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Effects of Chemical Properties of Different Soils on *Ganoderma* Disease in Oil Palm (*Elaeis guineensis*)

Yit Kheng Goh*; Choon Kiat Lim*; Cong Rong Cheng*; Suet Yee Tan*; Li Wen Cheah*; Petronella G Ah Tung*; You Keng Goh* and Kah Joo Goh*

ABSTRACT

Variations in the incidence of Ganoderma *basal stem rot disease in oil palm (*Elaeis guineensis*) on different soils had been observed in the field. Notably, acid sulphate coastal soils were observed to have higher* Ganoderma *incidence compared with inland soils. Therefore, the objective of this experiment was to study the effects of four different soils belonging to five soil series, namely Parit Botak* and Jawa** series - acid sulphate coastal soil, Bungor*** series - inland soil, Blenheim series - coastal shell deposit soil (*Typic Quartzipsamments*), and Kabu series - limestone-derived soil, coarse sandy loam (*Typic Paleudults*), on* Ganoderma *incidence and severity in oil palm seedlings in the nursery. The results show that the seedlings planted on both samples of acid sulphate coastal soils had the highest* Ganoderma *incidence and severity whereas those planted on coastal shell deposit soil had the lowest. Total number and weight of fruiting bodies produced at five months post-inoculation were exponentially correlated to percent reduction in dry weight of* Ganoderma*-inoculated rubber wood blocks (RWB). Inoculated RWB from Parit Botak series were the most friable and the softest, followed by those from Jawa and Kabu series. Three sets of symptoms of* Ganoderma *infection were observed: a) leaf symptoms prior to initiation of* Ganoderma *fruiting bodies (on Bungor and Jawa series); b) leaf symptoms after initiation of* Ganoderma *fruiting bodies (on Blenheim and Parit Botak series); and c) a combination of the two sets (on Kabu series). Discriminant analyses elucidated that zinc, copper and calcium were associated with more than 50% disease severity index (DSI) and necrotic bole tissues. In contrast, pH, iron, manganese and nitrogen were associated with less than or 50% of DSI and necrotic bole tissues. This study illustrates the impacts of soils with different chemical compositions on the incidence and severity of* Ganoderma *disease.*

* Advanced Agriecological Research Sdn Bhd
No. 11 Jalan Teknologi 3/6, Taman Sains Selangor 1,
Kota Damansara, 47810 Petaling Jaya,
Selangor, Malaysia.
E-mail: gohykheng@aarsb.com.my

ABSTRAK

Pemerhatian di ladang mendapati bahawa insiden penyakit Ganoderma *reput pangkal sawit (*Elaeis guineensis*) adalah berbeza pada tanah yang berlainan. Tanah asid sulfat pantai telah didapati mengalami insiden jangkitan* Ganoderma *yang lebih tinggi berbanding di tanah pedalaman. Oleh itu, objektif kajian ini adalah untuk mengkaji kemungkinan berlaku insiden jangkitan* Ganoderma *dan tahap kerosakan ke atas anak benih sawit di tapak semaian menggunakan tanah yang berlainan. Kajian ini menggunakan lima siri tanah dari empat taksonomi berbeza iaitu siri Parit Botak* dan siri Jawa** - tanah sulfat asid pantai, siri Bungor*** - tanah pedalaman, siri Blenheim - tanah mendapan shell pantai (*Typic Quartzipsamments*), dan siri Kabu - tanah terhasil dari batu kapur, lom berpasir kasar (*Typic Paleudults*). Keputusan mendapati bahawa anak benih yang ditanam di kedua-dua tanah asid sulfat pantai mengalami insiden jangkitan* Ganoderma *dan tahap kerosakan tertinggi, manakala yang ditanam di tanah mendapan shell pantai mengalami jangkitan terendah lima bulan selepas diinokulasi, jumlah bilangan dan berat jasad berbuah yang dihasilkan didapati mempunyai korelasi secara eksponensial dengan pengurangan peratus berat kering blok kayu getah terinokulasi. RWB yang diinokulasi dari siri Parit Botak adalah yang paling rapuh dan lembut, diikuti oleh RWB di tanah siri Jawa dan Kabu. Pemerhatian dilakukan ke atas tiga set simptom jangkitan* Ganoderma*: a) simptom daun sebelum terhasilnya jasad berbuah* Ganoderma *(pada siri Bungor dan Jawa); b) simptom daun selepas terhasilnya jasad berbuah* Ganoderma *(pada siri Blenheim dan Parit Botak); dan c) gabungan kedua-dua set (pada siri Kabu). Analisis diskriminasi menunjukkan bahawa zink, kuprum dan kalsium mempunyai kaitan dengan indeks kerosakan penyakit (DSI) dan nekrotik di*

Notes:
* Parit Botak (Typic Sulfaquepts)
** Jawa (Sulfic Tropaquepts)
*** Bungor (Typic Paleudults)

tisu bole melebihi 50%. Sebaliknya, pH, ferum, mangan dan nitrogen mempunyai kaitan dengan DSI dan nekrotik tisu bole pada tahap 50% atau kurang. Kajian ini menunjukan bahawa tanah yang mengandungi komposisi kimia yang berlainan akan memberi impak berbeza terhadap insiden dan kerosakan disebabkan penyakit Ganoderma.

