GRO-Cocoa

Issue 16
December
2009

Global Research On Cocoa - *working with and for farmers*

Taking Stock

Pesticides remain an essential tool for cocoa farmers, but legislative changes around the world are restricting what can be used, and how. In this issue we look at examples of how researchers are disseminating information about pesticide use, examining their impacts and continuing the long search for alternative control options.

Cocoa farmers can be isolated geographically and by poor knowledge flow from up-to-date information. An article from Nigeria describes how an initiative to educate farmers about adapting what and how they spray in order to accommodate changes in Nigerian government pesticide legislation has reached farmers in a remote cocoa-growing area of the country.

The next article is concerned with the ongoing changes in pesticide legislation around the world. The author explains how the second edition of the International Cocoa Organization (ICCO) sponsored *Pesticide Use in Cocoa* manual is being updated to help cocoa industry stakeholders cope with this.

Non-target impacts of copper-based (Cu) fungicides have been reported in various systems. We include a report on research in Cameroon that compared the effect of different Cu treatments on termite and earthworm faunas in cocoa, and drew conclusions about Cu use in this particular agroecosystem.

Turning to alternative strategies, plant extracts have been widely investigated for pest control, notably azadirachtin from the neem tree. We highlight a new review of azadirachtin, in which the author suggests why it has not been used or commercially exploited more despite excellent insecticidal properties.

Several articles in earlier issues of this newsletter dealt with endophytes, and parasitic *Trichoderma* fungi in particular, as they seem to hold promise for fungal disease control. Development of biological control technology is a lengthy process so the promise has yet to be fulfilled. However, in this issue, an article from Costa Rica indicates that progress with developing *Trichoderma* into a functional biocontrol agent for frosty pod rot (*Moniliophthora roreri*) is indeed being made.

Using a drama to explain pesticide regulations to cocoa farmers in a remote area of Nigeria. Here three men representing the Cocoa tree (middle), Black pod (right) and Mirid (left) showed how the cocoa tree was being tormented by Black pod and Mirid (Adeogun Stephen)

Making a Song and Dance about Pesticide MRLs

A dance–drama approach has been used by the Cocoa Research Institute of Nigeria (CRIN) to educate farmers in a remote area of Nigeria about which chemicals the Government has either banned, or approved for use, on cocoa.

The Federal Government of Nigeria recently reviewed which chemicals are permitted for use on cocoa (see [1]) because its cocoa industry needs to meet the requirements of the European Union (EU) Regulation on pesticide maximum residue levels (MRLs), 396/2005/EC[2], which came into force on 1 September 2008 (see Box, 'Why set MRLs?' p. 2). Europe is the main market for Nigeria's cocoa.

The EU Regulation is designed to safeguard the health of consumers of cocoa products, but by highlighting the need to use pesticides appropriately so that residue levels are not exceeded in cocoa destined for Europe, the Regulation will also help to protect those involved in production, handling and processing cocoa in producing countries[3].

The Nigerian government issued a directive banning some of the long-used chemicals because of their tendency to leave higher than acceptable residues, and approved the usage of others considered to result in acceptable residues (see below). The new order seems, however, to have posed an immediate major challenge among the mostly rural population of cocoa farmers. According to Asogwa & Dongo[1], the new legislation has left very few pesticides for use on cocoa in Nigeria, and this com-

In This Issue

Why Set MRLs?

Although EU Regulation 396/2005/EC has set MRLs for imported cocoa beans, this is not merely to appease nervous consumers and pesticide sceptics in importing countries – or even, as a hard-pressed farmer might reasonably think – to make life more difficult for cocoa growers. How they are set is a clue to why they are set.

An MRL is the maximum concentration of a pesticide (active ingredient) residue likely to occur in or on food or animal feed after use of the pesticide according to Good Agricultural Practice (GAP). The MRL is expressed in mg of residue per kg food/animal feed.

Field trials are conducted to establish how a particular pesticide should be used under GAP. Usually carried out by the agrochemical company wishing to register the pesticide for a crop (in our case, cocoa), these establish the application regime necessary for achieving effective pest control while minimising the pesticide residue on the crop. GAP and MRLs are therefore based on practical considerations and can benefit the farmer as well as consumer.

MRLs are adopted by bodies like the EU as legal limits to residues on crop products. GAP as it relates to use of a pesticide is set out on the pesticide label, so a farmer who follows the instructions on the label of a pesticide registered for use on cocoa should meet the new EU Regulation on MRLs. In addition, the farmer will be using a necessary but sufficient amount of pesticide, which brings economic and health/environmental safety benefits.

