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Testing local cacao selections in Sulawesi for resistance to vascular streak dieback

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ABSTRACT

Vascular streak dieback (VSD) causes serious losses for cacao (or cocoa) smallholders in Sulawesi, Indonesia. The disease is caused by a Cantharellales species, *Ceratobasidium theobromae*, which colonises the xylem, resulting in leaf chlorosis or necrosis, abscission and eventual branch dieback. The disease is most successfully managed by pruning infected branches and the propagation of resistant genotypes. In participatory trials located in three provinces in Sulawesi, 2.5-year-old trees, which had been clonally propagated from local genotypes or the hybrid progeny of resistant parents, were evaluated for disease severity from 2010 to 2012. Consistent resistance rankings were obtained for clones common to the trials; these were confirmed by re-evaluation in 2014. From plot averages of disease severity, broad-sense heritability was estimated as 0.67–0.92. Two progeny clones, KW617 and ICCRI03, from East Java, had similar levels of resistance in the trials as their respective (resistant) parental clones, PBC123 and Scavina 6. Among four clones monitored for 3 months in West Sulawesi, PBC123 had a higher proportion of healthy leaves on the branch tips and a more restricted spread of infection within the xylem. In contrast, disease symptoms reached the younger leaves in susceptible clones. Individual branches of KW617, monitored from an early stage of symptom development, had a significantly lower number of diseased leaves and higher ratio of new to infected leaves after 9–16 weeks than that in four other clones. Other cacao clones with a relatively high number of diseased leaves during this period overcame infections with the addition of new flushes. Resistance in farm selections did not usually co-exist with yield and bean quality. Deriving new genotypes from crosses between parents with VSD-resistance and high-yield and/or quality traits is required for the production of promising clones with good resistance, yield and quality.

1. Introduction

Vascular streak dieback (VSD) is a serious systemic disease of cacao (or cocoa, *Theobroma cacao* L.) with a distribution in the Southeast Asian and west Pacific regions (Keane and Prior, 1991; Guest and Keane, 2007). The disease is caused by a Cantharellales species, *Ceratobasidium theobromae*, syn. *Thanatephorus theobromae*, *Oncobasidium theobromae* (Talbot and Keane, 1971; Roberts and Royal Botanic Gardens, 1999; Samuels et al., 2012). VSD is characterised by either leaf chlorosis or necrosis, followed by abscission and, in susceptible genotypes, branch dieback. Severe impacts of this disease have been reported in Indonesia, including Sulawesi (Turner and Keane, 1985; Pawirosoemardjo et al., 1990; Pawirosoemardjo and Purwantara, 1992; Guest and Keane, 2007; Parawansa et al., 2013; McMahon and Purwantara, 2016). Indonesia produced 545,000 tonnes of cocoa beans in 2006/2007, but this had declined to 325,000 tonnes by 2014/15, with further declines predicted (http://www.icco.org/about-us/icco-annual-report.html). Among the factors contributing to lower farm productivity, including ageing trees, soil fertility decline and socio-economic trends such as the increasing average age of farmers, VSD has

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been one of the most important and has frequently influenced smallholders to replace their cacao with other crops. In their 2008/2009 annual report, the International Cocoa Organisation (ICCO) identified VSD as a major constraint to cacao production in the region.

Fungal isolates from infected cacao leaves and petioles in Sulawesi (Samuels et al., 2012) revealed the typical *Rhizoctonia*-like morphology of the VSD pathogen described in Papua New Guinea (Talbot and Keane, 1971; Keane et al., 1972) and Malaysia (Mainstone et al., 1983; Zainal Abidin et al., 1984). Furthermore, the morphology and dimensions of basidiospores in sporocarps are consistent with the descriptions of Talbot and Keane (McMahon and Purwantara, 2016). Internal transcribed spacer (ITS) and large subunit (LSU) sequences of Sulawesi isolates have 99–100% identity with isolates from VSD-infected tissues in Papua New Guinea (PNG), West Papua, Java, Malaysia and Vietnam (Samuels et al., 2012). Whilst the symptoms of VSD infection may become severe during drier periods, the disease spreads between trees in the rainy season, which provides the necessary conditions for sporulation. During wet periods, adherent sporocarps with monilioid hyphae and basidia are formed from mycelia emerging from vascular tissue on scars left by abscissed leaves or on cracked petioles or leaf mid-ribs. Basidiospores are produced at night under moist conditions (Keane et al., 1972; Dennis et al., 1992) and dispersed for up to 100 m by wind (Keane, 1981). Infection of flush leaves at the branch or seedling tip is followed by colonisation of the xylem. Hyphae then spread through vascular tissue to adjacent leaves, causing leaf drop and eventually killing the seedling or the branches of more susceptible cacao genotypes (Keane et al., 1972; Prior, 1978; Keane, 1981). Visible symptoms develop 3–5 months after infection. Branch death may occur around 6 months after the initial infection, although infected branches in cacao genotypes with partial resistance can remain alive for long periods and can even occlude infections and continue growing (Prior, 1979, 1985; Keane and Prior, 1991).

Until the 2000s, the most common VSD symptom resulting from infection of cacao trees was chlorosis, characterised by yellowing leaves with patches of green tissue, followed by rapid leaf abscission. In the last decade, a new set of VSD symptoms has become more common in cacao-growing areas throughout Southeast Asia and the west Pacific. Characteristically, distinct dark lesions beginning on leaf margins or tips expand until they cover all or most of the leaf lamina. Leaves with such necrotic symptoms also remain attached for a longer period of time than those with chlorotic symptoms. Moreover, sporulation may occur in cracks in the petiole or leaf and on leaf scars (Purwantara et al., 2009; Parawansa et al., 2013, 2014).