Keywords: BSR, fruiting body, soil nutrients, disease severity, disease incidence.

INTRODUCTION

Ganoderma boninense Pat., one of the main causal agents of basal stem rot (BSR) in oil palm in Malaysia, Indonesia and a few other countries, causes about RM 1.5 billion in economic losses annually in Malaysia alone (Arif *et al.*, 2011; Susanto, 2009; Singh, 1991). This basidiomycetous pathogen was observed to spread in the coastal areas before moving into inland soils in Peninsular Malaysia (Singh, 1991). In recent years, BSR has become more alarming in East Malaysia, namely Sabah (Chong *et al.*, 2011) and Sarawak (Rakib *et al*., 2014). A few decades ago, *Ganoderma* BSR was spotted mainly in mature and old mature oil palm over 10 years old. However, in recent years the disease has started to infect young palms aged 12 to 24 months in the field (Chung, 2011; Singh, 1991). Differences in pathogenicity level and *in vitro* growth rate were observed in *G. boninense* isolates collected from different states in Peninsular Malaysia (Kok *et al*., 2013). Furthermore, these *G. boninense* isolates caused slower vegetative growth and eventual death of the palms (Goh *et al*., 2014).

Biotic and abiotic factors can weaken plant vigour, and lead to higher incidence or severity of pest and disease attacks (Boa, 2003). Boa (2003) also highlighted five major abiotic factors, namely chemicals, mechanical agents, soil conditions, water and weather, that can affect the health of plants. Various soil factors and properties, namely pH, conductivity, water table, and nutrition, were proposed to be pre-disposing abiotic factors for *Ganoderma* infection (Ariffin *et al.*, 2000; Singh, 1991). Lower BSR incidence was observed in palms planted on coastal acid sulphate soils (previously also planted with oil palm) compared with those planted on soils without the acid sulphate layer (Singh, 1991). Contrary to this finding, another study reported that palms on acid sulphate soils had higher *Ganoderma* incidence compared with those on other soils (Parthiban *et al*., 2016). It was proposed that coastal soils with high clay content and water table or high water retention capacity stressed the palms, and, hence, led to *Ganoderma* infection. Furthermore, Singh (1991) reported that oil palm on a soil (previously planted with coconut) with a high level of salinity or conductivity (μs cm^{-1}) showed low *Ganoderma* incidence. In this preliminary study, the effects of five different soil series, namely Bungor, Jawa, Parit Botak, Blenheim and Kabu, representing four soil types, on *G. boninense* disease in two-month-old oil palm seedlings were assessed.

MATERIALS AND METHODS

Soil Series, Types and Origins

Coastal, inland and limestone-derived soils were selected for the study. Soils from Bungor series (*Typic Paleudults*, inland, fine sandy clay) (coordinates: 3°16′13.36″ N; 101°27′18.62″ E) and Jawa series (*Sulfic Tropaquepts*, coastal, clayey) (coordinates: 3°10′55.76″ N; 101°22′06.52″ E) were collected from Tuan Mee Estate in Selangor, in central Peninsular Malaysia. Soils from Parit Botak series (*Typic Sulfaquepts*, coastal, clayey) and Blenheim series (*Typic Quartzipsamments*, shell deposits, coastal, sandy clay loam) were sampled from Blenheim Estate (coordinates: 3°55′39.40″ N; 100°48′48.58″ E) in Perak, in northern Peninsular Malaysia. Soil from Kabu series (*Typic Paleudults*, coarse sandy loam, limestone) was collected from Menglembu Estate (coordinates: 4°33′40.77″ N; 101°02′07.39″ E) in Perak, also in northern Peninsular Malaysia. Samples of the five soils were analysed for pH and chemical properties according to methods described by {Kee *et al*. (1995), Teh *et al*. (2011) and Lee *et al*. (2012). Additional soil samples were collected and transported to Advanced Agriecological Research Sdn Bhd (AAR) in Kota Damansara, Selangor, for pathogenicity testing in the nursery.