MRLs for pesticides in various crop products imported into the EU, including cocoa beans, are set out in Annexes to regulation 396/2005/EC. The catch is that many chemicals formerly used on cocoa are not included. In some cases they were rejected on safety grounds, but others were simply not put forward for use in cocoa. A default MRL of 0.01 mg/kg applies to any chemical for which no specific MRL has been allocated.

The presence or absence of an MRL in the EU Regulation does not indicate whether a pesticide can or cannot be used in a producing country. That decision is the preserve of the national regulatory authority of the country itself. Nonetheless, unless residues are below the relevant MRLs when cocoa beans arrive at the EU port-of-entry, the beans will not be allowed into the EU.

Recognising that the EU Regulation was going to affect cocoa farmers, the ICCO and ECA/CAOBISCO* wrote to the relevant authorities in producing countries to alert them to how the EU Regulation would affect their industries, and explaining which chemicals had and had not been assigned MRLs. In addition, they invited members of the cocoa sector to contact them if they had concerns about inappropriately low MRLs or perceived that pesticides for which no MRLs had been established were essential for cocoa production in their country.

The ICCO commissioned a *Manual on Pesticide Use in Cocoa* (www.icco.org – also at www.dropdata.org/cocoa/index.htm) with the aim of equipping cocoa-producing countries to meet the EU Regulation. Lists of pesticides approved and not approved by the EU and elsewhere for use in cocoa are included (see, 'Pesticides: connecting farmer practice and regulation', pp. 3-5, this issue).

*European Cocoa Association/Association of Chocolate, Biscuit and Confectionery Industries of the EU.

Two cocoa farmers, one using banned chemicals and the other using approved chemicals (Adeogun Stephen)

Catch them young: the children in the village were also attracted by the performance of the drama group (Adeogun Stephen)

Cross-section of cocoa farmers in attendance during the programme (Adeogun Stephen)

pounds on-going problems with poor extension services and cocoa farmers' lack of knowledge about pesticide usage.

It was against this background that CRIN embarked on a sensitisation programme to educate cocoa farmers in Ondo State on chemical control at the production stage. The initiative aims to help Nigerian cocoa farmers produce cocoa to an acceptable specification so the Nigerian cocoa industry can meet the EU Regulation on MRLs.

On 29 July 2009, CRIN gathered some 270 cocoa farmers, officials of the state Ministry of Agriculture, and representatives of farmers' organisations and processing factories, together with religious leaders and the Chairman of Idanre Council for a day-long meeting in the village square of the remote village of Aseigbo, in Idanre Local Government Area. This 'Community-Based Awareness Programme (CBAP) on MRLs in line with the EU Regulation', aimed to sensitise the farmers to how the EU Regulation on MRLs affected use of chemicals on their cocoa farms.

During the meeting CRIN told the farmers that steps must be taken to ensure that cocoa destined for Europe complies with the EU Regulation. The representative of the Executive Director of CRIN at the forum, Dr E.O. Aigbekaen (Director Farming System Research and Extension), described the EU Regulation on MRLs as a serious warning to the cocoa industry in Nigeria. He urged all stakeholders to, "Take the matter with all the seriousness it deserves to avoid the looming doom on the country's cocoa economy." He described how CRIN had started to make efforts to address this by organising stakeholders' workshops in 2008 in Ondo and Cross River states, "To raise awareness to all those concerned in the Nigerian cocoa industry with respect to the EU regulation on MRLs." He added, however, that they recognised that these two locations were rather far from the "grassroots" of cocoa farming, and the difficulties of reaching the real farmers in remote locations.

Dr Aigbekaen said that the decision to take the programme to a remote area like Aseigbo was made in the hope it would encourage farmers to attend by removing constraints such as transport. The rural location did indeed draw in local farmers in good numbers. Dr Aigbekaen expressed the hope that the day's programme would go "a long way to enlighten the real farmers on issues bordering on chemical usage and

Men representing Approved Chemicals dancing to show their victory after overcoming Mirid and Black pod (Adeogun Stephen)

Dignitaries during the forum, from right: Executive of one of the farmers' organisations, Chief Affun Adegbulu (President, Cocoa Farmers Association of Nigeria) and Dr E.O. Aigbekaen (Director, Farming System Research and Extension, representing Executive Director of CRIN) (Adeogun Stephen)

the EU regulations. This is," he summed up neatly, "to ensure appropriate use of the approved chemicals." He set the aims of the day in a wider context by explaining that the meeting was "to determine the fate of our cocoa and the way forward in the international cocoa market," adding that, "The primary purpose of this programme is to raise awareness of this challenge."

