vascular system in specialized phloem parenchyma cells which are in contact with vascular bundles (Ross, 1998). Higher fruit drop intensity is generally associated with periods of lower auxin concentration in mango fruits (Singh, 2005). Thus, continuous auxin synthesis and basipetal transport to the abscission zone is critical for maintenance of plant organs, including the fruit (Davenport *et al.*, 1980; Roberts and Osborne, 1981). Activation of the abscission zones (AZ) of leaves, flowers and fruit appear to be governed by the interaction of auxin and ethylene (Gonzalez-Carranza *et al.*, 1998). The synthetic auxins which have been most effective in reducing mango fruit drop are naphthaleneacetic acid (NAA) and 2,4-dichlorophenoxyacetic acid (2,4-D). Together with auxins, cytokinins is another group of plant hormones which regulate cell division and cell expansion.

2. Cytokinins: Cytokinins (CKs) are adenine derivates represented by kinetin, zeatin and 6-benzylaminopurine. The most common cytokinin in plants is zeatin (Davies, 2004). The majority of adenine-type cytokinins are synthesized in the roots tips, cambium and developing seeds (Davies, 2004). CKs are a class of plant hormones that promote cell division in plant roots and shoots. They are primarily involved in cell growth and differentiation; however also affect apical dominance, auxillary buds growth, and leaf senescence. CKs transport is via the xylem from roots to shoots. CKs play an important role in inducing fruit set of several plants (Letham, 1967; Skoog and Armstrong, 1970). Also in mango, cytokinins play a key role in cell division and cell enlargement and have been found in both pericarp and seed, respectively (Ram *et al.,* 1983; Singh, 2005). Low cytokinin concentrations in developing mango fruits have been correlated with fruit drop and cessation of fruit growth (Ram, 1983). The application of synthetic cytokinin has also demonstrated improved fruit set and retention, suggesting that cytokinins play a role in establishing relatively high crop loads. Particularly post-bloom treatments with CPPU plus gibberellins (GA_3) at early stages of fruit growth promoted fruit set (Chen, 1983; Oosthuyse, 1995). Cytokinin is required for fruit development and retention, however, the role of cytokinin in abscission still remains inconclusive (Davenport and Nunez-Elisea, 1997).

3. Gibberellins: Gibberellic acid (GAs) are a family of compounds based on the *ent*-gibberelline structure. GA_3 was the first gibberellin to be structurally characterized. There a currently over 126 members identified and characterized from plants, fungus and bacteria (MacMillan, 2001). Biosynthesis of GAs takes place in seeds, young leaves, young shoots and also in roots and is probably transported in xylem and phloem to the target organs. In mango, major endogenous GAs have been reported in apical buds, leaves and vegetative shoots (Davenport *et al.*, 2001). Beside from being involved in growth and development of fruit, the direct role of GAs to the onset of abscission is ambiguous (Chacko *et al.,* 1970; Davenport and Nunez-Elisea, 1997; Ram, 1983). Several classes of plant growth retardants have been characterized to interfere with early and late gibberellin biosynthetic pathway (Rademacher, 2000) and have been therefore investigated to improve fruit retention in mango. However, the efficacy of exogenously applied GA_3 to pre- and post-anthesis inflorescences to enhance fruit set and retention have been inconclusive (Oosthuyse, 1995; Singh *et al.,* 2005).

1.2.5 Carbohydrate deficiency

In fruit trees a reduced carbohydrate availability due to heavy and prolonged flowering, subsequently limits nutrition to satisfy the demand on fruit set and development (Goldschmidt, 1999; Whiley *et al.*, 1996). Consequently, abscission of flowers and fruits at an early stage of development adjust fruit number to the capacity of carbohydrate supply which the tree can provide (Agusti *et al.*, 1982; Gomez-Cadenas *et al.*, 2000). In mango, carbohydrate storage capacity is one of the important factors that determine the number of fruit that the tree can nurture to maturity (Davie *et al.*, 2000; Normand *et al.*, 2009). However, competition for nutrient resources is also a factor for premature fruit abscission (Stephenson, 1981; Stephenson and Gallagher, 1986). Internal competition between vegetative growth, flowers and fruit have been proposed as a controlling factor in flower and fruit abscission (Kozlowski, 1992). In mango, the availability and distribution of photosynthates during fruit set and development was suggested (Davenport and Nunez-Elisea, 1997) as one of the reasons for fruit abscission and this is also reported for other fruit crops, such as apple (Berüter and Droz, 1991; Stopar, 1998).

1.3 Abscission zone

1.3.1 Morphology

Abscission of plant organs occur at predetermined positions called abscission zones (AZs) (Gonzalez-Carranza *et al.*, 1998; Sexton and Roberts, 1982; Taylor and Whitelaw, 2001) and previous microscopic studies of fruit AZs in several fruit crop species indicated, that cells affected by the separation process are morphologically distinguishable prior to abscission from neighboring cells (Pandita and Jindal, 1991; Rascio *et al.*, 1985; Stösser *et al.*, 1969a). In mango, the pedicel AZ exist from flowering stage to fruit maturity (Singh, 1961) and can often be ascertained externally as a thin circular groove at the pedicel close to fruit base (Baird and Webster, 1979; Barnell, 1939; Singh, 1961). Adventitious zones are defined as functional abscission zones (Addicott, 1982), and have been shown to form in stem, petiole, pedicel or phyllomorph tissue (Roberts *et al.*, 2000).

The terms “abscission zone” and “abscission layer” are used in accordance with the definition of Shiraishi and Yanagisawa (1988) and Gawadi and Avery (1950). Generally, “abscission zone” refers to the tissue region through which the “abscission layer” forms. The term “abscission layer” refers to one or two cell layers through which mechanical break occurs. “Abscission layer” and “separation layer” are synonymous terms (Roberts *et al.*, 2000; Taylor and Whitelaw, 2001); however, in this study “separation layer” is used.

The numbers of cell rows that comprise the AZ vary markedly with species, plant organ and the site of cell separation (Sexton and Roberts, 1982). Abscission zone in leaves e.g. of *Phaseolus* or *Olea* have 10 to 12 cell rows and *Sambucus nigra* have 20 to 30 cell rows (Osborne and Sargent, 1976), whilst AZ of tomato flower may consist of 5-10 rows of cells (Roberts *et al.*, 1984). In peach AZ consists of two rows of cells (Bonghi *et al.*, 2000) whilst Rascio *et al*,. (1985) indicated several rows of cells. According to Singh (1961) the AZ in the mango pedicel has 8 to 10 cell rows. The AZ of most fruit crops, including mango, have subtle characteristics that allow the general region to be distinguished before onset of abscission (Baird and Webster, 1979; Berüter and Droz, 1991; Rascio *et al.*, 1985; Stösser *et al.*, 1969b). Typically, cells of the AZ are small, square-shaped, closely packed and with enlarged nuclei and mitochondria and dense cytoplasm (Costa *et al.*, 2006; Henderson *et al.*, 2001; Osborne and Sargent, 1976; Sexton and Roberts, 1982), whereas vascular fibers are unusually small or absent (Baird and Webster, 1979). However, Singh (1961) reported of elliptical cells, rich of cytoplasm and arranged in irregular rows in the AZ of mango which widens with the development of the fruit. Moreover, the cells within the AZ of several plant species accumulate starch (Brown and Addicott, 1950; Gilliland *et al.*, 1976; Shiraishi and Yanagisawa, 1988).

1.3.2 Abscission process

Abscission of premature fruits in mango is a natural mechanism (Lam *et al.*, 1985; Singh, 1960) which is particularly high (90% fruit shedding) during the first 3 to 4 weeks after pollination (Nunez-Elisea and Davenport, 1986; Singh, 1960). Fruit drop might be caused by several factors, such as nutrient deficiency, disturbances in embryogensis and/or embryo abortion, sink competition between fruits, and abiotic and biotic stressors (Chadha, 1993). Generally, the cell separation process does not involve the entire AZ. The cells within the AZ that

are involved in the abscission process by rapid reduction in cell integrity (Sexton and Roberts, 1982) have been identified as separation layer (Addicott, 1982; Roberts *et al.,* 2000; Sexton, 1994). In mango, just the separation layer is mentioned to initiate fruit drop without precise information on how many cell rows are involved (Nunez-Elisea and Davenport, 1986); however, in cherry fruit and leaflet of olive 2 to 8 longitudinally rows of cells define the fracture line (Polito and Lavee, 1980; Stösser *et al.,* 1969a).

1.3.3 Role of phytohormones and assimilates

The activation of the separation layer involves several mechanisms in physiological response to such as e.g. auxin (Addicott, 1970). The reduction of the basipedal IAA transport results in the abscission of the organ e.g. fruit and leaf (Bangerth, 1989), which may be mediated by low assimilate allocation to the fruit, which was found in several fruit tree species *(*Agusti *et al.*, 2002; Berüter and Droz, 1991; Patrick, 1979; Stopar, 1998; Wertheim, 2000). Else *et al.*, (2004) assumed that increased basipetal auxin transport at periods of rapid cell expansion might promote translocation of assimilates into the fruit during extensive growth phases (Blanusa et al., 2005). Indeed, this has been intensively studied in apple (Berüter and Droz, 1991), pistacchio (Nzima *et al.*, 1999), cherry (Atkinson *et al.*, 2002) and orange (Ruiz *et al.*, 2001), however, starch content in the AZ of the pedicel is contradictorily discussed (Baird and Webster, 1979; Bornman *et al.*, 1966; Shiraishi and Yanagisawa, 1988).

1.4 Working hypothesis

The research focus is aimed at determining the plant processes which respond sensitively to specific environmental and/or crop management factors and consequently lead to fruit abscission. The basic working hypothesis is that premature fruit drop is caused by environmental stimuli like excessive air temperature and vapour pressure deficit (VPD) conditions which in turn result in a reduced basipetal auxin export from fruit and trigger the activation of the abscission layer in the pedicel.

The research objectives were:

1. To determine several environmental cues which induce the abscission process; particularly the hot, dry prevailing winds which occur at the beginning of fruit set in mango.

2. To examine the effect of irrigation and varying PGR spray applications, to evaluate their potential for controlling premature fruit drop.

3. To determine time-dependent diffusible IAA-export from fruits of irrigated and non-irrigated mango trees as a likely plant mechanism involved in fruit abscission.

4. To understand the morphological changes within the abscission zone during the fruit drop process.

The research activities were integrated in an international research cooperation "*Sonderforschungsbereich 564"* (DFG) entitled *"Sustainable land use and rural development in mountainous regions of Southeast Asia"* between Vietnam and Germany.

2. Material and Methods

2.1 Plant material and experimental design

This study was carried out on a mango (*Mangifera indica* L.) plantation in the Tu Nang commune at Yen Chau (20°56'10.66 N, 104°28'13.12 E) between mid-February and the end of May for the three consecutive growing seasons from 2007 to 2009. The soil was classified as Lixisol (FAO, 2006). The experimental design consisted of 40 randomly selected seedling mango trees, 20 trees each of cvs. 'Hôi' and 'Tròn', planted at an average distance of 10 x 10 m. Half of the selected mango trees of each cultivar were irrigated, the other half served as non-irrigated controls. The trees were regularly pruned and fertilized according to commercial practice. Pruning was carried out 3 months after harvest, just prior to the onset of the new vegetative flush. Fungal diseases were controlled by spraying biweekly Difenoconazole (75 ml 100 l^{-1}) and Propineb (0.1 kg 100 l^{-1}) throughout flowering and early fruit growth stages. The insect control from the 'pinhead' stage onwards was carried out by spraying Profenofos (125 ml 100 l^{-1}) and Lambda-cyhalothrin (50 ml 100 l^{-1}). Full bloom was recorded on February 15, 2007, March 28, 2008, and February 6, 2009, respectively. Each year, 10 inflorescences per tree were labeled at full bloom. Number of fruits within each labeled inflorescence was counted twice per week until the end of the fruit drop period at 20, 30 and 43 days prior to harvest in 2007, 2008 and 2009, respectively.