Artificial Inoculation of Oil Palm Seedlings and Pathogenicity Test

One of the most aggressive *G. boninense* isolates (G10) in our collection (giving the highest disease incidence and severity index) (Kok *et al*., 2013) was used to study pathogenicity of *G. boninense* in two-month-old seedlings of a Dumpy Yangambi Avros DxP cross grown on the five different soils. Rubber wood blocks (RWBs) of size 6 cm x 6 cm x 12 cm were used as the substrate for *G. boninense* and were prepared according to the method by Kok *et al*. (2013). The protocols used in the nursery experiment were conducted according to Kok *et al*. (2013) and Goh *et al*. (2014). The experiment was set up in a randomised complete block design (RCBD) in which each treatment (respective soil series sample) was replicated eight times, except for Kabu series, which had only six replicates due to the limited amount of soil collected.

Soil Water Volumetric Content and Temperature

Soil water volumetric content and temperature for all the treatments were determined using a WaterScout SM 100 soil moisture sensor and an IQ Scientific 150 pH meter & thermometer (Spectrum Technologies, Inc), respectively. Both parameters were measured at around 12.00 noon to 1.00 pm and approximately four hours after watering the oil palm seedlings in the nursery.

Disease Scores and Fruiting Body Formation

Non-destructive sampling: Disease incidence (DI) and disease severity index (DSI) were scored at three and five months post-inoculation (mpi) according to the approaches outlined by Ili Nadhrah *et al*. (2015), Kok *et al*. (2013), and Nur Ain Izzati and Abdullah (2008). Destructive sampling: The size of the bulb lesion or necrotic tissues in the stem bole was determined according to the grid method outlined by Idris *et al*. (2000). The bole was cut longitudinally at the end of the experiment, and the area of tissues with lesions or necrotic symptoms was recorded. The number of well-formed *Ganoderma* fruiting bodies and total weight of all the fruiting bodies obtained from each soil series were recorded at the end of the trial.

Decay of Rubber Wood by *G. boninense* in Different Soil Series

Ganoderma-inoculated RWBs (used to artificially infect the oil palm seedlings) from the respective soil series were collected and weighed to determine their fresh and dry weights. All RWBs were cleaned with double-distilled water to remove all soil particles and plant debris. Dry weight of the *Ganoderma*-inoculated RWBs as well as non-inoculated RWBs was determined by drying the pre-cleaned RWBs in an oven at 80°C for 12 hours (longer drying periods resulted in severe cracking of the wood blocks) prior to recording the relative dry weight readings. Percent reduction in dry weight of RWB was calculated based on the following formula:

$$\% \text{ reduction in dry weight} = \left[\frac{(DW_N - DW_I)}{(DW_N)} \right] \times 100$$

where DW_N refers to dry weight of non-inoculated RWB while DW_I refers to dry weight of *Ganoderma*-inoculated RWB (obtained from the different soil series). Isolation of *G. boninense* from infected or colonised RWB was conducted using *Ganoderma*-selective medium (GSM) (Ariffin and Idris, 1991).

Statistical Analysis

Means of soil nutrients, soil pH, and % reduction in dry weight of the decayed RWBs were normally distributed (with Anderson-Darling test). Differences in these means were analysed with Tukey's test at $P = 0.05$. However, means for both DSI and % of necrotic tissues in the bole were not normally distributed (with Anderson-Darling test). Therefore, in this case, differences in means were analysed with Kruskall-Wallis test and followed by Mann-Whitney U test at $P = 0.05$ (Minitab 16). Multivariate canonical discriminant analysis (CDA) was used to assess the effects of the various soil nutrients on DSI and % necrotic tissues in the bole using SPSS (SPSS 16.0) software. Prior to CDA analysis, disease scores were grouped into two categories: more than 50%, and 50% or less.

RESULTS AND DISCUSSION

Soil Analyses

Chemical analyses of the soil samples showed that pH, organic carbon (Org.C), nitrogen (N), iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), potassium (K), calcium (Ca), magnesium (Mg) and aluminium (Al) varied among the soil series (*Table 1*). However, canonical discriminant analysis identified only five chemical properties were required to distinguish the five soils. They were pH, organic carbon (OrgC), Mn, Zn and Cu (Supplementary data: *Table 1S*).

