BME Design-Fall 2020 - ADITYA AILIANI
Complete Notebook

PDF Version generated by

Cate FLYNN

on

Dec 09, 2020 @12:39 PM CST

Table of Contents

Project Information 2
- Team contact Information 2
- Project description 3

Team activities 4
- Client Meetings 4
  - 09/10/2020 - Client Meeting #1 4
  - 09/22/2020 - In Person Inspection 5
  - 09/23/2020 Client Meeting #2 6
  - 09/25/2020 Lead and Catheter Pickup 7
  - 10/7/2020 Client Meeting #3 8
  - 10/21/2020 Client Meeting #4 9
  - 11/4/2020 Client Meeting #5 10
  - 11/18/20 Client Meeting #6 11
  - 12/2/2020 Client Meeting #7 12
- Advisor Meetings 13
  - 09/11/2020 Advisor Meeting 13
  - 09/18/2020 Advisor Meeting 14
  - 09/25/2020 Advisor Meeting 15
  - 10/16/2020 Advisor Meeting 16
  - 10/23/2020 Advisor Meeting 17
  - 10/30/2020 Advisor Meeting 18
  - 11/6/2020 Advisor Meeting 19
  - 11/20/2020 Advisor Meeting 20
- Materials and Expenses 21
  - Project Expenditures Sheet 21
- Fabrication 22
  - Prototype Fabrication 22
  - Bubble Trap Fabrication 24
- Testing and Results 25
  - Protocols 25
    - Comparison of tubing connectors 25
    - Bubble trap effectiveness 27
  - Experimentation 28
    - Prototype Use with Machine 28
- Project Files 30
  - 20201005 - Terumo tubing set diagram, dimensions 30
- Team Meetings 32
  - 20200904 - Introductory Meeting 32
  - 09/07/2020-Questions for Client Meeting 33
  - 09/14/2020 - PDS Team Meeting 34
  - 09/21/2020 - Design Matrix Team Meeting 35
  - 09/28/2020 - Preliminary Presentation Meeting I 37
  - 10/1/2020 - Preliminary Presentation Meeting II 39
  - 10/2/2020 - Final Report Meeting 40
  - 11/16/2020 - Testing, Fabrication Discussion 41
  - 20201130 - Poster Planning Meeting 42

ADITYA AILIANI 44
Research Notes 44
Biology and Physiology 44
20200907 - Initial Research 44
20200914 - Client's PRP therapy 47
20201013 - Extracorporeal blood flow 48
20201007 - Introduction/Motivation Research 50
20201127 - Platelet, WEPLEX Physiology (Report Edits) 52
Competing Designs 54
20200914 - COBE Disposable tubing 54
20200916 - PDS - Standards and Specifications 56
20200930 - Tubing Set Patents 58
20201005 - ELP Tubing Set company specifications 60
20201006 - Testing Research 63
20201009 - Reusable Tubing Sets 65
20201026 - COBE Spectra associated patents 66
Design Ideas 68
20200910 - Client Meeting 1 68
20201001 - Max blood processing volume calculations 69
20201110 - Pressure Sensor Research/Ideas 71
TREVOR SILBER 73
Research Notes 73
Biology and Physiology 73
Project Description 73
Platelet Lysate 74
Platelets and Wound Healing 75
WEPLEX 76
COBE Spectra Apheresis Operator’s Manual 77
How to make adhesives work 79
Tubing Research 80
Competing Designs 81
Transfusion Medicine Apheresis Plateletpheresis 81
COBE Spectra Apheresis System 82
Design Ideas 83
Tubing Prices and Comparisions 83
Tubing Diagram 84
Tubing Pressure Sensor 85
Tubing Fabrication Idea 86
Other Information 87
Client Meeting 87
Project Selection Link 88
Client Meeting 2 89
PACT 90
Cole-Parmer Email 91
CATE FLYNN 93
Research Notes 93
Biology and Physiology 93
09/14/2020 Platelet Extract as an Anti-Inflammatory 93
09/17/2020 Existing Tubing Conditions 94
Design Ideas 95
09/22/2020 WIMR Visit #1 95
10/02/2020 Autoclave Impact on Silicone Stability 96
10/02/2020 Use of Ethylene Oxide for Sterilization 97
10/18/2020 Autoclave Sterilization Methods 98
10/19/2020 Hybrid Sterilization Technique 99
10/29/2020 WIMR Visit #2 100
11/09/2020 Selecting Tubing Material 102
11/09/2020 WIMR Visit #3 103
11/19/2020 WIMR Visit #4 104
11/20/2020 Testing Brainstorming 105
11/23/2020 Fabrication of Prototype 106

11/23/2020 Unforeseen Inner Diameter Issue 109
11/24/2020 WIMR Visit #5 110
11/30/2020 Large Connectors vs. Small Connectors Testing Ideas 113
12/03/2020 Connector Comparison Test Results 114
LOKESH KUMARAVEL 117
Research Notes 117
Biology and Physiology 117
20200906 Initial Research 117
20200908 Platelet Lysate 119
20201015 Real World Applications 121
20201017 Safety and Standards 122
Design Ideas 123
20200922 Initial Impression of Apheresis Machine 123
20200930 Hands on Investigation of Current Tubing 125
20201004 Materials Research 129
20201011 Material Cost Research 132
20201019 Microfluidic Tubing Research 134
20201024 Hardness of Tubing Research 137
20201029 Initial Loading of Device 138
20201109 Running Fluids Through the Tubing Set 140
20201119 Measurements of the Tubing Segments 142
20201124 Testing of Prototype 143
20201202 Fabricating the Bubble Trap 144
Training Documentation 146
Initial Startup of the COBE Apheresis Machine 146
NESYA GRAUPE 147
Research Notes 147
Biology and Physiology 147
09/06/20 Research Notes 147
09/14/2020 Research Notes 149
09/23/2020 Research Notes 150
10/04/20 Research Notes 151
10/16/20 Research Notes 152
10/19/20 Research Notes 154
11/05/2020 Research Notes 155
Design Ideas 157
10/4/2020 Design Notes 157
10/30/2020 Call to Action 160
11/10/2020 Bubble trap 162
11/17/2020 Bubble trap 163
2014/11/03-Entry guidelines 164
2014/11/03-Template 165

Team contact Information

LOKESH KUMARAVEL - Oct 05, 2020, 11:24 PM CDT

Last Name	First Name	Role	E-mail	Phone	Office Room/Building
Kinney	Melissa	Advisor	melissa.kinney@wisc.edu		
Brounts	Sabrina	Client	sabrina.brounts@wisc.edu	(608) 263-7600	
Galipeau	Jacques	Client (works w/ Prof. Brounts)	jgalipeau@wisc.edu		
Pennati	Andrea	Alternate Contact (hands-on guidance with device)			
Ailiani	Aditya	Leader	aailiani@wisc.edu	608-799-4012	
Flynn	Cate	Communicator	clflynn2@wisc.edu	651-583-4527	
Graupe	Nesya	BSAC	ngraupe@wisc.edu	414-828-1899	
Silber	Trevor	BWIG	tjsilber@wisc.edu	262-744-1426	
Lokesh	Kumaravel	BPAG	lkumaravel@wisc.edu	715-820-2884	

Project description

ADITYA AILIANI - Dec 09, 2020, 10:56 AM CST

Course Number: BME 200/300

Project Name: VETMED: CONVERSION OF HUMAN COBE PLATELETPHERESIS MACHINE FOR LARGE ANIMAL USE

Short Name: Plateletpheresis

Project description/problem statement:

Plateletpheresis uses centrifugal technology to separate platelets from other donor blood components, returning other components to the donor. Our clients, Professor Sabrina Brounts, Dr. Jacques Galipeau, and Dr. Andrea Pennati, have developed a new equine platelet therapy and would like to use plateletpheresis rather than whole blood extraction to extract platelets for further research [1]. The COBE© Spectra Apheresis System they are using costs $2000 - $2600 per use, mainly due to the expensive tubing sets that can't be reused. Our goal is to develop a system to decrease cost per use, allowing the client to reuse tubing sets or use cheaper tubing.

About the client:

Dr. Brounts is a veterinary sports medicine specialist at the UW Department of Veterinary Medicine, Dr. Galipeau runs the Program for Advanced Cell Therapy at the Wisconsin Institute of Medical Research, and Dr. Pennati is the associate director of research and development at PACT.

09/10/2020 - Client Meeting #1

Cate FLYNN - Sep 14, 2020, 12:55 PM CDT

Title: Client Meeting #1 Notes

Date: 9/10/2020

Content by: Cate Flynn

Present: Aditya, Lokesh, Nesya, Trevor & Cate

Goals: To ask the client questions to inform the next steps of the process

Content:

Link to the Teams meeting recording: https://web.microsoftstream.com/video/9e6cc71c-7129-408e-8465-21b765d7e87e

In this meeting we learned more about our client's priorities and expectations as well as more information about the current tubing and the problem statement. The budget for this project is $500 and, depending on if we pursue sterilization or produce a new tubing system ourselves, the client said that $100 to create tubing with 15-20 uses would be ideal. We also discussed going in person later next week to look at the machine and the current tubing. Depending on the cost of production, single use could be a viable option. The client was also very enthusiastic about contracting a third party to produce the tubing as there are concerns that the current supplier would phase out the tubing for the machine the client has as there is a newer version of the machine. The client discussed gas and steam as the two main sterilization techniques. The client also prefers bi-weekly meetings.

Conclusions/action items:

Send out the progress report to the client and advisor and do independent research into materials and sterilization techniques. We also need to begin work on our PDS.

09/22/2020 - In Person Inspection

Cate FLYNN - Oct 04, 2020, 10:51 PM CDT

Title: Meeting with Dr. Pennati at Wisconsin Institutes for Medical Research (WIMR) to see COBE machine

Date: 09/22/2020

Content by: Cate Flynn

Present: Cate and Lokesh

Goals: To inspect the COBE machine to inform the design process

Content:

Lokesh and I met with Dr. Pennati at WIMR to look over COBE machine so that we could understand the tubing and the machine better. The front panel that the tubing attaches to is more complicated than we had initially thought, but the meeting was informative. This meeting also prompted the question of accessibility for the machine. WIMR is technically closed to undergrads due to COVID conditions and may become so later on in the semester as well. If we want consistent access, it may be a good course of action to move the machine to a space where our advisor, Dr. Kinney, can always access it. I took a picture of the front panel for reference later on.

Conclusions/action items:

Speak with advisor about moving machine, continue to work on tubing replication.

09/23/2020 Client Meeting #2

ADITYA AILIANI - Sep 23, 2020, 5:28 PM CDT

Title: Client Meeting #2

Date: 09/23/2020

Content by: Nesya Graupe

Present: All

Goals: Meet with the client, better understand the problem

Content:

-14 gauge needles available at vet school, Dr. Brounts will look into 10 or 12 gauge needles because they are bigger and can take more blood

-Procedure should last for an hour (that is about how long horses can be sedated for)

-See if machine can cope with large volume of blood

-It is alright if we simplify the tubing

-If we use the autoclave the material will have to withstand a temperature of 120C. Gas sterilization is better because there won't be condensation but it is more expensive and trickier. If we create cheap tubing we won't have to worry about reusing/sterilizing it

-Machine should stay in Vet building now. Can possibly move it in future if needed.

-Horses have 50-60L of blood

-We can use large sinks in research lab for testing

-Try to find a substitute for blood to test

-Identify materials we need to order

Conclusions/action items: Make power point and Gant chart for next meeting. We will each take turns leading a meeting. Discuss our limitations and reasonable goals with our advisor - our previous understanding was that a sterilization system or a new tubing set or a redesigned set of parameters was the goal, but our clients would prefer all three.

09/25/2020 Lead and Catheter Pickup

Cate FLYNN - Oct 07, 2020, 9:44 AM CDT

Title: Lead and Catheter Pickup

Date: 09/25/2020

Content by: Cate Flynn

Present: Cate Flynn, Dr. Brounts

Goals: To pick up the donated leads and catheters from the client, Dr. Brounts

Content:

The client has donated 10, 12 and 14 gauge leads and catheters. The contents donated are as follows:

2 x 10 Ga Catheter
2 x 12 Ga Catheter
5 x 14 Ga Leads

Conclusions/action items:

These items are to be used to inform the flow rate calculations that will take place later in the design process.

