**Transcript – ACPPO webinar – MALDI-ToF**

15 May 2025, 04:01am

 **Vivian-Smith, Gabrielle** 0:17

So welcome everyone to the May Australian Chief Plant Protection Office webinar. Today's webinar is on MALDI-ToF, its benefits and how it'll make a difference before we get going, I'd like to first acknowledge the traditional custodians and traditional owners and custodians of country throughout Australia and I'd like to acknowledge their continuing connection to our land, waters and community, and pay my respects to the people, the cultures and the elders, past and present, and I extend that recognition to the traditional custodians of all the other lands on which we're gathered today, and also to all of our Aboriginal and Torres Strait Islander attendees today.
So in terms of housekeeping, if you could please just check that you have your video turned off and can you please mute your microphone?
This helps avoid any disruptions, but also improves bandwidth.
We’ll allow people to ask questions at the end and they can do that by either putting their hand up or turning their microphone on.
The webinar is going to be recorded today and it will be available afterwards through Teams and we will also share this link on LinkedIn.
And yeah, just put your questions in the chat as you have them.
So today's webinar is all about what MALDI-ToF is, the benefits it's brought to DAFF as a new technology and how this technology works. We'll also hear about experiences using MALDI-ToF mass spectrometry and the highs and lows associated with the using the technology associated with bacteria.
We have two speakers today.
We have Chris Nelson and Doctor Dominie Wright.
So about the speakers, Chris Nelson is the principal mass spectrometrist for the science and surveillance group at the Department of Agriculture, Fisheries and Forestry and he's played a key role in the recent development and deployment of MALDI-ToF mass spectrometry as a new diagnostic tool for biosecurity workflows. And we also are very pleased to have Doctor Dominie Wright.
Dominie is the principal laboratory scientist for the diagnostics and laboratory services at the Department of Primary Industries and Regional Development in WA, so the state agency and Dominie is a plant pathologist specializing in mycology and bacteriology.
Dominie is also a member of the National Subcommittee on Plant Health Diagnostics called SPHD here in Australia and is a coordinator of SPHD's diagnostic Protocol Working Group.
She's also the coordinator of the National Plant Proficiency Testing program.
So a very warm welcome.
To Chris and Dom.
And I just wanted to note, we've got an absolutely massive audience here today.
So that signals there's a huge amount of interest in this topic and I will now hand over to Chris and Dominie to take the floor. Thank you everyone.

 **Nellessen, Chris** 4:03
Thank you, Gabrielle.
Hi everyone.
My name is Chris Nellessen and I'm the principal mass spectrometrist, as Gabrielle said. And today I'm going to be talking a bit about MALDI-ToF.
So, what is MALDI-ToF?
Well, in brief, it's a type of mass spectrometer that can rapidly identify samples by matching the reference mass fingerprints.
So, you might think, what is a mass spectrometer?
Well, it's a tool that measures the exact mass of charged particles, which are generally called ions.
And, what is the mass fingerprint? It's a graph of all the exact masses of ions in a known sample within a specific mass range.
We can use these mass fingerprints to identify unknown samples by comparing known mass fingerprints to unknown ones.

 **Nellessen, Chris** 5:19
Mass spectrometry has been around a really long time, more than 100 years. We have a picture of the first mass spectrometer in the bottom right there and some 90 years later, the first MALDI came about and there's a picture of that on the top right.
The first time-of-flight mass spectrometer, which is the type of mass spectrometer we use with MALDI-ToF, was invented in 1948. It identifies these charged particles, or ions, through the time that it takes for it to move through a vacuum tube.
Mass fingerprinting, the method that we use to identify samples with MALDI-ToF, was first invented in 1975. It used pyrolysis for the ionization process, which was less effective than technologies that came later, as it would burn the sample down to component parts rather than keep proteins and peptides intact, such as with MALDI, or matrix assisted laser desorption ionization, which came in 1987, and was one of these first ionization methods that could keep the particles intact. When paired with ToF, as it was in commercially done in the early 2000s, we can get that mass fingerprinting that can get us identifications. It wasn't until about 2013 that commercial MALDI-ToF were approved on national levels for uses like human pathology, you can see that in the graphs to the right, that's when publication started to really pick up for these types of tools. Now, it's used pretty ubiquitously in both human and veterinary pathology as well as in pharmacology, food science and other fields.