2.2 Experimental treatments

2.2.1 Irrigation

An irrigation system was set up at the end of 2006 and started at the onset of flowering in 2007 56 days before flowering in 2008 and 37 days before flowering in 2009, respectively. The irrigation was discontinued after final fruit set. For the study year in 2007, soil moisture profile data was available starting from 25 dafb, respectively. Crop water requirement was calculated based on the Penman-Monteith equation by using the CROPWAT computer model (Allen *et al.*, 1998). The calculations assumed a crop coefficient (kc) of 0.8 for mango (Spreer *et al.*, 2007). Average evapotranspiration (ET_0) for 2007 was based on climatic data obtained

from FAO for Northern Vietnam, whereas for 2008 and 2009 it was derived from the data obtained from the weather station. The irrigation system consisted of a spring and rain water fed water tank and two main tubes (16 mm diameter, Netafilm, Israel) from the tank to the experimental site. Micro-sprinklers (Gyro Net LR 120, 0.15 MPa, Netafilm, Israel) were inserted to 6.5 mm diameter irrigation tubes (Netafilm, Israel) and those were connected to the main tubes and placed at 30 cm distance to the tree trunks. Irrigation was performed at 3-day-intervals for 45 minutes with a nominal rate of 90 L h^{-1}, respectively. The amount of rainwater was measured, but was not considered for the irrigation management. Soil moisture at a depth of 10, 20, 30 and 40 cm, respectively, was measured at 7-day-intervals throughout the fruit drop period in all experimental years by using a profile probe (PR2, Delta-t Devices Ltd, Cambridge; UK) at a distance of 50 cm to the tree trunk for 5 representative trees per treatment. The profile probe was connected to a hand-held data-logging device (Moisture meter HH2, Delta-t Devices Ltd, Cambridge; UK).

2.2.2 Plant growth regulators

For plant growth regulator spray application trial, 3 trees of each cultivar in 2008 and of each cultivar 6 trees in 2009 were used. The PGR experiment in 2008 was performed in the same orchard as the irrigation experiment; however, the PGR experiment in 2009 as the orchard used for the irrigation experiment was carried out in a neighbouring mango orchard with similar exposure and soil features. All trees were pruned and fertilized according to commercial practice. Plant protection against fungal diseases and pest injuries were conducted according to Huong (2004). The applications comprised following treatments to non-irrigated trees:

(1) CPPU 10 ppm

(2) CPPU 10 ppm + GA_3 40 ppm

(3) CPPU 10 ppm + GA_{4+7} 40 ppm

(4) NAA 40 ppm + Etalfix® Pro 5 ppm

(5) Combi I (1) + (2) + (4)

(6) Combi II (1) + (3) + (4)

(7) Control (distilled water + Etalfix® Pro 5 ppm).

The concentrations were as follows:

CPPU (Sitofex® 10 EC; AlzChem) 10 ppm equal to 1 ml^{-1} L H_20

GA_3 (ProGibb® 40; Valent) 40 ppm equal to $0.1g^{-1}$ L H_20

GA_{4+7} (ProVide® 10 SG, Valent) 40 ppm equal to $0.4g^{-1}$ L H_20

NAA (Rhodofix®; Syngenta) 40 ppm equal to $0.4g^{-1}$ L H_20

Application dates were in 2008 for CPPU at April 9 (12 dafb), GA at April 11 (14 dafb) and NAA at April 17 (20 dafb). In experimental year 2009, applications were conducted for CPPU at February 13 (7 dafb), GA at February 25 (19 dafb) and NAA at March 4 (26 dafb), respectively. In both years, applications of PGR dates were determined by semi-weekly fruit measurements. The chemicals were applied predawn due to decreased air temperature and duration of effect to selected inflorescences at flower and specific fruit size stages (Table 1) until run off using a pressure-compensated hand sprayer (Gloria, Typ 133, 1L/0.3 MPa, Witten, Germany). The pH of both gibberellin solutions was reduced by citric acid to a range between 4 and 5.5 pH. As wetting agent for NAA application 5ppm Etalfix® Pro (Syngenta) were used.

Table 1: Single and combined PGR applications in regard to specific fruit size stages.

Fruit stage	Fruit size (cm)	PGR application	Days after full bloom (dafb)
‚Pin-head'-stage	< 0.4 cm	(1), (5), (6)	12 dafb, 7 dafb
‚Pea'-stage	0.4 to 0.8 cm	(2), (3), (5), (6)	14 dafb, 19 dafb
‚Marble'-stage	1.5 to 2 cm	(4), (5), (6)	20 dafb, 26 dafb

2.3 Measurements

2.3.1 Environmental conditions

Ambient air temperature and relative humidity within the centre of the tree canopy were recorded with one micro-loggers (HOBO Onset, Pro v2; USA) in each of six irrigated and non-irrigated trees, respectively. The logger outputs were logged at 10-min intervals from full bloom until the end of the main fruit drop period. In addition, an automated weather station (Delta-T Devices Ltd. Cambridge, UK) was set up within the orchard for recording wind speed (A100L2), wind direction (W200P/L), pluvial precipitation (ARG100), global (total) and diffuse photosynthetic active radiation (PAR) and sunshine duration (BF3). Additionally, long-term records of air temperature and relative humidity from 1998 to 2009 for the months of the flowering and fruit drop period could be obtained from a weather station, based at Yen Chau, located 30 km from the study site. For determination of vapour pressure deficit (VPD) air temperature and relative humidity (RH), recorded by the HOBO loggers, were used to calculate the vapour pressure deficit according to the equations by Prenger and Ling (2000) and Snyder and Paw (2002).

(1) $VPD = VP_{sat} - VP_{air}$

with:

(2) $vp_{sat} = e^{(A/T + B + CT + DT2 + ET3 + FlnT)}$

where $A = -1.044x10^{4}$, $B = -1.129x10^{1}$, $C = -2.702x10^{-2}$, $D = 1.289x10^{-5}$, $E = -2.478x10^{-9}$ and $F = 6.456$ are constants, and T is air temperature in $T (°R) = T (°F) + 459.67$

and

(3) $vp_{air} = vp_{sat} * rh/100$

2.3.2 Soil moisture

Volumetric soil moisture content was measured using dielectric sensors (Theta Probe ML2x) at 10 and 20 cm soil depths at 30 cm distance to two irrigated and two non-irrigated trees, respectively. The output of each sensor was recorded at 1-min intervals and averaged over 30 min with a datalogger (Delta-T logger DL2e, Cambridge, UK). Manual soil moisture at a depth of 10, 20, 30 and 40 cm, respectively, was measured at 7-day-intervals throughout the fruit drop period in all experimental years by using a profile probe (PR2, Delta-t Devices Ltd, Cambridge; UK) at a distance of 50 cm to the tree trunk for 5 representative trees per treatment. The profile probe was connected to a hand-held data-logging device (Moisture meter HH2, Delta-t Devices Ltd, Cambridge; UK).

2.3.3 Fruit temperature

The surface temperature of 5 randomly selected fruit was measured on all treatment trees between 12 am to 2 pm at weekly intervals from full bloom until final fruit set by using a handheld infrared thermometer (Trotec TP6, Heinsberg, Germany).

2.3.4 Pedicel morphology

Fruit pedicels samples were cut longitudinally and trimmed with a razor blade to fit standard embedding moulds (1.5 x 1 cm). Samples were dehydrated in an ascending ethanol series (70 % for 2h, 90 % 3h, 96 % 3h and 96% overnight) and then by vacuum-infiltrated with glycol methacrylate (GMA) and methyl methacrylate (MMA). The embedding procedure was performed according to the methods described by Ruddell (1967) and Hermanns and Schulz (1981). Sections of 6 µm thicknesses were sliced using a rotation microtome (2050 Supercut, Reichert-Jung, Heidelberg, Germany) and placed with distilled water (stretching of sections) on glass microscope slides. The water was evaporated at 44.5 °C by a heating plate (Medax, Nagel GmbH Kiel, Germany) and slides were stored in a drying stove (Memmert GmbH Schwabach, Germany) at 41 °C. Afterwards, tissue sections were stained with haematoxylin for 30 min, rinsed with distilled water for 15 min and dried overnight at 35 °C. Sections were then immersed in xylene for two minutes and mounted with Eukitt (Kindler GmbH & Co. Freiburg, Germany). From total amount of 320 samples respectively, 160 fruit pedicels were

analyzed microscopically (Zeiss Axioskop, Germany) and micrographs were taken (Zeiss AxioCam HRc, Germany; software: Axiovision 3.1). Specimens were evaluated for pedicel thickness at the AZ, AZ length and AZ area, respectively by image analysis software Analysis 3.2 (Soft Imaging System GmbH Münster, Germany). Randomly, 100 cells in the AZ were evaluated for amount of starch grains and quantity was ranged in groups from 0 up to 10-12 starch grains per cell.

2.3.5 Plant hormones

To investigate IAA export of fruits of irrigated and non-irrigated mango trees, fruits were collected at weekly intervals from 18 to 60 dafb in 2008 and from 20 to 48 dafb for 2009, respectively. The fruits were placed with their stalk into the cavities of 24-well tissue culture plates (Greiner Bio-one, Germany), each cavity containing 3 ml phosphate buffer (0.05 M, pH 6.2). Assembled plates were incubated approximately close to 100% RH in darkness at approximately 20°C for 20 h. Individual fruit weights were recorded and diffusates were immediately frozen and kept at -20°C until further analysis. For IAA purification, frozen buffer solutions were gently melted, adjusted to pH 3 with acetic acid (HAc) and individually passed through previously activated Sep-Pak C_{18} cartridges (Waters, Germany). The cartridges were rinsed with 2 x 4 ml 0.01 M HAc, followed by 4ml 15% acidified methyl alcohol (MeOH) (0.01 M HAc). IAA fractions were eluted with 4 ml 40% acidified MeOH and aliquots of each sample were dried under vacuum, methylated with diazomethane and analysed in triplicates by Radio-Immuno-Assay (RIA) using polyclonal antibodies (Bohner and Bangerth, 1988).

2.4 Statistical analysis

Treatment effects on microclimatic soil and tree canopy parameters were evaluated by analysis of variance (ANOVA, P=0.05) using GenStat (13^{th} edition, Rothamsted, UK). Endogenous hormone concentrations were determined in 2 biological replications (4 fruits each) in 2008 and 12 biological replications (4 fruits each) per treatment and per sampling time, respectively. The effect irrigation treatments on IAA export was statistically evaluated by analyses of variance (ANOVA), P=0.05 using GenStat 10^{th} edition (Rothamsted, UK) to determine main treatment effects.

3. Results

3.1 Premature fruit drop induced by climatic factors

Premature fruit drop pattern: In 2007 irrigation had no effect on final number of fruit per inflorescence; however, fruit drop intensity was significantly different at most observation dates between the cultivars 'Hôi' and 'Tròn' (Fig. 2A). The cultivar 'Tròn' with averagely 24 fruits per inflorescence had a higher fruit set per inflorescence compared to 'Hôi'. In general, fruit abscised substantially from full bloom (15 February) until 'pinhead' to 'pea-size' stage at about 18 days after full bloom (dafb). 'Tròn' abscised approximately 70% of all fruits between 11 dafb and 18 dafb, while 'Hôi' in the same period, shed only 30%, followed by less intensive fruit drop for both cultivars until fruit reached marble-size at the end of March (46 dafb). Thereafter, fruit retention was relatively stable until harvest in mid May. At the end of the fruit drop period (53 dafb) 'Hôi' retained averagely threefold more fruits per inflorescence (0.6) than 'Tròn' (0.2) (Fig. 2A).

The season 2008 was characterized by a delayed flowering period (end of March) compared to flowering period normally occurring in early to mid February. Both cultivars reached full bloom at the same time. However, while flowering for 'Hôi' continued from 4 dafb to 11 dafb, weekly fruit counts revealed that 'Tròn' finished fruit set at 4 dafb respectively and started shedding (Fig. 2B). Generally, fruit abscission commenced at 'pinhead-size' for both cultivars. Unlike in 2007, irrigation reduced the intensity of fruit drop significantly in both cultivars. Crop load of non-irrigated 'Tròn' trees was reduced by about 75% at 39 dafb (0.4 fruits per inflorescence) whereas averagely 2 fruits per inflorescences were retained in irrigated trees. At harvest at the end of June (83 dafb in 'Tròn' and 90 dafb in 'Hôi'), irrigated trees of both cultivars had higher fruit retention with 1.6 fruits for 'Hôi' and 1.1 fruits for 'Tròn' per inflorescence compared to non-irrigated trees with 0.3 'Hôi' fruit and 0.1 'Tròn' fruit per inflorescence, respectively.

Full bloom in 2009 was at February 6, 9 days earlier as in 2007 and 59 days earlier as in 2008. The fruit drop period in 2009 was relatively short, being completed within 6 weeks after full bloom (Fig. 2C). With an average of 23 fruits per inflorescence at 6 dafb, cultivar 'Hôi' had

22% higher fruit set compared to ‘Tròn’ with 18 fruits per inflorescence. Generally, fruit drop was affected by cultivar and irrigation. Size of abscised fruit ranged from ‘pinhead’ to ‘pea’-size at the beginning of the fruit drop period until 20 dafb and reached ‘marble-size’ at 34 dafb. Throughout the fruit drop period the abscission pattern of irrigated and non-irrigated trees was similar, however, a significant difference between irrigated and non-irrigated trees occurred after 34 dafb. At harvest time, irrigated trees for both cultivars retained an average of 3 fruits per inflorescence, whereas non-irrigated ‘Hôi’ and ‘Tròn’ retained 0.6 and 0.4 fruits per inflorescence, respectively.

