Chapter 9

Appendices

Appendix A

Table A.1 API 20E TESTS FOR *ENTEROBACTERIACEAE*

ABREVIATION	TEST
ADH	Arginine
AMY	Amygdalin
ARA	Arabinose
CIT	Sodium citrate
GEL	Kohn's gelatin
GLU	Glucose
H_2S	Sodium thiosulphate
IND	Tryptophane
INO	Inositol
LDC	Lysine
MAN	Mannitol
MEL	Melibiose
ODC	Ornithine
ONPG	Ortho-nitro-phenyl-β - galactopyranoside
OX	Oxidase
RHA	Rhamnose
SAC	Sucrose
SOR	Sorbitol
TDA	Tryptophane
URE	Urea
VP	Creatine

Table A.2 API STAPH FOR STAPHALOCOCCI AND MICROCOCCI

ABREVIATION	TEST
ADH	Arginine
FRU	Fructose
GLU	Glucose
LAC	Lactose
MAL	Maltose
MAN	Mannitol
MDG	α- methyl-glucoside
MEL	Melbiose
MNE	Mannose
NAG	N-acetyl-glucosamine
NIT	Potassium nitrate
PAL	β-naphthyl-acid phosphate
RAF	Raffinose
SAC	Sucrose
TRE	Trehalose
URE	Urea
VP	Sodium pyruvate
XLT	Xylitol
XYL	Xylose

Table A.3: Results of the API tests undertaken for microbial identification. API Staph was used to identify Isolate in column labeled 6. Tests are indicated in braces

TEST	Isolates					
	1	2	3	4	5	6
ONPG	+	+	+	+	+	(O) -
ADH	-	-	-	+	-	(ADH) -
IDC	+	+	+	-	+	(FRU) +
ODC	+	+	+	+	+	(MNE) +
CIT	-	+	+	+	+	(MAL) +
H2S	-	-	-	-	-	(LAC) +
URE	-	-	-	-	+	(URE) -
TDA	-	-	-	-	-	(TRE) +
IND	+	-	-	+	+	(XLT) -
VP	-	+	+	-	+	(VP) +
GEL	-	+	+	-	-	(NIT) +
GLU	+	+	+	+	+	(GLU) +
MAN	+	+	+	+	+	(MAN) +
INO	-	+	+	-	+	(PAL) -
SOR	+	+	+	+	+	(RAF) +
RHA	+	-	-	+	+	(XYL) +
SAC	-	+	+	-	+	(SAC) +
MEL	+	+	+	-	+	(MEL)+
AMY	-	+	+	+	+	(MDG) -
ARA	+	-	-	+	+	(NAG) +
OX	-	-	-	-	-	

Appendix B

Data generated from Statistical analysis

Table B.1: Summary of Descriptive Statistics for Faecal coliforms

Time (weeks)	00	02	04	06	08	10	12
N	**8**	**8**	**8**	**8**	**8**	**8**	**8**
LMS 8 tons/ha							
Mean	3628.75	36575000	7525000	15775074	8525	7049.63	955
SD	1873.66	23419757	15785595	25629047	15533	14967	2245.78
P	0.0009	0.0031	**0.2196**	**0.1252**	**0.1645**	**0.2245**	**0.2682**
Median	2750	34000000	500000	3400000	0	0	0
P	0.0078	0.0078	0.0078	0.0156	**0.2500**	**0.2500**	**0.5000**
LMS 16 tons/ha							
Mean	26125	29937.5	14250000	14213225	870	327.87	395375
SD	10881.8	11724.33	12739590	18453896	1666.97	507.52	474287
P	0.0003	0.0002	0.0158	**0.0658**	**0.1834**	**0.1104**	**0.0505**
Median	31000	33500	11700000	3250000	30	69.5	195000
P	0.0078	0.0078	0.0078	0.0078	**0.0625**	0.0313	0.0313
HMS 8 tons/ha							
Mean	470	5861.25	47455	9175000	0	0	0
SD	1105.74	6546.56	89300.87	16803890	0	0	0
P	0.2684	0.0391	**0.1765**	**0.1664**	-	-	-
Median	75	3550	665	2800000	0	0	0
P	0.0313	0.0078	0.0078	0.0313	-	-	-
HMS 16 tons/ha							
Mean	1848.75	20162.50	7303750	6975056	7.5	6.25	0
SD	1195.04	10810.04	11065447	10933623	21.21	11.88	0
P	0.0033	0.0012	**0.1042**	**0.1141**	**0.3506**	**0.1803**	-
Median	1575	17600	4200000	2900000	0	0	0
P	0.0078	0.0078	0.0078	0.0313	**1.0000**	**0.5000**	-