So how does MALDI-ToF function within DAFF?
Well, DAFF protects Australia's environment and agriculture by making biosecurity risk decisions about pests and diseases. These need to be informed biosecurity decisions, which requires diagnostics that are accurate and quality assured or otherwise defensible, timely, and cost effective. Thanks in part, or largely in part, to the $22 million modern technologies and diagnostic tools budget measure, DAFF was able to upgrade its diagnostics and its expertise to better meet the modern benchmarks of these goals. MALDI-ToF is one of the new tools that was brought on board at this time and it provides easy, rapid, low cost diagnostics and its operational flexibility and workflow agility to DAFF’s diagnostics. Subsequently, to date, which is about two years in operation, MALDI-ToF has significantly impacted DAFF diagnostic output, performing more than 3500 tests for more than 350 goods and consignments.

So how does MALDI-ToF work?

Well on the right you can see an image of the instrument. It's a bit—it's an enclosed box, so you don't really get to see what's going on, but if we take a look inside, you get to see where the rapid automated sample analysis happens.
It begins with laser ionization, where a laser is shot as a mixture of the sample and a compound called a matrix, which is just a chemical term for something that's light absorbing, in this particular case, so it's a light absorbing chemical that makes it so that when the laser shoots this mixture, it doesn't just burn it and it actually absorbs the energy and transfers it to the sample to generate ions.
These ions are then accelerated with an electric field of known voltage.
This allows them to move up the vacuum tube that is empty and they'll separate based on their mass, since they have the same acceleration.
These are then detected at the end and their travel time will be recorded as well as relative amounts. This will be used to generate a spectrum, which is the output data, and the masses will be calculated for that [ion] based upon the potential in kinetic energy equations.
So this is then used during the identification process by being compared to the mass fingerprints we talked about earlier. That works because of proteomics, in large part. Since ribosomal proteins will show up in the mass range that we're measuring and their exact mass is conserved.
That's going to improve the consistency of the data compared to the fingerprints, so that we can match and be confident that the matches mean something.
Furthermore, the heights of the peaks are actually not quantitative. They have nothing to do with the concentration of that particular particle in the sample. They're largely governed by the ionization physics of that particular particle, which is physics laws. As you can see by the periodic table here [with ionisation energy requirements]. We also get multiple results for every output, so you get the top ten nearest matches and they're graded, so this allows scientists to do more dynamic analysis—as you would do for sequencing—where you can use your own background knowledge in the context of the diagnostics to generate the most reasonable result for any analysis.
And of course, it's easy to perform. On the right, you can see what a MALDI target plate looks like, and all you need is a toothpick to transfer your sample over.
And then, you add 1 droplet of the dissolved matrix, or light absorbing chemical, then you dry it so it can be ready to be shot with the laser.

So, what can it actually analyse?

Well off-the-shelf—and the key point here is off-the-shelf—the Bruker MALDI Biotyper, which is the MALDI-ToF we use at DAFF, can analyse some plant pests.
For bacteria, it's very consistent, and rapid, and very good at coming up with results quickly. And that preparation is just as you saw, very simple. Just takes a toothpick.
For fungi, the results are a bit more inconsistent. It has some difficulty with off-the-shelf analysis in terms of detecting the ions for the sample, and this has a bit to do with the thickness of cell walls for fungi in comparison to bacteria. And to combat that, manufacturer recommends various time-consuming procedures, whether it's addition of acids or crude extractions with mixtures of chemicals. That it works to some extent, but it's not perfect, so it's a bit problematic.
Similarly, off-the-shelf the MALDI Biotyper has lots of problems with invertebrates.
Unfortunately, there is no accepted approach by the manufacturer for analysing vertebrates, so there's nothing to go on there and you generally just have poor sample detection. There's also more variability—as the ribosomal proteins, while present, don't necessarily make up the majority of ions because there's so many different ions in these samples.

