Episode 90: Fentanyl: Toxicity, Testing and Interpretation

Transcript

Welcome back to the Tox Lab, I'm Rebecca and I'm Rob and this week we're finally two years in. I don't know how we haven't spoken about this subject. No idea. We're finally going to be looking at fentanyl.

I suspect of all of the subjects we've spoken about fentanyl is a word people will hear perhaps most often when they think about drugs, particularly in the US and fentanyl is interesting because it's both a legitimate medicine and also an illicit street drug. So before we dive too hard into science why don't we have a little bit of a general intro about what fentanyl is and what the word sort of means these days because it's probably street fentanyl is probably very different from medicinal fentanyl in the sense that one is pharmaceutical and one is maybe not but we'll kind of dive into that a little bit more later.

But fentanyl is a really powerful opioid drug. It acts like drugs like morphine, like codeine, really really powerful version of those drugs but it's completely synthetic. So morphine and codeine derived from the poppy, fentanyl you can make completely in a lab and it's got a rich history. It was originally synthesized in 1959 by Paul Janssen and I think we've probably mentioned Janssen before.

Yeah we have yeah. He is arguably one of the most famous and infamous people in fentanyl's entire history because if it weren't for him the fentanyl crisis would not have unfolded in the way it had but you know he wasn't a nefarious individual. Fentanyl was originally designed as a medicine and in the 1960s it became really an essential surgical drug. It was an intravenous anesthetic.

It led to this incredibly powerful pain-killing action and you know really high potency, really fast onset, relatively short duration as well. So people weren't out of it for very long compared to some compounds and it became really an essential medicine and still is today. You know really really commonly you go in for surgery, fentanyl is part of the cocktail they give you. But as we say there's this legitimate pharmaceutical fentanyl and this is produced with very high purity supplied at known doses and then you've got this illicit fentanyl, street fentanyl that will be a phenomenon many more people will be familiar with.

And this as I say is really big in places like the US, in Canada. We see a lot less of it in the UK although as we recently touched on the EUDA drug report possibly some signs that it might be emerging as an illicit drug in Europe too. And street fentanyl I say is quite different, it may be sold as powder, sometimes it's mixed with heroin or other cutting agents. It might be put into tablet form, it might be used to mimic authentic medicines, you know as a counterfeit.

Particularly big in the US and this is partly due to essentially supply lines. In order for heroin to be an abundant street drug you need to grow lots and lots of poppies, get that product across the world from places like Afghanistan where it's traditionally grown. Fentanyl you can make in a factory, you can buy in chemicals, you can mix chemicals together and produce this fentanyl. And so that's really the core difference, one is plant derived and one is synthetic.

But also it's so much more potent, you know potentially a hundred times more potent than morphine. So as I say it's a big topic that a lot of people will have heard on the news, I nearly said in the

newspaper but I don't know how many people read newspapers these days. I think some study. If you read newspapers you might have read about fentanyl in the newspaper.

So we've got a few little fact or fictions about fentanyl that we're going to sort of unpack a little bit. So first one and I've already given this one away fentanyl can be used as a medicine. True. True, yeah absolutely true.

The drug itself is neither good nor bad, it all depends on how it's used but it's routinely used in operations and for treatment of pain. Pharmaceutical fentanyl and illicit fentanyl are different things? I mean it's a tricky question because illicit fentanyl tends to be less pure than medicinal fentanyl. It's not just a tricky question, it's also a trick question.

Yeah. Because in theory the active ingredient in street fentanyl is the same as the active ingredient in medicinal fentanyl, fentanyl. The trouble is the word fentanyl for street fentanyl actually means a few different things and there's also a whole host of fentanyl analogues and they may get sold on the street as fentanyl. They're actually chemically different.