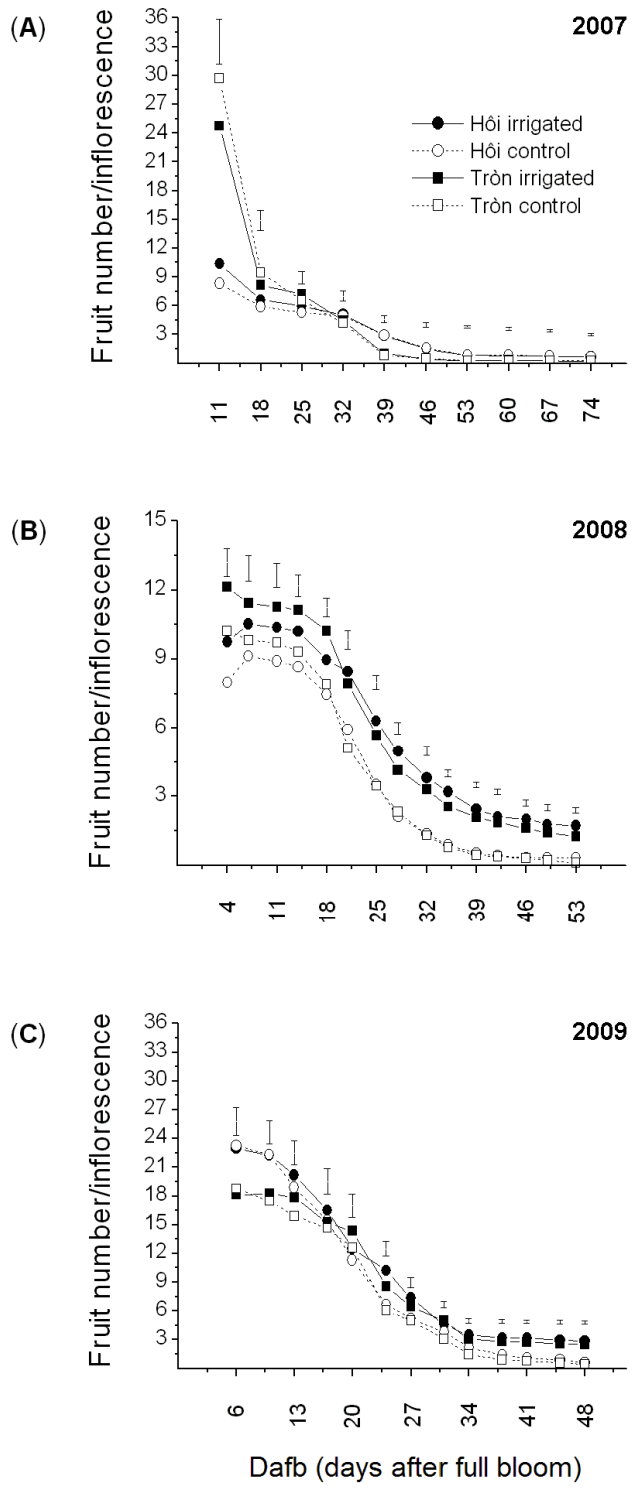

Figure 2: Seasonal fruit drop pattern of irrigated and non-irrigated mango cultivars 'Hôi' and 'Tròn', in 2007 (A), 2008 (B) and 2009 (C). Bars show the $LSD_{(0.05)}$ at each recording time.

Environmental condition: Wind direction and air temperature records over the 3-year seasonal fruit drop period indicated mainly hot winds prevailing from south-west to south-east direction. On 22% of all observation days, air temperature was ranging between 35 to 40°C with wind directions from southern direction (Fig. 3).

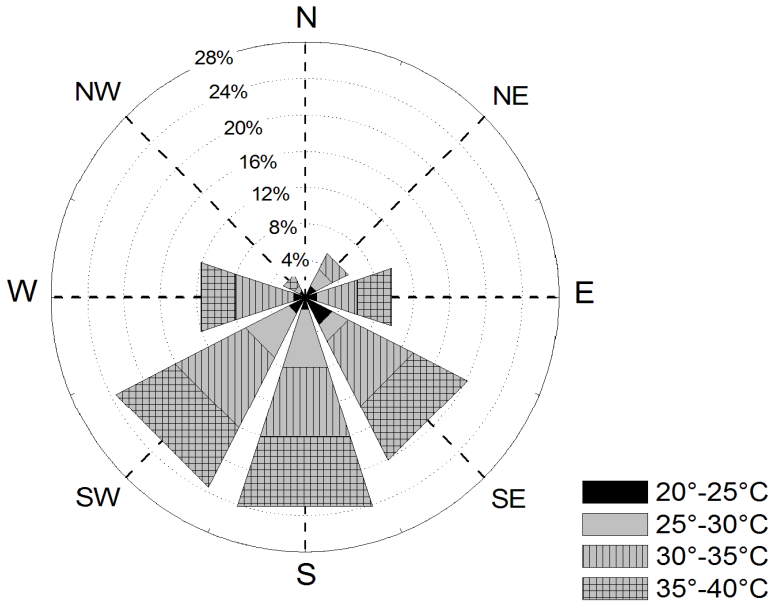

Figure 3: Average mid-day maximum air temperature during fruit drop period 2007, 2008 and 2009 (148 days). The wind rose depicts the relative frequency of wind direction on an 8-point compass with 4 main wind directions classified as N=0°, E=90°, S=180°, W=270° going clockwise. Each ring on the wi nd rose represents 4% of the total (148 days = 100%).

The beginning of the premature fruit drop at 11 dafb in 2007 was characterised by a mid-day average air temperature of 32°C (Fig. 4A). During the following week, a sudden drop of mid-day air temperature of approximately 15°C and a VPD decrease to 0.4 kPa was accompanied with strong winds of more than 10 m s^{-1} (data not shown). Thereafter VPD increased to 2.8 kPa and air temperature to 36°C between 25 to 32 dafb (Fig. 4A). From 32 to 46 dafb, air temperature reached 39°C; however, subsequently decreased to 21°C. The highest VPD value (3.3 kPa) was monitored within that period. High air temperatures in 2007 (Fig. 4A) coincided with high fruit temperature at 25 to 32, 39 to 46 and 60 to 67 dafb with 38.2°C, 42.8°C and 42.6°C, respectively (Fig. 4B). From 32 to 39 dafb, fruit temperature reduction (>2.5°C) was obtained in irrigated trees as compared to control. At 53 dafb fruit temperature decreased to

32°C irrespective of irrigation; however, within 60 to 67 dafb and air temperatures of 39° to 40°C, significant differences of 1°C between irrigated and non-irrigated trees could be measured.

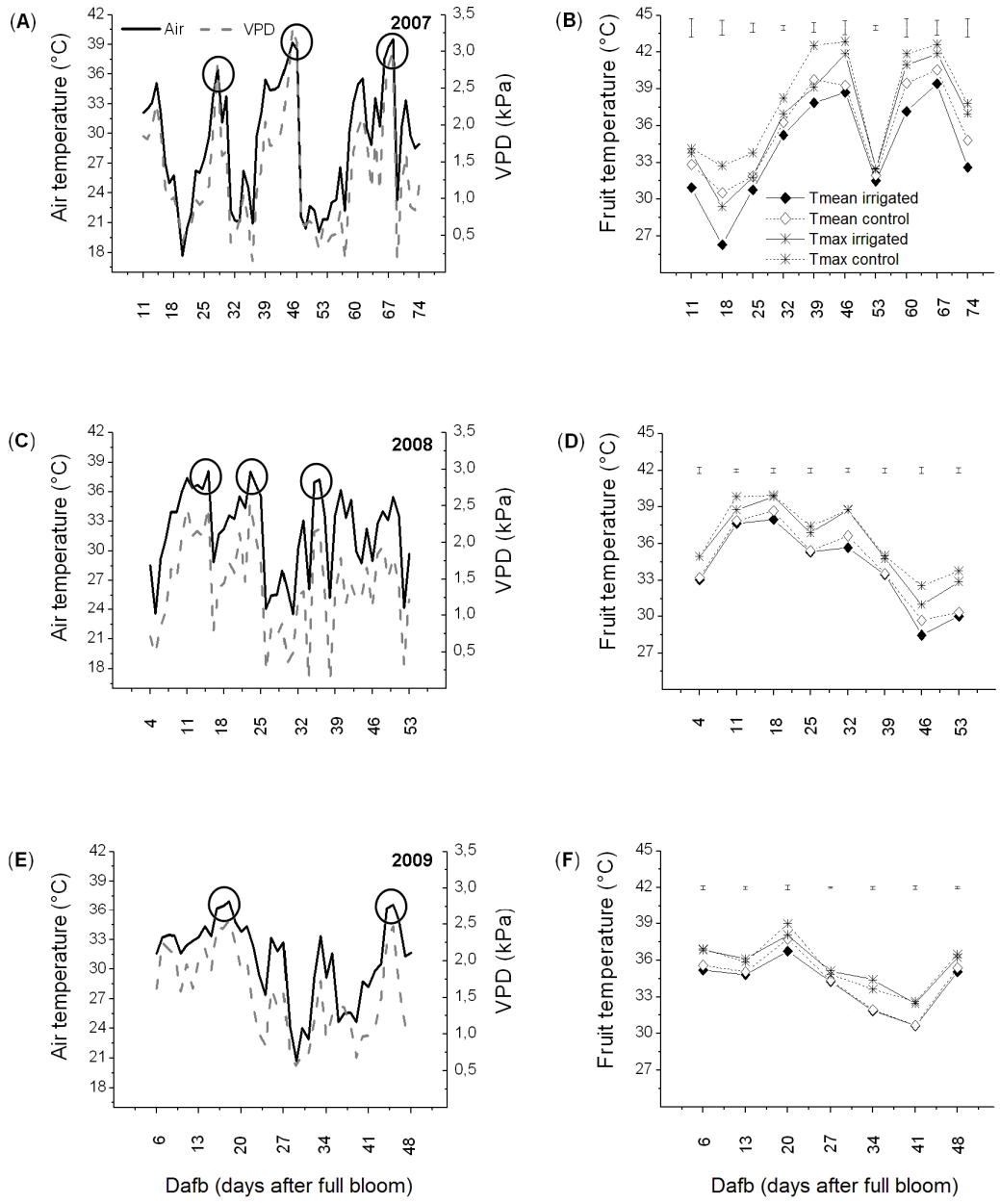

Figure 4: Mid-day mean air temperature and mid-day vapour pressure deficit VPD (A, C, E) and mean and maximum fruit temperature of irrigated and non-irrigated mango trees during the fruit drop period (B, D, F) for 2007, 2008 and 2009, respectively. Bars show the $LSD_{(0,05)}$ at each recording time. Circles indicate possible periods of environmental stress which might be associated with fruit drop periods.

In 2008 there was an early cold spell in February, resulting in delayed full bloom at March 28. At 4 dafb air temperature decreased by 5°C to 23.5°C followed by two short periods of high air temperature until 25 dafb; however, persisting high mid-day average air temperature compared to fruit drop season 2007 until the end of fruit drop period was monitored (Fig. 4A). The high air temperature indicated two distinct periods of 4 to 25 dafb and 32 to 53 dafb. Within the first three weeks, air temperature reached maximum values of 38°C, however, decreased by approximately 10°C between 11 to 18 dafb and 15°C at 25 dafb. High VPD values with 2.5 kPa were reached during 11 to 18 and 25 dafb, respectively (Fig. 4C). From 32 dafb, air temperature increased to 37°C while decreased to end of fruit drop period. Similar, VPD decreased from 2.2 to 0.13 kPa (39 dafb) throughout fruit drop period. Irrigation had no effect in reduced fruit temperature (Fig. 4D).

High air temperatures of over 30°C occurred before full bloom (February 6) and at the beginning of fruit drop 6 dafb in 2009 (Fig. 4E). With temperature peak of 37°C at 15 dafb, air temperature decreased below 21°C until beginning of March (27 dafb). A high VPD value of 2.5 kPa was at the temperature of 37°C (Fig. 4E). With lowest air temperature of 21°C and a VPD of 0.5 kPa from 27 to 34 dafb, air temperature increased again with 33°C at 34 dafb and 37°C at end of March, respectively. However, air temperature was generally higher and temperature fluctuations were less pronounced as compared to study year 2007. Fruit temperature seldom decreased below 30°C throughout the fruit drop period in 2009; however, fruit temperature was not significantly different in irrigated trees compared to controls (Fig. 4F). The results indicated maximum air temperatures of over 30°C in 2007 and 2009 at full bloom in February associated with occurrence of hot, dry winds (Fig. 4A, 4E). This climatic observation in February could be confirmed by 12-year climatic records from the weather station in Yen Chau (Fig. 5). Moreover, with over 30°C in 2007 and 32°C in February 2009, highest maximum air temperature peaks were measured from 1998 to 2009.