The dance–drama extension approach was adopted to ensure farmers left the venue with the intended message. The approach drove home the points in a way that was easily understood and remembered by the farmers, who are mostly illiterate. CRIN engaged the Ibadan-based Glad Tidings Drama Ministry to stage the comic drama which both educated and entertained the gathered cocoa farmers (see photos). It focused attention on compliance with the Federal Government's directive on banned substances and urged the audience to be wary of the economic consequences of non-compliance – the farmer is first and foremost a businessman. The drama also depicted the consequences of using the wrong chemicals on farmers' health and the environment.

To avoid confusion, the pesticide products that the sensitisation programme focuses on vary according to which chemicals the farmers in the area are familiar with.

Among the banned substances the farmers in Aseigbo were told about are chemicals like Cracker (active ingredients, a.i.'s = endolsulfan, deltamethrin [because endosulfan is on the Government banned list, although deltamethrin is not]), Unden (a.i. = propoxur), basudin (a.i. = diazinon), Blue Stone (a.i. = copper sulphate) and Gammalin 20 (a.i. = lindane). These were previously suspected or known to be used in cocoa.

The approved ones they learnt about are Dusban (a.i. = chloropyrifos), Actara (a.i. = thiamethomax), Caocobre Sandox (a.i. = copper oxide, Cu_2O), Ridomil Gold (a.i. = metalaxyl (M) + Cu_2O), Champ DP (a.i. = copper hydroxide, CuOH), Funguran OH (a.i. = CuOH), and Kocide 101 (a.i. = Cu_2O).

From participant reactions it was clear that the forum had conveyed the message that: "It is imperative that we take the matter of EU compliance very seriously now to avoid the backlash of non-compliance which could sound the death knell for the country's cocoa economy".

The farmers also grabbed the opportunity to put their views and problems to 'the men at the centre'. During the subsequent interactive session, several farmers asked for government to help in the form of a subsidy to enable farmers to cope with what they described as the high price of the chemicals now approved for use in cocoa, arguing that they are more expensive than the ones now banned. They complained that many farmers are not aware that some chemicals have been banned, and for those who are aware, the cost of the approved ones is ruinous. The farmers asked for the Government and other stakeholders in the cocoa chain to help make approved chemicals available at affordable prices. Amongst other issues they raised were low pricing and uncooperative actions of cocoa exporters who, they claimed, "always put the farmers at the receiving end of the negative effects of the international cocoa trade". The representative of the Cocoa Association of Nigeria (CAN), Chief Affun Adegbulu, together with the state officials promised to work together to resolve some of these issues raised by the farmers.

The urgency of the situation was acknowledged. It was observed that neighbouring cocoa-producing countries such as Côte d'Ivoire and Ghana have already made advances towards compliance with the EU Regulation. Fears were expressed that unless stakeholders in the Nigerian cocoa industry make significant steps in this direction, Nigeria's cocoa may end up being rejected by the EU.

[1]Asogwa, E.U. & Dongo, L.N. (2009) Problems associated with pesticide usage and application in Nigerian cocoa production: A review. *African Journal of Agricultural Research* **4**, 675–683. [August 2009 issue]
Web: www.academicjournals.org/AJAR

[2]Web: http://ec.europa.eu/food/plant/protection/pesticides/index_en.htm

[3]Rutherford, M. (2008) New EU pesticide regulations and West Africa. *GRO-Cocoa* No 13, pp. 5–8.
Web: www.cabi.org/default.aspx?site=170&page=1888

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Pesticides: Connecting Farmer Practice and Regulation

In *GRO-Cocoa* No. 13 (June 2008) I described how changes to legislation in the European Union (EU) and Japan have concentrated minds over crop protection practices in cocoa and other commodity crops. From 1 September 2008, with the implementation of EU Regulation 396/2005/EC, assessment of residues in imported commodities into the EU was brought into line with those of European crops. Thus for cocoa, for the first time this potentially included traces of substances that have been used upstream in the supply chain, including pesticides used on farms or in storage. The crop protection activities of farmers and middlemen have therefore become a matter of considerable concern to all in the cocoa trade.

Pesticides have a poor public image and are known to present dangers to both people and the environment. Nevertheless cocoa, like other tropical crops, is

EU Legislation by Numbers

Two sets of European legislation impact on pesticide use in cocoa, and it's worth reminding ourselves about these.

The most recent is Regulation 396/2005/EC, which came into force on 1 September 2008 and sets MRLs (maximum residue levels) for pesticide residues in food and animal feed produced, or being imported into, the EU area. This regulation directly affects use of pesticides by farmers and middlemen. EU MRLs were first published as Regulation 149/2008/EC in March 2008 in the form of Annexes to 396/2005/EC; these were updated before they came into force and continue to be subject to review.