Now, DAFF was able to overcome a lot of these challenges during the optimization that it performed during its pilot stage.
We found that off-the-shelf, the default analysis settings were largely optimized for speed, or high throughput—for human pathology labs and the like.
On the right we have an example of what the output data using these default settings would look like for a species called *Ewingella americana*.
And while this is actually very rapid to collect—it takes about 7 seconds to run a test on default settings—there's a high cutoff value for what it allows itself to detect, to prevent too much buildup of noise, and it collects only a certain amount of data, so it can be very rapid.
We found that we could extend the amount of data that collects, and decrease the cut off value for its detection, to greatly improve its sensitivity and filter out the noise that it was trying to cut off anyways through increased averaging. By doing this we were able to increase sensitivity more than 300 times, which is right about at the limit of what the instrument is actually capable of at a physical level, and this greatly increased the rates of identification, regardless of preparation method and sample type—expanding application for all things, and bringing a lot more benefit to the use of MALDI-ToF.
An example of this is it improved our fungi results, and even allows us to identify uncultured fungi. On the right you can see an uncultured fungi growing on a peach, and the results from that are the equivalent of ones that we would get from a culture.
We also can get better and more taxonomic data from impure samples. For instance, for bacteria of mixtures of four or less different genera, we can usually identify them as long as they make up more than 10% of that particular mixture in a colony.
And for invertebrates, it just generally increases the quality of results and allows you to capture results you otherwise wouldn't be able to, regardless of sample age, which is really problematic with off-the-shelf settings.

Challenges remain, though they are largely around fingerprint library gaps, though, for identification purposes. Now for bacteria, there's large commercial fingerprint libraries available. There's some 12,000 fingerprints in the Bruker commercial library, the instrument [type] that we use at DAFF. However, while plant pathogenic genera are well covered there, there's a lot of missing species, so still gaps remain.
Fungi libraries, commercially, are much smaller.
There's only about 1000 in the Bruker library, and as you might expect, this leaves some pathogenic fungi not well covered. And there's another problem, in that population differences can lead to some protein variation, which—in order to retain the confidence in identifications—means you need multiple fingerprints per species.
Which means the gaps are bigger than you might expect.
Invertebrates also face a number of challenges, and they can get more complex than you might see with bacteria and fungi.
The first one is that different body parts can result in lead to different results.
So, you need to maintain consistency with body parts you target—or just cover all the gaps and make fingerprints for each target that actually works well.
Different life stages also lead to different results. So, for truly comprehensive species identification, or the ability to comprehensively identify a target of that species, you need a lot of fingerprints—one of each life stage.
Different storage can also affect the results, so you need to make sure your workflows are consistent and manage that part well.
And different populations can also lead to somewhat different results, similar to like we saw with fungi as mentioned earlier, although depending on your context, this can actually be useful. In this particular case [image], we were able to group populations of mosquitoes together. However, in terms of identification, you would get lower scores for different populations when trying to match them against each other. And, there are no commercial libraries available for invertebrate fingerprints. So, there's definitely a lack of fingerprints for invertebrate pests.

Now, we've done quite a bit to overcome these challenges in terms of library gaps. So, the National Steering Committee on MALDI-ToF Library development recently within the past year published standards for library development, and it's the first multifaceted quantitative fingerprint standards in existence in the world.
Which allows consistent quality fingerprint results to be produced.
Using that, and ongoing state and territory collaboration, DAFF and states and territories have produced quite a number of fingerprints to meet the gaps that we talked about—DAFF has produced more than 500 custom fingerprints and contributing States and territories have produced more than 500 as well. And that's largely in NSW and WA Australia.
There's also an ongoing EUPHRESCO project that intends to research fingerprint inter-laboratory effectiveness and also build an open fingerprint library of planned pathogens to further expand the capabilities of multi path and address those gaps.
We also have an upcoming PhD project with the ARC Training Centre in ANU, and that'll explore the potential for MALDI-ToF to identify biomarkers in fruit flies:
to capture the different mating status or fertility status, the sources of those fruit flies, and whether or not they're a gamma radiated. It has to be explored to see if we're capable of doing that.
So MALDI-ToF can bring a lot of benefits, and while there were some challenges, a lot of them were overcome and there's ongoing work being done to address remaining gaps.
And with that, I'll take questions in the chat.
But for now, let's welcome Doctor Domini Wright, our next speaker.