Soils from the coastal areas, namely Parit Botak and Jawa series (with higher proportions of clay and silt compared to Kabu and Blenheim), had lower pH of 4.41 (4.31 to 4.54) and 4.06 (3.93 to 4.31), respectively, compared with the other sampled soils (*Table 1*). In one previous study, bentonite clay was found to improve the growth rate of the fungal pathogen, *Gaeumannomyces graminis*, and it was also proposed that this was due to the effect of water availability, and the clay being able to reduce antagonistic effects by antagonists (Campbell and Ephgrave, 1983). The effects of clay composition on *Ganoderma* growth and pathogenicity will require further studies. Bungor series inland soil had higher pH at 4.67 (4.37 to 5.01), compared with that of the coastal soils. Kabu series soil (which had a higher proportion of coarse sand than the other soils) had pH of 7.03 (6.71 to 7.30). The most alkaline soil was Blenheim (relatively higher in sand than silt and clay) at pH 7.90 (7.82 to 7.98) (*Table 1*). Low pH in Parit Botak and Jawa series soils was due to the presence of jarosite in their soil profile. High pH or the alkaline condition of Blenheim soil may be

TABLE 1. SOIL NUTRIENTS AND pH ANALYSES FOR JAWA, BUNGOR, PARIT BOTAK, BLENHEIM AND KABU SOIL SERIES SOILS

Soil series*	pH in water (2:5)	Org.C (%)	N (%)	Fe	Mn	Zn	Cu	Exchangeable cations (cmol(+) kg^{-1})				CEC (cmol(+)kg^{-1})
				(mg kg^{-1})				K	Ca	Mg	Al	
Jawa	4.06 e	1.39 b	0.25 a	11609 a	14.65 b	23.26 a	10.89 a	0.70 b	4.32 b	0.98 b	12.46 a	15.21 b
Bungor	4.67 c	0.90 c	0.12 c	11288 a	0.53 b	3.90 c	6.53 b	0.10 c	1.67 c	0.13 c	1.45 c	3.98 c
Parit Botak	4.41 d	1.32 b	0.17 bc	6002 b	25.02 b	15.95 b	7.24 ab	0.82 a	4.46 b	3.24 a	8.34 b	20.63 a
Blenheim	7.90 a	1.92 a	0.18 b	837 c	91.69 ab	6.08 c	8.51 ab	0.04 d	16.95 a	0.11 c	ND d	4.69 c
Kabu	7.03 b	0.24 d	0.03 d	2669 c	238.00 a	5.98 c	8.67 ab	0.03 d	0.39 c	0.02 c	ND d	2.93 c

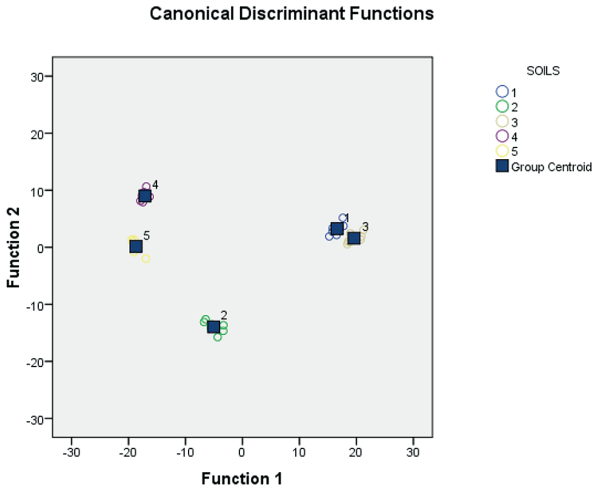
Note: *All the soil types were analyzed for 8 replicates, except Kabu which had only 6 replicates.
Abbreviations: Org.C = Organic carbon, N = nitrogen, Fe = iron, Mn = manganese, Zn = zinc, Cu = copper, K = potassium, Ca = calcium, Mg = magnesium, Al = aluminium, CEC = cation exchange capacity.
Means within each individual column followed by the same letter are not significantly different from one another at $P = 0.05$ according to Tukey's test.

SUPPLEMENTARY DATA

TABLE S1. STANDARDISED CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS FOR FIVE DIFFERENT SOIL SERIES BASED ON ANALYSED pH AND OTHER NUTRIENTS

Standardised canonical discriminant function coefficients				
	Function			
	1	2	3	4
pH	**-0.664**	1.126	0.666	0.122
Org.C	**-0.049**	0.130	-0.657	0.218
N	0.151	0.094	-0.253	-0.154
Mn	**-0.193**	**-0.049**	-0.136	0.965
Ca	0.299	0.033	-1.064	-0.027
Zn	**-0.296**	0.541	0.664	-1.549
Al	0.714	1.097	0.249	-0.018
K	0.638	0.024	-0.173	0.488
Mg	0.681	0.252	0.009	0.928
Cu	**-0.312**	**-0.156**	0.257	-0.181
CEC	0.171	0.355	0.736	-0.192

Note: *Abbreviations: Org.C = Organic carbon, N = nitrogen, Fe = iron, Mn = manganese, Zn = zinc, Cu = copper, K = potassium, Ca = calcium, Mg = magnesium, Al = aluminium, CEC = cation exchange capacity.



