NOTEBOOK NO. 46
ISSUED TO Gaxiola Estrada-Arroyo
ON 1/17/07 20 07
DEPARTMENT Media Prep
RETURNED 7/10 20 07

SCIENTIFIC NOTEBOOK COMPANY
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TLE Media Prep Training Book No. _____

m Page No. ____

bjective : To learn the overall activities ~~ground~~ g.d. of media preparation

Procedures

1. Media Lab Prep PPE
   - always wear safety glasses, gloves and lab coat during media preparation.
   - dust mask must be worn during preparing media
   - Take off the lab coat when leave the room.
   - Clean safety glasses by running through water and soap after use.
   - Wear close-toe shoes.
   - Take extensive awareness when dealing with chemicals.

2. Media ordering process
   (1) The requesters must submitted media request form 10 working days before they need.
   - Fill in the date requested, the name of requester, date needed (10 working days)
   - project or P.I name and account number of the project.
   - name of media or reagent
   - media recipe book SOP number (RCXXX)
   - volumn, quantity and other specificity of the media (The media that need to be prepared fresh will be prepared in the morning of the day needed)
   - PI signature.

   (2) Stamp the form with the date and time and leave the form in the "IN" drop box

   (3) Picking-up order
   - After the media are prepared, fill in the bottom part of the media request form
   - The batch number X.YYYY-Z ; X is the autoclave number.
     Y is the autoclave ~~to~~ run cycle.
     Z is the letter ~~of~~ specify each batch
   - After ~~sight~~ I.D. the requester sign the name and date when pick up the media, the form is put in the "Out" box.

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essed & Understood by me,	Date	Invented by:	Date
anon Richardson	1/23/2007	Viviana Lopez	1/30/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE _____

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3. Lab rules.
   - let the media lab know you will be working in the lab
   - Wear goggles and clean them with soap and put them into place.
   - Wash your glasses after use
   - Prepare 70% ethanol from 100% ethand under the hood
   - Contact media prep lab technician if there is any problem (when the button at the tank turns red)
   - Do not accumolate waste. Put the trash container outside to be picked up.
   - Always keep your area clean. Wipe the bench, balance or ~~if~~ media spill with 70% ethanol and paper towel.
   - Put the equipment into places.
   - Clean up anything dropped or spilled on the for except chemicals, notify technitian.
   - Ask when have question
   - Let the media technician know when supplies are running low.
   - Wash your hands before leaving the lab.

4. Walkin Cooler Policy
   - No inoculated plate store in a cooler (any inoculated items not allowed)
   - Tubes must be in a secondary container tray, and plates must be in secondary container box.

5. Media Preparation.
   - Follow the instruction in the sop
   - media sterilization is usually 121°C 45 minutes, unless other conditions are specify
   - measure the pH of media prepared before and after sterilization.
   - In broth, after autoclaving, let it cool on a bench. Also, cover the test tube broth with plastic wrap to prevent evapuration.
   - In agar, after autoclaving, put it in a water bath that has been set at 45°-48°C. to let cool to 45°C before pore plate.
   - After pour plate, let it cool over night

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Witnessed & Understood by me,	Date	Invented by:	Date
yanna Alcantara	1/23/2007	Uvrana Lanza	1/30/07
		Recorded by:	

Project No. ______

TITLE MEDIA AGAR PREPARATION
Book No. ______

From Page No. ____

Objective: To prepare media agar

Procedures.

1. ~~Look up~~ Y.D. ~~Locate the~~ Y.D. Take out the recipe binder and ~~look up~~ Y.D. locate the ingredients.
2. Prepare the ingredients on the table
3. Use the appropriate glassware for the appropriate volume (The media should be less than 2/3 of the maximum vol. of container)
4. Use cylinder to measure DI water that is needed in preparing the media. Then pour into the glassware prepared. Mark the level of that volume.
5. Discard some of the water.
6. Measure the media ingredients needed according to the recipe using weight boats.
7. Carefully pour the measured ingredients into the water.
8. ~~Measure~~ Y.D. Use cylinder to fill in the DI water into the glassware to the volume marked.
9. ~~Pull~~ Y.D. Drop a magnetic bar into the flask (the glassware)
10. Pull out some foil and cover the flask.
11. Pull out some autoclave labelling tape and put it on top of the flask. Label the batch letter
12. Stir and heat mix the agar media, watch it carefully to prevent boiling.
13. Fill in the Daily media batch sheet. (batch sheet binder)
14. Measure the pH of the agar media prepared.
15. Autoclave 121°C, 45 mins, liquid
16. Take out and put it in a waterbath which is set to 45°C.
17. Wait until the media cool down. Then, pour plate. (Stir before plating)

Activities

1. Prepare tryptic soy agar 900ml by following the procedures.
2. Prepare sorbitol Macconkey agar by following the procedures.
3. ~~Prepare slant~~ Y.D. pipette slants (batch number 3.1400H)

detailed description of how each ~~media~~ was prepared must be included

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Witnessed & Understood by me,	Date	Invented by:	Date
[signature]	1/24/2007	[signature] Recorded by:	1/30/07

Project No. ____________

Book No. ____________ TITLE Tryptic Soy AGAR PRERAPATION

From Page No. ____

objective. : To prepare tryptic soy agar. (700 ml)

Procedures.

~~1. Bring out the appropriate glassware and use cylinder to measure.~~ Y.D.

1. Bring out the appropriate glassware (flask 1 L)
2. Use cylinder to measure 700 ml ~~DI~~ and pour into the flask. Then, mark the volume. ~~of the glass~~
3. Discard some or about half of the water
4. Look at the recipe and calculate the amount of agar needed

1 L suspend 40 g of tryptic soy agar.

700 ml suspend $\frac{40}{1000} \times 700 = 28$ g of tryptic soy agar.