 **Dominie Wright** 18:41
Thank you, Chris.
Afternoon everyone.
So today I thought I would talk about the highs and lows of doing bacterial identifications.
So just a little bit of background, I'm a diagnostician for the Department of Primary Industries and Regional Development in WA.
So when we get plant samples that have bacterial symptoms in the old days, I used to use classical methods for identifying the bacteria.
So morphology, by clinical tests, Gram Stains.
Genetic testing, tobacco plant inoculations and other methods. Then we went on to in the early 2000s using BIOLOG.
And then from there we started using molecular methods and they all have their pluses and minuses for doing the different ways. And then along came the MALDI-ToF.
So when I first started getting interested in using the MALDI-ToF, we decided we needed to compare what was in the database and compare it against typed isolates from NSW and QLD.
These isolates have been typed by using fatty acid analysis, so again a very old way of identifying the bacteria.
And part of the reason for doing this was that when I got interested in using BIOLOG and started using it, I found that Australian isolates are quite different in bacteria, are quite different to the American isolates, and since then also the European ones.
So BIOLOG is based on American bacterial isolates, MALDI-ToF their database with Brooker tends to be based on European isolates.
So some of the comparisons that we've done, as we've done quite a lot of work with pectobacterium carotovorum through a veggie project that we had and we looked at the isolates that we got in.
I ran them through BIOLOG, and we did Q PCR.
And as you can see, with this particular isolate was determined to be carotovorum and BIOLOG showed it was atrosepticum, MALDI said it was atrosepticum and the Q PCR said it was atrosepticum.
And similar with the years that we can get consensus between BIOLOG and MALDI, but the Q PCR we weren't at the time.
We also went on to look at some of the Pseudomonas syringae group is an absolute nightmare, so that is something that Tony Chapman from the NSW department and I are going to be working on in the next 12 months.
So there's very little consensus between all of the results.
Bearing all of this background work that I had done.
We still go on to using it in our day-to-day work so we get a plant sample in.
We test for the bacteria using standard testing methods. Once we've isolated the bacteria, we'll sub it and make sure we've got pure colonies.
Then we'll go to the MALDI-ToF where Chris is able at the moment and Heidi sometimes quite often get results directly off the plant tissue.
We haven't been able to do that partly because we don't have a MALDI-ToF in our lab.
We go and use the one that we share with the Animal Health group and they're in a different laboratory and now currently they're on a different site to where we are. So it makes it a little bit more challenging for us to do our work.
But the MALDI-ToF after all, is seeing what the results are.
We will usually subculture that bacteria again and send them up for sequencing to confirm the identification that we've received.
And that means we're waiting for three to four weeks before we can give a result to the client.
Once we've got our database working and sorted, we'll be able to give the client a result on our plant diagnostic samples in 10 days maximum probably which is significantly shorter than when we used to do it in the past, which was four to six weeks using classical methods.
One of the examples that I just thought I'd pull out and show you was we're quite often, during the year, we'll get a whole lot of potatoes for checking before they're being exported, and it's usually 300 tubers in a sample.
These will all get cored. They're grouped into 40s or 50s.
They get soaked in a buffer solution and then we go through a process of spinning down and pulling out the bacteria.
The solution will also get sent up to molecular for them to go through PCR panels for dickeya and pectobacterium, but at the same time we'll play it out and check what we've got. In this case. The example shows that we detect a pectobacterium carotovorum when I got it sequenced, it was actually pectobacterium brasiliense.
Now we'll make a fingerprint from this and then label it and put it into the database.
And then we'll have to go back and check it against some other pectos and check that we can actually detect resiliency on the MALDI without having to always go up to molecular.
So the daunting aspect was learning how to make proof fingerprint profiles and certainly would not have been able to do this without Chris's help.
He's been really great in teaching us in how to understand to do this.
And I'll have to say it's really easy once you know how.
And once you get a handle on the software, it's actually very easy to use.
But the biggest difference, as Chris has put so much work into this, he's created this wonderful spreadsheet, which is a great CHEAT SHEET and I'll show you this as I go along for learning to make the fingerprints.
Some of the difficulties used in the MALDI is when you're having to share the machine with other groups, is getting time on it.
It does feel really daunting when you first start and the other aspect is all the software proprietary and sometimes it's a bit difficult to try and see if you can get extra licenses and things.
So I'm learning to make profiles in bold, starting off with pure cultures of course, and a known culture.
You need to do what they call as a full extraction on a single isolate, but using the method that Chris has developed which is called pit MALDI.
For getting a much better profile than the Brooker method.
So for example, on a target, we'll actually put 24 spots of a single isolate on that target, then run it through the MALDI, using the peak method and collect the profiles, and this is the profiles we collect and this one is with five of those particular spots that we run through on the target plate.
Then using some software that comes with the MALDI you go save all of that data.
Then you have to go and find the data and even that itself is quite daunting because when you go into looking for the data, you suddenly get this massive list and you think, Oh my God, what does this all mean?
Because it's all just numbers.
And then you think you have to open all of these different things, but you actually don't need to go down any further.
And you upload this information at this higher level.
Which is actually the fingerprint. Like those graphs I was showing you.
And then you go to Chris's workbook.
I know you won't be able to read this, but it actually just tells you step by step what you need to do.
And it makes it very, very easy to just follow this the instructions. The workbook has macros in it and, at the end you get this appearing - it tells you whether your profile thing, print that you've just made is, of what quality it is.
So whether it's a high medium, or low.
It gives a weighting and you can see where some things it might not have worked very well.
And then you fill in this other section, which is gathering all the information on the sample that you're making your Spectra on.
And this thing gives you your final name which.
I'll just point to here.
So the naming convention, that we've come up when we're working in the Standards Committee is quite long, but it lets you know that it was a full extraction done on a bacterial isolate.
The isolate comes from Kiwi vines in WA.
It's been molecularly identified correctly, and it's a vouchered specimen in our culture collection.
What are we currently doing?
Any bacteria that we're isolating, we're checking confirmation of the identification using molecular tests.
We're adding more isolates into our culture collection.
Each isolate is also having a fingerprint made.
These fingerprints will be then shared nationally.
But also doctor Tony Chapman from NSW department and I have got a small project funded by Jeff.
With this project, we will be validating more cultures, especially looking at the sub minus swimming group and a few others.
And we'll be sending cultures to each other and validating that our fingerprints that we're making are actually working.
And the aim is that I hope that I will be able to just identify a bacteria directly using the MALDI and not having to keep sending everything up for sequencing.
And yeah, if you have any questions, place them in the chat. Thank you.