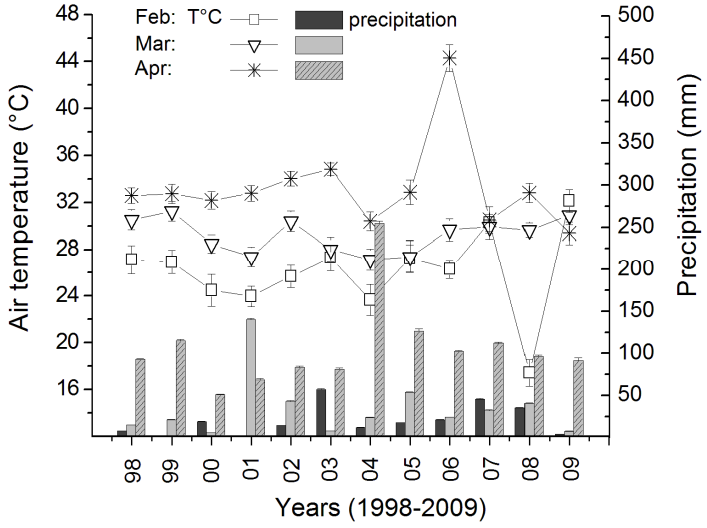

Figure 5: Average maximum daily air temperature (℃) and tot al precipitation (mm) recorded at the weather station in Yen Chau for February, March and April during 1998-2009.

Irrigation and soil moisture: With little precipitation (< 10 mm) at flowering time in 2007 (data not shown), the fruit drop period had two significant rain events. With total amount of 40 mm precipitation from 32 to 39 dafb, highest amount of 88 mm was measured from 53 to 74 dafb (Fig. 6A). Generally, soil moisture at non-irrigated trees rarely decreased below 30% throughout the study period, however, supplementary irrigation increased soil moisture in 10 to 40 cm soil depth compared to control. With 154 mm total amount of precipitation, the fruit drop period 2008 (Fig. 6B) obtained approximately 20% more precipitation than in 2007. However, with more than 80 mm precipitation from 4 to 18 dafb, approximately 50% of the total amount of the recorded precipitation occurred at the early phase of the fruit drop period. Highest soil moisture values with 35% and 37% at 10 and 40 cm depth, respectively, were recorded at irrigated trees from 4 to 18 dafb. With a total amount of 0.6 mm precipitation from 6 to 48 dafb, the fruit drop season 2009 indicated the lowest precipitation within the 3-year study (Fig. 6C) and even the lowest precipitation since March 2003 (Fig. 5). Soil moisture at control trees at any recorded soil depth was lower compared to the previous years. However, for irrigated trees, soil moisture records at 10 and 40 cm soil depth indicated higher soil moisture by average of 30% and 20% throughout the fruit drop period, respectively. Moreover, significantly increased fruit retention at harvest was observed for both cultivars

with an average of 2 fruits per inflorescence in 2008 and more than 3 fruits per inflorescence in 2009, respectively.

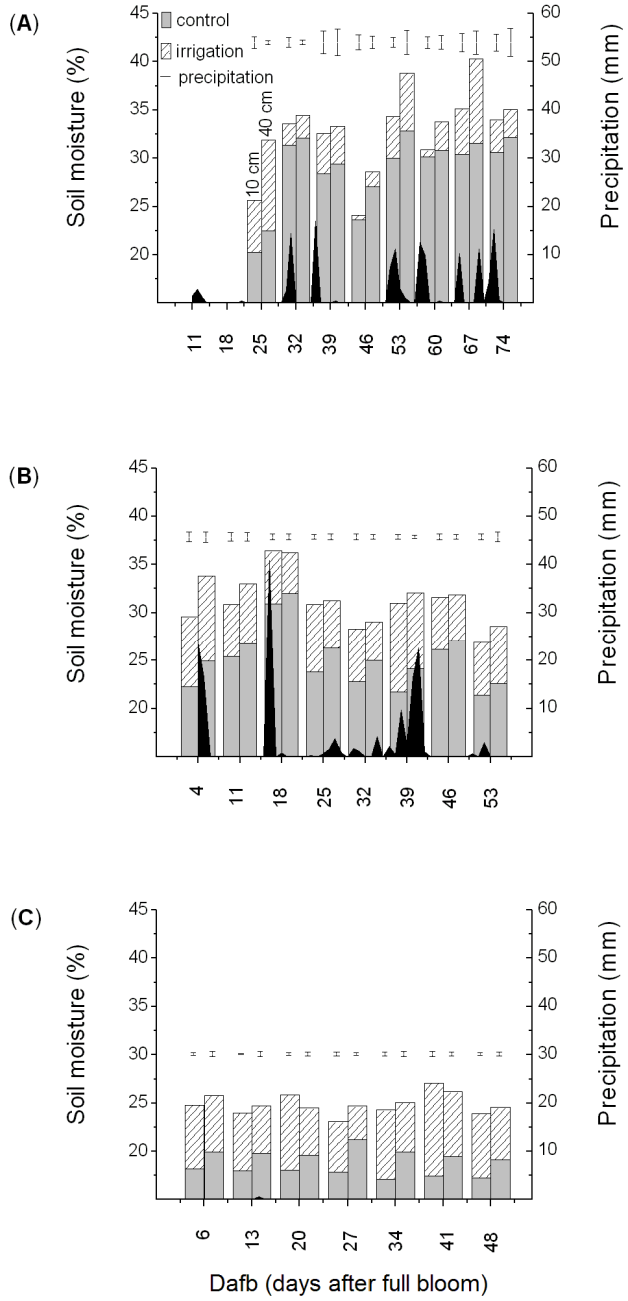

Figure 6: Daily mean soil moisture of irrigated and non-irrigated mango trees, measured at 10 and 40 cm depth and seasonal precipitation for the year 2007 (A), 2008 (B), and 2009 (C). Bars show the $LSD_{(0,05)}$ at each recording date.

3.2 Premature fruit drop alleviated by irrigation and the use of plant growth regulators

Fruit retention in response to irrigation: Full bloom for both cultivars occurred delayed at end of March in 2008, compared to flowering period normally occurring in early to mid February. The cultivar 'Hôi' had a higher fruit set per inflorescence compared to 'Tròn'. Flowering of 'Hôi' was ongoing from 4 dafb to 11 dafb; while cultivar 'Tròn' commenced fruit shedding, respectively (Fig. 7A). Generally, fruit abscission commenced at 'pinhead-size' for both cultivars. Irrigation reduced the intensity of fruit drop significantly in both cultivars. At the end of the fruit drop period irrigated trees had a five-fold increased fruit retention for cv. 'Hôi' and 12% increased fruit retention for cv. 'Tròn' in comparison to non-irrigated control trees (fruit retention of only 4% for 'Hôi' and less than 1% for 'Tròn'). Full bloom in 2009 occurred early to mid of February with fruit set at 6 dafb. Fruit drop pattern intensity was similar for both cultivars; however, fruit drop was affected by cultivar and irrigation (Fig. 7B). Final crop load was with approximately 13% fruit retention for irrigated trees compared to control trees with only 3% fruit retention higher in both cultivars (Fig. 7B).

Fruit retention and hormonal response: Seasonal changes of IAA-export in cultivar 'Hôi' and 'Tròn' and treatment throughout the fruit drop period in 2008 and 2009 are demonstrated in Figure 8. Higher IAA-export from fruits of non-irrigated trees could be measured at one date for each cultivar (Fig. 8A). IAA-export was highest with 1.2 ng $fruit^{-1} \cdot 20\ h^{-1}$ at 25 dafb for 'Hôi' compared to other cultivar and treatments below 0.8 ng $fruit^{-1} \cdot 20\ h^{-1}$. For 'Tròn' the maximum IAA-export was reached at 46 dafb with 1.6 ng $fruit^{-1} \cdot 20\ h^{-1}$ at 53 dafb. However, at 53 dafb which represents the last analyzed sample of the time course the level of IAA-export in irrigated and control trees of both cultivars was similar. A constant and significant increase of IAA-export from fruits after irrigation was not found in 2008. In fact, IAA-export was rather low from 18 to 32 dafb for irrigated trees of 'Hôi' with maximum values with 1.1 ng $fruit^{-1} \cdot 20\ h^{-1}$. However, after 32 dafb the IAA-export increased and stabilized at values around 1.1 ng $fruit^{-1} \cdot 20\ h^{-1}$ until 53 dafb. The IAA-export out of fruits from 'Tròn' was below 0.4 ng $fruit^{-1} \cdot 20\ h^{-1}$ until 32 dafb, thereafter IAA-export increased from fruits of both irrigated trees and controls. Samples from controls reached maximum IAA-export values of more than 1.6 ng $fruit^{-1} \cdot 20h^{-1}$ at 46 dafb.

The situation of IAA export of mango fruits in 2009 was different to the previous year (Fig. 8B). All trees indicated rather low IAA-export values throughout the first two weeks of sampling time until 34 dafb. Maximum values of 1.8 ng fruit^{-1}·20h^{-1} (Hôi) and 0.9 ng fruit^{-1}·20h^{-1} (Tròn) reached at 41 dafb for irrigated trees were significantly higher than the IAA-export values of the controls, respectively.

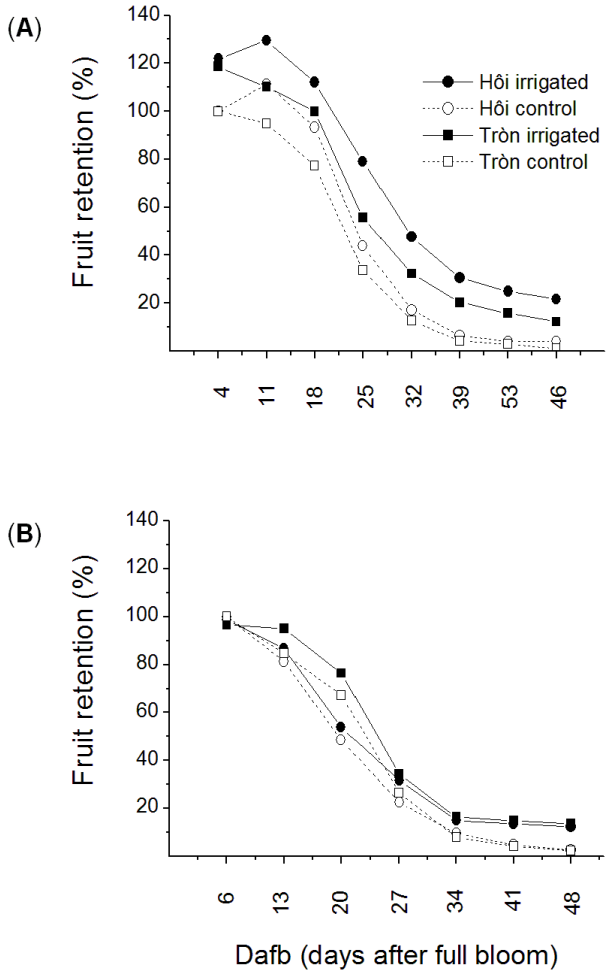

Figure 7: Seasonal mean percentage fruit retention of irrigated and non-irrigated mango trees cultivars ‘Hôi’ and ‘Tròn’ in 2008 (A) and 2009 (B).

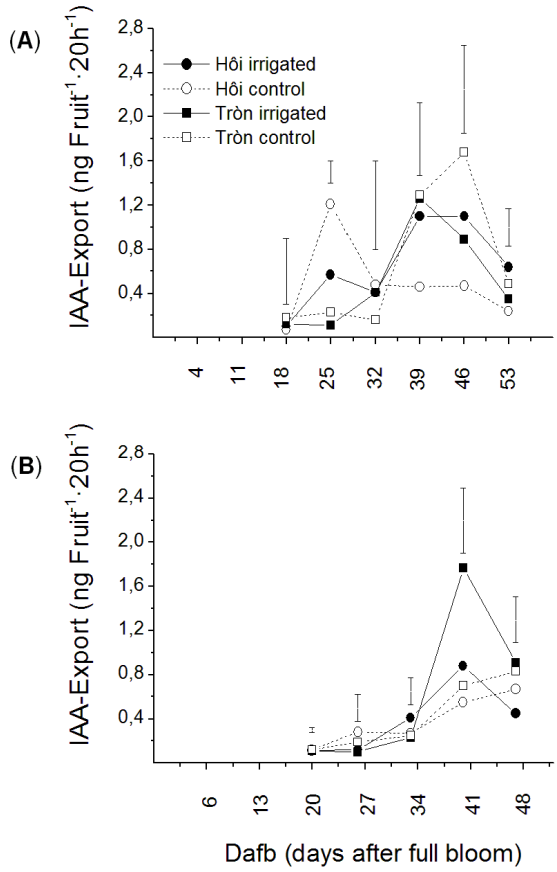

Figure 8: Diffusable IAA-export out of mango fruit, in response to irrigation treatment (irrigated, non-irrigated) and cultivar ('Hôi' and 'Tròn') in 2008 (A) and 2009 (B). Bars show the $LSD_{(0.05)}$ at each sampling time.