Legend: 1 = Jawa; 2 = Bungor; 3 = Parit Botak; 4 = Blenheim; 5 = Kabu.

Figure S1. Canonical discriminant functions for five different soil series.

caused by the calcareous shell deposits, which also contributed to the high soil Ca status. High levels of iron, aluminium and zinc in Parit Botak and Jawa soils could be due to the oxidation of jarosite (Shamshuddin, 2006; Bigham and Kirk Nordstrom, 2000; Shamshuddin and Auxtero, 1991). Manganese level was high in Kabu and Blenheim soils (*Table 1*).

During the soil sampling, differences in *Ganoderma* BSR incidence in palms growing on the

various soil series were noted in the field. Oil palm on Blenheim series soil (1998 planting) had less than 8% *Ganoderma* incidence. Palms on Bungor (2008 planting) and Kabu (1999 planting) had approximately 8% to 16% *Ganoderma* incidence, whereas those on Parit Botak (1997 planting) and Jawa (1996 planting) had more than 16% *Ganoderma* incidence. It was based on these variations in *Ganoderma* incidence, as well as their differences in soil pH and chemical properties, that these five soil series were selected for the *Ganoderma* pathogenicity experiment.

Disease Scores and Fruiting Body Formation on Different Soil Series

Disease incidence (DI) and disease severity index (DSI) of oil palm seedlings planted in polybags containing Kabu soil were highest at 3 mpi, followed by the seedlings on Jawa, Parit Botak, Bungor and, lastly, Blenheim series soils (*Table 2*). Furthermore, formation of fruiting bodies was relatively faster on Kabu soil, starting around 45 days after inoculation, compared with the other soils. Total number of fruiting bodies was also the highest on the surface of this soil (*Table 5*). The coarse sandy loam texture with low water volumetric content (<10%) or low water-holding capacity, higher soil temperature compared with the other soils (1°C to 2°C higher) (32°C for Kabu, and 29.8°C to 30°C for clayey soils), and low soil nutrient status in Kabu soil might have caused greater abiotic stress to the oil palm seedlings, resulting in early formation of fruiting bodies of *G. boninense*, and the high DI and DSI at 3 mpi. Abiotic factors, such as temperature, aeration, humidity, light radiation, wounding, acetic acid and nutrient depletion, have been proposed to induce the production of fruiting bodies in fungi (Döll *et al*., 2013; Lin *et al*., 2006). Moreover, the low water- and nutrient-holding capacity of Kabu soil (comprising mainly of coarse sand) could have increased the stress conditions experienced by the palm seedlings, and pre-disposing them to a higher risk of *Ganoderma* infection (*Table 2*). Furthermore, Narisawa *et al.* (2005) reported that *Heteroconium* disease development in Chinese cabbage was higher in a soil with low pH (5.5) and high soil moisture content compared with a soil with high pH (6.3 and 7.2) with low moisture content. These observations are in accordance with the current observations on *Ganoderma*-oil palm pathogenicity.

From 3 to 5 mpi, Jawa and Parit Botak series soils recorded the highest increment in both DI and DSI values, followed by Bungor and Kabu, and lastly, by Blenheim (*Table 2*). Furthermore, the lowest percent necrotic lesion tissue in the bole was noted among the seedlings planted on Blenheim soil. DI and DSI at 5 mpi correlated well with field observations on disease incidence as affected by the same soil series. For instance, DI and DSI were highest in acid sulphate soils, followed by Bungor, Kabu and Blenheim series soils. Observations from the nursery experiment were consistent with the field disease scores. Furthermore, in a recent study, oil palm growing on Jawa series soil (with an acid sulphate layer) were surveyed and found to have higher *Ganoderma* incidence and lower life expectancy compared with those on non-acid sulphate soils (*viz*. Briah and Selangor) (Chen *et al*., 2017).