Council Directive 91/414/EC currently regulates the placing on the market of plant protection products. Only substances included in an EU positive list (Annex 1 to the Directive) can be authorised by EU member states for sale and use in their countries. However, this legislation is set to be superseded by a new EU legislative framework on pesticides. This will comprise a new Pesticide Authorisation Regulation on placing plant protection products on the market, and a new Directive on the sustainable use of pesticides. Importantly (and contentiously), the new Regulation takes a hazard-based, rather than the existing risk-based, approach which it is anticipated will eventually lead to many compounds being withdrawn from use in the EU.

While the EU has no jurisdiction over what pesticides are permitted in other countries, compounds banned for use in the EU under the new Regulation are also likely to have residues banned under 396/2005/EC, which means their MRLs will be the limit of detection (LOD).

One other issue for cocoa is that little is grown on EU territory*. Some of the pesticides used in this crop are not used in EU agriculture and have thus not been considered within the framework of European legislation.

*French Guiana, as a French 'DOM' (Département Outre-Mer), is subject to EU legislation.

Sources/information: http://ec.europa.eu/food/plant/protection/index_en.htm

often ravaged by insects, diseases and other pests that must be controlled effectively as well as safely. With cocoa bean prices now regularly exceeding US$3000 per tonne, the readiness of farmers to protect their crops is likely to increase. Pesticides provide useful control solutions in many cases, but must be approved for use on the basis of Good Agricultural Practice (GAP). Unfortunately up-to-date GAP has yet to be established in many cocoa growing areas, so the International Cocoa Organization (ICCO) commissioned a manual in order to:

1. Summarise important underlying administrative and technical issues with pesticides, with specific reference to compounds that are, or may be, used on cocoa.
2. Help define a 'road map' for establishing good crop pest control, storage and distribution practices for bulk cocoa, for the assistance of trainers-of-trainers and research staff.

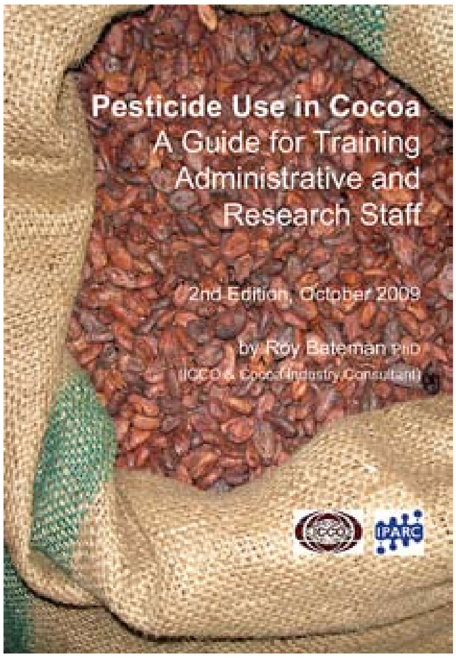
The first edition of this manual was published online in 2008, and a second edition, produced in October 2009, will be translated into at least three languages.

Only by regular interaction between farmers, researchers, extension workers, suppliers, buyers and administrators can the issues confronting the cocoa trade be overcome. Unfortunately, there has at times been confusion about pesticide issues which has led to misuse, safety concerns (or inappropriate lack of concern), poor crop protection and other difficulties. Perhaps because of the poor public image of pesticide science, the number of people with a good working knowledge of the subject has declined dramatically, to a stage where even recent scientific papers are marred by misconceptions and inaccuracies. To a certain extent, many in the cocoa industry were 'taken by surprise' by EU Regulation 396/2005/EC (see Box, 'EU legislation by numbers'), which itself continues to undergo amendment (i.e. to its Annexes). The original EU Regulation 91/414/EC was seen by many as just the start of a review process and in July 2008 EU agriculture ministers approved a proposal for even stricter controls, with a shift in emphasis from risk- to hazard-based assessment of pesticides[*].

With the advent of 396/2005/EC, the first edition of the manual inevitably focused on cocoa exports to the EU and West African production. The scope will be broadened in the second and future editions to give a more worldwide coverage of cocoa production and importation (e.g. to the USA and Japan). However, this makes the narrative even more complicated!

Although there are moves to harmonise trading standards (via the OECD; Organisation for Economic Co-operation and Development) significant differences remain. The well known disparity between the EU (where shelled beans are analysed) and Japan (where whole beans are analysed, which is more likely to result in residue violations) is compounded by differences between permitted lists and MRLs of the active substances themselves. Two of the few remaining organo-phosphorus (OP) compounds, diazinon and chlorpyrifos ethyl, provide an illustration. Although both have import tolerances of 0.05 ppm (parts per million) in Japan, in the EU diazinon has been rejected and chlorpyrifos has been assigned an MRL of 0.1 ppm.