10/7/2020 Client Meeting #3

ADITYA AILIANI - Dec 09, 2020, 10:22 AM CST

Title: Client Meeting

Date: 10/7/2020

Content by: Aditya

Present: Dr.Brounts, Dr. Galipeau, Dr. Pennati, Trevor, Lokesh, Cate, Nesya

Goals: To establish our goals for the semester, run through our preliminary design presentation with them to make sure we are all on the same page

Content:

We described our current research into the project:

Materials:

We informed the clients that our material of choice would depend on the sterilization method - for example, silicone tubing is more expensive but responds best to autoclave sterilization, while PVC is less expensive and can be gas sterilized but usually not autoclaved. The clients said they would prefer whichever overall method is cheapest - gas sterilization is more expensive that autoclaving, but they could make it work if PVC is significantly cheaper. We also described some new dimensions for the tubing set, increasing the diameter to 0.133 inches to almost double blood flow. However, the clients were thinking more along the lines of processing the entire blood volume of the horse in one hour, resulting in a flow rate of 50-60L / hour. They recommended going back to the calculations and scaling up the tubing radius as much as possible to get the maximum possible flow rate, not just the flow rate that had been used in equine studies previously.

Simplifications:

The clients were unaware that the tubing was so complicated and included so many hard plastic pieces that were needed for safety. They agreed with our assessment that the plasma collection bag and leukoreduction chamber could be removed (losing the LRS chamber would require an extra processing step after to remove white blood cells). We described how Trevor had been talking to Cole Parmer about a custom tubing set so we could vary the radius as much as we wanted, but the fact that the minimum order was for 10,000 feet was unfortunate. The clients said they would be fine with us working within the constraints of cheaper, non-custom bulk tubing.

Conclusions/action items:

We should conduct some more research into tubing costs before sending them materials to order, but we are close to having a conclusion about materials. The more pressing issue is deciding how to put all the tubing pieces together to make sure we have the safety functions (pressure sensors, air detectors). We will also need to redo flow rate calculations to maximize flow rate.

10/21/2020 Client Meeting #4

ADITYA AILIANI - Dec 09, 2020, 10:41 AM CST

Title: Client Meeting

Date: 10/21/2020

Content by: Aditya

Present: Dr. Brounts, Dr. Galipeau, Dr. Pennati, Cate, Nesya, Lokesh, Trevor

Goals: To review our testing plan with the clients and see if they have any concerns

Content:

Our current plan is to make cheap tubing sets to test - we will have to cut the centrifuge loop from our original tubing set to make this work because this component is too difficult to manufacture. We showed the client an image from the centrifuge loop's patent and explained that, short of casting our own tubing, we weren't sure how to vary the tubing diameter and included a dam on the inside of the tubing. The clients agreed that the centrifuge loop could be a project for next semester. They did request that we include air sensors in the tubing set, but could maybe get away with tricking the pressure sensors because they have ways of clinically measuring horse blood pressure outside of the device.

They decided that testing three different materials (Nylon, PVC w/o phthalates, and silicone) would be cheap enough. We also decided to set up a time to go in to test the tubing set with saline.

Conclusions/action items:

We had to swap silicone out for a fluoropolymer because the silicone tubing ended up being the wrong diameter when we went to confirm our order. Other than that, the next steps are to collect measurements of the original tubing set and order materials, including autoclavable/gas sterilizable connectors.

11/4/2020 Client Meeting #5

ADITYA AILIANI - Dec 09, 2020, 10:48 AM CST

Title: Client Meeting

Date: 11/4/2020

Content by: Aditya

Present: Dr. Brounts, Dr. Galipeau, Dr. Pennati, Trevor, Nesya, Cate, Lokesh

Goals: To update our client on progress and set up a time to test the tubing

Content:

We had to let our clients know that we hadn't found a feasible way to get past the sensors - Cate and Lokesh had seen that the device was really picky about having the pressure and air slots filled in the front panel. We described out plan to manufacture a small, cheap bubble trap based on microfluidics and described that the only material we would need aside from 3D printed plastic from the Makerspace was a PTFE membrane. The clients approved this design once we showed that the membrane could be reusable if it was washed in ethanol.

We mainly described what we would need for the next testing session, including a saline bag to load the tubing set, the proper centrifuge cap for the ELP tubing set, and maybe an operator's manual of the device. I had called Terumo and they said that they do not distribute operator's manuals or technical information about the COBE Spectra anymore. The clients asked that we send this information in an email to them so that they could try to email the original owner of the device and see where some of those additional components could have gone. They said that if they didn't get an affirmative answer by next week, they were ok with us purchasing the cap off EBay.

Conclusions/action items:

We'll need to send the clients all the information we would like to have for testing next week. In addition, we should decide how we are going to use the two saline bags Dr. Pennati might have, since the original tubing set has 3 fluid inlets aside from whole blood and each bag can only be spiked once.

11/18/20 Client Meeting #6

ADITYA AILIANI - Dec 09, 2020, 10:53 AM CST

Title: Client Meeting 7

Date: 11/18/20

Content by: Aditya

Present: Dr. Galipeau, Dr. Pennati, Nesya, Lokesh, Cate, Trevor, Aditya

Goals: To plan dates for testing and sterilization

Content:

We first talked to the client about the centrifuge cap,and they told us to order it off EBay. The original device owner was not getting back to them. We mainly just went over our testing plan to compare fluid carrying in the COBE Spectra between the original tubing set and our manufactured one. Our main goal was to schedule a time to test the device with Dr. Pennati around so he could make sure we were doing things correctly. He agreed to bring saline and observe the testing.

Conclusions/action items:

This meeting was brief, and the main goal was to set up testing. We couldn't get a sterilization testing date because Dr. Brounts is in charge of the gas sterilizer, so we will have to email her about if it would be possible for us to test material properties (bend radius) before and after gas sterilization of our tubing set.

12/2/2020 Client Meeting #7

ADITYA AILIANI - Dec 02, 2020, 6:59 PM CST

Title: Last Client Meeting

Date: 12/2/2020

Content by: Aditya

Present: Dr. Brounts, Dr. Pennati, Nesya, Lokesh, Cate, Trevor, Aditya

Goals: To describe our final testing results and future work with the clients

Content:

The centrifuge cap that goes with the platelet collection tubing set couldn't be delivered - the sellers did not have it and ended up reimbursing our clients (this can be taken out of our expenses report but should still be described so the next team knows what they'll need).

The clients approved of the testing Cate and Lokesh had done to get a partially functional tubing set into the machine without leaks, with some components clamped off. We described that some of the tubing briefly shifted out of place in the peristaltic pumps, and they agreed that this could be improved using glue.

We described the different types of connectors (large and small) we were planning to test for reliability. Based on Cate's observations so far (had to heat the tubing to get the connectors to stay), Dr. Brounts predicted that the large straight connectors would be more reliable and form a tighter seal.

Dr. Brounts also requested a per-use cost estimate based on our design so far. This would include the cost of tubing proportional to the length we used, the connectors, collection bags, needles, and fabricated bubble traps. She also acknowledged that the cost of the centrifuge loop and pressure sensors that we cut from the device would be difficult to estimate, and we could make a note of this in our final report. She also said that cost of sterilization at this point was not going to make or break anything, so we could just focus on material costs.

In the team's meeting after the clients left, we decided to include an additional section in the poster for per-use expenses, since that was an important part of the client's request. In terms of diagrams, we wanted to include a photo of the fabricated tubing set, a photo or diagram of the original tubing set (depending on available poster space and legibility), a CAD drawing of the bubble trap design, and graphs from the connector leak testing. Finally, Cate noted that parts of the tubing have extremely narrow inner diameters (around the size of a mechanical pencil lead) and connected to adapters to match the rest of the tubing set. The most notable examples were connected to the centrifuge loop (possibly the whole blood line) and the inlet manifold (not sure which fluid line yet). These sections were incorporated into the fabricated tubing set by making connections upstream of the adapters, but we weren't sure why the diameter had to decrease so drastically.

Conclusions/action items:

The decrease in inner diameter is an interesting find that we can look into (maybe the centrifuge loop patent would have a clue?), but the poster is our first priority. After putting together our images and text, we will go over it a final time and record in Zoom. The recording and poster pdf will be sent to the client, so they can see the cost estimate soon.

09/11/2020 Advisor Meeting

NESYA GRAUPE - Sep 11, 2020, 2:00 PM CDT

Title: Advisor Meeting

Date: 09/11/2020

Content by: Nesya Graupe

Present: All

Goals: Meet with our advisor, review the project

Content:

-Introduced ourselves to Dr. Kinney

-Talked over the project and possible methods of action if campus shuts down

-Dr. Kinney recommended that we conduct very thorough research on our designs at the beginning of the semester so that there will be no need for last minute changes at the end of the semester.

-Dr. Kinney requested that our research notes have sources sited properly (instead or website links) and notes in our own words (instead of excerpts from the internet) to help prepare for the final paper.

-Talked over possible need for biosafety or animal testing training if testing with horse blood our live horses

Conclusions/action items: Conduct individual research, work on project design specifications, meet as a group

09/18/2020 Advisor Meeting

ADITYA AILIANI - Sep 30, 2020, 11:52 AM CDT

Title: Advisor Meeting 2

Date: 9/18/20

Content by: Aditya

Present: Lokesh, Cate, Nesya, Trevor, Aditya

Goals: To review our PDS and establish goals for our design matrix

Content:

Before our advisor meeting, we received an email from Prof. Brounts saying that, if campus shut down, we could work on optimizing the COBE Spectra's parameters for horses as a virtual project. Prof. Kinney thought that it would be best to focus more on tubing, since we had done our research with that in mind and needed to commit to a design idea soon. For our design matrix, she suggested including several design routes, including 1) manufacturing cheaper tubing 2) reusable tubing/sterilization process 3) parameter adjusting. We could choose categories for our design matrix that weighed practicality more heavily. Even if we end up not being able to acquire materials, Prof Kinney said we could still use computer simulations or existing material parameters for autoclave/EtO processes to virtually test our ideas.

We were also told to think carefully about how to prove our design met the specifications. For example, we will need to identify the device's range of tolerance for tubing diameter and look for known sterilization parameters (how many cycles does tubing with X dimensions made of Y material need?). We may be able to use the BME Design Lab to test fluid flow through a model tubing set, although Prof Kinney said we may need to find a blood-like substance (not just water).

Questions for our client that came up from this meeting:

-Do we need to provide tubing alone or include other disposable parts (connectors, collection bags)?

-Does the tubing set require some kind of anticoagulant lining?

-To what extent could we change the device's parameters/controls?

Conclusions/action items:

We will meet on Monday to decide on our design matrix. Before then, we should do some research to help us decide what is feasible and look more carefully at the tubing setup outlined in the COBE Essentials Guide our client sent us. Cate is planning to set up a recurring meeting with our client, and she and Lokesh are thinking about times they can see the tubing in person (Trevor and Nesya may be able to come as well). Trevor is contacting TBL plastics to see what kind of tubing is available - Terumo only provides information to their clients, so Prof Brounts or Dr. Pennati may need to request specifications for us.

09/25/2020 Advisor Meeting

ADITYA AILIANI - Sep 30, 2020, 12:10 PM CDT

Title: Advisor Meeting 3

Date: 9/25/2020

Content by: Aditya

Present: Lokesh, Nesya, Trevor, Cate, Aditya

Goals: To reconsider our client's new expectations and decide how to manage upcoming tasks (new design ideas, preliminary presentation, report)

Content:

Professor Kinney addressed some concerns that came up earlier in the week:

-The 10 gauge needle that Prof. Brounts gave us has a 39 L/hr flow rate written on the packaging. What does this mean?

->Likely a maximum burst flow rate. We will still need to do flow rate calculations using our known parameters.

-Our client has requested new project management tasks: a Gantt chart and update slides for each biweekly meeting. Is this feasible, considering the project management we already have to do for BME 200/300?