So a little bit of a trick question I'm afraid. Fentanyl is so potent that merely touching it can lead to an overdose? I think that's one of those myths that you know has become very over-exaggerated in the news and in headlines. It gets thrown around headlines a lot and to be fair you know you do hear stories of law enforcement people raiding fentanyl factories where there's kilograms of powder floating around and powder flying around the air and I think the evidence is that in general skin contact isn't sufficient to cause fentanyl overdose.

That said if there is a lot of powder in the air because there's been a shootout in a warehouse filled with fentanyl or something like that then it's certainly plausible. Yeah. But you are right and there are quite a lot of headlines and I think yeah I hate using this word hysteria but I think the thought that it can lead to an overdose can sometimes lead to symptoms consistent with an overdose. Panic attacks and loss of breath and so on.

Yeah. But in general it's not absorbed through the skin. And it's one of those myths that I think prevents people from acting when someone is experiencing an overdose because they're worried about coming into contact with it and you know being at risk of an overdose themselves. You're absolutely right yeah no you're absolutely right and I think that does that does put people off actually.

That said you know just for context you don't need very much fentanyl for a dose it is really potent you know we are talking milligram doses which are if you've ever tried to weigh out a milligram it's such a pain most people haven't but you know can confirm we have and it is really hard yes that's not overhype the risk there but also let's not underestimate its potency either. Yeah. So fentanyl's really got this dual identity it's this mix of medicine but also illicit drug and this week we're going to be looking at a few different areas of fentanyl.

Our first paper actually segues quite nicely really; those boundaries blur a little bit because we're firstly going to be looking at medicinal fentanyl that maybe goes wrong. Before we dive into today's episode if you want to stay up to date with our content you can find us on Instagram at @the_tox_lab we're also over on LinkedIn and Reddit and for those of you who don't use social media but still want to get in touch with episode suggestions or comments on episodes why not contact us through our new website at thetoxlab.co.uk.

So this first paper comes out of the National Programme on Substance Use Mortality. Why don't you take us through the paper, Rebecca? So this paper is a systematic case series of Prevention of Future Death reports published in England and Wales. So this study aimed to conduct the first data linkage case series of coroners' and toxicology reports using the NPSUM database and the Preventable Death

Tracker to identify safety issues reported by coroners following fentanyl patch fatalities and assess the differences in deaths reported to each database. So before we go any further let's unpack this a little bit.

So for those who are not aware the death investigation system in the UK is a coronial system, so there is essentially a judicial figure, a coroner, who oversees the investigation and makes a determination. One of the powers that those coroners have is to issue what they call a Prevention of Future Death report, so if they identify circumstances in which intervention could reduce a future death or prevent a future death they issue these reports.

So the data relating to fentanyl patch fatalities extracted from the NPSAM covers 1997 to the 1st July 2024 and the deaths from the preventable death track evolved deaths from fentanyl patches occurring between July 2013 and November 2024. Before we go any further I just want to unpack fentanyl patches. So I talked about fentanyl being used in surgery and it's used in intravenous fentanyl in hospital stays and so on. Another form of fentanyl is fentanyl in a transdermal patch and so this is why actually my original question about fentanyl traveling through the skin is almost a little bit of a trick question too.

That is true yeah. Because these patches are specially formulated to allow fentanyl to cross the skin and so people are prescribed a patch they put it on for a number of days and it continuously releases low levels of fentanyl throughout the lifetime of the patch. And fentanyl patches have come under a bit of controversy so the MHRA and the FDA have both issued warnings and guidance over safety concerns following accidental exposure and the safe disposal of fentanyl patches and the risk they kind of pose to others.

So moving back to the paper there were 99 unique fentanyl patch related fatalities with 89 unique cases identified in the NPSAM database and 12 from the preventable death tracker with the first death occurring in 1999 and the majority of deaths occurring in 2019. But it is also important to note that deaths being reported to these databases are quite delayed. They were expecting at the time of writing this paper that there'd be more deaths occurring in 2023 and 2024 that had yet to make entry into these databases so that is just something to bear in mind.