Fewer basidiocarps were observed on Bungor soil with only two well-formed fruiting bodies

TABLE 2. DISEASE SCORES AND PERCENT OF NECROTIC TISSUES IN BOLE OF OIL PALM SEEDLINGS PLANTED IN FIVE DIFFERENT SERIES SOILS

Treatment (soil series)	Disease score*				% of necrotic tissues†
	3 mpi**		5 mpi**		
	DI	DSIϕ	DI	DSIϕ	
Jawa	42.9	7.1 ab	100	88.1 a	55 a
Bungor	25	4.2 ab	75	54.2 ab	56.88 a
Parit Botak	37.5	6.3 ab	100	70.8 ab	60 a
Blenheim	12.5	2.1 b	50	37.5 b	38.75 a
Kabu	66.7	11.1 a	66.7	50 ab	50.83 a

Note: *Two disease scores were adopted: DI (disease incidence) and DSI (disease severity index).
**mpi refers to months post-inoculation.
†Percent of necrotic tissues was observed in the oil palm bole.
ϕDSI for the respective treatments at 3 and 5 mpi is presented as the mean of 8 replications (except Kabu soil, as the mean of only 6 replications). DSI at different mpi and % of necrotic tissues were analysed separately.
Means within each column for DSI and % of necrotic tissues followed by the same letter are not significantly different from one another at $P = 0.05$ according to Kruskal-Wallis test, followed by Mann-Whitney test.

(*Figure 1c*) and on Jawa soil with four well-formed fruiting bodies (*Figure 1b*) at 5 mpi (*Figure 1*), although signs of basidiocarp initiation around the bole of the dead seedlings were noted. Infection symptoms varied among palms on the different soils (*Figure 2*) as follows:

a) Chlorotic leaf symptoms appeared prior to the production of basidiocarps on seedlings in Bungor and Jawa soils, thus resulting in a lower number of well-formed basidiocarps. These chlorotic leaf symptoms resulted in earlier seedling death (*Figure 2b*).
b) Chlorotic leaf symptoms appeared after the formation of basidiocarps on the seedlings in Parit Botak and Blenheim soils (*Figures 2a and 2c*).
c) A combination of both sets of symptoms was observed in seedlings raised in Kabu soil (*Figure 2d*).

Variations in basidiocarp morphology were noted among the different soils (*Figure 1*). At the end of the experiment, both Jawa and Parit Botak soils produced higher DI and DSI than the rest (*Table 2*). Heavy soils, which are clayey and silty, with high water-retention capability had been suggested to allow for the proliferation of *G. boninense*, while the high moisture conditions had been proposed to be unfavourable to the growth of antagonistic fungi (Singh, 1991). In the current experiment, water volumetric content for Parit Botak, Jawa and Bungor soils was 30%-37%, 25%-30% and 13%-20%, respectively. Among all the soils, Kabu and Blenheim soils had lower water volumetric content (below 10%) compared with the clayey soils. *Ganoderma* inoculation in Blenheim soil resulted in the lowest DI, DSI, and % of necrotic tissues in the bole of the seedlings (*Table* 2). The high pH of Blenheim soil (*Table 1*) could be one of the reasons for the lower *Ganoderma* infection compared with the other soils with lower pH.

Based on canonical discriminant analyses, the soil nutrients responsible for high (more than 50%) or low (50% or lower) DSI and % of necrotic tissues in the bole are summarised in *Tables 3* and *4*. High pH, Mn and Fe could have contributed to the low DSI and % of necrotic tissues in the bole, whereas high Ca, Zn and Cu might be responsible for the high DSI and % of necrotic tissues in the bole (*Tables 3* and *4*). Soil pH does play an important role in suppressing or increasing fungal diseases. If both acidic (Jawa, Bungor, and Parit Botak) and alkaline (Blenheim and Kabu) soils were analysed separately, DI and DSI at 3 mpi were inversely correlated to an increase in soil pH ($r^2 > 0.90$) (*Tables 1* and *2*).

Ganoderma-infected RWBs from Different Soils

Data on percent reduction in dry weight of *Ganoderma*-inoculated RWBs in the different

