

Microbial Flora of Personnel

Introduction

This series of experiments are designed to show the ubiquity of bacteria on personnel in a food factory and domestic situation, and routes of their dispersal. Take your time read through what you are going to do, think of additional sampling sites, and label the plates before you start.

Part 1 Handwashing

- 1) Label PCA, VRBGA and BP as 'Before washing', 'After washing' and 'End of practical'.
- 2) Place your fingers of your writing hand on the surfaces of the predried PCA, VRBGA and BP.
- 3) Wash and dry your hands THOROUGHLY with an antimicrobial soap THREE times. Note : you can do some of Part 3 at this time as well.
- 4) Repeat finger sampling.
- 5) Record the ingredients in the antimicrobial soap.
- 6) When you have finished your practical work, again place the fingers of your writing hand on the final set of agar plates.

Part 2 Skin flora

- 1) Label the plates; 'forehead', 'ring', 'watch', 'fingers'
- 2) Moisten the swabs as demonstrated.
- 3) Swab the following locations; forehead just below hairline, under ring, under a watch and between the fingers.
- 4) Use the swabs to inoculate PCA, bifidobacteria, MRS, VRBGA and BP plates.

Part 3 Aerosol dispersal

- 1) Whilst someone is drying their hands, hold an open PCA plate in the stream of air to collect any aerosols.
- 2) Cough onto the surface of a TSA plate.
- 3) Using the air sampler for bacteria and yeast & moulds test for 8 minutes the air either;
bench, window sill, sinks, corridor.

Part 4 Transmission via fomites

Hygiene monitoring

- 1) Moisten 12 swabs with saline as demonstrated (10 ml volume provided).
- 2) Find 4 potential places where you might expect to find faecal bacteria (other than the obvious!). For example; toilet seat, sink tap, door handle, floor.
- 3) Sample by holding three swabs in your hand each time. Remember to label where the sample was taken from.
- 4) Streak one swab from each sampling point on a PCA, Oxford or VRBGA plate as if you were streaking for single colonies.

Worksurface monitoring – note there are two types of swabs being used.

Version 1 Benches E, F & G are using Cottonwool swabs

- 1) Moisten 5 cotton wool swabs in saline and return to their packaging.
- 2) Swab a work surface (10x10 cm area) and streak on
PCA, Pseudomonas agar, *B. cereus*, Perfringens and
VRBGA agar plates.
- 3) Wipe the surface clean with the cleaning cloth provided.
- 4) Moisten 5 cotton wool swabs in saline and return to their packaging.
Reswab and streak onto
PCA, Pseudomonas agar, *B. cereus*, Perfringens and
VRBGA agar plates.
- 5) Put Hycolin onto the surface and allow to act for 2 minutes.
- 6) Moisten 5 cotton wool swabs in saline and return to their packaging.
Reswab and streak onto
PCA, Pseudomonas agar, *B. cereus*, Perfringens and
VRBGA agar plates.
- 7) Place the cleaning cloth in a Stomacher bag with 20ml saline and
'homogenise' for 30 seconds.

- 8) Dilute the Stomacher bag contents to 10^{-8} in 9ml saline.

In the sequence 10^{-8} to 10^{-1} , inoculate predried nutrient agar, *B. cereus*,
Perfringens, Pseudomonas agar and VRBGA with 0.1ml of each dilution.
Remember the 4ml overlay of the VRBGA and incubate 37°C aerobically.

Bacterial Transference

You are provided with :

Knife, fork and pen.

Swab the knife, fork and pen and streak on PCA.

Everyone on the bench use the pen to sign in on the register.

Re-swab the pen and streak on PCA.

NOTES:

- 1) At the end of the session remember to overlay the VRGBA plates with molten
VRGBA in order to exclude air from the colonies.
- 2) Remember to do the final finger plates

Worksurface monitoring – note there are two types of swabs being used.

Version 2 Benches B, C & D are using ATP swabs

Note ATP swabs can be 'stored' for up to 30 min before activating. But once activated they must be read within 5 seconds.

- 1) Using an ATP swab,
Swab a work surface (10x10 cm area) and determine the RLU value.
- 2) Wipe the surface clean with the cleaning cloth provided.
- 4) Reswab the surface and determine the RLU value.
- 5) Put Hycolin onto the surface and allow to act for 2 minutes.
- 6) Reswab the surface and determine the RLU value.
- 7) Place the cleaning cloth in a Stomacher bag with 20ml saline and 'homogenise' for 30 seconds.
- 8) Dilute the Stomacher bag contents to 10^{-8} in 9ml saline.

In the sequence 10^{-8} to 10^{-1} , inoculate predried nutrient agar, *B. cereus*, Perfringens, Pseudomonas agar and VRBGA with 0.1ml of each dilution. Remember the 4ml overlay of the VRBGA and incubate 37°C aerobically.

Bacterial Transference

You are provided with :

Knife, fork and pen.

Swab the knife, fork and pen and determine the RLU.

Everyone on the bench use the pen to sign in on the register.

Re-swab the pen and re-determine the RLU.

NOTES:

- 1) At the end of the session remember to overlay the VRGBA plates with molten VRGBA in order to exclude air from the colonies.
- 2) Remember to do the final finger plates