



© School of Allied Health Sciences, De Montfort University UK

### Transcript of API strip videos

#### Setting up the API strip and inoculating with a stock solution

Right I'm going to firstly label the API strip and so you just label it with the organism that is unknown in that right hand corner, and then what we do is take the lid off and in the base of the well you can see all these corrugated areas. And what I'm going to do is just fill those with water. If there is any excess water, in order to find out if you've got any excess, if you move it up and down until you fill most of those wells, and if there is any excess get rid of it in a pot. And then that provides a humidified chamber for your API strip. And that (the lid) just sits in there like that and then I'm just going to place it at a slight angle and what I'm going to do next is take one colony from here (picks up a Petri dish), and I'm probably going to take this colony there (locating a round colony), inoculate it into 5 mls of sterile distilled water and then use that to fill these all the way across.

So I'm going to move the Bunsen flame to the hottest flame and what I want to do is now flame this straight wire until it is red hot to sterilise it and get rid of any bacteria on there. Give it a few seconds to cool down and then pick that colony there (pointing to a colony) into the sterile distilled water. I've got quite a bit on the end of that straight wire, and then I'm going to inoculate the distilled water.

So what you need to do is bring the straight wire inside the inside of the universal and then emulsify a little bit in the distilled water and then on the inside of that glass container. And then you take it out, pop the lid back on and flame your straight wire.

And then give that a nice shake so it has nicely mixed in the bacteria in the distilled water. And I'll stop there.

#### Pipetting inoculum into the API microtubules

(Proceeds to fill up the API strip with inoculum).

So I'm just filling up the microtubules with the bacterial suspension, only up to that level all the way across to the very last one. I'm just going to stop there because I've run out of suspension. So then go back and get more suspension into your pipette and fill in the rest of the cupules starting with INDOLE, always doing it in that left hand corner which minimises the air bubbles you will get. Up until the very last one ARABINOSE and then I'm going to stop there. I'm going to pick it up to see if there are any air bubbles, which there aren't, so that is OK, so I'm going to put it down flat on the bench.

## Completing the strip ready for incubation

So topping up the CITRATE cupule with some more bacterial suspension, topping up your VP with some more bacterial suspension and topping up the GELATIN with some more bacterial suspension. Then we're going to use the liquid paraffin to top up some of the cupules, and the ones we are going to top up are the ones that are underlined so ADH, LDC, ODC, H<sub>2</sub>S and urea. And what we are doing is simulating anaerobic conditions by topping up with paraffin oil. So pop the lid back on so you don't contaminate with any bacteria available in the environment. And then we are going to do a purity plate with our bacterial suspension just to make sure what we've put in there is pure so we are just going to take one drop and drop it on there (onto the agar plate) and that will be sufficient.

Then using your bacteriological loop and using the hottest part of the flame, sterilising it again, and then we are going to streak out that drop we've put onto the agar plate all the way across, so eventually next time we can see nice single bacteria colonies. So going back to originally where we inoculated it, spreading it out, then I've got to heat the loop each time, moving the plate around, take some there across to the next area, flame the loop, taking a little bit more from that sector to the next area, and we're going to do that two more times so that eventually right at the very end we are going to have nice pure colonies that we can then do further work on if we need to. So this will be the last one and what you need to do with the last one is do a squiggle so that we can see those nice single colonies next time.

And then flaming the bacteriological loop when we have finished, pop the lid back on, and then it is going to be incubated that plate, with the API strip.