 **Vivian-Smith, Gabrielle** 31:15
Great. Thank.
Thank you very much Dominie.
And part of that, a big thank you to Chris.
So if you if you want to please extend a virtual round of applause.
We've now got time for some questions, although I note that Chris has been doing an absolutely magnificent job in the chat, addressing many, many, many questions.
So I might just go to a couple that haven't been.
Well, maybe they've all been answered.
But maybe I'll throw to you. Chris, are there any of the questions that are in the chat that you wanna highlight a bit more or explain in more detail or you haven't been able to explain in the chat?

 **Nellessen, Chris** 32:00
Sure. I'll answer a couple.
So Vera ask if we have to do isolation and culturing. So we often do for fungi, it's the one that you generally don't have to if, our role at DAFF, which you're familiar with, Vera, where we're doing screening for bio security.
We can get away with not necessarily doing so if we're screening for specific things and kind of keeping an eye out on certain pathways.
So for fungi, we're quite able to do an identification of the species level.
Quite easily for bacteria you have the issues directly off the sample of it ends up being a mixture and there tends to be bacterial films on the sample and there's lots of bacteria there.
You'll pick out the most populous species fairly easily, but as you are quite aware of, that's often not necessarily the one we necessarily have to worry about, because there's lots of bacteria out there, so we'll often have to culture to capture.
All the other things that might be there so.
We don't usually get to skip culturing, but sometimes we do for fungi.
For at least, yes, profiles and fingerprints out.
Those are the same. I think I might have answered that analysis is about 30 seconds per test. 37 I believe was the number I got to last time.
But there's a slight amount of variation of about 5 seconds depending on the sample, due to just how the laser moves around.
Sharing, we're organizing through our steering committee.
We've got a process now at DAFF to request access to the fingerprint libraries we have. So that has to be requested.
And we are hoping to plan a project in terms of expanding the libraries more, but we both the States and territories and our Council are trying to update the library with things that we come across and intercept and not working on.
I think that's a lot of them.