->We can choose progress reports or slides, based on our client's requirements. A Gantt chart can similarly replace the project timeline at the bottom of our progress reports. We were advised to go through the class requirements for BME 200/300 to assure the client that we have project milestones to clear just as part of the class. In addition, we should try to accommodate the client's request to change the device's parameters, but remind them that the project can be continued in another semester. The scope of the project can be moved from semester to semester, so we can still focus on tubing and propose some changes to the device that another team can pick up.

-What assumptions can we make for blood flow calculations?

->Because the device uses peristaltic pumps, laminar flow can be assumed. There are likely some adjustments to the classic flow through a tube equation made for peristaltic pumps, so we were advised to look for literature on the subject.

-We have some concerns with accessing the device, as we would have to go to the medical school or veterinary medicine school to test tubing on the COBE Spectra. Our clients were worried about moving the device out of these departments in case the university shut down.

-> We were advised to move the device into the BME Design Lab. We should submit a request to Dr. Puccinelli for storage space in ECB. Even if campus gets shut down again, professors will still have access, so the BME Department should be able to arrange to return the device to our clients.

In addition, our preliminary presentation is next Friday. If we get our slides to Professor Kinney by Wednesday, she may be able to give us feedback before then. The presentation should be recorded and submitted by 10am on Friday. Trevor said a useful tubing system manufacturer may be Medical Instrumentation for Animals (MILA).

Conclusions/action items:

We should focus on delivering a tubing set in the coming weeks, with sterilization and flow calculations as our next two concerns. Having the COBE Spectra on hand will be useful to make sure our tubing will fit (there should be information in the manual on a range of tubing diameters that will lock into the device's sensors).

-Use peristaltic pump corrected calculations to figure out how large our tubing should be

-Figure out how to make a Gantt chart and make biweekly update slides (this week's should be pretty much the same as our preliminary presentation)

-Request room in ECB

10/16/2020 Advisor Meeting

ADITYA AILIANI - Oct 16, 2020, 1:53 PM CDT

Title: Advisor Meeting

Date: 10/16/2020

Content by: Aditya

Present: Nesya, Lokesh, Trevor, Cate

Goals: To discuss difficulties from the past week

Content:

We were concerned about putting pieces of tubing together because the branched structure was complicated, but Prof Kinney said that we should be able to use Y connectors for most of the tubing connections and just use multiple segments. For testing, she recommended having a couple backup testing options in case the tubing didn't work in performance testing. These would include passing fluid through the tubing post-sterilization and seeing if bacterial growth occurred in the fluid. We also could look into testing for residual blood components. Some other ideas for showing our product meets specifications are looking through research protocols or autoclave manuals to see how far steam can permeate in a given material. Apparently, tubing is easy to autoclave because the walls are thin, and it is reused in research all the time.

She also recommended researching the components on the interior of the centrifuge tubing because those might not be reusable. If they are reusable, we could cut the centrifuge loop out of the original and connect it to our tubing set.

Those of us who are not on campus should take on more of the research and planning (she recommended evaluating our assumptions about materials and dimensions using math/chemistry) to make this less painful for people on campus.

Conclusions/action items:

We still might be able to deliver a functioning product for our client. Currently, our goals are to develop a fabrication plan using tubing of different sizes and cheap connectors and develop a testing plan. The testing plan should include performance testing, in which we may need to silence the ultrasonic air sensors and pressure sensors, and some kind of sterilization testing (could be substituted with relevant literature, but good to have).

-General tasks for next week:

-> Look into microfluidic tubing sterilization (Lokesh,)

-> Research components of centrifuge loop (Nesya,)

-> Research testing options (Aditya,)

-> Fabrication, testing plans (everyone at our next meeting)

10/23/2020 Advisor Meeting

ADITYA AILIANI - Oct 26, 2020, 12:17 PM CDT

Title: Advisor Meeting

Date: 10/23/2020

Content by: Aditya

Present: Nesya, Cate, Lokesh, Aditya

Goals: To establish goals for the upcoming week in fabrication, testing, and show and tell

Content:

Fabrication

At our client meeting, Prof. Brounts requested that we keep the air detectors in the tubing set (pressure sensors can be worked around). Prof. Kinney suggested 3D printing cylinders out of a compatible, autoclavable material. To do this, we can use a SolidWorks model of the air chamber cylinder that interfaces with the ultrasonic air detectors on the device. The Makerspace can cheaply 3D print this for us if we send it to them as a [file type?] file.

Terumo said they could send us 3 (!) non-sterile tubing sets, so if those arrive soon (within 2 weeks) we can cut them apart to have backup centrifuge loops and make it easier to measure specific segments. We also might be able to reverse-engineer the parts connecting to detectors.

Modelling

Prof. Kinney said we do not need a SolidWorks model of the whole tubing set - a digital drawing would be preferred over a hand-drawn one, but it depends how clearly we can show the important parts. She recommended trying the free trial of BioRender (can only make one picture) or editing Terumo's drawing.

Try to use computational fluid dynamics to predict maximum flow through the device based on tubing dimensions.

Testing

It would be good to get baseline results for flow rate through the tubing and even just to see how the tubing fits into the moving pumps on the COBE Spectra. Outside of physical testing, Prof. Kinney recommended digging into the specifications of the pumps used in the device to see how much they limit tubing dimensions.

Show and Tell

Show and Tell will be a week-long Piazza event. We will post a picture of our design (or a photo of the device+tubing depending on when Andrea is available), a brief description, and a call to action statement requesting specific feedback, and teams will respond with feedback over the following week. We will have to post information by Friday (10/30) at midnight, so we can discuss our call to action statement in next week's advisor meeting (should have a draft on our progress report for next week).

Conclusions/action items:

We are planning to split up research again this week, with less of it going to Cate and Lokesh because they will be testing the device soon. Cate is emailing Andrea to see when he is available to supervise testing at WIMR (ideally late next week). On Monday, we will discuss our testing plan and what measurements we need to complete the fabrication plan.

Research/Design Work:

-CFD (Nesya)

-Air detector, pump specifications (Aditya)

-Testing protocols (Trevor, Cate)

-BioRender, drawing ideas (Lokesh)

10/30/2020 Advisor Meeting

ADITYA AILIANI - Dec 09, 2020, 9:55 AM CST

Title: Advisor Meeting

Date: 10/30/2020

Content by: Aditya

Present: Cate, Lokesh, Nesya, Trevor

Goals: To discuss advice from Show and Tell and plan for new testing

Content:

Cate and Lokesh had gone into WIMR to test out the original tubing set on the device, but couldn't get very far because the tubing set needed saline to pump through. They did note that the device was very finicky about making sure the pressure sensors and air chambers fit in directly.

Most of the advice we got from show and tell was for integrating pressure transducers directly into the tubing, but the pressure transducers are already in the COBE Spectra.

In terms of testing, Prof. Kinney discussed options such as putting colored water into the saline and anticoagulant inlets to make sure our Y connectors would result in adequate mixing of the inlet fluids. She also recommended trying to get around the issue of pressure sensors by placing a dummy sensor into the device's front panel so we could just focus on the tubing.

Another major difficulty was that the tubing we have (ELP Platelet set) did not fit into the centrifuge when Cate and Lokesh went to test it. Looking back through the manual, we found that this was because there are different "caps" that go on the centrifuge depending on the type of tubing set. Prof. Kinney recommended emailing the client immediately and following up on the centrifuge cap at our next client meeting.

Conclusions/action items:

We should dig into the research for how the device uses data from pressure sensors to see if there's a way we can get around the pressure sensor alarm. For now, our materials are starting to come in, so we need a plan to fabricate a basic skeleton of the tubing set. We have a simplified diagram, but we need to think about the specifics of fabrication.

11/6/2020 Advisor Meeting

ADITYA AILIANI - Dec 09, 2020, 9:57 AM CST

Title: Advisor Meeting

Date: 11/6/2020

Content by: Aditya

Present: Cate, Lokesh, Nesya, Trevor

Goals: To take stock of our project so far and figure out a timeline for the next few weeks

Content:

Our clients were unaware of the centrifuge cap issue and got back to us saying that they would look around the lab to see if other centrifuge caps came with the device. Prof. Kinney recommended looking for an outside source where we could purchase it just in case.

The three free tubing sets from Terumo arrived, so we can use those for measurements and to cannibalize parts now. We hope to have measurements done in the next few days so Cate can start cutting the bulk tubing to the right pieces.

We expressed concern about being able to manufacture bubble traps and pressure sensor components in time to test the device before Thanksgiving. Prof. Kinney advised us to look into manufacturing a bubble trap to test independently of the rest of the tubing set and plan to cut those components from the original tubing set if necessary.

Conclusions/action items:

We will need to make measurements so we can start to fabricate the tubing. For research, we should try to focus on how the air chambers interface with the device's front panel, since we are working on manufacturing bubble traps. We may be able to figure out a design for pressure sensors based on the patents Trevor and I found, but we would have to figure out materials and order them soon, and it may be difficult to establish a seal to keep blood from leaking into the device. Nesya has a CAD design for a cheap microfluidic bubble trap, which is a promising start to manufacturing the bubble trap.

11/20/2020 Advisor Meeting

ADITYA AILIANI - Dec 09, 2020, 10:05 AM CST

Title: Advisor Meeting

Date: 11/20/2020

Content by: Aditya

Present: Trevor, Cate, Lokesh, Nesya

Goals: To outline our currently fabricated items and identify how much testing we can get done before Thanksgiving

Content:

We have received all the materials needed to make a tubing set without pressure or air sensors. We have a design for a disposable 3D-printed part for the air sensor and are working on a design for a disposable 3D-printed part for the pressure sensor. We are planning to begin fabrication of the basic tubing set using the PVC tubing set (ruled out Nylon and Fluoropolymer because the products are too stiff and not clear) and tubing connectors. We met with our client, who agreed to order additional parts to make the disposable pressure and air sensor parts and requested an overall per-use price estimate based on these disposables. We are running saline through the COBE Spectra with the intact existing tubing set and may get access to a gas sterilizer to test material properties before and after sterilization. We met with Dr. Galipeau and Dr. Pennati to confirm our plans for testing this week and early next week (confirm w/ Prof. Brounts for gas sterilization).

One difficulty we had earlier was getting our large tubing connectors (1/8" OD) to fit into our tubing set - the straight connectors managed to fit once Cate heated them with a hair dryer for 5 minutes and maintained a strong seal. The large Y connectors did not fit at all, so we ended up ordering 1/16" versions of both connectors. We are hoping to test the seal provided by each one and compare them. Prof. Kinney advised us to consider gravity-loading the tubing, which we could do at home in case the connectors didn't arrive until after Thanksgiving. She also recommended that we ask the client what their preferences are about Cate and Lokesh going in to WIMR after Thanksgiving.

Prof. Kinney approved Cate and Lokesh's initial testing of the manufactured tubing set, which ran the tubing through the saline rinse cycle on the COBE Spectra and did not trigger any alarms.

Conclusions/action items:

Try to get testing done in the next week - we should all try to research backup testing options in case we aren't able to finish testing or go back into WIMR after Thanksgiving.

Project Expenditures Sheet

LOKESH KUMARAVEL - Dec 09, 202(

Title: BME 200/300 Large Animal Plateletpheresis Expenditures

Date: 12/07/2020

Content by: Lokesh Kumaravel

Goals: To explain all materials purchased within the course of the semester.