~~5.~~ Y.D

5. Weigh 28 g of tryptic soy agar and pour in the flask
6. Add DI water to the volume needed (700 ml), indicated by the line marked.
7. Drop a magnetic bar into a flask.
8. Cover the flask with ~~floi~~ Y.D foil and label with autoclave tape.
9. Stir and heat mix the agar media. Watch carefully to prevent boiling
10. Fill in the daily media batch sheet.
11. Pour some media into a plastic basket to measure pH later.
12. ~~A~~ Y.D. Sterilize by autoclaving for 121°C, 45 minutes liquid.
13. ~~After~~ Y.D. Let the media cool down in the 45°C water bath
14. Pour plate using aseptic technique. Stir first
15. Take small sample to test pH after autoclaving

Witnessed & Understood by me, Yewon Dilsaltry | Date 2/3/07 | Invented by: Viviana Souza | Recorded by: | Date 2/8/07

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TITLE Sorbitol Macconkey agar preparation

Project No. _____

Book No. _____

From Page No. ____

Objective : To be able to prepare sorbital MacConkey agar by the standard operating procedures.

Procedures

1. Prepare 700 ml of Macconkey agar.
2. Bring 1 L flask and use DI water and cylinder to measure the 700 ml scale of the flask
3. D^{4D}. Discard some water
4. Look at the recipe and calculate the amount of agar needed.

   1 L Suspend 50 g of sorbitol MacConkey agar.

   700 ml Suspend $\frac{50}{1000} \times 700 = 35$ g

5. Weigh 35 g of sorbitol MacConkey agar and carefully pour into the flask.
6. Follow the procedures 6 - 14 of the tryptic soy agar preparation in page (4)

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Witnessed & Understood by me,	Date	Invented by:	Date
	2/3/07		2/8/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE UVM broth preparation, weekly and monthly housekeeping

From Page No. ____

Objective :- To be able to prepare uvm broth
- prepare 5 racks of uvm broth ; each contains 9.5 ml

1) UVM PREPARATION

Procedures.

1) Calculate the amount of media needed to prepare.
(5 × 75 × 9.5) + 300 = 3.7 L. (make extra 3 tubes for each rack for QC. check)
2) Bring out the appropriate glassware and use DI cylinder to measure DI water and mark the right volume.
3) Discard some of the water
4) Weight the correct amount of agar needed (52 g UVM media per 1 L water)
5) carefully pour into the glass container
6) Use cylinder to fill in DI water to the volume marked
7) Use magnetic bar to help mixing the media under heat.
8) After mixing the media thoughroughly, transfer into tubes by using Omnispens Dispenses.

8.1) Operating Omnispense Dispenser.
a) wearing clean gloves turn on the pump (from the back)
b) manual should come up on the view screen, press auto/MAN until "Automatic" is shown.
c) press SEL 9 times until Recall ✱ comes up and press the program you want (Program 3: 9.5 ml). Then, press ENTER.
On the screen you should now see the volume you selected
(IF you press the wrong program press SEL twice until you see the "RECALL" on the screen. Press CLEAR once followed by the program you want then ENTER
d) IF you see DI. pressed the right program and you can verify it by looking at the volume shown on the screen press AUTO/MAN 3 times until you see AUTOMATIC on the screen.
e) Press START when you are ready to begin and STOP when you are done

8.2) Prepare 2 flasks :- one for DI water and one for waste disposal
8.3) The tube with the glass tip will take the water in and the tube with plastic tip will be the route that water comes out.
(This procedure is to clean the dispenser's tubes) ≈ 150 ml of water
8.4) Lift the water-in tube to let all the water in the tube comes out
8.5) Dip the tube with the glass tip in the media broth, Click start and let it run once into the media container before Dip it into the glass tubes, keep some media for pH measurement. To Page No. ____

Witnessed & Understood by me,	Date	Invented by:	Date
yan Delany	2/4/07	Recorded by:	

8.6) Cap the glass tubes. Not pushed down.
8.7) After finish, run with water again (~300 ml)
8.8) Turn off the pump.
8.9) Clean the bench and area with EtOH.
) ~~Label~~ Y.D. Put the auto clave tape on top of the rack and label with letters.
). Sterilize by autoclave at 121°C 45 mins
) Cool on the bench overnight. Label the batch number on each racks. Push down caps
) Wrap the racks with plastic wrap.
) ~~Label~~ Y.D. Put a labelling tape on the top.
) Store it in a cooler.

) Weekly housekeeping. Y.D.
1) Clean outside the water ~~batch~~ bath with EtOH
2) Clean the pH meter and balances with EtOH

) Monthly housekeeping
1) Take out all the water in water bath. Clean with EtOH. Fill in clean DI water.
2) Dust the shelves ~~and~~ Y.D., cabinets, desk.
3) Check the media in the refrigerator for expired media.

essed & Understood by me, [signature] | Date 2/4/07 | Invented by: [signature] | Recorded by: | Date 2/8/07

Project No. ____________

Book No. ____________ TITLE 0.01 % peptone broth preparation.

From Page No. ____

Objective : Prepare 4 l of 0.01 % peptone broth (9 ml tubes) ~

Procedures

1) Calculate the amount of media needed to prepare
   (5 racks × 75 × 9.5 ml) + 300 = 3.8 L. ≈ 4 L.
2) Bring out the appropriate glassware and marked the right volume by using cylinder and DI water to measure the water.
3) Discard some water
4) Weight ~~0.04~~ Y.D 0.4 g into 4 l of water.
5) Follow the CVM procedures from no. 5 (~~*~~ Y.D see page 6)

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Witnessed & Understood by me,	Date	Invented by:	Date
Yanan [signature]	2/4/07	[signature] Recorded by:	2/8/07

Project No. _____

TLE ~~BH~~ Q.D. Brain Heart Infusion Agar preparation Book No. _____

m Page No. ____

Objective : Prepare 5x 100 ml of BHIA ~~4n plates~~ Q.D.