 **Vivian-Smith, Gabrielle** 34:05
Dominie over to you.
You might want to add to some of those.

 **Dominie Wright** 34:10
I think the gap in the plant pathogenic bacteria is also going to be filled with the project that Tony and I have got going now.
And with EUPHRESCO that the fact that we've joined up with the EUPHRESCO project and we're hoping to share some of the libraries between us and they've got.
Some of the exotics that we don't have in Australia, so we're hoping that we can do that work as well.
And bring those libraries across. If we can show, that they'll work on the way that we've developed the standard.
Because the standards between the two groups are very different.

 **Vivian-Smith, Gabrielle** 34:54
Oh, that's good.
There is one question about is the data located in a central location? The validated data from the project.

 **Dominie Wright** 35:09
All those isolates are in either the NSW Culture Collection or in the WA Culture Collection.
So now be because they all have to be vouched.
All the fingerprints that we make have to be vouchered specimens.
We've made that a standard.
That that has to happen so.

 **Vivian-Smith, Gabrielle** 35:35
Great. Thank you.
And Doris, thanks for circling back to David Neil's question.
So can you explain a bit where MALDI-ToF fits in the diagnostic workflow?

 **Nellessen, Chris** 35:50
So it can fit in multiple stages.
That's kind of why it's a great agile and flexible technology.
It shines really well in the beginning because it's really fast.
You can easily screen a bunch of samples.
And whether it's cultured or not, you can do a lot of introductory testing and then when it is cultured, you can screen those cultures, separate them out.
So once you might be more interested, you might want to have to follow up with sequencing because it is generally considered a golden standard and the international basis.
That we can be quite confident in our answers with the MALDI. We like to increase that confidence just by also doing the tests and we'll know it's already that particular thing we're concerned about. So we'll already act, but then we'll follow up anyways just to add those layers of confidence.
So all the stages is where it can fit, but we find the most use in the early stages because of its speed.

 **Vivian-Smith, Gabrielle** 36:50
There's also a question about the time frame. So you answered the time frame. I think for the test.
But there's a question there about how long does it take after that?
The process after the sample is analyzed.
How long does that take?

**Nellessen, Chris** 37:10
I'm curious what Dominie says.
For me it takes a couple of minutes maybe to look at the results and turn that into a report for people, but I'm curious on how long it takes Dominie to look at results and be confident in answers there.

 **Dominie Wright** 37:26
So as we developing our confidence in that the identification is correct and I don't have to send it off for confirmation, like Chris a couple of minutes.
But going through the whole isolation process and subculturing.
That cause the bacteria will either be four or six days to get our initial isolation results.
We're looking at 8 to 10 day turn around time on receiving the sample, but reading the results here once you get the results out, it's a couple of minutes and then you're ready to go with your answers once you know so there's quite a lot of bacteria now that come up that I know is correct and I don't have to send it off for checking.
So that includes things like Pseudomonas put it up, syringae still always comes up with sybastenoise, so we're still trying to sort that one out and other things, but yeah.
And the pectobacteriums are usually pretty correct now and the dickeya are correct. So yeah.

 **Vivian-Smith, Gabrielle** 38:39
All right.
So we might just take one or two more questions.
There's a lot of answers, questions and answers going into the chat as we speak.
There's a couple of sort of high level questions ones around.
What are the standards that we're talking about?
Are they Australia standards and then there's another question of a similar sort of general nature and that's like is this used in the private sector?