66% of fentanyl patch related deaths involved males with a median age of 45 years and deaths where a coroner wrote a preventing future death report occurred mainly in females and these reports were written involving older individuals so on average age between 50 and 64 compared to the age range of majority of deaths reported to the NPSAM database which were individuals aged between 35 and 49. Presumably and maybe you're going to come on to that there was a difference in circumstance. Yeah I might touch on that a bit later.

They also calculated years of life lost and the value of statistical life which is an economic value used to quantify the benefit of avoiding a fatality and a median of 39 years were lost for every fentanyl patch death costing 2.73 million pounds per death. I really struggle with conceptualizing concepts like financial cost of life loss. I do get health economics in the sense that it's a thing and I appreciate it. I do struggle with it.

It feels very inhuman. Yeah no it does. 45% of the inquest conclusions were that of a drug related death and 96% of cases involved multiple drugs. In 72% of the fentanyl patch deaths resulted from misuse with the remainder being accidental deaths.

And for those deaths where a preventing future death report was written the majority of cases were accidental deaths rather than misuse. And I guess that makes sense. Yeah. To a degree I mean I suppose you could argue that misuse potential could also be subject to a preventing future death

report but in general terms these are cases where something bad has happened and there may be potential for a manufacturer or a prescriber or somebody to have prevented that death.

So 59% of fentanyl patch deaths reported to the MPSum database involved patches that were prescribed to the deceased and all the prevention of future death reports involved prescribed fentanyl to the deceased. There were seven deaths where fentanyl patches were obtained from family members such as parents or siblings and three deaths occurred where fentanyl patches were purchased illicitly. Okay. When looking at indications for fentanyl patch prescribing these weren't known in the majority of cases but chronic pain and cancer pain were common indications where it was reported which fits with the fentanyl patch use.

There's not many indications for people being prescribed a fentanyl patch beyond chronic pain or chronic pain related to cancer. Safety events were reported in 77 cases by coroners and the most common was to do with the adherence and use of fentanyl patches and involve cases of individuals wearing multiple patches at once, one case of three patches being worn in an individual then taking a hot bath or shower and when individuals do this this causes a rapid release of fentanyl from the patch through the skin into the blood. There were also administration issues where 20 individuals had used the fentanyl patch orally.

It doesn't break down whether this was intentional lecturing of fentanyl patches or whether directions given weren't super clear. I have come across cases where people have either eaten a patch or deliberately cut the corner off and sucked the contents out. Seven had heated, inhaled or smoked the fentanyl and five had injected the fentanyl from the patch.

Prescribing safety events involved a lack of medication review, concomitant and prescribing with other drugs resulting in interactions, incorrect doses being prescribed, inappropriate prescribing and one case involved the off-label use in a 10-year-old opioid naive child for pain associated with cerebral palsy. That's a harsh case. Yeah. There were also some dispensing and aftercare concerns.

For example, the dispensing was not reviewed which may have resulted in the incorrect dose being given and this would have been spotted if it had been reviewed. The use of an outdated or defected fentanyl patch was also seen in a couple of cases and I think there was also at least one case where there was a failure to return unused patches to nursing staff which then meant they were accessible to individuals who weren't being prescribed them. Okay, yeah. And these deaths occurred in 40 percent of cranial jurisdictions across the Inland Wales and Northern Ireland with a fifth occurring Northern Island which I was quite surprised about.

Wow. Yeah. That is surprising. So I don't know whether they just happen to prescribe more fentanyl patches in that area.

It'd be interesting to see the spread of prescribing. Prescribing patterns, yeah. Alongside that, yeah. And whether it's like it's weighted that way because they prescribe more or whether something else is going on.

And if they prescribe more, do they have more indications or are they using fentanyl patches? More readily. Who knows? Yeah, that's really interesting.

So 12 coroner's raised 52 concerns across 15 domains for the prevention of future death reports with poor communication, information systems and processes being common domains that concerns were reported about. 19 organisations received the 12 reports and 58% responded. Six organisations had responses outstanding and these include NHS England, the GMC and the Royal College of GPs alongside some local NHS trusts.