Content:

Total Expenditures

Item	Description	Manufacturer	Part Number	Date	QTY	Cost Each	Total	Link
Tubing								
Nylon Tubing	100 ft. Black Nylon Tubing, 5/32 in Outside Dia., 7/64 in Inside Dia.	Grainger	1PCC7	10/22/2020	1	$17.59	$17.59	https://www.grainger.com/product/GRAINGER-APPROVED-100-ft-Black-Nylon-Tubing-1PCC7?breadcrumbCatId=CC_1041_1001014_15251
PVC Tubing	PVC Tubing, Metric, 2 mm Inside Dia., 4 mm Outside Dia.	Grainger	22XH86	10/22/2020	2	$23.50	$47.00	https://www.grainger.com/product/TYGON-PVC-Tubing-22XH86?breadcrumbCatId=CC_1041_1001014_18429
Fluropolymer Tubing	50 ft. PFA Fluoropolymer Tubing, 3/16 in Outside Dia., 1/8 in Inside Dia.	Granger	2VLU5	10/21/2020	2	$79.30	$158.60	https://www.grainger.com/product/GRAINGER-APPROVED-50-ft-PFA-Fluoropolymer-Tubing-2VLU5
Tubing Connectors								
Y Connector	Polypropolene Y-tubing connector; 0.125– 0.156in.; 4mm (20 pack)	Fischer Scientific	S50701A	10/21/2020	1	$30.29	$30.29	https://www.fishersci.com/externalappproductdisplay?catnum=S50701A&catalogId=&isChemical=false&langId=-1&storeId=10652&isItem=Yes
Straight Connector	Staight Coupler 1/8 In ID, -Natural Kynar-QC (Case of 10 Pack) (Pack of 100)	Fischer Scientific	01-000-511	10/21/2020	1	$69.30	$69.30	https://www.fishersci.com/itemdetail?catnum=01000511
Y Connector	PVDF Barbed Y Connector, 1/16 In. ID, 1/32 In., 1/2 In., 3/8 In., Pack Of 10, 3	Fischer Scientific	NC9866519	11/14/2020	1	$36.92	$36.92	http://www.fishersci.com/itemdetail?catnum=NC9866519
Straight Connector	Fisherbrand™ Straight Coupler with 1/16 in. ID - Polypropylene - QC (pack of	Fischer Scientific	01-000-506	11/14/2020	1	$52.00	$52.00	https://www.fishersci.com/shop/products/straight-coupler-1-16-in-id-polypropylene-qc/01000506#?keyword=barbed+connector+1%2F16in+straight
Bubble Trap								
Membrane Filter	Azzota Corp Membrane Filter For Standard Flow Bubble Trap Material: 10 Um Ptfe Qty: 5/Pk	Fischer Scientific	50-195-3960	11/14/2020	1	$53.29	$53.29	https://www.fishersci.com/shop/products/m-filter-for-stand-f-bubb-trap/50195396O#?keyword=bubble+trap
C-Clamp	Cast Iron C-Clamp with 3 in. Jaw Opening	Fischer Scientific	S81060	11/14/2020	1	$4.23	$4.23	https://www.fishersci.com/shop/products/cast-iron-c-clamp-3-in-jaw-opening/s81060?searchHijack=true&searchTerm=S81060&searchType=RAPID8
Flangeless Fitting	Flangeless Fitting PEEK 5/16-24 Flat-Bottom, for 4mm OD	IDEX	XP-132	11/14/2020	3	$6.29	$18.87	https://www.idex-hs.com/store/fluidics/fluidic-connections/fittings/flat-bottom-fittings/flangeless-fittings/flangeless-fitting-peek-5-16-24-flat-bottom
Bubble Trap Housing	PLA 3D printed housing for the bubble trap	UW Makerspace		11/16/2020	1	$0.96	$0.96	
						Total	$489.05	

Prototype Fabrication

Cate FLYNN - Dec 09, 2020, 2:20 AM CST

Title: Prototype Fabrication

Date: 11/23/2020

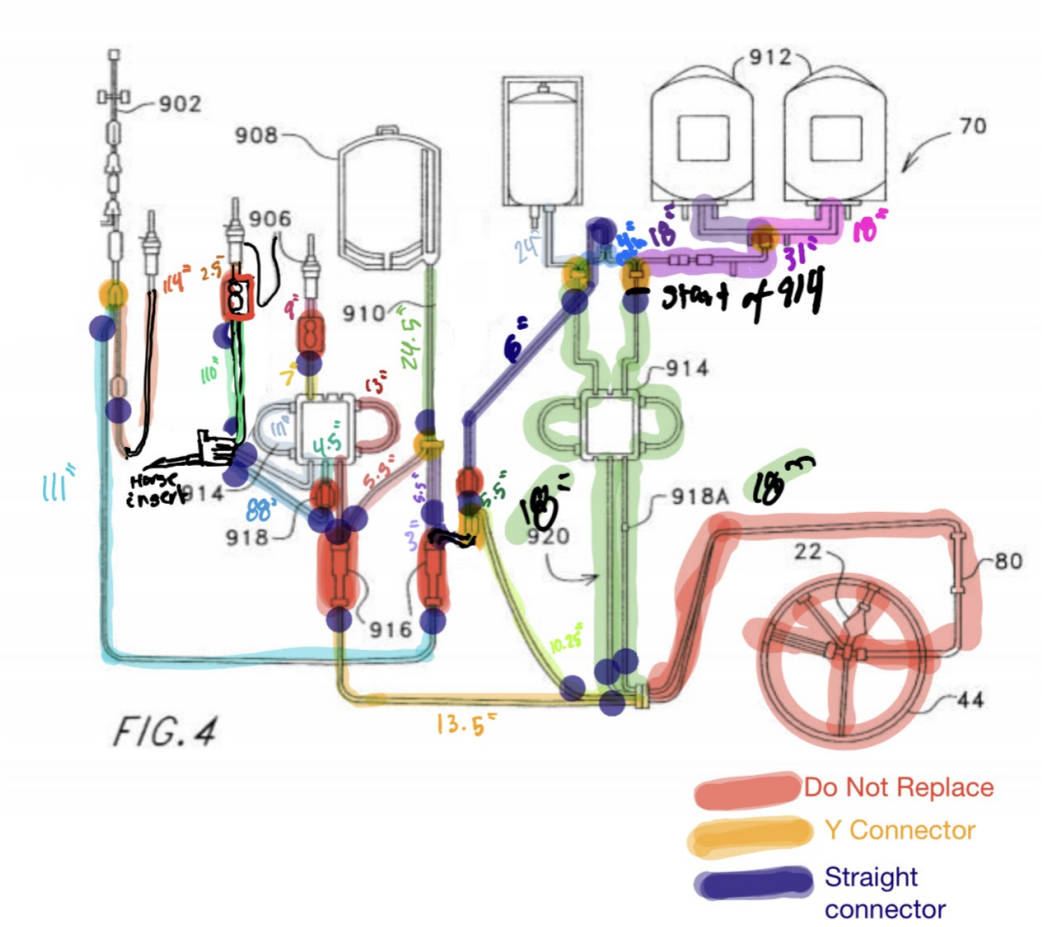
Content by: Cate Flynn

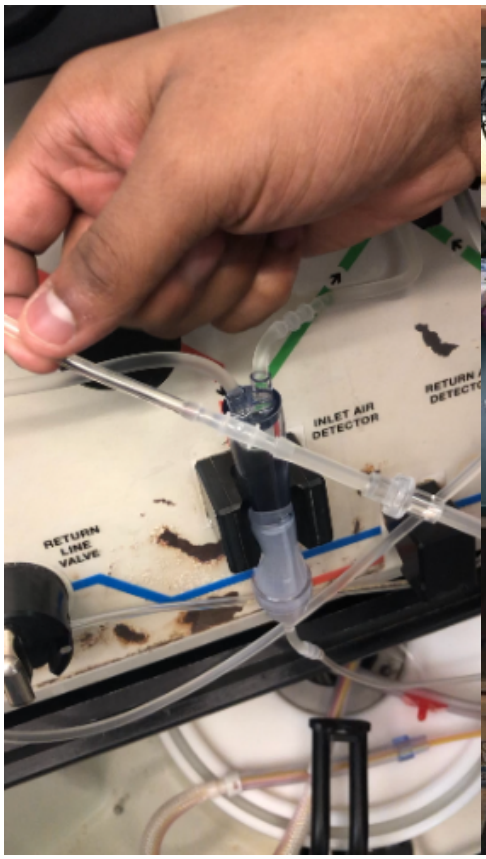
Present: Cate Flynn

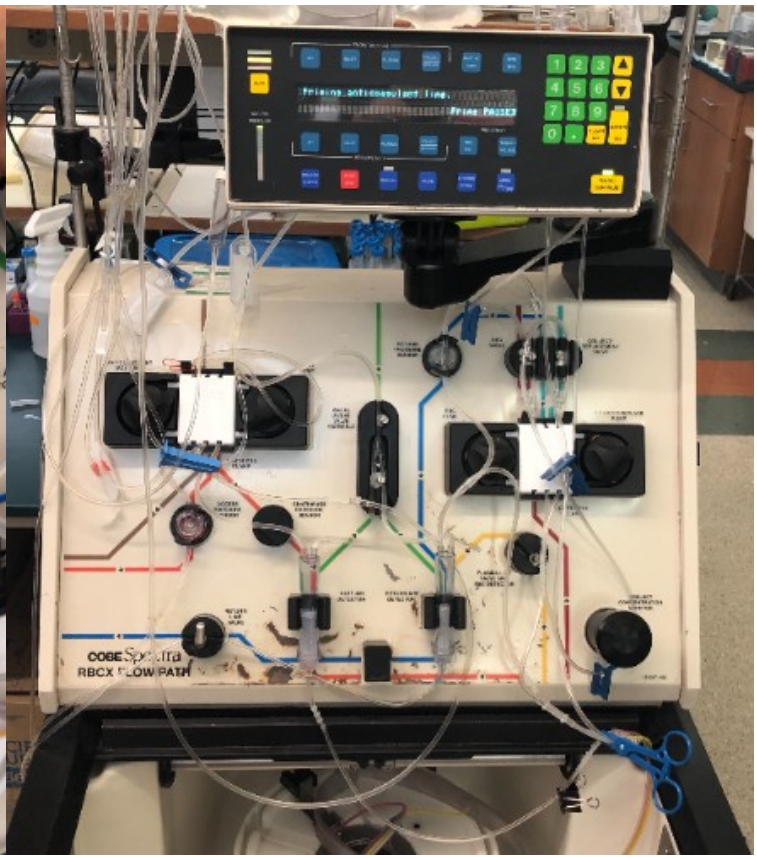
Goals: Create a prototype tubing set by replacing existing lengths of tubing with our own PVC tubing and connectors.

Content:

In fabricating this prototype, I measured out the tubing segments to cut from our PVC tubing as shown in the measurement document created during our fourth trip to Wisconsin Institutes of Medical Research (WIMR). I have attached this image below for reference. Once I had measured out the indicated lengths of tubing, I used a hairdryer on medium heat to warm the PVC tubing. I held each section of tubing under the hair dryer for about 5 seconds before pushing the 1/8" straight connectors into the warmed section of tubing as indicated on the measurement graphic. Once the tubing had cooled, a strong seal formed. I continued this process, leaving the parts that we could not replace as they were. I attempted this same procedure with the 1/8" Y connectors, but was unable to get the connector to fit into the tubing regardless of how long the tubing was heated for. This will be something to consider moving forward, as smaller Y connectors are likely necessary. Due to this complication, I left the original Y connectors intact. I have attached a close up image of one of the seals below as well as an image of our prototype in the apheresis machine itself.







Conclusions/action items:

The prototype has been fabricated through the use of a hairdryer, PVC tubing and 1/8" inner diameter straight connectors. The next step will be testing it with the machine at WIMR in order to verify the quality of the seals and ensure that there aren't any pressure issues.

Bubble Trap Fabrication

NESYA GRAUPE - Dec 07, 2020, 3:24 PM CST

Title: Bubble Trap Fabrication

Date: 12/7/2020

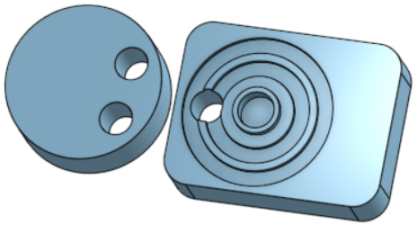
Content by: Nesya

Present: Nesya

Goals: Fabricate a device to remove bubbles from the tubing

Content:

- The bubble trap consists of a 3D printed base, a PTFE membrane, and a clamp. In addition, flangeless fittings are used to secure tubing. The membrane is inserted into the grooves on the rectangular portion (bottom) of the base. The circular (top) portion of the base is placed on top of the membrane. The system is fastened together using the clamp or a screw. The two holes in the circular portion of the base are for the inlet and outlet tubing, which is affixed to the trap using the flangeless fittings. Air collection tubing is attached to the back side of the rectangular portion of the base using the flangeless fittings. When fluid enters the trap through the inlet port, the air across through the membrane to the air collection tubing while the fluid cannot pass the barrier and flows to the outlet port.