[*] Anon. (5/7/2008) A balance of risk. *The Economist* **387** pp. 100–101.



Inevitably perhaps, by far the most controversial part of the first edition of the manual was Appendix 3, which lists pesticides commonly used in cocoa. Simply dividing pesticides into 'positive and negative' lists has proved over-simplistic and, as illustrated above, impossible to maintain with the new wider geographical scope. For the second edition Appendix 3 has therefore been divided into four categories:

A. Strategic/recorded pesticides for use in cocoa which:

- Are known to be on 91/414/EC Annex 1 and have Japanese/US import tolerances
- Have proven efficacious against an important pest species of cocoa, as published in (preferably refereed) literature
- Show acceptable levels of low mammalian toxicity and environmental impact (which will have to be defended in the EU within a limited time period) and *do not* belong to the highest toxicity group WHO/EPA Class I – defined in the manual

Sampling cocoa beans from a sack in Ghana (Roy Bateman)

B. Compounds to be used with CAUTION (limited time span). These active substances:

- Have permitted MRLs in some markets, but not others and/or …
- Are likely to be phased out within 2–3 years, but …
- Have shown demonstrable efficacy in at least one regional cocoa growing country
- Do not belong to WHO/EPA toxicity Class I

C. Pesticides that MUST NOT BE USED FOR COCOA. These substances:

- Have been recorded as used on cocoa (e.g. as listed in *GRO-Cocoa* No. 13) but have been rejected by major importing countries (usually for toxicological/eco-toxicological reasons)

D. Experimental control agents for possible future inclusion in category 'A'. These control agents:

- Are known to be on 91/414/EC Annex 1; compounds for inclusion continue to be reviewed, and special care should be taken with any compound that remains on the 'pending' list
- Are subjects of current or recent field tests and could well conform to criteria in category 'A'
- *Do not* belong to WHO/EPA toxicity Class I and are preferably in Class III or better

It is perhaps the development of the 'D' list that will be of greatest interest from a technical point of view, since it will include the control agents that could substitute for the lengthening list of withdrawn chemicals. Where possible, reference is made to these in the main text: e.g. novel neonicotinoid insecticides for mirids (but there is a potential bee toxicity issue) and carboxylic acid amide (CAA) compounds for black pod (*Phytophthora*) control. Many cocoa growers and procurement managers complain about the increased cost of most of the newer products – although I would remind readers that pyrethroids were considered very expensive in their early days of development. The most obvious way to mitigate this problem is to apply less by applying more efficiently, so emphasis is placed on optimising pesticide application methods.

Wherever possible, new information in future editions of the manual will be provided in 'boxes' (for ease of translation!); in the second edition, new boxes include Southeast Asian problems (especially the cocoa pod borer, *Conopomorpha cramerella*) and herbicides.

My main message is that the cocoa industry must 'stay ahead of the game' and not just try to keep up with existing pesticide legislation: not least because of the continuing and developing technical challenges in pest management (invasive species, to name but one).

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Unearthing How Cocoa and Copper Affect Soil Fauna

The literature on the impact of cocoa agroforestry on biodiversity tends to focus on macrofauna (birds in particular) and management practices to optimise its diversity. In contrast, a 4-year study in southern Cameroon looked at the effects of fungicide regimes in cocoa on the soil fauna[1,2]. The effects of fungicide on soil are important for two reasons: overwhelming disease pressure and absence of fertiliser inputs in this agroforestry system.

In southern Cameroon, 70% of smallholder farmers grow cocoa, predominantly under shade provided by upper-canopy forest trees, retained for timber, fruits, nuts or medicinal products. The main production constraint is black pod disease caused by the virulent *Phytophthora* species, *P. megakarya*. Despite efforts to find alternative control measures, fungicides remain the mainstay of disease control. In this study, almost no crop was harvested if fungicides were withheld. Farmers most commonly use copper-based (Cu) fungicides, generally at lower doses than manufacturers recommend.

Microbial and soil fauna-mediated nutrient cycling through decomposition of leaf litter and dead plant material is crucial to the sustainability of cocoa farming because fertilisers are rarely used by these smallholder farmers. None of the 200 farmers surveyed for this study applied fertiliser. Some other studies, that include cocoa and coffee, have found elevated exchangeable soil Cu concentrations associated with Cu fungicide use. Studies in temperate agricultural crops have suggested that high concentrations of heavy metals including Cu in soil have a negative impact on the soil fauna, particularly earthworms.

What is the impact of the Cu fungicides applied against black pod disease of cocoa in southern Cameroon on the soil fauna and nutrient cycling process?