Procedures.

1) Bring 5 of 1 L flasks and marked the point ~~where~~ Q.D where 100 ml lies.
2) Discard some amount of water.
3) Calculate the amount of agar needed. (1000 ml → 52 g / 100 ml → 36.4 g)
4) Weight 36.4 g of BHIA ~~on~~ Q.D using weighboats and carefully pour it into the water
5) Fill in the water to ~~the~~ Q.D 100 ml in volume (to the marked line)
6) Drop in the magnetic bar, cover with foil, label the name of agar and batch letter on the autoclave tape and put it on the ~~f~~ covered foil.
7) Heat mix until the agar melt and homogenize, pour some agar to measure pH.
8) Autoclave 121°C, 45 mins, liquid.
9) ~~Put in~~ Q.D. Cool down in 50°C waterbath. (around 1 hr.)
10) pour plate using aseptic technique, and measure the pH of ~~the~~ Q.D each batch.
11) ~~Cool down~~ Q.D. & Q.D Let agar solidify overnight

Clean the pH meter, waterbath, and balances.
Clean Desks, bench, laminar flow, waterbath in room 314.

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essed & Understood by me,	Date	Invented by:	Date
[signature]	2/6/07	[signature]	2/6/07
		Recorded by:	

Project No. _____

Book No. _____

Y.D.

TITLE ~~Buffer peptone water preparation~~ BHIA preparation

From Page No. ____

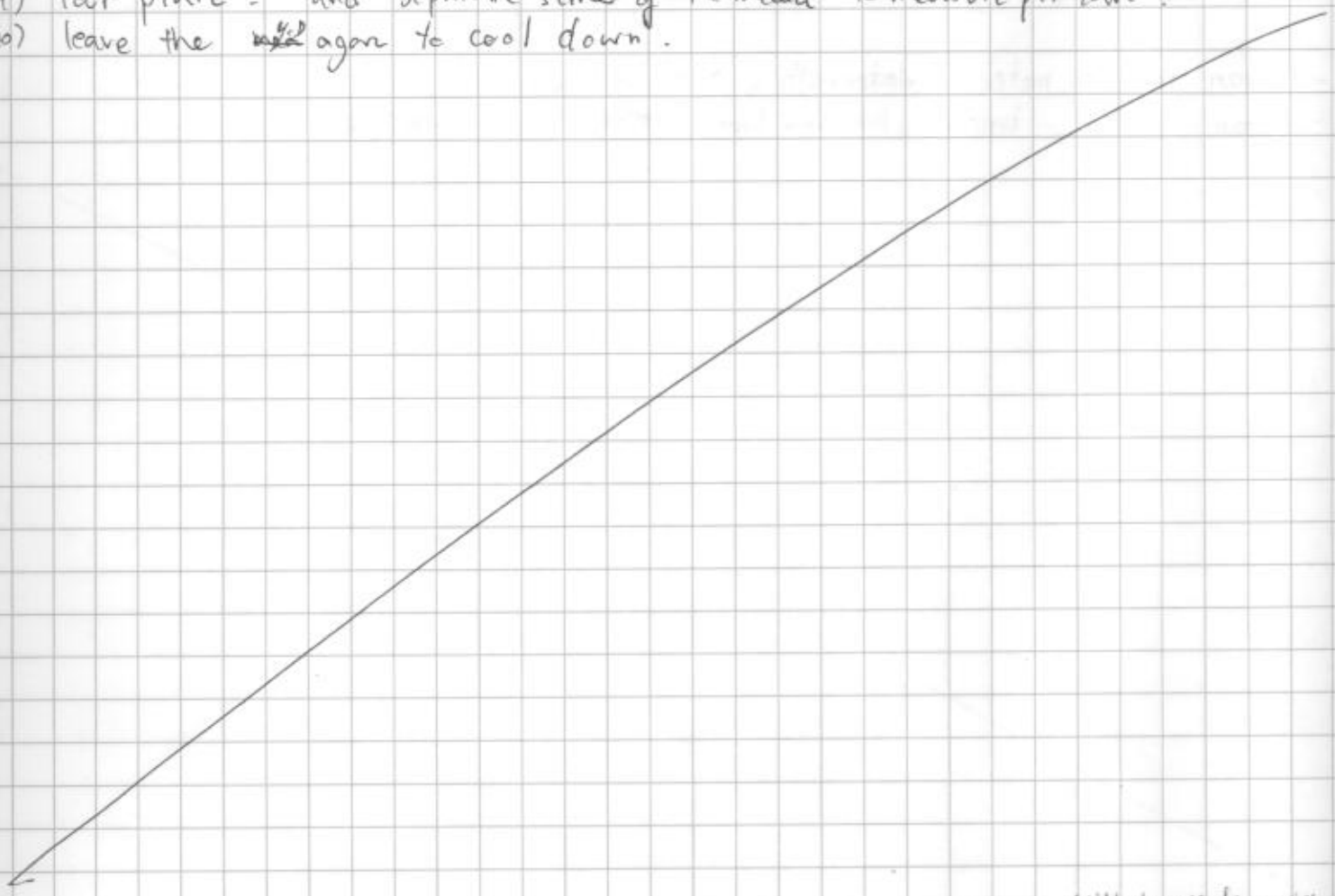
~~Objective: To prepare 3 racks of 9 ml of buffer peptone water broth.~~

~~Procedures.~~

Objective: To prepare 5x 700 ml Brain Heart Infusion Agar.

Procedures.