Fruit retention in response to PGR application: As shown in Table 2, significantly increased final fruit numbers for both cultivars were identified for all treatments compared to control for each experimental year 2008 and 2009. Dependent on the treatment, final fruit numbers vary significantly within each study year. Application of CPPU and NAA generally indicated highest fruit retention compared to all other treatments. Spraying with NAA resulted to maximum crop load at final fruit counting. Comparison between the study year 2008 and 2009 indicated; that the study year 2008 had a significantly higher fruit number in both cultivars. However, this included all treatments and the control, except the treatment CPPU (2) in 'Hôi' which showed no significant difference in fruit number per inflorescence in 2008 and 2009.

Table 2: Final fruit number per inflorescence for cultivars ‘Hôi’ and ‘Tròn’ in 2008 at 53 dafb (May 20), and 2009 at 48 dafb (March 26) for each PGR treatment.

Treatment	Final fruit number				LSD (0.05)
	Hôi		Tròn		
	2008	2009	2008	2009	
1. Control	0.8	0.5	1.5	0.1	0.16
2. CPPU	3	2.8	3.4	1.7	0.21
3. CPPU + GA_3	2.1	2.7	2.5	1.1	0.22
4. CPPU + GA_{4+7}	2.2	1	2.6	0.6	0.23
5. Combi I	2.1	2.8	2.8	1.5	0.21
6. Combi II	2.3	1.6	2.4	0.3	0.24
7. NAA	3.5	2.3	3.7	1.8	0.23
LSD 0.05	0.46	0.3	0.51	0.27	

3.3 Morpho-physiological changes in the abscission zone of Mango fruit pedicel

Abscission zone morphology of mango pedicel: A groove-shaped indentation at the base of the mango fruit pedicel indicates the abscission zone (Fig. 9A). Increased susceptibility to fruitlet abscission is correlated with colour changes of the pedicel from deep green to green yellow or even brownish. Fruitlet abscission was also closely linked to the yellowing of the dorsal fruitlet shoulder and visible shrivelling of the fruit skin (data not shown). When pedicel and fruit are green the fruit is tightly attached; however, if yellowing was found, a small mechanical impact e.g. bending of the pedicel, resulted in detachment of the fruit. The morphological characteristics across a longitudinal section of the abscission zone in mango, cv. ‘Tròn’ at 60 dafb (n=64) is shown in Figure 9B.

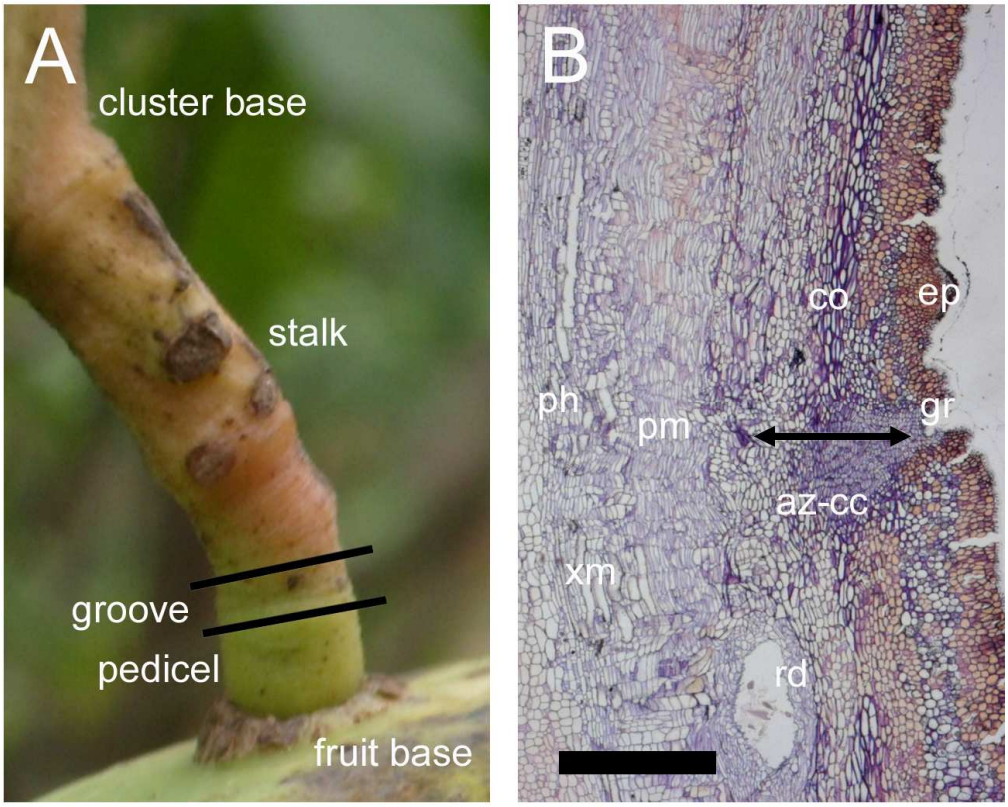

Figure 9: Morphological characteristics of mango fruit pedicel (A). Longitudinal section of the abscission zone with groove (gr), abscission zone cortex cells (az-cc), epidermis (ep), piths (ph), cortex cells (co), phloem (pm), xylem (xm) and resin ducts (rd) (B). Arrows show the selected area used for microscopically evaluating starch grain accumulation. Scale bar = 500 µm.

The groove area marks the abscission zone, crossing the pedicel as a disk-like layer consisting of several thin-walled cell rows. The cortex parenchyma (co) cells are peripherally surrounded by the epidermis (ep). The parenchyma tissue extends to phloem vessels (pm) and is randomly interrupted by resin ducts (rd) which occur in both the exocarp and the inner region of the mesocarp. Bands of xylem vessels (xm), large parenchyma cells of the pith (ph) can be found. Pith and cortex are connected by parenchyma rays that transect the vascular ring. Cortex tissue determines specific area of AZ cortex cells (AZ-cc).

Measurement of pedicel thickness at the abscission zone: The thickness of fruit pedicel was significantly increased in cultivar and treatment (Fig. 10A). Irrigated trees had a thicker fruit pedicel in both cultivars compared to non-irrigated controls. From 32 to 74 dafb, irrigated trees of 'Hôi' indicated increased pedicel growth with 3.1 to 3.8 mm diameter, respectively. Conversely, pedicel growth of 'Trôn' at 32 dafb were below the control; but increased from less than 1.6 mm to 3 mm diameter within 28 days in irrigated trees. In Figure 10B, the AZ in 'Hôi' with 0.2 mm length was greater than in 'Tròn'. Notably, AZ in 'Tròn' was roundish-shaped while being broader in 'Hôi' (data not shown).

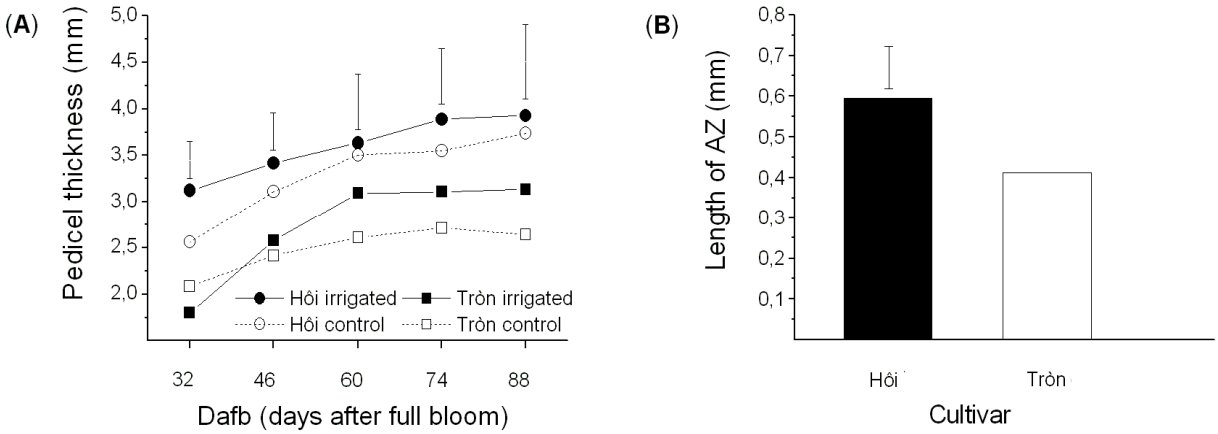

Figure 10: Maximum pedicel thickness of irrigated and non-irrigated mango cultivars throughout the sampling period (A). Length of the abscission zone in cultivar 'Hôi' and 'Tròn' (B). The bars in (A) and (B) show the $LSD_{(0,05)}$.

Cell separation: Prior to the activation of the abscission process and cell separation, a potential site of fracture crossing the pedicel was not detected. Due to results of the microscopically studies the abscission process was initiated in the cortex, phloem and at the pith, but was not found starting at the periphery of the pedicel. If sections indicated an early stage of abscission, separation commenced in AZ-cc horizontally, but also perpendicularly over several cells layers (Fig. 11A, B).

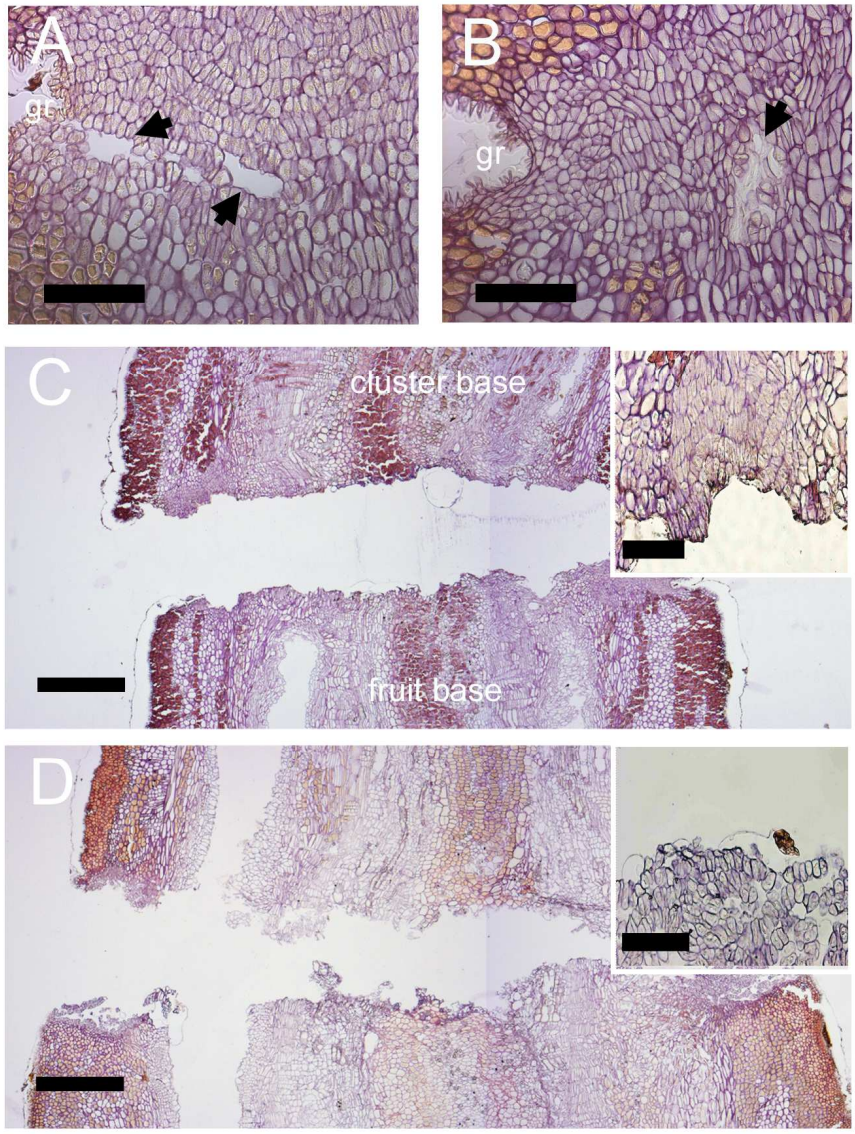

Figure 11: Horizontal (A) and vertical (B) cavities within the separation layer in close proximity to the groove (gr). Morphological characteristics of the fruit abscission zone resulting from natural fruit drop (C) and fruit drop artificially by hand (D). Insets in both figures provide a close-up view of the fracture line. Scale bar: C, D = 500 µm; A, B, inset C, inset D = 100 µm.