I didn't mention this at the start when I talked about the preventing future death reports but not only do reports get sent to people who are relevant to a PFD we call them, but also there is an expectation that they will respond as to what they will do to prevent the future death and this highlights the power of the preventing future deaths tracker because it keeps track of all of these reports and how many people have or haven't responded. There are quite a few on there that are getting on a bit or are quite old that haven't been responded to and I guess in some situations there isn't a response.

That's not like a systemic reason that they can fix I guess. You could still respond and say that though. Well, yeah. So this does show one of the drawbacks of fentanyl in that it is incredibly potent and it is a medication that can get misused.

I'm interested to know what the relative numbers are. We've heard quite a number of cases where things have gone wrong, but actually, is this 1% of all fentanyl prescriptions or is this 10% of all fentanyl prescriptions? I suspect it's not. It's probably less than 1% and so it's a powerful medication that has its role.

I think some of these issues are, as you say, things like medication review or even just giving a powerful medication to an individual that might be at risk of not following the instructions because they're elderly or they've got confusion and so on. There are always a subset of these cases where people intentionally stick multiple patches on themselves to take their own life and that is a risk with fentanyl patches. A risk with any medication.

What is an interesting fact that I didn't mention at the beginning is that there seems to be a decline in fentanyl being prescribed in England and Ireland between 2014 and 2022, but deaths from fentanyl have increased. There was one, I think reported to MPs in 1999 and this increased to 41 in 2023. So despite it being prescribed less, deaths seem to be climbing. That is interesting.

That's outpatient prescribing. Fentanyl patches have decreased. These were just deaths from fentanyl. Okay, yeah.

As a whole, say. That's interesting. So we are seeing the occasional illicit fentanyl in the UK. Pretty few and far between though.

Yeah, it is interesting that the prescribing seems to be going down and the death seems to be going up. I'm not really sure I understand why. Is it just people who are on fentanyl patches are getting older? I mean, possibly.

So that's actually something we haven't touched on and I'm not sure we're going to touch on the rest of the episode. Post-mortem fentanyl concentrations in somebody on a fentanyl patch. So challenging to interpret. Yeah, they're a bit of a nightmare.

You get continual diffusion from the patch in the post-mortem interval. Virtually anybody who is on a fentanyl patch will have a fentanyl post-mortem that is higher than in inverted commas what could be considered fatal because actually the threshold for fatality for fentanyl is pretty low. It's hugely tolerance dependent. And again, another paper that we're probably not going to talk on, but maybe we should definitely touch on this in a future episode.

That paper out of the US where people were taking grams of fentanyl because they're just so tolerant. And so it is a big factor to consider tolerance in that. But also just the interpretation of a post-mortem fentanyl on somebody who's on a fentanyl patch can be really tricky. And so a lot of people are possibly going to be in that in inverted commas fatal range.

Yeah, that's true. And it all depends on what gets put down as the cause of death. It's like the coroner feels at the conclusion of inquest that fentanyl was involved or may have played a role, makes it into

the things listed under the cause of death. And that also potentially skews the stats if they were on a fentanyl patch, they were on the correct dose, but because of post-mortem diffusion.

Or they were also taking diazepam and pregabalin and a load of other things that, as you say, perhaps in retrospect, maybe somebody should have reviewed their prescriptions. Okay, so now let's switch it up a bit. And we're going to look a little bit at the analysis of fentanyl and how we decide who's positive for fentanyl. And this is particularly important in places like the US, where you may want to know if somebody's taking illicit fentanyl.

So illicit fentanyl use has increased in the States. And there are now guidelines by the association for diagnostics and laboratory medicine, and some laws for common in certain States that fentanyl should be included on a urine drug screen panel. So a lot of urine drug screen testing involves immunoassays with set cutoffs. So in workplace drug testing settings, the cutoffs are set to legally mandated thresholds aimed at reducing false positive detections.