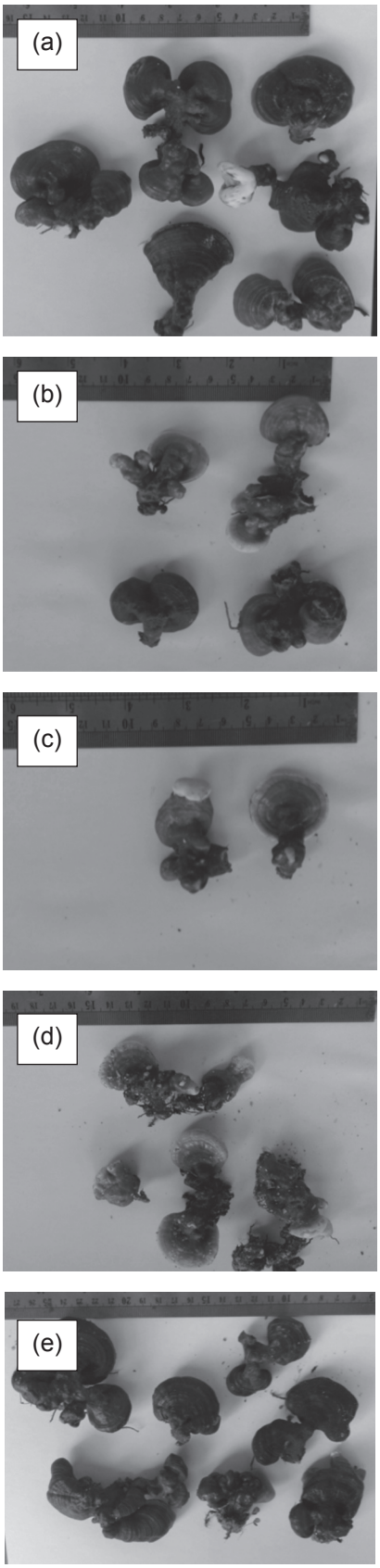


Figure 1. Morphology of fruiting bodies produced by Ganoderma boninense *on oil palm seedlings planted on different soil series, harvested at 5 months post-infection – Parit Botak (a), Jawa (b), Bungor with the least amount of fruiting bodies (c), Blenheim (d), and Kabu (e) (appeared to be darker – blackish brown – and thicker, and higher number of fruiting bodies).*

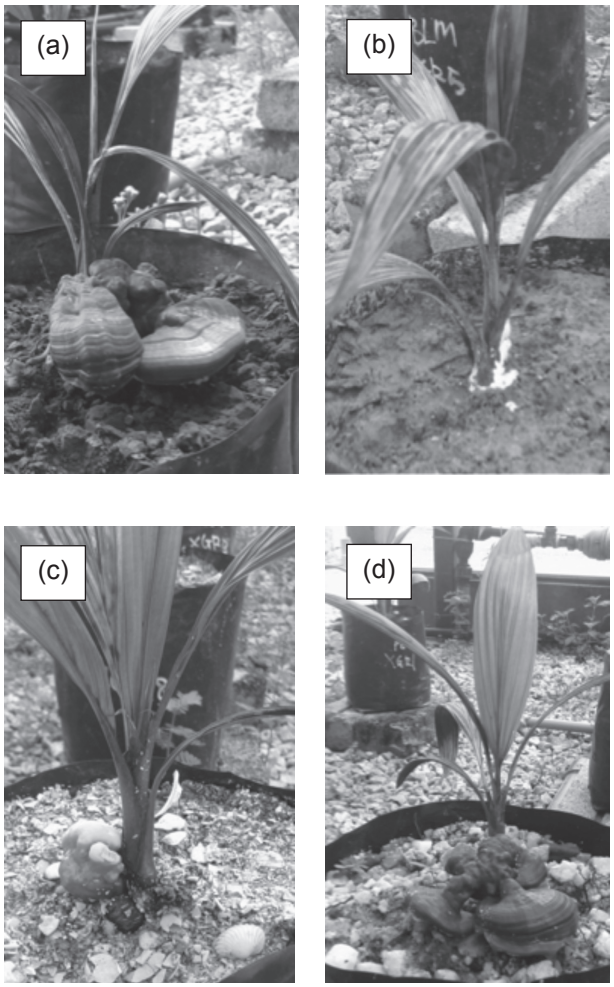


Figure 2. Morphology and formation of Ganoderma boninense *fruiting bodies on oil palm seedlings planted on different soil series at 5 months post-infection – Parit Botak with well-formed fruiting bodies on dead seedling (a), Bungor with fungal mass on dead seedling (e), Blenheim with initial stage of fruiting body formation on chlorotic seedling (c), and Kabu with well-formed fruiting body on chlorotic seedling (d).*

soils compared with non-inoculated RWBs are summarised in *Table 5*. *Ganoderma*-inoculated RWBs in Bungor soil had the lowest percent reduction in dry weight (22.6%) compared with the others. On the other hand, inoculated blocks from Kabu soil had the highest reduction in dry weight (50.2%). There was a high exponential relationship between total weight of *Ganoderma* fruiting bodies produced (y) and percent reduction in RWB dry weight (x), reflected by the equation $y = 3.456^{0.0645x}$ ($r^2 = 0.94$), and a high correlation between total number of fruiting bodies and percent of reduction in RWB dry weight ($r^2 = 0.97$). Losses in RWB dry weight could be due to degradation of RWB by *G. boninense* to produce fruiting bodies. Rees (2006) reported that enzyme activities were involved in the degradation of lignin and other cell wall components. Cell wall degrading enzymes produced by *G. boninense* might be useful for the pathogen to colonise and exploit the food source from RWBs. RWBs from Parit Botak soil were the most friable compared with those from Blenheim, Kabu and Bungor soils.