During and after abscission, intact cells showed a roundish shape along the separation line (Fig. 11C, inset) conversely, hand-broken non-abscising mango pedicels result in ruptured tissue (Fig. 11D) with many separated or destroyed cells at the edge (Fig. 11D, inset).

Starch accumulation and IAA analysis: Starch grains could be found in all parenchyma cells but were mainly concentrated in the cortex cells, and, to some extent, in the cells of pith and pith rays. From 32 to 60 days after full bloom (dafb) increased starch accumulation in AZ-cc was observed in both cultivars; however, non-irrigated trees showed not significant starch grain accumulation per cell (Fig. 12A). In general, 'Hôi' showed a higher starch grain accumulation than 'Tròn'. At 60 to 74 dafb, irrigated 'Hôi' trees indicated the highest grain number with 2.9 grains per cell on average, while 'Tròn' had a maximum of 2.7 grains per cell on average. Thereafter, starch grain number decreased in both cultivars, irrespective of the treatment. Figure 12B shows the percentage of cells with varying numbers of starch grains per cell. At 32 dafb 'Tròn' had a higher percentage of cells without starch grains compared to 'Hôi'. In Hôi 40% (1 to 3 grains per cell) and 15% (4 to 6 grains per cell) of the cells indicated the highest starch content at 32 dafb. At 88 dafb more than 50% of the cells were without starch accumulation in both cultivars (Fig. 12C).

Seasonal changes of IAA-export in both cultivars and treatments throughout the observation period are shown in Figure 13. A positive effect of irrigation on IAA-export at certain dates could be found for both cultivars. Overall, IAA-export of 'Hôi' fruit was higher compared to 'Tròn' throughout the sampling period (Fig. 13). In detail, the highest IAA-export of 6.4 ng IAA $fruit^{-1} \cdot 20h^{-1}$ was found in IAA diffusates of irrigated 'Hôi' at 60 dafb, whereas in irrigated 'Tròn' the enhanced IAA-export only reached 2.6 ng IAA $fruit^{-1} \cdot 20h^{-1}$ at 40 dafb (Fig. 13). From 60 dafb to 74 dafb a steep decline in IAA-export was observed in both cultivars.

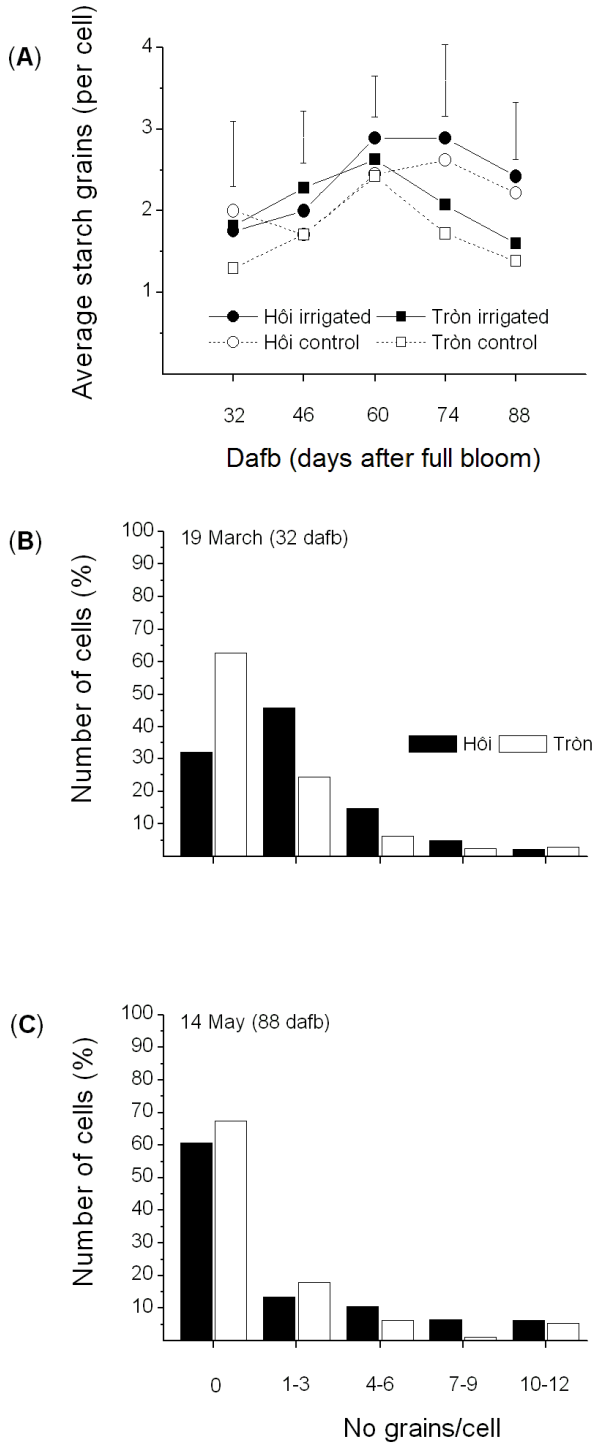

Figure 12: Average number of starch grains within cortex cells of the abscission zone in irrigated and controls trees throughout the sampling period (A). Occurrence of starch grains at 32 dafb (B) and 88 dafb (C) in cultivar 'Hôi' and 'Tròn'; 32 to 60 dafb indicate termination of intensive fruit drop. The bars indicate the $LSD_{(0,05)}$ at each recording time.

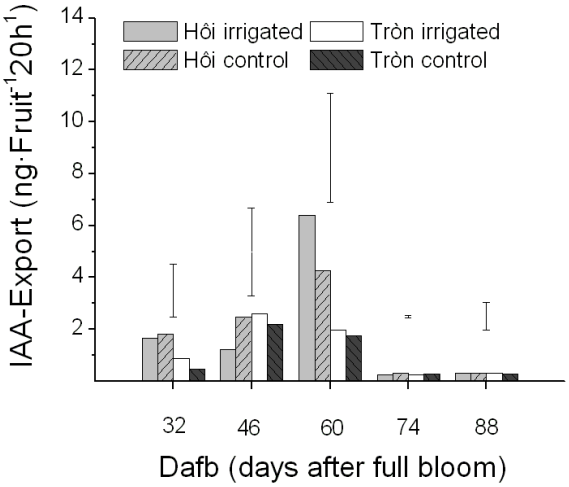

Figure 13: Seasonal IAA-export from mango fruit for irrigated and non-irrigated 'Hôi' and 'Tròn'. Bars show the $LSD_{(0,05)}$.

Discussion

3.4 Premature fruit drop induced by climatic factors

General climatic conditions: The effect of climatic conditions in Northern Vietnam on the intensity of premature fruit drop was analyzed throughout a 3-year experimental period. It seems to be evident that the variable flowering period of the cultivars Hôi and Tròn was based on the specific weather conditions within a given year. The dry season, occurring between November to April, generally begins with a cool period from November to end of January followed by a hot period from February and/or March to April (Phan, 2005). Furthermore, the hot period in February and March is typically accompanied with strong and dry prevailing southern winds, with less than 12% relative humidity (Do Anh, 2004). These data together with our own results might indicate that the dry season with prevailing winds induce the onset of the fruit drop. A similar weather pattern in Southern California, known as the hot and dry 'Santa Ana winds' (Raphael, 2003; Westerling *et al.*, 2004) affect the flowering and fruit set of avocado (Witney and Bender, 1992). As the combination of high air temperature and low humidity was also found in Northern Vietnam, it is proposed that this might be the causal factor for the premature fruit drop and reduction in yield in mango.

Premature fruit drop: Fruit counts in this study revealed considerable differences in fruit drop pattern of the cultivars 'Hôi' and 'Tròn', which mirrors previous descriptions for cultivar specific fruit drop patterns in mango by Guzman-Estrada (1996). In our study, mangos initially abscised from full bloom to early 'pinhead' stage, which occurs in all mango cultivars and is considered as a natural thinning mechanism. The second phase of fruit drop between 18 to 46 dafb at 'pinhead' to 'marble-size' (Fig. 2A) in 2007 is regarded as the premature fruit drop, induced by environmental cues. Similar patterns of fruit drop, most pronounced during 3 to 4 weeks after fruit set, have been reported for mango (Searle *et al.*, 1995; Singh and Arora, 1965). In 2008 with delay in flowering and fruit set possibly caused by the cold weather conditions before and during the flowering period, premature fruit drop occurred at 11 to 46 dafb,

whereas for 2009 period of flowering and fruit drop followed the season 2007. Fruit abscission intensity of both cultivars was remarkably different for all years, possibly due to environmental conditions and genotype interaction. It is known that many cultivars, e.g., 'Kensington', 'Tommy Atkins', and 'Haden' usually bear only one fruit per inflorescence to maturity, whereas 'Sensation', 'Irwin', and 'Nam Dok Mai' often carry two or more fruits per inflorescence; thus intensity of fruit drop seems to be influenced by genotype (Singh, 2005). According to Singh and Arora (1965), Krisanapook *et al.* (2000) and Searle *et al.* (1995), fruit drop is most pronounced during first 28 days after fruit set, whereas Guzman-Estrada (1996) timed the main fruit abscission period between 25 and 50 days after fruit set. Consequently, our results for 'Hôi' and 'Tròn' indicate a fruit drop period from 18 to 46 dafb which corresponds well with other studies of mango cultivars.

Effect of air and fruit temperature on fruit drop: Poor fruit set in mango in the subtropics due to unfavourably cool temperatures during floral anthesis and fruit set was described previously (Whiley *et al*. 1988; Tsai *et al.* 1996). Generally, air temperatures of 15°C/5°C day/night cause damages of stigmas and ovaries in various mango cultivars, resulting in embryo abortion and fruit drop (Sukhvibul *et al.*,1999, Issarakraisila *et al.*, 1992). Moreover an increased susceptibility of generative organs to low air temperature of polyembryonic cultivars as compared to monoembryonic cultivars was reported (Sukhvibul *et al.*, 2005; Sukhvibul *et al.*, 1999b). Indeed, Sukhvibul *et al.* (1999) concluded that monoembryonic cultivars appeared to be more tolerant to low temperatures during flowering. Although 'Hôi' and 'Tròn' are polyembryonic and hence potentially at greater risk to unseasonably cool conditions occurring during anthesis, fruit samples at marble-stage in 2008 had fully developed seeds (data not shown). Generally, air temperatures above 30°C promote vegetative growth in mango (Whiley *et al.*, 1989), and can negatively affect fruit development. However, several studies indicate different air temperatures detrimentally to mango fruit set and development (Issarakraisila and Considine, 1994; Lam *et al.*, 1985; Nunez-Elisea and Davenport, 1983). In contrast to seasonal warm years in 2007 and 2009 with mean value of 17°C, 2008 had lowest air temperature in February, compared to 12-year climate records in Yen Chau (Fig. 5).

Fruit temperature is dependent on ambient air temperature but also on direct exposure to the sunlight (Ferguson *et al.*, 1998; Wünsche *et al.*, 2001). According to Ferguson *et al.* (1998)

bulky organs such as fruit may act as heat sink, and flesh and skin temperatures vary distinctively to air temperature. The surface temperature of fruit from irrigated trees in 2007 and 2008 was randomly lower with 1.2°C compared to control trees, which might indicate an increased transpiration rate as mechanism for evaporative cooling. Moreover, high wind speed in 2007 might additionally decrease temperature at the fruit surface. Similar effect is reported by Thorpe (1974) in apple, where wind speed increase from 0.3 to 4.0 m s^{-1} resulted in temperature drop of 5°C at fruit surface. For 2009; however, this observation could not be confirmed and heat presumably induced closure of stomatal closure apertures (Wilson and Bunce, 1997) would reduce the rate of leaf transpiration (Bunce, 1996). High temperatures can also directly affect the photosynthetic processes in mango and thus reduce carbohydrate availability for supporting fruit growth processes (Yamada *et al.*, 1996). Further, strong fruit abscission during hot and dry season could be a response to reduced leaf photosynthesis in mango (Chacko *et al.*, 1995).