Conclusions/action items: Finish the final report

Comparison of tubing connectors

ADITYA AILIANI - Dec 09, 2020, 9:36 AM CST

Title: Comparison of tubing connectors test

Date: 12/3/2020

Content by: Aditya

Present: Cate

Goals: To record the procedure used to compare tubing connector seals

Content:

-4 lengths of 2mm ID 4mm OD PVC tubing were cut 1.5 ft. each

-A 1/8" polypropylene straight connector and 1/16" Kynar straight connector were inserted to connect each pair of 1.5 ft lengths, resulting in 2 3 feet lengths of tubing

*The polypropylene (large) connector required the tubing to be heated for about 5 minutes using a hair dryer to expand and fit, resulting in a tighter seal. They 1/16" connector fit without this manipulation (heating might have widened the tubing too much)

-A control length of PVC was cut (3 ft length)

-In random order (treatment numbers 1-3), each tubing set was loaded with 2.5 mL tap water from a 2.5 mL syringe, with water exiting into a container of known mass (using a kitchen scale)

*Loading time was measured to verify a roughly equal loading flow rate

*This took about 10 seconds each - this was done slowly so the syringe plunger would not back up due to resistance from the tubing

-In each trial, the mass of water in mg was measured using a kitchen scale and converted to mL (1mg water = 1 mL)

The following MATLAB script was used to record and analyze data

```
% Time
% "smaller" refers to the 1/16" connectors and the "larger" refers to the
% 1/8" connectors
Timesmallerdata = [10.42, 10.43, 10.44];
Timelargerdata = [10.40, 10.41, 10.40];
tubinglengthft = 3;
tubinglengthm = tubinglengthft*(1/3.28084);
avsmall = mean(Timesmallerdata);
avlarge = mean(Timelargerdata);
stdsmaller = std(Timesmallerdata);
stdlarger = std(Timelargerdata);
velocitysmaller = tubinglengthm/avsmall;
velocitylarger = tubinglengthm/avlarge;

% Starting water for all tests is 2.5 mL, the data included below
% represents the water reaquired after being ran through each tubing set
watersmallerdata = [2.32, 2.35, 2.34];
waterlargerdata = [2.33, 2.36, 2.34];
watercontroldata = [2.37, 2.34, 2.32];

watersmallerdiff = [2.5, 2.5, 2.5] - watersmallerdata;
waterlargerdiff = [2.5, 2.5, 2.5] - waterlargerdata;
watercontroldiff = [2.5, 2.5, 2.5] - watercontroldata;

waterdata = vertcat(watersmallerdiff, waterlargerdiff, watercontroldiff);
[p, anovatab, stats] = anova1(waterdata.'); %anova1 treats columns as different groups, need to transpose waterdata
multcompare(stats);
```

->Flow rate for the 1/8" connectors is 16.57 mL/min and the flow rate for the 1/16” connectors is 16.53 mL/min
Standard deviations for the path times for the 1/8" and 1/16” connectors are 0.0058 and 0.0100 respectively, the standard error values are 0.0033 and 0.0058 respectively

Conclusions/action items:

The ANOVA reported no significant difference between water recovered from each connector and the control, so the seals seem adequate. It should be noted that none of the tubing sets actually let all the water out (2.3-2.4 mL actually recovered). It was observed that droplets of water were still stuck inside the tubing, even when air was pushed through with a syringe to dry it. This could be important for cleaning the tubing set later - it might need to be vacuum dried.

A 95% confidence interval of the loading times (avg +/- t*std error, t = 4.3 for 95% confidence and 2 degrees of freedom) showed no significant difference (overlap) in loading times, so that assumption was adequate. The flow rate was about 16.5 mL/min, which is within the COBE Spectra pump range but is much lower than the max flow rate we were hoping to test.

Ultimately, there wasn't a difference in seal quality, but just by trying to pull the connections apart we saw that the heat-sealed 1/8" connector was much stronger and required several days of pulling to come apart, while the 1/16" connector came apart easily. If there was a big difference in seal quality we might have looked into adhesives or other options to support the 1/16" connector strength, but that doesn't seem necessary.

Bubble trap effectiveness

NESYA GRAUPE - Dec 07, 2020, 3:27 PM CST

Title: Bubble Trap Testing

Date: 12/7/2020

Content by: Nesya

Present: Nesya

Goals: Determine the effectiveness of the bubble trap

Content:

Since the membrane that was needed for testing the bubble trap did not arrive in time, a proposed method to test the bubble trap is described. The bubble trap should be assembled and then primed with water. 1 mL of air should be inserted into the tubing via a syringe. Water should be continuously added to the tubing until the air has moved through the bubble trap. The amount of air remaining in the outlet tubing after passing through the bubble trap should be recorded. This should be repeated for a total of 5 trials with 1mL, 3 mL, and 5mL of air.

Conclusions/action items: I would have liked to be able to actually test the bubble trap. Hopefully the next group that gets this project will be able to do so.

Prototype Use with Machine

LOKESH KUMARAVEL - Dec 09, 2020, 12:14 AM CST

Title: Prototype Experiments with the Machine

Date: 11/24/2020

Content by: Lokesh Kumaravel

Present: Cate and Lokesh

Goals: To test the prototypes capabilities to be integrated with the COBE machine

Content:

Procedure:

- Load prototype into the tubing as if it were the original
- Run the power up tests and attach saline bags
- Clamp off the return portion of the tubing as well as the centrifuge loop
    - We do not have an effective way of draining the saline out of the tubing set. Normally, after a procedure, all of the fluids would be returned back into the donor leaving an empty tubing set, but we are unable to run the full procedure, so it was in our best interest to keep the saline away from certain parts of the set
- Begin the priming phase of the plateletpheresis process
- Unload the tubing and remove the saline that is in the prototype
    - We did this by cutting below the inlet air chamber and letting the saline drain from the access portion of the tubing
    - We then later joined the severed set again

Questions/Issues we were looking for

- Will the tubing set properly load onto the the machine?
    - The prototype's outer diameter is a tad smaller than the original and we were concerned that the fitting would be too loose for the machine to properly operate
- Will the machine detect a non-original tubing set via pressure sensors
    - The pressure sensors tend to be very precise as when we ran the original tubing through we encountered some problems. Our concern is that a smaller diameter tubing might lead to different pressure reading that the machine will not accept
- Can fluid be retained in the tubing while flowing throughout the set?
    - We are looking to see if fluids can be transported through the prototype without leaking

Results:

The machine was able to successfully accept our prototype without major complications. The machine was unable to notice that a non-original tubing set was being used and the pressure sensors did not indicate any errors. Saline ran through the tubing without any problems and no leaking was visible. We ran the machine for approximately 15 minutes and no saline leaks were detected. A minor problem occurred when loading the prototype set onto the machine. Specifically, the tubing that ran through the access pump cartridge was small enough to slip through the guiding ports. When the pumps rotated during the loading processes, the tubing slipped creating slack around the pump. This slack made it so the tubing could not properly sit in the peristaltic pumps as shown in Fig. 2. This was easily solved by holding the tubing and applying tension so when the tubing loads into the pump there is no slack. Once the tubing is loaded there was no problems.

Figure 1: The prototype fully loaded onto the machine

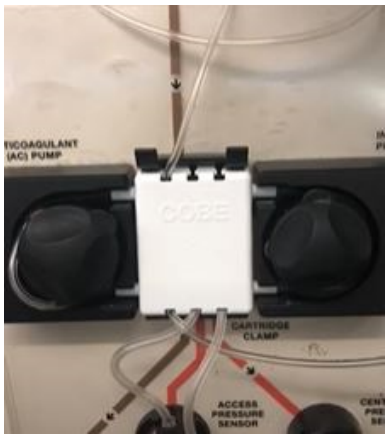


Figure 2: Partially loaded tubing set with too much slack around the pumps

Figure 3: Close-up of tubing connectors (There is currently saline running through the joint)

Conclusions/action items:

The prototype experiments we did with the COBE machine were a success. It proved that a tubing set manufactured by someone else other than Terumo could be integrated onto the machine. Moreover, it showed that our material, size, and connecting options were the correct ones as the machine was able to pump saline through the tubing without any major problems.

20201005 - Terumo tubing set diagram, dimensions

ADITYA AILIANI - Oct 05, 2020, 10:05 AM CDT

Title: Terumo BCT tubing set diagram, dimensions

Date: 10/5/2020

Content by: Aditya

Goals: We received an email from Angela Richardson, a Terumo BCT representative, with the following data (text is copied from the email, pdf is attached).

Content:

COBE BCT PLATELET AND PLASMA COLLECT BAG TUBING SPECIFICATIONS

OUTSIDE TUBING DIAMETER		WALL THICKNESS	
MINIMUM	**MAXIMUM**	**MINIMUM**	**MAXIMUM**
0.157 inches	0.163 inches	0.020 inches	0.024 inches
3.99 mm	4.14 mm	0.51 mm	0.61 mm

CLEARED TUBING SPECIFICATION FOR

USE WITH THE SCD 312 STERILE TUBING WELDER

OUTSIDE TUBING DIAMETER		WALL THICKNESS	
MINIMUM	**MAXIMUM**	**MINIMUM**	**MAXIMUM**
0.152 inches	0.220 inches	0.020 inches	0.043 inches
3.86 mm	5.6 mm	0.508 mm	1.1 mm

PLATELET COLLECT BAG TARE WEIGHT:

Section 10, the “Helpful Hints” section, of the COBE Spectra Apheresis Operator’s Manual, provides instructions for “How to Calculate Collect/Plasma Bag Tare Weights”. The weights listed in the Operator’s manual are accurate for the slide clamp and tubing which will be included in disposable sets beginning in August (lot numbers beginning approximately 07D). The table below summarizes the component weights (grams):

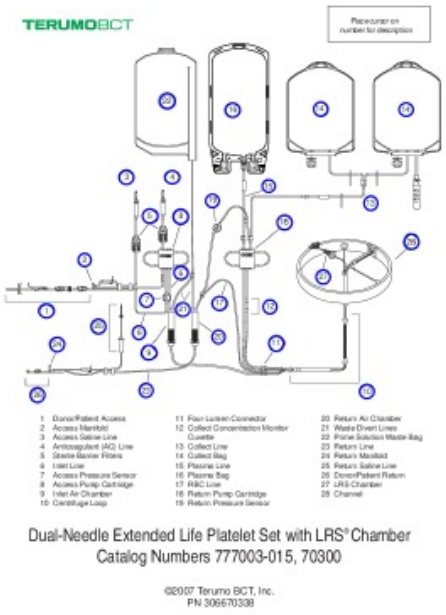
COMPONENT	OPERATORS MANUAL	01D-07D lots	after 07D lots

		(approximately)	(approximately)
Platelet Collect Bag	36	33	33
Slide Clamp	1.4 (Blue)	1.5 (White)	1.4 (Blue)
Weight of tubing (per inch)	0.20 (thin wall)	0.35 (thicker wall)	0.20 (thin wall)

Conclusions/action items:

This information will be used in individual notebooks to decide which aspects of the tubing can be simplified or purchased elsewhere. We have noted the large number of hard plastic parts that are not feasible to fabricate (flow controllers, needle guards, sensor attachments, clamps, filters). The COBE Essentials Guide states that this tubing set has an inner diameter of 0.113 in, but it seems that the outer diameter varies considerably within the tubing set. We will have to visually inspect the tubing to see these changes.

ADITYA AILIANI - Oct 05, 2020, 10:03 AM CDT



Spectra_Dual_Needle_ELP_LRS_1_.pdf(193.4 KB) - download An interactive pdf diagram of the Dual-Needle Extended Life Platelet disposable tubing set with Leukoreduction chamber.