Whereas in a clinical setting, urine drug screening cutoffs are often set at a lower threshold to expand the detection window and increase sensitivity, but aren't evaluated alongside the clinical utility of the test. Commercially available assays have fentanyl cutoffs ranging from one to 3.8 nanograms per millilitre with differing degrees of cross-reactivity with fentanyl analogues. And some second generation assays have been designed to have an enhanced cross-reactivity with norefentanyl, some of the metabolite norfentanyl, aimed at increasing detections. So firstly, we're looking at relatively low numbers.

Yeah. We are looking at nanograms per millilitre or micrograms per liter, so relatively low concentrations. But also, yeah, you hit the nail on the head there when you start talking about norefentanyl. Fentanyl is cleared relatively quickly.

That's actually a combination of two things. Firstly, it is probably quite rapidly absorbed into fatty tissue. So it probably leaves the blood fairly fast, but also it has a relatively short mode of action. And so you're right, if you're drug screening somebody, there is a world in which they don't take their drugs that morning on the morning of their drug test and will pass the test because they were still using the day before, but they had dropped below the threshold that morning.

And this is the logic then for looking for the metabolite because the norefentanyl hangs around for a lot longer. And the big issue with fentanyl that we've touched upon is that it's widely used in inpatient hospital settings for analgesia, and it's frequently used during labor in spinal and epidural anesthesia. And this also has the ability to cross the placental barrier leading to detected fentanyl in neonatal urine samples, which can lead to quite serious consequences, including child welfare reports if legitimate medical administration is not recognized as the cause of this positive result.

So this study aimed to evaluate urine drug screening utilization and to retrospectively assess the suitability of fentanyl positivity thresholds for high volume populations, including the adult emergency department, labor and delivery and neonatal units. And this analysis was carried out at the Johns Hopkins Hospital Central Laboratory, and they assessed samples across 2024 and then looked specifically at samples from high volume units that were selected between September 2024 and February 2025 for further analysis.

So these samples were analyzed using the ARC fentanyl 2 homogenous enzyme immunoassay on a Roche C502 analyzer, and this has a cutoff of 1 nanogram per mil and has a 7% cross-reactivity with norefentanyl. So 15 nanograms per millilitre of norefentanyl will produce a positive result on this immunoassay. Samples were then also screened using LC high res MS and quantitation of fentanyl was conducted using LC MS MS. So over a year 17,801 urine drug screens were performed and 76.1% of

these were from inpatient units.

The overall positivity rate for a urine drug screen, excluding fentanyl, so when looking for other drugs, was 43.9% with higher rates being observed with inpatients compared to outpatients. And the overall positivity rate for fentanyl was 15.1% and was again higher among inpatients compared to outpatients, which kind of makes sense. Patients in an inpatient study are going to be on drugs that will cause them to test positive on a urine drug screen. Makes sense.

The highest volume of samples came from the adult emergency department with 5,901 samples followed by labour and delivery with 2,803 and general medicine units at 1,192. The highest fentanyl positivity rate was observed in neonatal units at 33.8% followed by the emergency department at 26.6%, general medicine at 25% and labour and delivery at 2.5%. So neonatal, they're testing positive not because they've been given fentanyl, but because mum's been given fentanyl and labour. We'll get onto that in a little bit.

Ah, sorry, skipping ahead. So sample screen and immunoassay were then screened using high res and the samples were quantitated using a triple quad. So the quantitative screen had a limit of quantitation at five nanograms per millilitre compared to the one nanogram per millilitre limit of detection for the high res mass spec and the immunoassay. So there is a bit of a difference and that did cause some dis-concordant results.

So samples across the emergency department, labour and delivery and the neonatal samples were dis-concordant and they did look at norfentanyl and this was quantified and was detectable in all but two samples. So that kind of accounts for that positivity we're seeing on the immunoassay, but we're not seeing fentanyl on the triple quad or the high res instrument because it's below the limit of detection fentanyl, but norfentanyl is there and that's causing the cross-reactivity with the immunoassay. And that here shows the power of mass spec versus the immunoassay, doesn't it?