Particularly the fruit drop season 2007 (Fig. 4A) indicated high VPD values (>3 kPa) which might have caused leaf transpiration. Similar high values in 2008 and 2009 (>2 kPa) were observed (Fig. 4C,E); however, days with low VPD (< 0.5 kPa) might indicated less stress potential for mango trees. Stomatal response has been reported for many plants (George *et al.*, 1990; Lange *et al.*, 1971; Leuschner, 2002; Menzel and Simpson, 1986). According to several studies, stomatal conductance decreases exponentially with increasing air temperature (Massman and Kaufmann, 1991; Monteith, 1995), in response to an increased leaf to air VPD (Higuchi *et al.*, 1999). Moreover, closure of stomata caused midday depression of photosynthesis and transpiration in the subtropical evergreen *sclerophyllis* (Beyschlag *et al.*, 1992), and this kind of response could suppress photosynthesis in mango as well, even if mango is adapted to (sub)tropical climates.

Effect of irrigation and soil moisture on fruit drop: High air temperature mostly associated with high VPD values indicated a possible stress potential for trees, however, drought stress has been suggested as an additional factor triggering fruit drop. Precipitation of less than 285 mm during each fruit drop period of all experimental years might detrimentally affect fruit set in non-irrigated controls. Adequate water supply is of major importance during flowering and fruit set for many fruit crops (Domingo *et al.*, 1996; Lahav and Kalmar, 1977; Li *et al.*,

1989b). According to Ponce de Leon *et al.* (2000) fruit growth in mango is divided in three stages; the first two weeks after fruit set with a slow growth and high cell division rate, followed by increased growth rate due to cell expansion from third to the eighth week, and a final stage of slower growth when fruits proceed to maturity. Water deficiency at early mango fruit development can seriously affect yield and especially the first 20 days of fruit growth under water limiting conditions affect yield by 20% (Coelho and Borges, 2004; Schaffer *et al.*, 1994). Although mango is considered to be drought-resistant, supplementary irrigation has a positive effect on productivity when rainfall is scarce or poorly distributed during the generative phase. The beneficial effect of irrigation on the increased final fruit number per inflorescence was confirmed in 2008 and 2009. For the season 2007, the setup of the irrigation system might have been too late, resulting in an insufficient period of water supply before flowering.

The results indicate that premature fruit drop in the mango cultivars 'Hôi' and 'Tròn' in Northern Vietnam is caused by unfavorable weather conditions during the period of flowering and fruit set. The seasonal cycle of flowering, fruit set and fruit development of mango in this study area is shown in Figure 14 which are all affected by environmental cues. Additional water supply by irrigation had an effect on reducing stress potential and increasing orchard productivity; however, irrigation has to be adjusted to the phenological cycle of mango tree such as flower initiation and flowering and particularly early fruit set and development.

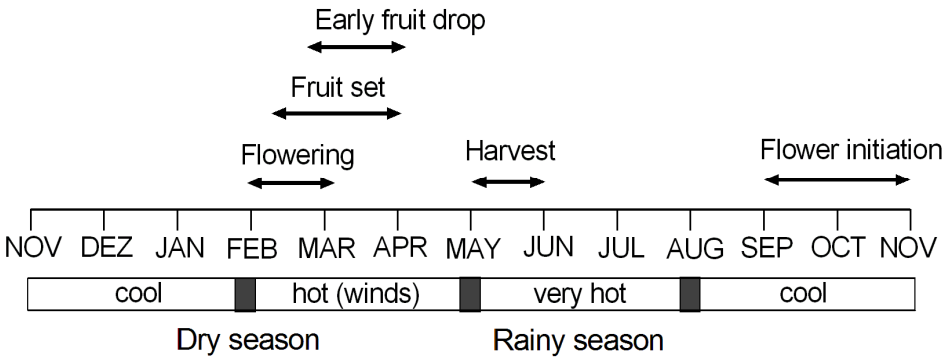

Figure 14: Seasonal fruit phonological stages of mango in Yen Chau and prevailing environmental conditions throughout the growth cycle.

3.5 Premature fruit drop alleviated by irrigation and the use of plant growth regulators

Effect of environmental conditions: Throughout the 2-year study, the year 2008 indicated a delay in flowering and fruit set due to cold weather conditions before and during the onset of flowering. In contrast, the experimental year 2009 constitute mean warm air temperatures which facilitated timely onset of full bloom. However, fruit counting indicated differences in fruit drop pattern of 'Hôi' and 'Tròn', which is confirmed by other studies in mango, respectively (Guzman-Estrada, 1996; Singh, 1960). Abscission occurs from full bloom throughout all stages of fruit development (Krisanapook *et al.,* 2000; Ram *et al.,* 1983); however, is intensified during 2 to 4 weeks after full bloom (Bhuyan and Irabagon, 1993; Nunez-Elisea and Davenport, 1983) similar to results in our study. The total amount of precipitation differs significantly throughout the fruit drop period of each experimental year. According to 12-year climate records, the lack of precipitation during early fruit development associated with hot and dry winds which are common for the study area. With 0.6 mm precipitation from full bloom throughout the fruit drop period respectively, the year 2009 indicated lowest precipitation within the study period. However, the year 2008 marked a severe cold spell in February (maximum and minimum ambient temperature of 17°C and 11°C respectively) including a total amount of 154 mm precipitation within the period fruits abscised.

Effect of irrigation: Mango is generally considered as a drought tolerant fruit crop (Chacko, 1984; Urban and Jannoyer, 2004), however, water requirements vary according to the phenological stage. Sufficient water supply is most critical during the first 6 weeks (Schaffer *et al.,* 1994), and lack of precipitation might affect photosynthetic activity, pollination and fruit set (Gonzalez *et al.,* 2004; Lu and Chako, 1997). Water deficiency during early stage of fruit development is assumed to induce fruit drop intensity (Spreer *et al.,* 2007). Similarly for other fruit trees like citrus adequate water supply is primarily important during flowering and fruit set to guarantee high crop load (Chalmers and Wilson, 1978; Domingo *et al.,* 1996). For both experimental years and both cultivars, fruit set and number of fruits per tree were significantly increased by irrigation compared to control (Fig. 7A, B). Moreover, harvest in 2008 showed

increased fruit size (data not shown). This result correlates partly with studies of mango, where higher yield due to increased fruit number rather to enlarged fruits were obtained (Pavel and de Villiers, 2004; Spreer *et al.*, 2007). It is recognized that the distribution of precipitation rather than the total quantity of water is a crucial factor for the productivity of fruit crops (Chalmers and Wilson, 1978; Lechaudel *et al.*, 2004). Precipitations from 4 to 39 dafb in 2008 were abundant; while it was presumably sufficient in 2009 with only 0.6 mm rainfall to replenish soil moisture. Little rainfall during rapid fruit growth coincides with decreased water stress (Li *et al.*, 1989a; Li *et al.*, 1989b); moreover, decreased photosynthetic capacity in mango rapidly reverses due to irrigation (Damour *et al.*, 2009). According to Spreer *et al.* (2009), in years with precipitation, well-irrigated trees indicate higher yields; contrarily, in dry years this effect is more pronounced to non-irrigated trees.

Water application availability presumably increased fruit set in 2008 compared to control; however, compared to higher fruit set in 2009, additional precipitation might also detrimental affect flowering and early fruit set in both cultivars. Water stress may help to stimulate trees to flower but with complete emergence of inflorescences supplementary irrigation is recommended (Coelho and Borges, 2004). Moreover, Bally *et al.* (2000) suggested that water stress might improve floral induction. Contrarily, heavy precipitation and high humidity conditions during this critical stage heavily damage early fruit set, or furthermore stimulate vegetative growth (Chacko, 1984). Hence, reduced fruit set in 2008 might associated with inadequate environmental conditions which act as natural fruit thinning mechanism in mango.

Hormonal response: Plant hormones have been suggested as dominating signals inducing early fruit drop in mango (Chen, 1983, Nunez-Elisea and Davenport, 1983). However, continuous auxin basipetal transport through the abscission zone is critical for retention of plant organs including fruit (Davenport *et al.*, 1980; Roberts and Osborne, 1981). The hormone IAA (indole-3-acetic acid) is postulated to inhibit AZ activation by reducing the sensitivity of cells in the AZ to ethylene, an abscission promoter (Blanusa *et al.*, 2005; Taylor and Whitelaw, 2001). In our study, from full bloom to 18 dafb, fruit abscission might be considered as natural thinning mechanism; however, the period of low IAA-concentrations (18 dafb to 32 dafb) in 2008 might corresponded with excessive fruit drop in mango with the reported period of high rate of mango fruit drop (Prakash and Ram, 1984). Notably, from 32 dafb to final fruit

set, the increased IAA-export might result in higher amount of sampled fruits which are less susceptible to abscise. Similarly, sampling in 2009 showed stabilizing effect of fruit retention at 34 dafb to the end of fruit drop period (48 dafb), which might be correlated with increased IAA-export (Bangerth, 2000). Shedding intensity in both years indicated increased IAA-export values at mid or close to end of fruit drop period. Thus, pollination and embryo degeneration as observed in young fruitlets described as potential causes of abscission can be excluded (Chadha, 1993; Singh and Singh, 1995). However, in cherry a specific role for IAA in regulating abscission was assumed when late-formed fruit detached preferentially to first-formed fruit, respectively (Blanusa *et al.*, 2005). According to Bangerth (1989) the fruit that is set first and develops first dominates over later developing lateral fruit and induce their abscission. At hormonal level, the strong polar IAA transport pathway of a dominant fruit thus affect weaker IAA pathway of a dominated fruit, which causes an autoinhibition of the auxin transport thus fruit shedding (Li and Bangerth, 1999). Thus, as periods of high IAA-export might link with low fruit drop, it is suggested that fruits which have survived the main shedding period can resist abscission. Water stress may therefore manipulate nutrient supply to fruits which determine the auxin status and/or supply of the abscission zone.

Effect of PGR: Chemical application in each study year increased fruit set. It is well known in many fruit crops, including mango, that retention of a fruit e.g. the capacity of a fruit to prevent itself from being shed, relates positively of the fruit ability to produce growth promoting hormones (Berüter and Droz, 1991; Buban, 2000; Chen, 1983; Prakash and Ram, 1984). The applications of synthetic PRGs such as CPPU, GAs and NAA have been reported to enhance fruit retention in mango (Burondkar *et al.*, 2009; Oosthuyse, 1995, 1997; Singh and Ram, 1983) and might suggest a correlation of deficiency or metabolic and or transformational alterations of natural occurring hormone values at early stages of fruit development with fruit drop in mango (Malik and Singh, 2003; Ram *et al.*, 1983).

Several studies in fruit crops such as apple, citrus, guave, macadamia, persimmon and mango (Black *et al.*, 1995; Kassem *et al.*, 2010; Masia *et al.*, 1998; Oosthuyse, 1997; Ortola *et al.*, 1998; Singh and Ram, 1983; Williams, 1980) indicated enhanced crop load by CPPU and NAA, respectively. In grapes combined effects of CPPU and GA_3 were better than single chemical treatment (Han and Lee, 2004). In our study, however, single application of CPPU

applied directly after full bloom promote mango fruit retention, with maximum of 3 fruits per inflorescence in 2008 and 3.4 fruits per inflorescence in 2009. This contradicts results by Oosthuyse (1997) where CPPU alone was not found to increase fruit retention in mango applied shortly after flowering.

The specific role of carbohydrates in fruit abscission and phytohormones are not completely understood. It has been suggested that carbohydrates and hormones participate in a complex signal transduction system (Roitsch, 1999) and in citrus; abscission induced by carbohydrate shortage was triggered by increases in the levels of ABA, ACC as precursor of ethylene, respectively. Auxins have been reported to promote variable effects (Agusti *et al.,* 2002); however, capacity modifying fruit abscission has been described as well (Yuan *et al.,* 2002). In mango, highest cytokinins levels were measured at 7 to 21 and 42 to 70 days of fruit growth, respectively (Ram *et al.,* 1983); whereas Chen (1983) defined maximum concentration of cytokinins 5 to 10 days after full bloom, respectively which decrease rapidly thereafter. In kiwi the effect of CPPU might be explained by increased cell divisions reducing flower shedding, respectively (Iwahori *et al.,* 1988). Thus, Oosthuyse (1995) concluded, that cytokinin is important during initial stages of fruit growth in mango; thereafter, increasing concentrations of auxin and gibberellins become significant.

In view of the results in CPPU, spray application of NAA increased significantly fruit retention; however, the degree of efficiency depends on the kind of auxin used and the weather conditions of application time and additives (Agusti *et al.,* 2000; Meland, 1998). The data show that when NAA was applied at marble stage, maximum increase over control with 3.5 fruits per inflorescence in 2008 and 3.7 fruits per inflorescence in 2009 for cultivar 'Hôi' whereas similar, however lower fruit retention were obtained in both years for 'Tròn' respectively. Therefore, the use of NAA to improve retention appears to be linked with developmental fruit stage. Indeed, similar studies mentioned inhibitory or delaying treatment effect of NAA on apple at late growth stage (Masia *et al.,* 1998); however, for mango Naqvi *et al.* (1990) specified, spraying with NAA to premature fruit at pea size stage and two weeks after marble stage significantly increased fruit retention. Similar studies confirm; period of application; however, NAA concentration and application repetition might significantly enhance increasing fruit retention in mango, respectively (Naqvi *et al.,* 1990; Oosthuyse, 1995, 1997).