20200904 - Introductory Meeting

ADITYA AILIANI - Sep 04, 2020, 4:51 PM CDT

Title: Introductory Meeting

Date: 9/4/2020

Content by: Aditya

Present: Nesya, Lokesh, Cate

Goals: To decide on team roles, methods of communication, and initial goals

Content:

Current roles (will confirm with Trevor and Professor Kinney):

Leader - Aditya

Communicator - Cate

BSAC - Nesya

BWIG - Trevor

BPAG - Lokesh

->We plan to meet again with the full group on Monday and use our Slack channel until then. We may switch to Snapchat, depending on how well Slack works. Cate will email the client after we all meet on Monday, so we can prepare questions before then. We are planning to meet with Professor Kinney for about 25 min between 1-2pm on Fridays. For non-urgent communication and collaborative reports, we will use the Google Drive that Nesya set up.

Conclusions/action items:

Conduct preliminary research using the sources in our project description and brainstorm questions for the client. Decide on a meeting time for Monday.

09/07/2020-Questions for Client Meeting

Cate FLYNN - Sep 07, 2020, 5:03 PM CDT

Title: Questions for Client

Date: 09/07/2020

Content by: Cate

Present: Cate, Aditya, Nesya, Lokesh & Trevor

Goals: Confirm team roles and outline preliminary questions for first client meeting.

Content:

We agreed on a 1:30 to 2 pm meeting time with our advisor and our team roles. We discussed setting up a recurring team meeting on Mondays at 1 pm. We discussed a couple of questions that would be good to ask our client in the first meeting including:

- What materials the client is willing to provide
- Are there other parts of the machine we should look at besides the tubing
- What makes animal use different than human use in terms of reusability
- Are there other factors that increase cost per use
- Does the client have more detailed documentation on the device
- What is the material that the tubing is made from now and what makes it so costly
- What is the intended use of the platelets
- Are there any machine sensitivities we should be aware of
- Are there any legal considerations with the materials for this project
- Is there a designated workspace for us to see the machine
- What does the budget look like for this project

We want to meet with our client by the 11th. We need to upload our photos to the team website and send the link to our client.

Conclusions/action items:

Trevor will upload the photos to the team website and Cate will send emails to set up meetings with our client and our advisor. We will all continue to do individual research and contribute to the progress report.

09/14/2020 - PDS Team Meeting

ADITYA AILIANI - Sep 14, 2020, 5:48 PM CDT

Title: PDS Team Meeting

Date: 9/14/2020

Content by: Aditya

Present: Nesya, Lokesh, Cate, Trevor

Goals: To review what we had learned from the client meeting and plan for the PDS

Content:

We discussed potential in-person meeting times to look at the disposable tubing set and COBE device. From initial research, it looked like the tubing set included more than just simple tubes, so we agreed that we should wait to see the tubing set before reaching out to Trevor's contact. We hadn't heard back from Prof. Brounts yet regarding in-person meeting times or her article in Tissue Engineering (although Nesya was able to find a full-text pdf).

We split up the PDS as follows:

Aditya: Function, Life in Service, Weight, Standards/Specifications

Trevor:Client Requirements, Shelf Life, Materials, Customer Considerations

Cate: Performance Requirements, Operating Environment, Aesthetics, Patient-related Concerns

Lokesh: Safety, Ergonomics, Quantity, Competition

Nesya: Accuracy/Reliability, Size, Target Product Cost

We decided to try to have the PDS done by 4PM on Thursday.

Conclusions/action items:

Everyone is going to work on their PDS sections and submit questions/concerns in our groupchat. Some sections may overlap or be less relevant, but we will notice that more as we are working on our sections.

09/21/2020 - Design Matrix Team Meeting

ADITYA AILIANI - Sep 21, 2020, 2:59 PM CDT

Title: Design Matrix Team Meeting

Date: 9/21/2020

Content by: Aditya

Present: Trevor, Cate, Lokesh, Nesya, Aditya

Goals: To decide on criteria to evaluate preliminary design ideas and identify the most feasible

Content:

Design Ideas:

Based on our client's and advisor's suggestions, we had three preliminary design ideas: Fabricating cheap disposable tubing, Sterilizing the current tubing set for reuse, and Modifying apheresis parameters.

-Fabricating cheap disposable tubing would require us to buy bulk tubing of an appropriate cheap material and use it to construct a tubing set that could be used in the COBE Spectra.

-Sterilizing the current tubing set for reuse would require us to research and test feasible sterilization techniques (EtO, autoclave, EM radiation?). The main work would be establishing reasonable parameters for the sterilization technique. Our immediate concern with this technique was that our client expected around 20 uses out of the tubing initially, and there are no longer any available disposable tubing sets for the COBE Spectra.

-Modifying apheresis parameters was our client's idea for a virtual project if campus shut down. We would research the properties of equine blood as they compare to human blood (what the machine was designed for) and propose changes to the device's parameters to maximize platelet concentration.

Criteria (weight out of 100):

-Cost per Use (26): Our client's goal was to make equine plateletpheresis a viable alternative to whole-blood donation for her research by reducing the cost per use. This will be the main metric our client uses to judge our project. Therefore, cost per use was the highest weighted category.

-Sterility (21): This category was a close second to cost per use because tubing is only considered viable for medical purposes if it can be adequately sterilized before use.

-Safety of patient (19): This category differs from sterility because it considers factors other than bloodborne pathogens. For instance, the tubing system would have to be resistant to kinking, not induce clot formation, and adequately ensure a closed system. Although safety is an important consideration for designing our system, these options were all suggested by the client, a professor of veterinary medicine. We therefore thought all the ideas would be safe for the horses in principle, and ranked designs based on how easily we could ensure safety.

-Durability (18): This category describes how often the system would need to be changed or replaced. The idea is straightforward for tubing, as more durable tubing would be more resistant to mechanical failures or degradation. The idea of durability for adjusted device parameters would consider how often the user would need to tweak the parameters and if the parameters might lead to increased wear and tear.

-Ease of Fabrication (10): We may need to weight this higher if it turns out that we don't have access to the design lab, since that will become a major factor in our design process. For now, we know that we have a few virtual testing options, so ease of fabrication is not the most important category.

-Ease of Use (6): Our design ideas are similar in terms of ease of use, as they do not affect the workings of the device too much. Ease of use is important for the client, but it was not weighted heavily because it is less helpful for deciding between the three ideas.

Rankings:

-Cost per use: We felt that sterilizing existing tubing would most greatly impact the cost per use, as it wouldn't require the client to buy multiple sets of disposable tubing, even if they were cheap. This came with the caveat that, eventually, the current tubing set would not be reusable under multiple sterilization cycles, so fabricating cheap tubing was a close second. Ideally, we could fabricate cheap tubing that can be sterilized. Adjusting device parameters may increase platelet concentrations per use, but would not significantly impact cost per use.

Fabrication: 9/10, Sterilization: 10/10, Parameters: 6/10

-Sterility: Fabricating cheap, disposable tubing would be the best option for sterility, as it could be discarded after each use. An adequate sterilization protocol would also result in sterility, but it would require more confirmation through testing. Adjusting device parameters would not

affect sterility.

Fabrication: 10/10, Sterilization: 7/10, Parameters: 0/10

- Safety of Patient: The current tubing has been demonstrated to be safe for use in horses, so deviating as little as possible from that design would ensure safety of the patient during apheresis. Under the oversight of our client, fabricated tubing could be safe enough for use in horses. Changing device parameters has the most potential to be unsafe, as we have only seen sources that test existing protocols (for a 700lb woman).

Fabrication: 8/10, Sterilization: 10/10, Parameters: 7/10

-Durability: Cheap disposable tubing would rank highest in durability because it could be made from more durable materials than the current tubing set. The current tubing set is supposed to be unusable after multiple sterilizations, so it ranked lower in durability. Modifyng apheresis parameters could result in increased wear and tear to the device and would need to go through several rounds of adjustment and testing.

Fabrication: 7/10, Sterilization: 6/10, Parameters: 6/10

-Ease of Fabrication: Modifying apheresis parameters requires no fabrication and would be the easiest to complete without access to the design lab. Sterilization would require testing using the Department of Veterinary Medicine's resources, and fabricating tubing would require us to build in the design lab.

Fabrication: 7/10, Sterilization: 8/10, Parameters: 10/10

-Ease of Use: Cheap disposable tubing would be easiest to use, as it would require minimal changes to the existing protocol. Sterilization would add an additional step each time an apheresis had to be done, and modifying parameters could significantly change the protocols for setting up a plateletpheresis run.

Fabrication: 10/10, Sterilization: 6/10, Parameters: 1/10

Conclusions/action items:

Based on the final score out of 100 for each idea, we will try to fabricate our own cheap tubing set from bulk tubing. We will need to research viable materials and their costs in addition to making sure we understand the connections between pumps in the device and the collection bags.

ADITYA AILIANI - Sep 21, 2020, 1:46 PM CDT

			Fabricate cheap tubing		Sterilization protocol for current tubing		Modify apheresis parameters	
	Criteria	Weight	Score (max 10)	Weighted Score	Score (max 10)	Weighted Score	Score (max 10)	Weighted Score
1	Cost per Use	26	9	23.4	10	26	6	15.6
2	Sterility	21	10	21	7	14.7	0	0
3	Safety of patient	19	8	15.2	10	19	7	13.3
4	Durability	18	7	12.6	6	10.8	6	10.8
5	Ease of Fabrication	10	7	7	8	8	10	10
6	Ease of Use	6	10	6	6	3.6	1	0.6
	Sum	100	Sum	85.2	Sum	82.1	Sum	50.3

DesignMatrix_1.JPG(71.2 KB) - download Preliminary design matrix with weighted scores

09/28/2020 - Preliminary Presentation Meeting I

ADITYA AILIANI - Oct 01, 2020, 10:56 PM CDT

Title: First Preliminary Presentation Meeting

Date: 09/28/2020

Content by: Aditya

Present: Lokesh, Cate, Trevor, Aditya

Goals: To split up roles for the preliminary presentation and make changes to the design matrix

Content:

-Design Matrix Changes:

We had to change our previous design matrix because it became clear from the client meeting that changing COBE apheresis parameters was not a viable alternative to our tubing project. It seems that Dr. Galipeau viewed this as a supplement to the project, but tubing is necessary. The remote component of our project will have to be a proposal detailing tubing materials and maybe using fluid modelling as a preliminary test. Therefore, we decided to critically evaluate the "tubing fabrication" design option.

We split the "tubing fabrication" design into two different ideas: fabricating a tubing set based entirely on the tubing set that Cate and Lokesh had picked up from Dr. Pennati or redesigning the tubing set to be simpler. Dr. Galipeau had mentioned that we should simplify the tubing, but it is not clear to us which components should be removed/combined. However, if we are able to simplify the tubing, it would be cheaper just by virtue of needing fewer materials, and it would be easier for the client to use.

We were concerned that a simplified option might not be easier to fabricate because we would have to do significantly more research and testing to validate a new model of tubing. However, it was a better option than fabricating tubing without changes, because that option would have a high cost per use, potentially low sterility and safety to the patient (if we are trying to re-create a complex tubing system with several hard plastic components, we are likely to make errors in fabrication that the device won't tolerate). A re-created tubing set could also be challenging to use, as it would be similar (but could not possibly be exactly the same) to the original complex tubing set. Therefore, our primary goal was still to make simpler tubing. We agreed that we should still consider sterilization because we may not be able to get each set under $20, which Professor Brounts recommended as the upper price limit for disposable (non-sterilizable) tubing.

			Simplify cheap tubing		Sterilization protocol for current tubing		Fabricate cheap tubing (no changes)	
	Criteria	Weight	Score (max 10)	Weighted Score	Score (max 10)	Weighted Score	Score (max 10)	Weighted Score
1	Cost per Use	26	9	23.4	10	26	6	15.6
2	Sterility	21	10	21	7	14.7	6	12.6
3	Safety of patient	19	8	15.2	10	19	7	13.3
4	Durability	18	7	12.6	6	10.8	6	10.8
5	Ease of Fabrication	10	7	7	8	8	10	10
6	Ease of Use	6	10	6	6	3.6	5	3
	Sum	100	**Sum**	85.2	**Sum**	82.1	**Sum**	65.3

Presentation Roles:

Introductions and Background: Trevor

PDS Summary: Lokesh

Preliminary Designs: Nesya

Design Matrix: Aditya

Future Work: Cate

Other notes:

Lokesh showed us the tubing set over the Teams call, and it seems very complicated, with plastic meshes, flow controllers, spikes, collection bags, and hard plastic bits (interface w/ sensors?). We thought we'd try to measure the length of the whole blood circuit, but it was too hard to immediately identify where the blood would actually flow just from the tubing set (Dr. Pennati said it would be fine to remove it from the packaging, but that its sterility would be compromised). We decided to try finding some schematics online to help us.