It lets you see that difference between the positivity caused by the parent drug and the positivity caused by the metabolite, the breakdown product. So there were two of these results that didn't have a quantifiable norfentanyl, but both individuals were on labattilol, which is a positive interferon in the fentanyl immunoassay, so did lead to a false positive result. That's interesting. So we're looking at positive patients in the different departments.

There were 12 positive patients in the emergency department and 11 did not receive any fentanyl analgesia to explain their result. 6 had reported recent illicit use. 2 denied recent use and 3 had an unknown use status. 11 of the patients had documented history of polysubstance use, including opioids, cocaine, benzos, alcohol, cannabis and methamphetamine.

One patient in the emergency department was receiving IV fentanyl analgesia and also reported illicit recent use. 71% of the 14 positive patients in labor and delivery had been given fentanyl. 2 had a fentanyl that was quantifiable and these were individuals who were not receiving fentanyl but had a history of polysubstance use involving opioids, cocaine and cannabis. For the neonatal samples that were positive, 32% were from neonatal IV administration.

41% were maternal epidural administration. 23% were maternal IV administration. So we're looking at the fentanyl concentrations in quantifiable samples. So in the emergency department, there were 9 samples that had a quantifiable fentanyl, so above that 5 nanograms per millilitre concentration.

And the median concentration was 83 nanograms per millilitre. And the majority of these were from individuals who were not being given fentanyl but had reported recent illicit use. And that one individual who was given IV fentanyl but had also reported illicit use. 2 LND patients had quantifiable fentanyls and the average result was 41.6 nanograms per millilitre.

And again, these were both individuals who were not given fentanyl in hospital but had history of polysubstance use. So all in all, it looks like this screening test works fairly well, right? Yes, the issue is the cutoff. So when you're using a cutoff of 1 nanograms per millilitre, which is the limit of detection on the immunoassay, you do get a lot of positive results.

But when you look at the quantifiable samples where the limit of quantitation is 5 nanograms per millilitre, anyone who's above that cutoff hasn't been given fentanyl in hospital and it's coming from illicit use. Okay, so actually using the higher cutoff, you will pick up the illicit fentanyl and not the medicinal fentanyl. Yeah, and by using that lower cutoff, especially in settings such as labor and delivery and in neonates, you then risk causing quite a lot of downstream harm where the drug result ends up in a report.

And I guess that could be particularly problematic if somebody is, for example, has a history but isn't currently using and has just given birth, there may be reason to doubt the denial, for example. Yeah, I can see why that's a problem. And medicinal administration isn't always captured very easily in medical records. And it might take a while for that information to be pulled out, but by that time, the balls already started rolling and then neonates can be taken away from their parents and things like that.

So there are quite significant severe consequences. This paper was quite interesting at looking at changing that cutoff. I don't know whether they went ahead and did change it. I think a lot more work needs to be done across different populations to work out whether if you increase the limit of detection from one to five nanograms per millilitre, do you always capture the cases that it's important or do you start missing things?

Did they do any rock analysis? I'm going to have to unpack this for our listeners. I've kind of nerded out now. I didn't see any rock curves in the paper, sadly.

So as you make a test more or less sensitive, you will make it more or less specific in the sense that sensitivity goes up, specificity goes down. If you move that cutoff to five, you're going to stop pulling in all of the borderline ones and you're going to report fewer positives, but you are going to miss those people in that bottom curve. And so it's going to be more specific, but it's going to be less sensitive. And a rock curve really looks at the balance between the two and tries to work out the maximal area under the curve, if we call it.

I didn't think we'd be talking about rock curves to say so. I didn't think so either. So when they looked at their proportion of patients that are positive, when they increased that cutoff from one to five, they still captured all the individuals who had illicit or environmental exposure to fentanyl, but they didn't flag all those patients that were positive from iatrogenic exposure. So being given fentanyl in hospital.