3.6 Morpho-physiological changes in the abscission zone of Mango fruit pedicel

Abscission morphology and cellular changes: The morphology of the mango pedicel was similar to that described earlier by Barnell (1939); however, lignified pith cells proximal to the groove were not found in the present study. The grooves represent the site where the abscission process ultimately occurs (Barnell, 1939). Singh (1961) also reported lignified pedicel cells during late stages of fruit development; thus, sampling was discontinued prior to lignifications of the pedicel. No varietal difference could be observed in the shape of the grooves between 'Hôi' and 'Tròn'; however, it was shown that 'Hôi' with a bigger fruit size, had an increased fruit pedicel thickness compared to 'Tròn', which represents a cultivar specific feature. This was also observed in several citrus cultivars, where pedicel diameter was also linked to fruit size (Bustan *et al.*, 1995; El-Otmani *et al.*, 1993; Stewart *et al.*, 1952). The AZ of tightly packed small cell rows and the transition from AZ cortex cells (AZ-cc) to neighbouring tissue is clearly distinguishable. The smallest number of AZ-cc cell rows of investigated pedicels was 4 and ranged to a maximum of 16 at the broadest point of the AZ-cc. Throughout the sampling period, the number of rows of cells within the AZ steadily increased, whereby 'Tròn' tended to have more cell rows compared to 'Hôi'. These observations may correspond with studies in plums where the ease of fruit detachment was positively correlated with the number of cell rows (Al-Jaru and Stösser, 1973). The cultivar 'Hôi' indicates a larger AZ thickness; however, this did not result in enhanced fruit retention. According to Bonghi *et al.* (2000) AZ-cc in peach are grouped in cell units and transverse sections in our study on mango indicate similar cell formations. Moreover, sectioning through the AZ indicated that resin ducts are perforating the pedicel AZ. Resin ducts are non-uniformly arranged in fruits of various *Anacardiaceae* species (Grundwag, 1976; Joel, 1980; Joel and Fahn, 1980). The resin ducts occur both in the exocarp and the inner region of the mesocarp and the duct system enters the pedicel and also penetrates the abscission zone (Joel, 1980, 1981). It is unclear to what extent the incidence of resin duct systems in mango may contribute to the instability of the abscission zone; however, as shown in Figure 11D, it is obvious that resin ducts in the AZ reduce the area of cellular connection and may support indirectly the abscission process due to destabilization of the pedicel.

Separation process: With a reduced amount of samples it might be suggested that fruit abscission in mango occurred fast. This is supported by several studies, reporting that the abscission of plant organs occur within hours after induction of the abscission process. Polito and Lavee (1980) and Brown (1997) indicated abscission of fruits and leaves typically within 12-60 h after AZ induction; however, for tomato, flower abscission was already found within 4 h (Roberts et al., 1984) or even 1 h (Abeles *et al.*, 1971). In mango the beginning of abscission was first noticeable by cell separation occurring in the AZ-cc adjacent to vascular tissue and progressing to the periphery. Similarly, Baird and Webster (1979) described the abscission process of sour cherry, beginning in the central part of the AZ and progressing towards the periphery. In apple it was shown that abscission starts at epidermis cells continuing through collenchyma, vascular tissue and sclerenchyma cells of the pith (McCown, 1943). However, according to Pandita and Jindal (1991) separation of cells in the AZ of apple commenced in the cortex and spread to the vascular tissue. This correlates with results on flower abscission in tomato, where cell separation occurred at the distal side of the AZ with parenchyma cells responding first (Roberts *et al.*, 1984). Conversely, Polito and Lavee (1980) reported that the separation processes in olive leaves occurred first in abaxial cortical cells adjacent to the vascular tissue and progressing toward the epidermis. Stösser *et al.* (1969a) identified the AZ-layer in cherry by its low affinity to haematoxylin, indicating that cell walls of different tissues showed different staining intensity. Further, he suggested low affinity through alteration of cell walls in the AZ-layer and degradation of cell-wall constituents such as polysaccharides and pectin (Stösser *et al.*, 1969a). In mango, staining of cell walls of the separation layer could not be observed prior to abscission; however, pedicels indicated staining in AZ-cc after abscission differing from neighbouring tissue.

The even fracture surface with rounded cells (Figure 11C) suggests that the cell separation is based on cell wall lysis due to middle lamella degradation and increased turgor of the AZ cells (Brown, 1997; 2002). Sexton (1979) stated that rounded cells are found at the edge of abscised tissue. Cell enlargement in association with abscission is considered as an important component of the abscission process (Osborne and Morgan, 1989; Rascio *et al.*, 1987; Sexton and Redshaw, 1981). Brown and Addicott (1950) suggested leaflet abscission caused by shearing forces which presumably resulted in different tension and compression of vascular vessels in the AZ tissue, respectively. Similar was observed in apple, where mechanical rup-

turing of the vascular strands enlarge the break in the separation layer leading to complete separation of the cells (Pandita and Jindal, 1991; Sexton and Redshaw, 1981).

Inducing factor of abscission process:

1. Indole-3-acetic acid (IAA): Sampling for fruit IAA-export analysis at 32 dafb (19 March) likely excluded lack of pollination and embryo degeneration as potential causes of abscission (Chadha, 1993; Singh and Singh, 1995). Several reports suggest that IAA is reduced in about-to-abscise fruits due to suppression by a stronger auxin export from adjacent, more dominant fruit or shoot tips (Agusti *et al.,* 2000; Bangerth, 1989). Conclusively, it has been postulated that, if the IAA flux through the AZ is maintained, cell separation is inhibited and thus abscission prevented (Berüter and Droz, 1991). Moreover, the balance model proposes induction of abscission depends on a complex interplay of IAA and ethylene concentration (Beyer and Morgan, 1971; Sexton, 1998; Taylor and Whitelaw, 2001). Although Nunez-Elisea and Davenport (1986) suggested antagonistic relationship between ethylene and auxin is evident for abscission processes (Taylor and Whitelaw, 2001). In our study, 'Hôi' fruit exported more IAA compared to 'Tròn' fruit throughout the sampling period, which is correlated with greater fruit retention for 'Hôi'. Higher IAA export from fruitlets might control the sensitivity of the AZ to ethylene and thus abscission susceptibility. IAA is postulated to have a direct effect on assimilate partitioning (Agusti *et al.,* 2002; Patrick, 1979) i.e. the regulation of the flow of assimilates to developing fruits, thus might be an important factor in determining whether or not fruits are retained (Else *et al.,* 2004).

2. Carbohydrate: It was shown that 'Hôi' had an increased fruit pedicel thickness compared to 'Tròn', which might be linked to bigger fruit size and represents a cultivar specific feature between both cultivars. Similarly, it was observed in several citrus cultivars, where pedicel diameter was also linked to fruit size (Bustan *et al.,* 1995; El-Otmani *et al.,* 1993; Stewart *et al.,* 1952). Relationship between fruit set and carbohydrate availability are reported for several fruit trees, including mango (Chacko, 1991). A low carbohydrate level is often mentioned as one possible reason for premature fruit drop in mango (Davie *et al.,* 2000; Ram

et al., 1983). Generally, the photosynthetic capacity of the tree regulates the supply of carbohydrate with a high percentage of the accumulated photosynthates being primarily utilised for growth and development followed by the respiration process of the tree (Kozlowski, 1992). The surplus of carbohydrates is then stored, usually in the form of starch (Davie *et al.,* 2000; Kozlowski, 1992; Normand *et al.,* 2009). In our study, pedicels from irrigated trees indicated increased starch accumulation in AZ-cc; and starch grains were also found in cells of pith area, which was also reported for *Citrus* (Wilson and Hendershott, 1968). However, starch grains were not present in cells of the AZ separation layer itself, which was reported for tomato flower AZ (Roberts *et al.,* 1984). In different species starch accumulation was found in cortical cells of the leaf abscission zone, as well as in flower pedicels (Gilliland *et al.,* 1976; Roberts *et al.,* 1984). However, there are ambiguous observations of abscission susceptibility which might be linked with low starch levels (Kozlowski, 1992; Scott *et al.,* 1948). Further, Shiraishi and Yanagisawa suggested (1988) that starch was hydrolyzed in cells of the distal parenchyma and resynthesized during AZ formation suggesting an increase of starch concentration, respectively. According to Davie and Stassen (1997) mango trees set very large numbers of fruits which the tree nurtures before natural fruitlet abscission reduces crop load to levels the tree is capable to support. Further, it was concluded, that slow-growing small fruits were prone to abscission possibly due to competition for photosynthates and lower production of endogenous hormones (Krisanapook *et al.,* 2000). Referring to early stages of fruit growth in apple, a reduction of glucose below critical thresholds in the pedicel may induce the abscission process of fruits (Berüter and Droz, 1991). In this study, where the number of starch grains decreased in the AZ and the adjacent tissue an increased susceptibility of mangos to abscise might be linked to lack of energy source.

3. Water relations: According to Bally *et al.* (2000) water is the crucial factor, affecting flowering, fruit set, fruit size and total yield in mango, which is also supported in studies on apple (Berüter and Droz, 1991) and peach (Chalmers and Wilson, 1978; Li *et al.,* 1989b). Initial fruit growth within the first 6 weeks following full bloom represents a critical phase in mango fruit development when water deficiency might strongly affect crop load (Coelho and Borges, 2004). Further, fruit transpiration and the water movement through the pedicel affect the fruit water content (Jones and Higgs, 1982). However control and treated trees of both

cultivars indicated fruit abscission, water deficiency of control trees might affect negatively pedicel growth, respectively. Similar is confirmed in citrus by increased vascularisation of the pedicel due to transport of water and photosynthates to vigorously growing fruitlets (Garcia-Luis *et al.*, 2002). Generally, drought stress and related water deficits have been reported to cause abscission processes (Apelbaum and Yang, 1981; Taylor and Whitelaw, 2001). Further, plant water deficits associated by rehydration and shrinking of plant organs generating wound ethylene (McMichael *et al.*, 1972; Morgan *et al.*, 1990; Taylor and Whitelaw, 2001) have been linked to abscission and determine the IAA biosynthesis and metabolism thus inducing abscission process.

4. Conclusion and Outlook

Fruit drop can be regarded as one of the most serious problems for mango production in Northern Vietnam which affects the performance of the two main local cultivars ‘Hôi’ and ‘Tròn’. Both mango cultivars respond with enhanced fruit shedding during fruit set and early fruit development which falls into the dry season with low humidity and excessively high ambient temperatures.

Irrigation during flowering and early fruit growth stages improved final fruit retention and irrigated trees had higher crop load per inflorescence close to harvest compared to non-irrigated trees. Additionally, morphological changes in AZ tissue in mango showed, that irrigation had a positive effect on fruit retention in both cultivars, which might be influenced by increased pedicel thickness thus increased nutritional status of the fruit during critical environmental periods.

However, in northern Vietnam water resources especially on the hillside plantations are increasingly scarce resource and irrigation systems are costly investments for local farmer. As an alternative to reduce stress-induced fruit drop in mango, application of plant growth regulators at specific stages of fruit development are studied. In considering reducing fruit drop all chemical treatment indicated increased fruit retention. However, as shown in this study, single spray application of NAA and CPPU can effectively reduce fruit drop when sprayed shortly after flowering and ‘marble’-stage fruit size. Further, with low concentration and one-time application, PGR treatments indicate an economic viability and ease of handling for farmers.

However, we assume that shortage of carbohydrate may be associated with hormonal changes as signals triggering the abscission process. Here, more studies will be needed to elucidate the abscission process in mango, particularly the interplay between activation of the AZ, hormonal changes and carbohydrate reserves.

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Declaration of Originality

Hereby I declare that this doctoral thesis is independently written by myself. In addition, I confirm that no other sources than those specified in the thesis have been used. I assure that this thesis, in the current or similar format, has not been submitted to any other institution in order to obtain a Ph.D. or any other academic degree.

Ich erkläre hiermit, dass ich diese Dissertation selbständig angefertigt habe. Es wurden nur die im Literaturverzeichnis aufgeführten Hilfsmittel benutzt und fremdes Gedankengut als solches kenntlich gemacht. Ich versichere, dass ich diese Arbeit in gleicher oder ähnlicher Form noch keiner anderen Institution zur Prüfung vorgelegt habe.

Hohenheim, May 2011 Malte Gerhard Roemer

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