Conclusions/action items:

Fabricating the tubing is not trivial, and more research will need to be done to even think about simplifying the design. We have to know how blood flows in the tubing and what all the additional pieces are for. It is not clear to us right now if our client wants parts or all of the tubing set replaced - we will need to ask at our next meeting.

Complete the presentation, Cate will send slides to Prof. Kinney for feedback by Wednesday, and plan to record on Thursday.

10/1/2020 - Preliminary Presentation Meeting II

ADITYA AILIANI - Oct 05, 2020, 10:08 AM CDT

Title: Second Preliminary Presentation Meeting

Date: 10/1/2020

Content by: Aditya

Present: Lokesh, Nesya, Cate, Trevor, Aditya

Goals: To record our presentation and discuss any new findings or ideas

Content:

We discussed the actual tubing in more detail, and Lokesh had given Trevor some pictures to send to potential vendors. Our main question was what could be simplified or removed in the current tubing set. Based on our research, we had found some platelet extraction tubing patents assigned to Terumo (aka Gambro BCT aka Cardian BCT) [1]. Unfortunately, all the labelled parts seemed important for safety (optical detectors for platelet concentrations, flow controllers for replacement fluid and anticoagulant, parts to connect to pressure sensors). We will bring these concerns to our next client meeting.

We recorded the presentation over Teams. After the recording, a Terumo BCT representative responded to our email requesting a schematic of the Dual-Needle ELP Disposable Tubing Set with a schematic and some company data with tubing sizes. These will be analyzed in more detail in individual research folders.

Conclusions/action items:

We are in a better position to understand what the tubing does now that we have a more defined set of design options (from the preliminary report and design matrix), the tubing set itself, and an accurate, labelled schematic. We will need to get familiar with the different tubing elements so we can discuss these in detail at our client meeting. Also, prepare for the preliminary report.

10/2/2020 - Final Report Meeting

ADITYA AILIANI - Oct 05, 2020, 10:13 AM CDT

Title: Final Report Meeting

Date: 10/2/2020

Content by: Aditya

Present: Nesya, Trevor, Cate, Lokesh, Aditya

Goals: To plan research for the final report meeting

Content:

After our presentation, Professor Block recommended that we consider the amount of time we spend fabricating the new tubing set in our final cost assessment. We also received a question on what aspects of the tubing we plan to simplify, which is fair because we were wondering that ourselves.

We decided to each focus on different areas of research. I will look through tubing patents to categorize all the extra tubing parts, Nesya will research sterilization protocols to see which parameters will be important for us to consider, Lokesh will try to measure or calculate tubing dimensions that could be used in case we have to order our own tubing, Trevor will look for tubing vendors who might do custom work for us, and Cate will research materials we can use. We also split up parts for the final report.

Our next client meeting is next Wednesday, so we will meet on Monday to plan questions and slides for then.

Conclusions/action items:

Document research for the final report, start writing up sections. Meanwhile, we will have to think about some smaller project milestones we can hit, because our clients want a gantt chart to track our progress.

11/16/2020 - Testing, Fabrication Discussion

ADITYA AILIANI - Nov 16, 2020, 1:46 PM CST

Title: Testing, Fabrication Discussion

Date: 11/16/20

Content by: Aditya

Present: Trevor, Nesya, Lokesh, Cate, Aditya

Goals: To discuss research so far and plan testing and fabrication in the next two weeks

Content:

We will have a client meeting this Wednesday. We need to ask the following questions:

-Can we test a tubing set in the gas sterilization chamber early next week? Is there an established protocol the clients want us to use?

-Would we be allowed to come back to WIMR after Thanksgiving?

-Do you have preferences for how we test the bubble trap or advice on how to quantify results?

-Confirm other testing options: 1) Run water through each tubing set with auxiliary branches (saline, AC, collect lines) closed to see how much the inlet flow differs from outlet flow. Compare between unchanged tubing sets and our tubing set. 2) Put tubing sets through gas sterilizer and measure bend radius, inlet/outlet flow difference before and after sterilization.

In other news, we have received the three free tubing sets that we can run in the device, and we can test them. Cate may be able to fit the current tubing connectors in the PVC set - they are slightly too large but might fit if the tubing is heated with a hair dryer. Prof Kinney advised us against cutting slits in the tubing because the cracks could expand along the length of the tubing. We have ordered smaller tubing connectors that could also fit, but we might not need them this semester.

Making our own pressure diaphragms could be very difficult - the silicone rubber would need to be cut to precise circles to prevent leaking between chambers (this is usually done with a laser cutter), we would need a strong adhesive to keep the parts together (Trevor found that, for PVC, this may need a UV-cured adhesive), and the pod would have to lock into the apparatus on the device, which is already sensitive to small misalignment. For our project, we will use pressure pods from an existing tubing set.

The air detectors on the device fit tubing wider than the one we use with our bubble trap. We are considering keeping the air chamber from the original device and attaching the bubble remover downstream. This would have the benefit of maintaining the tubing's interface with the air detector while also meaning we will have a bubble remover/filter assembly we can access more easily than the device's air chamber. We will need to do more research and ask Prof Kinney and our clients to see if we can have quantifiable results from the bubble trap without using an air detector.

Conclusions/action items:

We have a set of questions for our client and a rough testing schedule that will be refined at our client meeting. Looking ahead, we should have some research on the Spectra Optia, the newer apheresis device, to add to our future work section, and, depending on how our client meeting goes, may need research on gas sterilization protocols for tubing. Trevor is looking back at our preliminary report to make changes based on feedback.

20201130 - Poster Planning Meeting

ADITYA AILIANI - Nov 30, 2020, 1:42 PM CST

Title: Poster Planning Meeting

Date: 11/30/2020

Content by: Aditya

Present: Nesya, Cate, Lokesh, Trevor, Aditya

Goals: To decide on testing results and procedures to include in our final poster

Content:

Cate and Lokesh were able to fit a prototype of the tubing (including pressure sensors, air detectors from the original tubing set) into the device, clamping off the centrifuge loop section, and completed the saline rinse protocol. No leaks were detected, and no alarms went off during this protocol.

Straight and Y connectors came in, so we can test the effectiveness of the seal provided by each connector.

Testing results we would like to include:

-comparison of seal (fluid out - fluid in / fluid in) for straight and y connectors, compare between dimensions (maybe present as a bar graph)

-qualitative analysis of how well the bubble trap performs (feed in turbulent water, observe output OR take pictures of the assembled bubble trap (*we don't have the filter membranes yet)*

-Qualitative evaluation of material properties of three tubing set materials we have (Nylon, fluoropolymer, PVC) to identify reasons for using a particular material. We can incorporate the actual Young's Modulus or minimum bend radii of these materials to quantitatively show what values are considered too stiff.

Conclusions/action items:

We will meet with our clients again on Wednesday and update them on the testing we are able to complete. We can each work on sections of the poster to describe the information we've gathered about feasible designs.

20200907 - Initial Research

ADITYA AILIANI - Sep 09, 2020, 7:29 PM CDT

Title: Initial Research

Date: 9/7/2020

Content by: Aditya

Goals: To understand goals of the project and develop initial questions

Content:

From the Project Description (BME Design website) [1]:

Apheresis equipment uses centrifugal technology to separate blood components, return uncollected components to donor (plateletpheresis is specifically extraction of platelets). "Potentially safer and more effective method of platelet collection" (what alternative methods are there?). Platelet lysate is used in regenerative medicine. Plateletpheresis is feasible and effective in dogs and horses, but expensive. Goal is to design a multiple use, easy to use version of a COBE apheresis device to be used in large animals. COBE device is available for us to view thanks to the client.

Questions:Plateletpheresis is "safer and more effective" than what? Why are disposable tubes used for human apheresis (materials, safety)? What modifications were needed in the studies demonstrating plateletpheresis in horses and dogs? Does the client have a price range for the modified apheresis device (project description claims $2000-$2600 PER USE of human apheresis)? Where does the cost of human apheresis come from (tubing, additional supplies)? What is platelet lysate used for?

From "Relevant Sources and Publications" (BME Design website):

COBE Spectra Apheresis System [2]:

-The system is highly customizable, with 4 defined protocols: Therapeutic Plasma Exchange (likely our focus), Red Blood Cell Exchange and Depletions, Stem Cell, Mononuclear Cell and Granulocyte Collections, Platelet and White Blood Cell Depletions. Flow rate and centrifuge speed are adjusted depending on donor, cell types.

-"The system draws whole blood from a donor or patient, adds anticoagulant, separates the blood components, collects or removes specific components and returns uncollected components to the donor or patient. In therapeutic plasma exchange and red blood cell exchange procedures, appropriate replacement fluid is continuously returned." => Anticoagulant could be a per use cost factor.

-Includes four pumps, five valves, and optical sensors to monitor density. Website requires registration to look at technical documentation - maybe a general block diagram is available elsewhere?

Platelet lysate obtained via plateletpheresis performed in standing and awake equine donors (Sumner et al) [3]:

-Platelet lysate has the potential to replace fetal bovine serum as a source of growth factors for mesenchymal stem cell culture (MSCs in PL maintain "differentiation potential and immunophenotypical profile"). This has been demonstrated in human medicine, but (at the time of this paper's publication) not in veterinary medicine. Plateletpheresis is safer than whole blood collection (take blood, extract platelets). At the time, studies had shown viability of apheresis in dogs, but not horses (still using whole blood collection).

-Used a 2005 version of the COBE Spectra apheresis device. Modified by using the largest body size parameters: 7 ft, 500 lb female.

-"Preliminary blood work (platelet count and hematocrit values) from each horse was used to manually input baseline platelet count and total blood volume of the equine donor for calculation of a more appropriate infusion rate, pump rate, and inlet flow rate" => What calculations are needed?

-Used a dual-needle procedure **Come back to this study for a detailed look at apheresis procedure**

-Varying degrees of hypothermia were seen in horses - all temps returned to normal after 4 hours. (Something we should be concerned about?)

-Measured prepheresis and post hematologic values (iron, white blood cells, platelet count, lymphocytes, calcium, coagulation factors) - all values returned to normal in the 48 hour window except platelet count for 1 horse (returned to normal after 1 week). No horses were at risk for "bleeding tendencies."

-Equine donors showed lower platelet concentration (1-2.3x normal blood vs. 2.6-7x in humans/dogs/swine). May be due to smaller platelet size, higher viscosity and different pigments in equine blood (optical sensors designed for human blood) =>Something we can modify?

Apheresis: Man vs Machine (Rock and Sutton) [4]:

-Editorial giving an overview of modern (in 1997) apheresis techniques/features. Apheresis is now widespread, with 116,802 therapuetic apheresis procedures in 1996 (American Association of Blood Banks).

-Usually citrate is used as an anticoagulant. Apparently concept of separation is similar to a cream separator. "

-"The extracorporeal volume varies somewhat (from approx. 140-315 mL), but all of the machines require at least a minimal hematocrit for processing the blood into components, a step that is, for the most part, carried out by differential centrifugation."=> Hematocrit is the % by volume of red blood cells in blood

-Apheresis generally considered safe - mostly side effects are mild chills, fever, urticaria (hives), muscle cramps, and paresthesia (pins and needles). Few patients were unstable and required intervention, and improvements were seen between two studies cited in this review. **Come back to this source for stats on apheresis safety/widespread use**

-Review goes on to describe complications due to return of highly citrated blood in patients with impaired liver function (need calcium), deaths due to errors (misplaced subclavian line, early granulocyte collection. Modern safety features help, but bring challenges. For example, modern machines have lower blood:anticoagulant ratios, which means kinks in tubing could cause clots. Review cites another example (Uhl et al, Transfusion) where users deliberately loading tubing incorrectly and alarm did not sound. Conclusion was that, despite safety features, users must be careful.