1) Marked the glasswares used at the level 700 ml. using DI water.
2) Pour out some water to Y.D.
3) Weight 36.4 g of BHIA (100.7157) in each 700 ml (since 1 L need 52 g)
4) Add DI water to 400 ml as marked on the glasswares.
5) ~~Cover~~ to Y.D. Drop a stir bar in, and cover with aluminium foil, labeling tape. Then, Heat Mix.
6) Pour some media to take pH later.
7) Actoclave liquid 45 min, 121° C
8) Put in a waterbath (48° C) Wait for cool down 40 mins.
9) Pour plate. and separate some of the media to measure pH later.
10) leave the ~~media~~ agar to cool down.



* Note: Also include the other activities you do: Kill, housekeeping, etc

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Witnessed & Understood by me,	Date	Invented by:	Date
[signature]	2/7/07	[signature]	2/15/07
		Recorded by:	

TITLE BHIA and peptone water preparation. Project No. _____ Book No. _____

From Page No. ____

Objective: To prepare 5×700 ml BHIA and 3 racks of 9 ml.

~~X~~ G.D-(I) Brain Heart Infusion Agar. 5×700 ml.

Procedure.

1). Use cylinder to measure 700 ml DI water and mark the flask.
2) Pour some DI water out. G.D.
3) Weight 36.4 g of media in to each ~~700 ml~~ flasks.
4) Pour ~~some~~ G.D. DI water until reach 700 ml.
5). Cover with foil and heat mix.
6) Pour some media out to measure pH before autoclave.
7) Autoclave 121°C 45 min, liq. G.D.
8) After taking out from the ~~aut~~ G.D. autoclave, leave it in ~~an~~ G.D. a water bath to cool down for 40 minutes.
9) ~~Be~~ G.D. Quick stir for 20 sec.
10) Pour plates by hand. Measure pH
11) leave the plate on the bench to cool down.

(II) Buffer Peptone water 3 racks ; 9 ml/tube.

1) Calculate the broth needed to prepare. ((9.5 × 3 × 75) + 300 ml) ≈ 2,500 ml
· Prepare 1.25 L × 2. could prepare all together in larger G.D. flask
2) Calculate the ~~agar~~ broth needed. (25 g / 1.25 L) since ~~1L~~ 20 g/L.
3) marked the bottle where 1.25 L lies. Pour out DI water.
4) Weight 25 g using weighboats. carefully pour into the ~~flat~~ G.D. bottle.
5) Heat mix doesn't need mag stir wrath
6) Use the dispenser to distribute 9 ml into each tube.
7) Autoclave 121°C 45 min Liq.
8) Push the cap down, Leave the broth on the bench to cool down.

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Witnessed & Understood by me,	Date G.D. 02/08/07	Invented by: Vincaxa Lopez	Date 2/15/07
Carson Delaney		Recorded by:	

Project No. _____

Book No. _____ TITLE BHIA preparation.

From Page No. ____

Objective : To prepare 5 × 700 ml of Brain Heart Infusion Agar.

Procedure.

1) Marked the line where 700 ml lies using cylinder and DI water.
2) Weight 96.4 g and pour into each flask.
3) Fill the DI water to 700 ml.
4) ~~Hot~~ Y.D. ~~Heat Y.D. mix~~. Cover mit foil and heat mix.
5) Autoclave 121°C, 45 min, liq.
6) leave it in the waterbath (50°C), for 40 mins.
7) Pour plate. Measure pH.
8) leave it on the bench overnight to ~~cool down~~ Y.D. solidify.

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Witnessed & Understood by me,	Date	Invented by:	Date
yanon Bin	02/13/07	Viviana Laza	2/15/07
		Recorded by:	

Project No. _____

TITLE BHIA preparation Book No. _____

From Page No. ____

Objective : To prepare 7 x 700 ml flasks of BHIA.

~~S.O.P.~~ Procedure

1) Use cylinder to measure 700 ml DI water and use it to mark the volume of the flask at 700 ml.
2) Discard some of the water
3) Weight 36.4 g into each flask.
4) Fill in DI-water until it is 700 ml.
5) Drop a stir bar and cover with aluminium foil. Heat mix.
6) measure pH before autoclave.
7) Autoclave 121°C, 45 min, Liq.
8) Leave it in a waterbath (50°C) to cool down.
9) Pour plate. measure pH of the media.
10) Leave it over night to solidify.

II) Weekly housekeeping.
1) Clean the waterbath.
2) Clean the balance and pH meter.

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Witnessed & Understood by me,	Date	Invented by:	Date
Yaman Dabaj	02/14/07	Viviana Loaiza	2/15/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE Prepare 6 racks of 0.01% Peptone

From Page No. ____

Object: To prepare 6 racks of 0.01% Peptone. (9ml/tube)

1) Calculate the amount needed to prepare.
   (9.5 × 6 × 75) + 800 = 4.6 L.
2) Calculate the media (Peptone) needed.
   1 L need 0.1 g.
   4.6 L need 0.46 g.
3) ~~Weig~~ ~~mark~~ Use L Flask to prepare the media
4) mark the glassware ~~tube~~ at 4.6 L. by ~~use~~ using DI water and cylinder
5) Discard some of the water
6) Weight 0.46 g using small weigh boat.
7) Fill ~~the~~ DI water to the mark ~~of~~ 4.6 L.
8) Heat mix using magnetic bar to help mixing
9) Distribute to each tube by using dispenser
10) autoclave 121°C, 45 mins.
11) Leave them to cool down on the bench.

What else was done this day?

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Witnessed & Understood by me,	Date	Invented by:	Date
yanan Dubang	02/15/07	Recorded by:	3/5/07

Project No. _____

LE _____ Book No. Prepare 7x700ml of BHIA

n Page No. ____

bject: To prepare 7 x 700 of Brain Heart Infusion Agar.