Sampling decomposition bags and earthworm casts in a cocoa farm in the village of Nkomotou III in central Cameroon (Lindsey Norgrove)

Earthworm casts expelled on the soil surface in a cocoa farm in central Cameroon (Lindsey Norgrove)

Termite mound rebuilt (darker area) following damage, Essong Mintsang in central Cameroon (Lindsey Norgrove)

Lindsey Norgrove and colleagues compared the soil fauna from 35-year-old cocoa farms and secondary forest of the same age. On the cocoa farms abandoned 3 years earlier, they established plots that were treated each year for 4 years with recommended and low doses (6.4 and 2.13 kg/ha/year, respectively, applied as eight treatments) of the fungicide Ridomil (active ingredients: 600 g/kg Cu_2O + 120 g/kg metalaxyl) and compared them to untreated control plots. They recorded information about the termite and earthworm faunas and the soil.

For termites, species diversity was lower in cocoa than in secondary forest, although differences were not as great as has been reported elsewhere (possibly because this study compared cocoa and secondary forest of the same age). There was, however, a loss of specialist soil-only and wood-only feeders from cocoa in favour of generalist soil and wood feeders. For earthworms, there were more adults in the forest than in cocoa farms, although total densities (adults + juveniles) were similar. Apart from the deep-burrowing ('endogeic') fauna, which was similar in cocoa farms and secondary forest, there were significant differences in diversity. The only upper-soil layer and litter-feeding ('anecic') forest species was present in lower densities in cocoa farms than in secondary forest (for fungicide impacts, see below). This has implications for nutrient cycling in cocoa as it is the main cast producer and its casts are richer in nutrients than those of endogeic earthworms. Of litter-feeding (epigeic) earthworms, the dominant forest species was absent from cocoa farms; two minor species were present in cocoa farms in higher numbers than in the forest (where they were possibly outcompeted by the dominant species). The loss of epigeic earthworms probably reflects the inferior nature of cocoa leaf litter. Norgrove[1] suggests that adding legume cover crops and mulching would improve litter quality, reducing species loss and also possibly reducing the *P. megakarya* soil inoculum, the most important source of pod infection.

Fungicide treatments had no significant effect on termites, but earthworm numbers were lower in fungicide-treated cocoa than in control plots or secondary forest. The only anecic earthworm species was completely absent from the higher (recommended) dose fungicide treatment in cocoa and reduced to 20% of forest density at the lower dose. Earthworm cast production was higher in the forest than in all cocoa treatments. In the fourth year, cast production in cocoa was significantly lower in the higher fungicide treatment than in the lower dose or control treatments. Yet there was no evidence that Cu fungicide had affected litter or soil decomposition and soil compaction over the same period. In addition, the recommended, higher, fungicide dose gave a 2.5× greater cocoa yield than the lower dose – and as we have noted above, yield was negligible where fungicide was not used.

All the same, might sustained use of Cu fungicides do long-term harm in these cocoa agroforests? Before drawing conclusions, Norgrove[1] considered that the background concentration of exchangeable Cu in these tropical soils is very low. Therefore even the highest concentrations found in sprayed plots were still low compared with global levels. This is due to two reasons, First, total Cu concentrations are lower and, second, soil pH is lower therefore Cu is less available and more likely to be complexed with organic acids in the soil rather than being in the soil solution. So adding a fixed amount of Cu to a system would have a lower impact in these tropical, unfertilised soils than in temperate soils. However, if soils were to receive lime to increase soil pH, then soil exchangeable Cu would be much higher and toxicity might be a problem.

Biodiversity considerations aside, in this area where cocoa is under severe pressure from *P. megakarya* and farmers are not in a position to apply fertilisers to soil, is the use of Cu fungicides sustainable? The conclusion drawn from this research in this context is that application of Cu fungicides at recommended doses provides best current crop protection and does not appear to have a detrimental effect on soil fertility.

[1]Norgrove, L. (2007) Effects of different copper fungicide application rates upon earthworm activity and impacts on cocoa yield over four years. *European Journal of Soil Ecology* **43**, S303–S310.

[2]Norgrove, L., Csuzdi, C., Forzi, F., Canet, M. & Gounes, J. (2009) Shifts in soil faunal community structure in shaded cacao agroforests and consequences for ecosystem function in Central Africa. *Tropical Ecology* **50**(1), 71–78.

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All about Azadirachtin

'Azadirachtin, a scientific gold mine' by E. David Morgan[1] packs a wealth of information drawn from 40 years' research on this compound into ten pages. The author also explores why a compound that has apparently outstanding potential for insect pest control has not been more widely used or commercialised. The paper therefore makes useful reading for researchers involved in the practical development and use of neem extracts for pest control.