Table B.2: Summary of Descriptive Statistics for *E. coli*

Time (weeks)	**00**	**02**	**04**	**06**	**08**	**10**	**12**
N	**8**	**8**	**8**	**8**	**8**	**8**	**8**
LMS 8 tons/ha							
Mean	2178.75	26362500	4625000	0	3750	6775	226.25
SD	736.91	23884959	10825730	0	10606.6	14315	447.63
P	0.0001	0.0168	**0.2667**	-	**0.3506**	**0.2225**	**0.1959**
Median	2000	20000000	0	0	0	0	0
P	0.0078	0.0078	**0.2500**	-	**1.0000**	**0.5000**	**0.5000**
LMS 16 tons/ha							
Mean	25250	29937.5	10200000	1629475	684.5	164.38	101625
SD	10361.47	11724.33	13249690	459475	1676.66	348.04	216955.5
P	0.0002	0.0002	**0.0659**	**0.3492**	**0.2861**	0.2234	**0.2268**
Median	29000	33500	5500000	0	5	6.5	0
P	0.0078	0.0078	0.0078	**0.2500**	**0.1250**	**0.1250**	**0.2500**
HMS 8 tons/ha							
Mean	446.25	5861.25	1572.5	1762500	0	0	0
SD	1113.64	6546.55	3575.49	3624495	0	0	0
P	0.2944	0.0391	**0.2536**	**0.2114**	-	-	-
Median	50	3550	315	0	0	0	0
P	0.0313	0.0078	0.0313	0.5000	-	-	-
HMS 16 tons/ha							
Mean	1547.5	19912.5	6991250	2137556	7.5	6.25	0
SD	1359.76	11092.78	11149352	4914364	21.21	11.88	0
P	0.0147	0.0014	**0.1194**	**0.2583**	**0.3506**	**0.1803**	-
Median	1450	17600	3600000	0	0	0	0
P	0.0078	0.0078	0.0078	**0.2500**	**1.0000**	**0.5000**	-

Appendix C

Reagents

LB Medium

10g Bacto-tryptone

5g Bacto-yeast extract

5g NaCl

Adjust pH to 7.0 with NaoH

Ampicilin

Ampicilin 0.25g

Sterile dH20 5ml

Filter sterilise, make aliquots and store at –20 ^{o}C

LB plates with Ampicilin

Add 12g of agar to 1L of LB medium

Autoclave

Allow the medium to cool at 50^{o}C before adding ampicilin to a final concentration of 100 μg/ml

Pour 30-35 ml of medium into 85 mm petri dishes

Can be stored at 4 ^{o}C for up to a month or at room temp for a week

Nutrient Agar

20 g of nutrient agar in 1L of distilled water. Autoclaved at 121 ^{o}C

TE Buffer

10 mM Tris.HCl

1mM EDTA

pH 8.0

50X TAE Buffer

40 mM Tris.HCl

20 mM NaoAc

1mM EDTA

pH 8.5

Dilute 1:50 in dH_2O before use (1 X TAE)

IPTG stock

1.2g IPTG

Add water to 50 ml final volume

Filter sterilise and store at -4^oC

X-Gal

100 mg 5-bromo-4-chloro-3-indolyl-β -D-galactoside

Dissolve in 2 ml N,N'-dimethyl-formamide

Cover with aluminium foil and store at -20^oC.

Solution I

50 mM Glucose

10mM EDTA

25 mM Tris.HCl

pH 8.0

Solution II

0.2 N NaoH

1% SDS

Must be prepared fresh

Solution III

3 M NaoAC

pH 4.8