 **Nellessen, Chris** 39:09
So MALDI-ToF is extensively used in the private sector in human pathology and in veterinary pathology. There's not a huge amount of time pathology private sector, but it has been introduced there in the past as well.
And there's a number of publications out about that.
It's so common. Every hospital you've been to in the past 10 years has at least one MALDI-ToF.
That's generally how they do those bacteria swabs that you get, that they have the answer for you in 20 minutes.
That's a MALDI-ToF doing that.
Unless if it's a PCR, which they do for some things where they have the test for like COVID, but otherwise it's a MALDI.
Dominie, I'm happy for you to talk to the standards if you want to and how good they are across Australia in terms of what they are.

 **Dominie Wright** 40:01
So under the ARMSFIT committee, we developed a Standards committee for MALDI-ToF with reps from each state.
The majority of the work was yours, Chris.
You're the one who put it up.
A lot of it together, so you should be taking.

 **Nellessen, Chris** 40:19
They're very comprehensive standards for the data itself that comes out.

 **Dominie Wright** 40:21
Yeah.

 **Nellessen, Chris** 40:25
It's something like, there's a lot of mass behind it.
About 18 quantitative points are listed for quality.

 **Dominie Wright** 40:30
Yeah.

 **Nellessen, Chris** 40:36
But yeah, they've been endorsed by SPHD, so they're Australian standards for quality.
On the data that's put out, you still need to make sure that what you're actually grabbing the fingerprint from is a good sample, and that's on whoever's making it.
But we were able to make this, so it's more easily applied by just looking at the data because it's pretty easy to say, make sure your sample is vouchered and identified correctly, but then checking the data down the line is a bit harder to do so.
We introduce those standards for that purpose.

 **Vivian-Smith, Gabrielle** 41:10
Right. One final question is, can the detection identification go down to subspecies or race level for bacteria? It seems for Pseudomonas that stays at the species level?

 **Nellessen, Chris** 41:29
There's conflicting information there.
At DAFF, we haven't tried to really go down to sub species level very much.
I've definitely read a lot of people have difficulty with that.
I know with peak MALDI there's the potential to be able to do that more easily.
Dominie might be able to speak to this more, but I know it's difficult.
Definitely with off the shelf settings.
I don't think you can do it so.

 **Dominie Wright** 41:53
Doesn't work with off the shelf settings.
And we I don't think there's enough.
We're still trying to work it out, but I have the sneaky suspicion we won't be able to go that far because I don't think the proteomics will be different enough between the subspecies level, because if you think about the history of bacteria, the subspecies levels were only given to identify which plant it came from originally.
And especially in the Pseudomonas syringae group, which is so broad.
That's why all the subspaces levels arrived.
So it'll be interesting to see if we can, but I suspect we won't be able to get to that level.

 **Vivian-Smith, Gabrielle** 42:44
Right. Thank you, Dominie.
So we might just finish up there today.
It's been a really interesting webinar so a huge thank you to our two speakers.
Chris Nelson from Department of Agriculture, Fisheries and Forestry, Commonwealth Department of Australia and Dominie Wright from our counterpart department in WA.
It was really interesting and thank you so much for addressing all of those questions that really added to the discussion.
And I think helped enlighten us and make help us understand a bit more about this new application in plant health diagnostics in Australia.
So before we wrap up, we, I just want to also mention that the nominations for the 2025 Australian Biosecurity Awards are opening very soon and we encourage everyone to have a think about whether you know any biosecurity champions in your networks who you might like to nominate for one of those awards.
Please keep an eye out on agriculture.gov.au/ABA for more information and on how to submit a nomination.
So before we go just a huge thank you Dominie and also Chris.
I found it fascinating.
I keep hearing about MALDI-ToF and hear little snippets.
I'm not a diagnostician myself, but you did a great job of explaining it to those of us who are not diagnosticians so.
It's really good.

 **Dominie Wright** 44:19
Thank you.

 **Nellessen, Chris** 44:20
Thank you.

 **Vivian-Smith, Gabrielle** 44:22
Thanks everyone. That brings us to the end of our monthly ACPO webinar.