Azadirachtin is a plant-derived chemical, in the limonoid group of triterpenoids, extracted from *Azadirachta indica*, the neem tree. The 176 references in the review (marred only by the journal's policy of not including titles of papers) demonstrate the large number of previous reviews on aspects related to the chemistry and antifeedant/insecticidal effects of the compound, which reflect the scientific interest and amount of research it has generated. As the author says, "The neem tree must be one of the most intensively studied sources of natural products." However, he points out, there is an imbalance between this large body of research and the use to which it has been put.

There is a lot of chemistry in this review, with accounts of research into the complex structure and equally complicated synthesis of azadirachtin, as well as its extraction and analysis. Although azadirachtin is the most abundant and biologically active of the triterpenoids in neem extracts, more than 150 others have been found, and the paper touches on the structure and significance of some of these. The author outlines the current limited knowledge of how azadirachtin is biosynthesised in the plant, and notes that much has yet to be explained about the compound's mode of action and structure–activity relationships.

Neem is native to South Asia, where its insecticidal/repellent properties have long been known, including the antifeedant properties of its leaves against desert locust, *Schistocerca gregaria*. However, as the literature reviewed in this paper indicates, neem extracts exhibit insecticidal properties against a very broad range of insects, and at far lower doses than those producing antifeedant effects. It is now planted in many parts of the semi-arid tropics, and is often used as a source of azadirachtin or crude neem extract, but also has value as a rapidly growing tree that is tolerant of harsh conditions, suitable for windbreaks, combating desertification and as a source of firewood. The trees begin to bear fruit at 3–5 years old and at maturity can produce up to 50 kg of dried seed annually.

Azadirachtin is found in all parts of the neem tree, but highest concentrations are found and most effectively extracted from seeds. The amount that can be extracted varies, which the author suggests is at least partly due to the precise extraction process. The impact of environmental, soil and seasonal factors on seed azadirachtin content is also unknown.

Morgan says that variable results from experimental use of neem extracts have occurred partly because the term 'neem extract' is very imprecise: it might mean an extract of leaves, or of seeds, or the seed oil, and so on; these will have quite different contents and are in no way equivalent. It is often unclear precisely what compounds have been tested, and how much. Nonetheless, although testing ill-defined extracts is not helpful in developing the technology, in the longer term the use of a mixture of compounds is beneficial in preventing development of resistance in the target pest. Morgan explains that he coined the term 'azadirex' "for the insecticidally active extract of neem seeds, however obtained, containing azadirachtin as its principal active component, with other biologically active limonoids."

Like many natural pesticides, regulatory hurdles have hindered the commercialisation of neem products [and this has also been the subject of an anti-biopiracy campaign]. However, the author makes an interesting comparison between azadirex and another much more successfully commercialised plant-derived insecticide, pyrethrum, identifying from this "some of the advantages and disadvantages of azadirex in production and use."

In early commercialisation efforts, the author suggests, too little attention was paid to the stability of the product to light, temperature and pH, which meant results were variable and potential users discouraged. Nowadays far more importance is attached to formulation of biological pesticides in general. There is potential for improving the stability of azadirachtin products (e.g. with UV screens) and its formulation (e.g. one possible avenue is forming complexes with certain sugars to increase water solubility).

The author identifies the high cost of raw material and therefore the final price, as well as licensing fees, as continuing obstacles to commercialising neem-based products. He suggests the high cost of commercial neem production might be tackled via mechanical harvesting, or finding commercial outlets for neem by-products.

Although poor uptake of neem-based control measures has been ascribed to apparent unreliability, as well as supply problems, Morgan suggests an additional and quite different reason: cheap and easy availability of crude neem seed extracts – coupled with slower action against pests than more expensive synthetic pesticides – made farmers undervalue the 'homemade' neem-based product in early farm-based initiatives. With farmers facing growing restrictions on use of synthetic chemicals, a new window of opportunity may have opened for developing and promoting neem-based technology.

[1]Morgan, E.D. (2009) Azadirachtin, a scientific gold mine. *Bioorganic & Medicinal Chemistry* **17**, 4096–4105.
Web: www.elsevier.com/locate/bmc

Editors' note: A temporary MRL of 0.01 mg/kg is set in the new EU Regulation 396/2005/EC (see Box, 'EU legislation by numbers', p. 4) for azadirachtin in cocoa beans (nibs), determined after removal of the shells. Azadirachtin residues in cocoa beans imported into the EU must not exceed this level.

Endophytes: Is their Potential for Disease Control to be Realised?