~~1) Mark 700~~ Y.D

1) Use Cylinder to measure 700 ml of DI water and use it to mark the glassware

~~2) Weigh 36.4 g of BHIA~~ Y.D

2) Discard some amount of water

3) Weight 36.4 g of BHIA and carefully pour the media in to the flask.

4) Fill the water to the marked line.

5) Drop a magnetic bar and cover the flask with foil.

6) Heat mix til the agar Y.D ~~melt and~~ P.D ~~homogenize with water~~. media homogenize.

7) Pour some of the media into a small container to measure the pH.

8) autoclave 121°C 45 min

9) Cool down in ~~Waterbath~~ Y.D 45°C water bath. for 40 mins.

10) Stir for 20 seconds.

11) Pour plate

12) leave it to cool down on the bench.

Do daily housekeeping for lab 314 (Clean ~~desktop~~ Y.D.
Clean pH meter, balance, waterbath (weekly housekeeping lab 315)
Clean table in lab 305

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essed & Understood by me,	Date	Inverited by:	Date
	02/20/07	Recorded by:	2/5/07

Project No. _____

Book No. _____ TITLE Prepare 3 racks of 0.01% peptone

From Page No. ____

Objective: To prepare 3 racks of 0.01% peptone.

Procedures

1) 3 racks, the volume needed is (3×75×9.5)+300 = 2.5 L.
2) Weight 0.25 g of peptone into the 4 L. flask that marked the 2.5 L volume and contain some DI water (around 1.8 L)
3) After carefully pour the media, fill water to the volume of 2.5 L
4) heat mix
5) distribute to tubes by using automatic dispenser
6) Autoclave 121°C, 45 min
7) Leave on the bench to cool down.

what else was done this day?

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Witnessed & Understood by me,	Date	Invented by:	Date
Yanan Dabdoub	2/20/07	Juan Lara	3/5/07
		Recorded by:	

Project No. ______

TITLE Prepare 10 X 700 ml BHIA — Book No. ______

From Page No. ____

objective: To prepare 10 × 700 ml BHIA.

1) Mark the 1 L flask by using cylinder and DI water to mark the volume at 700 ml.
2) Discard some amount of water
3) Weight 36.4 g of BHIA
4) Carefully pour the agar into the flask
5) Fill the water into the volume marked.
6) Drop in magnetic bar and cover with foil. label ~~with~~ ZD on an autoclave tape.
7) Heat mix
8) Pour some amount into a basket to measure pH
9) Autoclave 45 min 121° C.
10) Put it in ~~an~~ ZD a waterbath (45°C) for 40 mins.
11) Mix 20 s.
12) Pour plate

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Witnessed & Understood by me,	Date	Invented by:	Date
~~2/21/07~~ ZD [signature]	2/21/07	[signature]	
		Recorded by:	

Project No. ______

Book No. ______ TITLE Prepare 8x 700 ml of BHIA

From Page No. ____

Objective: To prepare 8 ~ 700 ml BHIA

1) Use ~~4~~ 4D Cylinder to measure 700 ml of DI water
2) Mark each flask with 4D at the level of 700 ml. Discard some water
3) Weight 36.4g of BHIA (1L use 52 g)
4) Carefully pour the media in to the flask
5) ~~heat mix and place~~ cover with foil and heat Mix
6) Autoclave 121°C, 45 min
7) Cool in water bath (45°C) for 40 min.
8) Mix 20 sec.
9) Pour plate.

- Do house keeping for each lab.

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Witnessed & Understood by me,	Date	Invented by: / Recorded by:	Date
Yenom [signature]	2/22/07	Vireana Loeza	3/5/07

Prepare BHIA 7× 700 ml

TLE Prepare buffer phosphate buffer (PPB), 4 rack. Project No. _____ Book No. _____

m Page No. _____

Objective: To prepare 4 racks of buffer phosphate buffer.

1) Prepare ~~(3×9.5×75)+300~~ =4.25 L of PPB (4×9.5×75)+300 ≈ 4 L.
2) Bring out the phosphate buffer stock solution
3) Since 1.25 ml : 1 L need 5 ml
4) Use cylinder to measure ~~5~~ 4 L the DI-water and use it to emulsified 4 L of 6 L flask
5) Use small cylinder to measure 5 ml ~~30~~ and fill in the flask
6) Heat mix
7) Distribute to each tube by using dispenser.
8) Autoclave 121°C, 45 min.
9) Cool on bench.

II Prepare BHIA 7 × 100 ml

1) Mark the point of 100 ml by using cylinder and DI water
2) Pour out some amount of water.
3) Weight 86.4 g of BHIA and carefully pour into the flask
4) Fill the water to 100 ml
5) Drop magnetic bar and cover with ~~flask~~ foil.
6) Label on an autoclave tape a batch letter.
7) Heat mix
8) Autoclave 121° C, 45 min
9) Cool in water bath (48°C) 40 min
10) Pour plate.

- Do housekeeping for lab 314, 314, 305

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nessed & Understood by me,	Date 2/27/07	Invented by: Viviana Lozano	Date 3/5/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE Prepare 2x 700 TSA, 2x 700 MOX, 4 racks of BPW.

From Page No. ____

Objective: ① Prepare 2x 700 ml TSA
② Prepare 2x 700 ml MOX
③ Prepare 4 racks 9.5 ml of Buffer peptone water.

Procedures.

I Prepare 2x 700 ml TSA.