It sounds like in this data set at least, it was a good call. Interesting. So I actually have a fun fact about fentanyl and norfentanil for all those mass spec nerds out there. And I know there's a few of you listening, don't be shy.

Another drug that's commonly given in epidurals is bupivicaine. It's a local anesthetic nerve blocking agent. And bupivicaine is broken down to norbupivicaine. And that is isobaric with norfentanil.

And that can cross react with your norfentanil assay. So that's one to watch as well, particularly in people who are given epidurals because they're often given both. Yeah. You can get kind of chaotic pictures of what looks like a massive norfentanil peak.

And actually it's not, it's norbupivicaine. They look very similar structurally as well. So one other way of trying to identify the difference between illicit medicinal fentanyl and illicit fentanyl is by looking for

other markers of the street fentanyl. Because as we said earlier, it's not pure.

It's not pharmaceutical grade. And one such marker is this compound called 4ANPP. And we thought we'd have a little bit of a look at that. So there are different methods for synthesizing illicit fentanyl, including the Janssen method, which tends to produce a high purity product, and the Siegfried method, which produces a larger amount, but often leads to impurities.

And by 2019 in the US, the majority of illicit fentanyl that's being produced was via the Siegfried method. As this requires less expertise, but it does also allow for the detection of these impurities alongside the fentanyl. And these will vary depending on the starting materials that are used and the formation of pre-cursed analytes in the synthesis pathway. And one of these is 4ANPP or 4-anilino-nphenol-etha-peridine.

This is also a monomythabolite fentanyl, but has no in vivo activity, but it's a major precursor in the Siegfried method and is cast as a schedule two control substance in the states. So the aim of the study from NMS Labs was to evaluate four ANPP concentrations and four ANPP fentanyl ratios between clinical and post-mortem cases to identify patterns that could help determine the source of the fentanyl. And they used their cases from 2023, so from January to December. And this is completely logical, right?

Let's think about this for a moment. In medicinal fentanyl, you should have a nice fentanyl peak and you're gonna have a very, very small four ANPP because it's a minor metabolite. In illicit fentanyl, we should be seeing the fentanyl peak but we should also see a much bigger four ANPP because it's a synthesis precursor. So there is a limitation that I want to flag before we start.

There was an assumption that the clinical cases that they were looking at, those individuals were being prescribed fentanyl and the post-mortem cases, it was purely from illicit use. And as we've seen in the previous paper, there is gonna be an overlap in clinical settings where people are coming in because they've taken the overdose of fentanyl, for example, or they're being given both and same, you're gonna get post-mortem cases where they were in hospital and they've been given fentanyl and that source was medicinal and not illicit. So 32,723 forensic cases and 1,015 clinical cases were positive for fentanyl in 2023.

91% of forensic samples had a reportable for ANPP concentration with a median of 2.1 nanograms per millilitre and the range was from 0.2 to 4,100 nanograms per millilitre. Huge range. Huge range. 44% of the clinical samples had a reportable for ANPP concentration with a median concentration of 0.96 nanograms per millilitre, ranging from 0.2 to 306 nanograms per millilitre.

So that range is definitely smaller. There were significantly less cases, though that is another point. So the ratio between fentanyl and 4ANPP was also investigated and there was significant overlap between the forensic and the clinical populations. Forensic samples had a median ratio of 0.141 and this ranged from 0.00078 to 140.

Compared to the clinical samples with a median ratio of 0.105 ranging from 0.005 to 60. There is a big problem here, isn't there? Because the ratio will vary depending on when somebody took the fentanyl. Yeah, that is a good point.

When looking at ratios of less than 0.1, 49% of clinical cases fell within this range compared to 33% of forensic cases, suggesting that clinical cases had proportionally lower 4ANPP concentrations relative to the fentanyl. And that's logical, that's what we were expecting. Yep, so as we've talked about, there was that big limitation that they didn't have access to the case history for the clinical samples. So the source of the fentanyl in those cases might be up for debate.