The fungal pathogen of cocoa, frosty pod rot (*Moniliophthora roreri*), continues to consolidate its coverage in Latin America. The threat of it escaping its current geographical limits into other regions of South America (Brazil and Bolivia) and other continents, in particular Africa, increases as South–South trade and technology exchanges become more prevalent. As a result it is becoming imperative that a mechanism to control the disease is developed. In a previous article in *GRO-Cocoa* by Bryan Bailey (' Improving efficacy of cocoa disease biocontrol: a way forward ', No 12 [December 2007], pp. 1–3) the use of endophytic *Trichoderma* species was highlighted as possibly providing a way forward for biocontrol of fungal diseases of cocoa.

Field assistant Valex Adonijah assessing disease severity of cocoa pods (Jayne Crozier)

A number of similar conclusions from biocontrol field trials carried out in Latin America and West Africa were noted:

- *Trichoderma* applications enhance flowering and fruit set
- The application of *Trichoderma* shows only limited influence on the reduction of diseased pods in the short term
- Yield is increased because of the increase in initial pod set
- Accumulating evidence indicates that repeated application of *Trichoderma*, over multiple years, tends to lead to continual improvements in disease control

Key questions were raised as to whether the effect of *Trichoderma* is cumulative and how the efficacy of *Trichoderma* could be enhanced to improve colonisation and associated disease control. Main targets for future assessment to improve efficacy were species/strain selection and formulation.

Two years on, we report how a continuation of a long-term collaboration between

Cocoa pods in the Trichoderma *colonisation study showing varying degrees of frosty pod rot (Jayne Crozier)*

CABI, USDA-ARS (US Department of Agriculture – Agricultural Research Service) and CATIE (Centro Agronómico Tropical de Investigación y Enseñanza) is addressing these questions and targets through field trials in Costa Rica.

A large-scale field trial was carried out at CATIE's field station at La Lola during the 2008/2009 growing season with the aim of finding out whether there was a cumulative effect of application of *Trichoderma* on disease control; i.e. whether a limited number of applications early in the season would provide protection throughout the entire season.

Encouragingly, reduction in frosty pod rot was achieved in comparison to controls by monthly application of *Trichoderma*. This was comparable to the reduction in disease incidence achieved with application of a copper fungicide. However, early season application alone did not provide sufficient protection throughout the growing season. It would appear that the *Trichoderma* formulation we are currently using needs to be applied at monthly or regular intervals during the growing season to offer protection to the pods.

Better selection of *Trichoderma* species or strains and improved formulation may well improve colonisation and persistence of *Trichoderma* on pods in the field. A programme was therefore initiated to assess approximately 80 endophytic *Trichoderma* species/strains and select the most promising to take to the field for testing in subsequent large-scale field trials. The selection of isolates was based on performance in a number of laboratory-based screening tests which assessed a number of different mechanisms thought to be the most important for biocontrol: antibiosis, mycoparasitism, colonisation, competitive exclusion and induced resistance. It is often the case with isolate selection for biocontrol that those showing most promise in the laboratory can fail to have any impact in the field under high disease pressure. By using this combination of screening methods we hope to avoid these problems.

Fourteen *Trichoderma* strains were selected based on efficacy in the various screening tests and these were included in small-scale studies in the field at CATIE to assess their ability to colonise cocoa pods. Varying degrees of colonisation ability in cocoa pods were observed, with key species (*T. ovalisporum* and *T. harzianum*) showing improved ability to colonise cocoa pods compared to the other strains screened.

A further field trial is currently underway using *T. ovalisporum* and *T. harzianum* with a focus on improving the formulation to enhance germination of *Trichoderma* on the pod surface and increase colonisation of the pod, thereby increasing protection from disease. In past field trials *Trichoderma* spores have been applied to cocoa in water-based formulations, often with the addition of surfactants to enhance spread on the pod surface. However, it has been observed in studies at USDA-ARS and CATIE that the germination rate of *Trichoderma* spores is very limited when they are applied to cocoa in a water-based formulation. This season's field trial is investigating whether germination and colonisation can be improved by providing additional water for the spores to germinate on the pod surface using humectants and oil-based formulations and by providing additional sources of nutrients.

Studies continue to improve *Trichoderma* strains for control, and application technology is being developed to enhance the endophytic agent's ability to colonise and persist on the cocoa pods in the field. Given the advances we have made, we anticipate that these developments will ultimately lead to a reduction in the impact of fungal pathogens on cocoa production. As our knowledge of the interactions between *Trichoderma*, cocoa and its pathogens increases we expect to be able to improve the efficacy of *Trichoderma* not only as a biocontrol agent of fungal disease of cocoa such as frosty pod rot but also of other pod pathogens such as *Phytophthora*.

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Global Research On Cocoa (GRO-Cocoa) is produced biannually (June and December) with financial assistance from the US Department of Agriculture (USDA) by:

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