1) Use cylinder to weight DI water 700 ml and pour the water into the flask.
2) Mark the glass where the wate lies.
3) Discard some water.
4) Use weight boat to weight 28 g of TSA media (1 L use 40
0.7 L use 28)
5) Carefully pour the media into the flask.
6) Pour DI water to the marked line.
7) Drop a stir bar and cover with foil
8) heat # and throughly mix
9) Pour some media into pH for pH measurement.
10) Sterilize by autoclave at 121°C, 45 min
11) Cool down in waterbath (48°C) for 40 mins.
12) Pour plate

II Prepare 2x 700 ml MOX (oxford medium base)

1) Use cylinder to weight DI water 700 ml and pour the water into the flask
2) Mark the water line and discard some water
3) Weight 40.29 g of media on a weight boat (57.5 g/1 L , 0.7 L → 40.25)
4) Drop in a magnetic bar, ~~and~~ cover with foil, heat mix
5) Sterilize by autoclaving at 121°C, 45 min.
6) Cool down in water bath (48°C)
7) Prepare modified oxford medium supplement by rehydrate the supplement by adding 10 ml of DI water.
8) Add supplement to the 700 media after the media cool down (10 ml/L → 7 ml in
9) Pour plate
10) Leave to cool down on the bench.

IV. Prepare 4 racks of Buffered peptone water (9 ml)

1) Calculate the amount of media (9.5 x 4 x 75) + 300 = 3.2 L.
2) Mared 3.2 L ~~mark~~ line by using cylinder and DI water
3) Weight 64 g of BPW (20 g/1 L of water)

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Witnessed & Understood by me,	Date	Invented by:	Date
	3/7/07		3/26/07
		Recorded by:	

Project No. _____

LE _____ Book No. _____

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4.) heat mix, 5.D.

5.) Distribute into tubes by using dispenser.

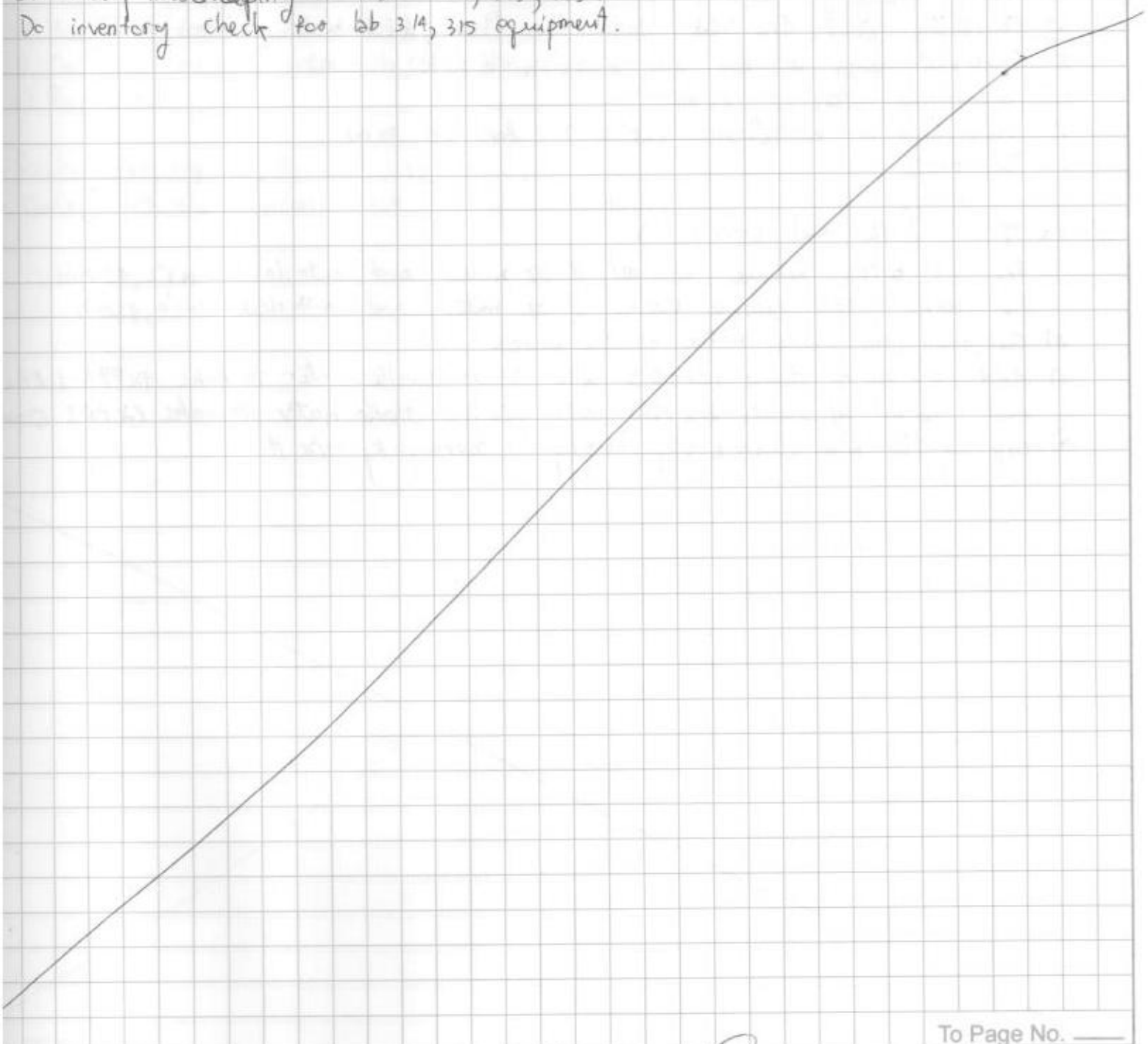
6) Autoclave 121°C, 45 min.

7) Leave on a bench to cool down.

Do daily housekeeping in lab ~~315~~ 4.D 315

Do weekly housekeeping in lab 314, 315, 305

Do inventory check for lab 314, 315 equipment.