Another point to consider with the post-mortem samples is that post-mortem redistribution of 4ANPP isn't really well understood. We know that fentanyl undergoes post-mortem redistribution, but how that then affects the ratio between fentanyl and 4ANPP is something that needs some further work. This wasn't an interesting paper, but I think it's only, when looking at the ratios, it's only really useful if your ratio is less than 0.1 and it kind of guides you a little bit either way. Well, let's not think of it like that.

Think of it in the sense that it's useful because it tells us that that information isn't useful. Yeah, that's also true. It shows us the limitation of the evidence as much as anything, doesn't it, which is really helpful? It'd be interesting if you could go back through the clinical samples and identify cases where they were definitely given fentanyl and cases where they definitely weren't and see what the ratios look like for those ones.

I suspect that the overlap we are seeing here in the ratios is not just because there's patient overlap in the clinical samples. No, but it'd be interesting to see whether that patient overlap helps to clear up a little bit. Yeah, so just thinking in terms of pure ratios, this is my, run with me on this mathematically in my head and tell me if I'm making sense. If I take fentanyl, my blood concentration of fentanyl will increase quite fast.

My 4ANPP will very gradually lag behind it. As the fentanyl is cleared, my fentanyl will go down, the 4ANPP will go up. So there will be a point very early on in that administration of fentanyl where I have a very high fentanyl and an undetectable 4ANPP. Yeah.

Conversely, there will be a point in that curve where I have an undetectable fentanyl and a relatively high 4ANPP. Yeah. So a high 4ANPP to fentanyl ratio doesn't tell you anything. Yeah.

You need to know when it was taken in order to interpret that ratio. Yeah. Does that make sense? Yeah, I think the expectation was because illicit fentanyl produced by the Siegfried method contains a lot of 4ANPP that there would be a very obvious data set where that 4ANPP is ridiculously high because it's all coming from impurities in the fentanyl that you could then tell them apart.

Okay, but it turns out you can't really. Reliably, you can't, no. It is interesting, isn't it, looking at the fentanyl picture in the US versus what we're more familiar with in the UK and Europe. Because when we see fentanyl in our day-to-day, virtually always, it's been given in hospital.

Yeah. The idea of needing to have a marker to distinguish between illicit and illicit fentanyl, circumstances usually tell you what's going on, right? Yeah. Whereas clearly in the US, that's not the case, there's a lot of overlap.

And it's interesting just to see this very different perspective on a drug that we see, but not really, not like US seem to. Yeah, it is interesting how there's that difference that the US do see a lot of fentanyl, whereas we see a lot more heroin than they do. And that's kind of the, like, opioid of choice here. Yeah, that's it, that's it.

Yeah, very true. So is there some other magical marker that would let us tell the two apart? What we need is a synthesis intermediate that isn't also a metabolite, right? Yes.

That's the dream. I don't know if one exists. I don't know, there might be something, I guess it depends on, it's so variable depending on starting materials and the different intermediates that different methods will produce that I don't know where there's like a reliable enough marker from like the Siegfried method that can help you identify illicit fentanyl. Because it'd have to be present in high enough concentrations for you to spot it and be consistently produced in that method as well.

Yeah. Even if you're like starting materials might be slightly different. And actually that's another thought I have just had. Presumably there's a lot of batch to batch variation of purity and how much of the precursor and synthesis products are there versus end product.

I guess it depends on how good of a chemist you are. Well, yes, exactly. So really in conclusion then, fentanyl really is a compound of many, many different faces. The legitimate pharmaceutical, as we've heard, a street drug, and also there's gonna be quite a lot of overlap with people in the middle.

And I hope you've enjoyed listening to us this week as we have a look at fentanyl. I've certainly really learned something actually looking at these papers. If, as always, you've enjoyed listening, do give us a like, give us a share. We really appreciate that.

Hope you all have an amazing week and we'll see you next time. Bye.