Management of VSD is achieved largely by the cultural methods of pruning diseased branches, raising seedlings in nurseries under UV-resistant plastic sheeting (to prevent infection by the wind-borne spores) and the use of resistant plant material (Keane and Prior, 1991). Some triazolic systemic fungicides have been shown to be effective in controlling VSD (Prior, 1987; Singh, 1989; Holderness, 1990), but their high cost makes them uneconomic in most cacao farming situations. Frequent and regular pruning programmes have been shown to be effective on estates in West Java (Pawirosoemardjo et al., 1990; Pawirosoemardjo and Purwantara, 1992) and Malaysia (Jayawaderna et al., 1978) but are extremely labour intensive. Branches are cut about 30 cm below the extent of visible infection – pruned branches can be left on the farm as sporulation only occurs on attached, living branches (Prior, 1985; Keane and Prior, 1991). Such widespread programmes are difficult to implement on smallholdings, in which investment in management may vary greatly from farm to farm. Lack of management (especially sanitation pruning) and the propagation of relatively susceptible genotypes results in a higher rate of sporulation and infection of neighbouring trees (Keane, 1981; and see Ploetz, 2007). Therefore, on smallholdings in particular, the use of resistant genotypes, combined with adequate cultural management, is the most promising strategy for reducing the impacts of this disease.

Cacao genotypes surviving severe epidemics of VSD in PNG and Malaysia in the 1960s and 70s (Shaw, 1962; Bridgeland et al., 1966; Mainstone et al., 1983) demonstrated that VSD resistance occurred among diverse genotypes on farms (Keane, 1974; Prior, 1979; Chong and Shepherd, 1986; Keane and Prior, 1991; Efron et al., 2002). Resistant clones, such as KA2101, KEE1, PBC123 and KKM22, initially selected in PNG and Malaysia, are still propagated or used in breeding programmes (Chong and Shepherd, 1986; Efron et al., 2002). Two clones, PBC123 and BR25, were selected in the 1980s in Malaysia by screening the progeny of crosses for resistant, high-yielding types. These clones are presently the most widely planted clones in Sulawesi, Indonesia, where they have been renamed Sulawesi 1 and 2, respectively. Tan and Tan (1988) concluded the genetic basis of VSD resistance to be mainly additive and quantitative, in line with a horizontal type of resistance (Vanderplank, 1963; Keane, 2012). This is supported by the durability of resistance occurring in resistant genotypes (Prior, 1979; Keane and Prior, 1991).

In Sulawesi, a strategy of selecting and testing local cacao genotypes on smallholder farms, which enables the identification of genotypes with promising resistance and other characteristics, demonstrated the potential of a participatory farmer approach to select from genetic resources available on smallholdings (McMahon et al., 2009, 2010; 2015). The potential yield, quality characteristics and resistance to cacao pod borer (*Conopomorpha cramerella*) and *Phytophthora palmivora* of local clones tested in farm-based trials in three provinces in Sulawesi has been reported previously (McMahon et al., 2015; Purwantara et al., 2015). The clones tested in these trials included local farm selections and hybrid progeny identified with possible VSD resistance, in addition to susceptible clones. Here we report evaluations of the response of these clones to VSD infections transmitted under field conditions and identify cacao genotypes with potential resistance to the disease.

2. Materials and methods

2.1. Trial establishment

Trials were established in March 2008 at four sites in Sulawesi, each testing 11–12 local cacao clones. Clones were selected from working farms or from progeny of hybrid crosses. A few of these clones were common to two or more of the trials. Clones common to all four trials were the Malaysian clone, PBC123, also known as 'Sulawesi 1' (widely propagated for its VSD resistance and high yield) and the high-yielding M01, now officially released and renamed Masamba Cacao Clone 1 (MCC01). Three trials established on farms in early 2008 were located in West Sulawesi in the village of Beluak in Anreapi sub-district, Polewali-Mandar (Polman) District, South Sulawesi (Fakkie Village, Tiroang, Pinrang District) and Southeast Sulawesi (Pakue, North Kolaka District). The trial design, latitude/longitude coordinates and the clones tested in these trials have been reported previously (McMahon et al., 2015). A fourth trial, also established in early 2008, was located on a working farm in Bone-Bone subdistrict, North Luwu: latitude 02°38′587″S and longitude 120°37′163″E and an elevation of 67 m above sea level. The trial in North Luwu tested 11 cacao genotypes, including clones in common with the other trials, additional on-farm selections and a hybrid cross, ICCRI04 (see Table 1). To establish the trials, budwood obtained from mature trees of selected genotypes was grafted onto rootstock seedlings in a nursery and after 4–5 months planted at the trial sites. Each clone was propagated in 8-tree plots, with four replicate blocks, providing a total of 384 trees for the trials testing 12 clones and 352 trees in the North Luwu trial. In the North Luwu trial, some trees were lost to root diseases or had stunted growth so that finally 4–7 trees per plot were assessed for VSD severity. VSD, endemic to most cacao-growing areas in Sulawesi, was transmitted naturally under field conditions. As for the other trials, pesticides were not applied to the trees in North Luwu except for fungicidal treatment at the early stages of establishment after planting. No insecticides or fungicides were applied during the period of evaluation. Monthly rainfall