To Page No. _____

essed & Understood by me,	Date 3/7 07	Invented by:	Date 5/26/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE Prepare 9x700ml TSA, 8x1LPBS, 5x500ml 1X PBS

From Page No. ____

Objective: To prepare 9x 700 ml of TSA. (3.148 & A-I)
To prepare 5x 1L of 1x PBS and 5x 500ml of 1XPBS

I) Prepare 9x700 ml of TSA

1) Use cylinder to measure 700 ml of DI water and pour it into 1L flask.
2) Mark the line of water and pour out some water
3) Weight 28 g of media using weighboat and balance. (40 g/L)
4) Carefully pour the TSA into the flask, Add water to 700 ml.
5) Heat mix using stir bar, also cover with foil.
6) Autoclave 121°C, 45 min.
7) Cool down in waterbath (48°C) for 40 min.
8) Pour plate

II 1x PBS. (10 to 1L and 500 ml)

1) for 1L 1X PBS measure 900 ml of DI water and autoclave 121°C, 45 min
for 500ml 1X PBS measure 450 ml of DI water and autoclave 121°C, 45min.
2) Cool down the sterile water on the bench.
3) Add 100 ml of sterile 10x PBS into 900 ml sterile water to make 1x PBS 1 litre
Add 50 ml of sterile 10x PBS into 450 ml sterile water to make 1X PBS 500
4) cap tightly and shake vigorously to thoroughly mix it.

To Page No. _

Witnessed & Understood by me	Date	Invented by:	Date
	3/7/07	Recorded by:	3/7/07

Project No. _____

LE Prepare 9 x 700ml of XLD. Book No. _____

Page No. ____

Objective: Prepare 9 x 700 ml of XLD. (3/6/07)

Procedure

1) Use cylinder to measure 700 ml of DI water and pour it into 1L flask
2) ~~Dissolve~~ Mark the line of the water and discard some amount of water
3) Weight 38.5 g of XLD agar (55g/L) and carefully pour it into the flask
4) Pour DI water to ~~the~~ XLD 700 ml
5) Drop stir bar and cover with foil, label
6) heat mix until media boils.
7) Cool down on the water bath (50°C) 40mins.
8) Pour plate

- Do housekeeping for the lab 315.
- Continue with the inventory check of the equipment

To Page No. ____

Read & Understood by me,	Date	Invented by:	Date
[signature]	3/12/07	[signature]	3/24/07
		Recorded by:	

Project No. _____
Book No. _____ TITLE Prepare 1×700 TSA, 1×700 MOX, 2×9 ml×2 racks BPW (3

From Page No. ____

Objective. To prepare 1×700 ml TSA, 1×700 ml MOX, 2×9 ml×2 racks of Buffer peptone wate

Procedures.

I Prepare 1×700 ml of TSA
1) Use cylinder to measure 700 ml DI water and mark the water line
2) Discard some water
3) Weight 28 g of TSA and pour into the flask (1 L → 40g)
4) Add water to the marked line 0.7 L → 28 g
5) Heat mix using stir bar while covering with foil.
6) Autoclave, liq. 121°C 45 mins
7) Cool down in water bath (50°C) for 40 mins.
8) Pour plate using aseptic technique

II. Prepare 1×700 ml MOX.
1) Mark the flask at the volume of 700 ml using DI water and cylinder. to measure the amount of water
2) Discard some water
3) Weight 40.25 g of oxford medium base and pour it into a flask (57.5 g / 1L, 0.7 L → 40.27 g)
4) Add DI water to 700 ml
5) Autoclave, liq 121C 45 mins
6) Cool down in water bath for 40 mins (50°C)
7) Add 7 ml of modified oxford medium supplement
8) Mix 20 sec.
9) Pour plate

III Prepare 2 racks of BPW 9ml.
1) Calculate the amount of media needed to prepare (9.5 × 75 × 2) + 300 = 1.5 L
2) Use cylinder to measure 1.5 L DI water, mark the line.
3) Discard some water
4) Weight 30 g of BPW media (20 g/L) ∴ 1.5 L you need 30 g
5) Drop in magnetic bar and heat mix while cover the flask with foil
6) Cool down on bench
7) Distribute into tubes. using dispenser.
8) Autoclave liq 121°C, 45 mins.
9) Cool down on bench.
- Collect the hazard disposal.

To Page No. ____

Witnessed & Understood by me,	Date	Invented by:	Date
	3/12/09	Recorded by:	5/24/09

Project No. _____

Book No. _____

Prepare 15×700ml of TSA

Title : To prepare ~~15 flask~~ 15×700 ml of TSA

Procedures

Use Cylinder to measure 700 ml of DI-water, Use marker to mark the line. Discard some water

Weight 28 g of TSA media (40g/L)

Add water to 700 ml

Heat mix

Autoclave 121°C, 45 mins.

Cool down in a water bath (50°C) 40 mins.

Pour plate

Do Housekeeping for labs 314, 315, 305.

Collects kills

To Page No. ____

ed & Understood by me,	Date	Invented by:	Date
	3/12/07		3/24/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE Prepare 7×200ml of XLD

From Page No. ____

Objective: To prepare 7×700 ml of XLD.

Procedure:

1. Use cylinder to measure 700 ml of DI water. Use label to mark the line
2) Discard some amount of water
3) Weight 38.5 g of XLD media and carefully pour into the flask. (55g/L)
4) Turn on the water bath.
5) Heat mix, ~~stir~~ until the media boils
6) Cool down in a water bath. 40 mins (50°C)
7) Pour plate

- Do the housekeeping for labs 314, 315, 305.
- Do kill run.