Isolation of *Ganoderma* from Inoculated RWBs

Small pieces of RWB of size 1 to 2 cm in length were inoculated onto GSM medium to retrieve viable *G. boninense* cultures. *Ganoderma* cultures were revived from most of RWBs, except the blocks from Blenheim soil. The latter did not produce viable *Ganoderma* cultures, probably because of the presence of bacteria in the soil which competed with or were parasitic to *Ganoderma*. However, further studies are required to verify the effects of other soil microbes, namely bacteria, on the survival of *Ganoderma* in RWBs.

CONCLUSION

Among all the soils tested, oil palm seedlings planted on acid sulphate coastal soils and Bungor series soil had higher *Ganoderma* incidence and severity, as well as higher percentage of necrotic tissues in the bole. Seedlings planted on Blenheim soil (coastal soil with shell deposits) had the lowest *Ganoderma* incidence and severity. In contrast, Kabu soil showed high disease incidence and severity at 3 mpi, but the increment in disease scores was slower from 3 to 5 mpi compared with that for Jawa, Parit Botak and Bungor soils. Both total number and weight of *Ganoderma* fruiting bodies were correlated to percent reduction in RWB dry weight. Three sets of symptoms of *Ganoderma* infection were noted in this experiment: a) leaf symptoms prior to formation of fruiting bodies; b) leaf symptoms after the production of fruiting bodies; and c) a combination of both. Further research on the effects of soil components or properties on both the palms and *Ganoderma* infection is crucial to understand both the epidemiology and aetiology aspects of *Ganoderma* disease.

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TABLE 3. STANDARDISED CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS FOR DISEASE SEVERITY INDEX (DSI) (TWO CATEGORIES: MORE THAN 50%, AND = OR LESS THAN 50%) FOR SEEDLINGS IN FIVE SOIL SERIES BASED ON ANALYSED pH AND SOIL NUTRIENTS

		Function coefficients for different soil parameters							
Less than or 50%	Function 1: Group centroid	**pH**	**Fe**	**Mg**	**OrgC**	**Mn**	**N**	**K**	**Al**
	0.773	2.02	1.23	0.98	0.70	0.53	0.41	0.03	0.02
More than 50%	Function 2: Group centroid	**Ca**	**Zn**	**Cu**					
	-0.527	-1.01	-0.77	-0.41					

Note: *Abbreviations: Org.C = Organic carbon, N = nitrogen, Fe = iron, Mn = manganese, Zn = zinc, Cu = copper, K = potassium, Ca = calcium, Mg = magnesium, Al = aluminium.

TABLE 4. STANDARDIZED CANONICAL DISCRIMINANT FUNCTION COEFFICIENTS FOR PERCENT NECROTIC TISSUES IN THE BOLE (two categories: more than 50%, and = or less than 50%) FOR SEEDLINGS IN FIVE SOIL SERIES BASED ON ANALYSED pH AND SOIL NUTRIENTS

		Function coefficients for different soil parameters					
Less than or 50%	Function 1: Group centroid	**pH**	**Al**	**Mn**	**N**	**Fe**	**Mg**
	0.442	0.87	0.78	0.75	0.63	0.56	0.18
More than 50%	Function 2: Group centroid	**Zn**	**Cu**	**Ca**	**K**	**OrgC**	
	-0.376	-0.85	-0.60	-0.12	-0.08	-0.04	

Note: *Abbreviations: Org.C = Organic carbon, N = nitrogen, Fe = iron, Mn = manganese, Zn = zinc, Cu = copper, K = potassium, Ca = calcium, Mg = magnesium, Al = aluminium.

TABLE 5. NUMBER AND TOTAL WEIGHT OF *Ganoderma* FRUITING BODIES PRODUCED IN RESPECTIVE TREATMENTS AND PERCENT OF REDUCTION IN DRY WEIGHT FOR *Ganoderma*-INOCULATED RUBBER WOOD BLOCKS COMPARED WITH THE SAME IN NON-INOCULATED BLOCKS

Treatment (soil series)	Number *Ganoderma* fruiting bodies	Total weight of *Ganoderma* fruiting bodies (grams)	Percent reduction in dry weight of rubber wood block (%)*
Jawa	4	36.14	40.54 a
Bungor	2	16.11	22.65 b
Parit Botak	6	78.58	49.84 a
Blenheim	5	52.31	41.23 a
Kabu	6	111.04	50.23 a

Note: *Means (of 8 replications, except Kabu soil with only 6 replications) within the column of % reduction in dry weight of rubber wood block (compared with the same for non-inoculated block) for the respective treatments followed by the same letter are not significantly different from one another at $P = 0.05$ according to Tukey's test.

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