To Page No. ____

Witnessed & Understood by me,	Date	Invented by:	Date
jaman Dang	3/13/07	Recorded by:	3/26/07

Project No. _____

TITLE Prepare 2×700 ml TSA, Prepare 2×700 ml MOX Book No. _____

From Page No. ____

Objective: To prepare 2×700 ml TSA and 2×700 MOX

Procedure. I) 2×700 ml TSA

1. Use cylinder to measure 700 ml of DI water.
2. Marke the water line.
3. Pour out some water.
4. Weight 28 g of TSA media (40 g/L) using weight boat.
5. Carefully pour the media into the flask, Add DI water to 700 ml
6. Drop stir bar and Caver with foil.
7. Heat mix
8. Autoclave, Liq 45 min.
9. Cool down in a water bath 40 mins (50°C)
7. Pour plate

II 2×700 MOX

1) Use cylinder to measure 700 ml of DI water, use marker to mark the volume.
2) ~~X~~ 40. Discard some water
3) Weight 40.25 of media on a weight boat (57.5 g/L)
4) Add DI water to 700 ml
5) Autoclave, Liq 121°C 45 mins.
6) Cool down in water bath for 40 mins (50°C)
7) Add 7 ml of modified oxford medium sopplement in to the ~~agar~~ 40 flask.
8) Mix for 20 sec.
9) Pour plate

- Do the housekeeping for lab 314, 315, 305
- Collect hazard bells.

To Page No. ____

Witnessed & Understood by me,	Date 3/28/07	Invented by: Viviana Lezza	Date 4/5/07
		Recorded by:	

Project No. _____

Book No. _____ TITLE To Prepare 4 racks of BPW.

From Page No. ____

Objective: To prepare 9ml of buffer peptone water

1. Calculate the ~~media~~ amount of media that is needed to prepare ~~(9×7.5~~ 9.5×75×4) +300 ml.
2. Use Cylinder to measure 3.2 L water and mark where the water lies.
3. Discard some amount of water.
4. Using weighboat to ~~me~~ weight 64 g of BPW (20 g/L)
5. Carefully pour the media into the flask
6. Add DI water to 3.2 L.
7. heat mix until the media dissolve. - broths don't need to be heated
8. Distribute into tubes using dispenser.
9. Autoclave 121°C 45 min.
10. Cool down on bench.

- Do housekeeping room 3/4
- Do kill run.
- Do inventory check of the expired media.

To Page No. ____

Witnessed & Understood by me	Date	Invented by:	Date
	3/28/07	Recorded by:	4/5/07

TITLE To prepare 15× 900 ml TSA, Prepare 3 racks of 10% sodium Thiosulfate   Project No. ___ Book No. ___

From Page No. ___

Objective 2) To prepare 15 × 900 ml TSA
1) Use cylinder to measure 900 ml DI water, use marker to mark the volume
2) Discard some water
3) Weight 28 g. of TSA media (40 g/L)
4) carefully pour the media in to the flask
5) Add DI water to 900 ml
6) Autoclave 121°C 45 min.
7) Cool in 50°C water bath. for 45 min.
8) Pour plate.
9) Cool down on bench.

II Prepare 10% sodium thiosulfate 3 racks. (9 ml)

1) ~~Use cy~~ calculate the amount of media needed. (3 × 7.5 × 9.5) + 300 = 2.5 L.
2) Use cylinder to measure the volume and mark the flask.
3) Weight 250 grams of sodium thiosulfate (100 g/L) and add it into flask.
4) ~~heat~~ Mix thoroughly.
5) Distribute into tubes using dispenser.
6) Autoclave 121°C 45 min
7) Cool down on bench.
~~8)~~ Do housekeeping in lab 315

To Page No. ___

Witnessed & Understood by me,	Date	Invented by:	Date
[signature]	4/5/07	[signature] Recorded by:	4/5/07

Project No. _____

Book No. _____ TITLE ~~Report~~ , 3 weeks of BPN.

From Page No. ____

- 3/28/07 :- The media is preparod but the pressure of the autoclave has not down to zero.
- Do the housekeeping for room 314, 315, 505

Should have informed me of the situation

Witnessed & Understood by me,	Date	Invented by:	Date
	4/5/07	Recorded by:	4/5/07

03-30-07 AM0:27 IN

Project No. _____

TITLE Prepare 10×TSA (700 ml) Book No. _____

From Page No. ____

Objective : To prepare 10 × 700 ml TSA

Procedures.

1) Use cylinder to measure 700 ml of DI water, pour into flask, and mark the vol
2) Discard some water.
3) Weight 28 g of TSA into the flask (40g/L)
4) Add water to 700 ml
5) Heat mix. while the flask is cover with foil.
6) Autoclave liq 45 121°C.
7) Cool down in 50°C water bath for 45 mins.
8) Pour plate.

- Do monthly housekeeping for lab 314
- Do daily housekeeping for lab 315 & 310

To Page No. ____

Witnessed & Understood by me,	Date	Invented by:	Date
	4/5/07	Recorded by:	9/5/07

04-04-07 A09:08 IN

Project No. _____

Book No. _____ TITLE Prepare 10×700 ml TSA.

From Page No. ____

Objective : To prepare 10 x 700 ml TSA.

1) Mark the volume of 700 ml of the flask using DI water and cylinder.
2) Discard some amount of water.
3) Weight 28 g of TSA and pour into the flask.
4) Add water to 700 ml
5) Drop stir bar into the flask and cover with foil.
6) heat mix
7) Autoclave 121°C, 45 min
8) Cooldown in a 50°C waterbath, 45 min.
9) Pour plate.
- Do hazard disposal.
- Do housekeeping for Job 314, 315, 305

04-04-07 A09:08 IN

04-04-07 P04:17 IN

To Page No. _____

Witnessed & Understood by me,	Date	Invented by:	Date
	04/05/07	Recorded by:	4/5/07