Clinical Indications and Adverse Reactions of Platelet Apheresis (Amanat et al) [5]:

-Observational study of plateletpheresis procedures in Pakistan Atomic Energy Commission General Hosptial

-"A single unit of platelet concentrate produced from a unit of whole blood contains, on the average, 7.5 x 10^10 platelets and should increase the platelet count by 5 to 10 x 10^9/L (5,000 - 10,000/µL) in a 70 kg recipient. Apheresis platelet concentrates generally contain 3 - 6 x 10^11 platelets, depending on collection practice."

-Few severe adverse effects (from citrate toxicity, anticoagulant intoxication, vasovagal reaction (dizziness, low blood pressure in response to trigger), hypovolemia)

-Used a dual needle procedure, "After donor selection, according to above criteria, disposable kits were fitted on cell separators and priming was done. As per the manufacturer's recommendations, we used a **double venous access with a C5L kit in a dual-needle procedure (program PLT5d DN).** The machine parameters were set as per protocol. The following data were entered into the cell separator program for instrument: **donor's height, weight, sex, hematocrit (Htc) and pre-apheresis peripheral blood platelet count**."

-Paper lists several indications for platelet transfusion (come back to this for Epidemiology/Impact)

Clinical and clinicopathologic effects of plateletpheresis on healthy donor dogs (Callan et al.) [6]:

-Used COBE Spectra Apheresis system to obtain platelet concentrate in dogs

-Mostly tolerated well (no hypotension) with some side effects (lip licking, tremors, ventricular ectopy [extra hearbeats starting in ventricles]) - authors concluded that hypocalcemia is a major risk for canine apheresis and users should supplement with calcium

Come back to this source to compare canine/equine methods (will need full access through UW Libraries)

Googled "COBE Spectra Apheresis System tech specs":

COBE Apheresis System Essentials Guide [7]:

-Useful reference (has a section on disposable tubing sets), review more completely in a different notebook entry.

Conclusions/action items:

Apheresis seems to be a safe, widely available method for taking platelets from donor humans, dogs, and horses. Concerns seem to primarily be about how donors respond to removal of blood components and anticoagulants, which can be addressed with procedural changes. We must see how the client would like to change things and which features are non-negotiable for safety reasons.

Discuss questions from this document with the group at our next meeting, see if they found similar obstacles/points of confusion. Review COBE Essentials Guide, methods of canine study as soon as possible. Look for sources on basic anatomy/physiology for information on blood composition, MSCs and how they differ between animals.

Sources:

[1] S Brounts. *VETMED: CONVERSION OF HUMAN COBE PLATELETPHERESIS MACHINE FOR LARGE ANIMAL USE,* UW Madison BME Department. 2020. Accessed Sept. 7, 2020. Available: https://bmedesign.engr.wisc.edu/selection/projects/25de148e-1405-47a7-9894-c76d2366793d.

[2]https://www.terumobct.com/cobe-spectra

[3] https://onlinelibrary.wiley.com/doi/full/10.1111/trf.14124

[4]https://onlinelibrary.wiley.com/doi/pdf/10.1046/j.1537-2995.1997.371098016435.x

[5]https://pdfs.semanticscholar.org/0388/dd2690ca2e7589815a98744ff4b2333130cf.pdf

[6]https://onlinelibrary.wiley.com/doi/abs/10.1111/j.1537-2995.2008.01803.x

[7] http://startrinity3.com/mssn/04/Apheresis%20System%20Essentials%20Guide.pdf

20200914 - Client's PRP therapy

ADITYA AILIANI - Dec 09, 2020, 10:57 AM CST

Title: Client's PRP therapy

Date: 9/14/2020

Content by: Aditya

Present: Aditya

Goals: To read the client's recent publication and understand what the platelets will be used for. This may not necessarily guide our tubing design, but it provides information on the potential impact of decreasing cost per use and understanding of how the client used blood previously.

Content:

This paper used equine platelets derived from pooled blood samples (2L per horse) that were centrifuged to get platelet-rich plasma. Platelets could be obtained directly from apheresis instead, which is what our clients are looking for. This went through several centrifugation and washing steps before becoming "WEPLEX." The authors tested its anti-inflammatory properties in mice with experimentally-induced colitis.

Conclusions/action items:

It will be important for future reports to go back through the methods of this paper to highlight inefficiencies in the whole blood collection process and how plateletpheresis is an improvement.

Source: Pennati A, Apfelbeck TM, Brounts SH, Galipeau J. Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical [published online ahead of print, 2020 Aug 28]. Tissue Eng Part A. 2020;10.1089/ten.TEA.2020.0160. doi:10.1089/ten.TEA.2020.0160

20201013 - Extracorporeal blood flow

ADITYA AILIANI - Dec 09, 2020, 11:11 AM CST

Title: Extracorporeal blood flow

Date: 9/16/20

Content by: Aditya

Present: Aditya

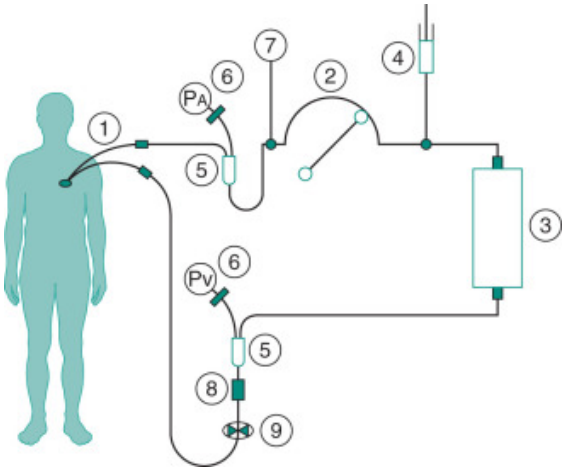
Goals: To look through the following source for potential specifications for medical tubing designed to carry blood

Content:

Zaccaria, Marta, Mauro Neri, Francesco Garzotto, and Claudio Ronco. “Principles of Extracorporeal Circulation and Transport Phenomena.” In *Critical Care Nephrology*, 3rd ed., 841-847.e1. Philadelphia, PA: Elsevier, 2019. http://www.clinicalkey.com/#!/content/book/3-s2.0-B9780323449427001394.

This source is mainly about hemodialysis but describes extracorporeal circulation in general. I'm mainly using it to think about which tubing components are essential for blood flow and safety.

This source also confirms that plasticized PVC is the most common tubing material, but does not say they should be reused (one-time sterilization with EtO or gamma radiation is preferred). There is an interesting section on pressure variation within the tubing. These calculations are very tied up in the pressure across the filtration membrane, which we don't use for apheresis, so it might take more time to revise them. I also don't see the purpose right now in finding that information, since the device pre-calculates an expected pressure at each pressure sensor and has alarms if that pressure is too high or low.



1. Blood inlet line (catheter) - could be one double-lumen catheter ("single needle procedure") or two single-lumen catheters ("double needle" - what the COBE Spectra uses)

2. Blood pump (peristaltic)

3. Filtration device - that's for hemodialysis, this would probably be replaced with the centrifuge loop + collect line

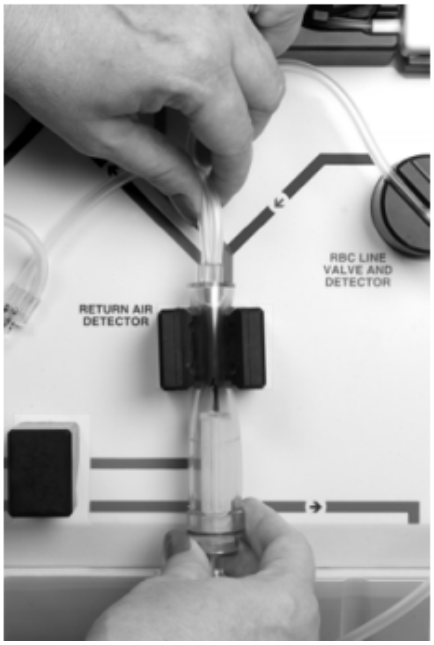
4. Anticoagulant infusion pump

5. Air-capture chambers (return and inlet)

6. Pressure monitor (return and inlet)- this source describes it as a pressure-transmitting sterile barrier that separates blood from the pressure transducer

7. Saline priming line

8. Ultrasonic air detector - this design has it downstream of the air chamber, while the COBE Spectra seems to have it in the middle of the air chamber (image from the COBE Spectra Essentials Guide). The black piece houses the ultrasonic air detector upstream of the air filter?



9. Clamp - should be easy enough to purchase by itself

Conclusions/action items:

These components are all essential for safety - my misconception about the pressure sensors was that they were both for measuring the donor's blood pressure, but they are actually needed to detect if the tubing kinks or leaks or gets blocked, so they are necessary in those exact locations. The air chamber design of the COBE Spectra is a bit odd - the manual says it detects air when the 20mL container is 3/4 full but can also detect foam. Our bubble trap has not been integrated yet, but we may need to consider including it upstream of the air detector housing. Maybe it could rest on top of the black housing? Will need to physically see the device to be sure.

20201007 - Introduction/Motivation Research

ADITYA AILIANI - Oct 07, 2020, 11:50 AM CDT

Title: Introduction/Motivation Research

Date: 10/7/2020

Content by: Aditya

Present: Aditya

Goals: To research a broader motivation for our tubing project for the preliminary design report.

Content:

Motivations include:

-Cheaper tubing for veterinary use of plateletpheresis devices

-Also because equine plateletpheresis can be used for human therapies in the future(client's WEPLEX work)

[1] This source establishes some general information about plateletpheresis. In particular, it mentions that 75% of platelet doses used in the US are from apheresis donations. I did research on this for another class, and found that this review from Transfusion shows that, in 2017, over 2.5 million platelet doses were distributed in the US [2].

[3] This source shows that apheresis can be used in veterinary medicine, but it is still emerging. In the Introduction, I will also cite Sumner et al and the small dog plateletpheresis study (see Initial Research) to show that plateletpheresis has been validated in animals, extracting platelets for different uses. Our client's WEPLEX study claims that equine blood components have a number of benefits for cross-species therapies. This should be a point of further research for the final report.

I will describe the main barriers to equine plateletpheresis as cost (according to our client - all the cost-effectiveness studies I have seen are for human plateletpheresis) and suboptimal performance (see .Max blood processing Calculations).

Because the WEPLEX study examined the anti-inflammatory properties of WEPLEX in mice with experimentally-induced colitis, it would be reasonable to include some information on widespread impact of inflammatory bowel diseases in humans to give some scope to the project. Veterinary medicine might be more relevant without the WEPLEX study, but this study's conclusion emphasized the cross-species use of lyophilized WEPLEX. I will use CDC data for how many adults were impacted in 2015 [4]. It seems that IBD mostly affects Hispanic/non-Hispanic whites who live in poverty in America, but there were a lot of other populations listed as well. Based on feedback for this report, I might include the population in the Introduction for the final report.

Finally, there is a previous designs section of the Introduction. I will briefly describe the research in Tubing Set Patents (20200930) from the Competing Designs notebook. One important factor that I'm unsure about is that the 2012 patent for disposable tubing (most recent) has a different design from the 2000 tubing our client is using. Some differences are to be expected because the 2012 set is primarily for plasma protein extraction, not platelet extraction, but the patent says a similar approach can be used for both. In addition, the 2012 patent seems to have one block with 4 pumps rather than the 2 cartridges with 2 pumps each that we have seen in the COBE Spectra.

Conclusions/action items:

Remaining questions: how prevalent are diseases that require platelet therapies in veterinary medicine? Why does the 2012 tubing patent design use a four-pump manifold as opposed to the older two-pump cartridge?

Action Items - summarize this information in the Introduction of the preliminary design report, see if other patents can give clues about where to simplify tubing designs. My research in that area has been inadequate - I know there are more patented designs, but it takes a long time to sift through the patents that primarily focus on process control. Once the preliminary deliverables are done, I will see if the rest of the team thinks this is a useful path forward and maybe more people can work on this.

Sources:

[1]https://www-sciencedirect-com.ezproxy.library.wisc.edu/science/article/pii/B9780123744326000063

[2]Jones, Jefferson M., Mathew R. P. Sapiano, Alexandra A. Savinkina, Kathryn A. Haass, Misha L. Baker, Richard A. Henry, James J. Berger, and Sridhar V. Basavaraju. “Slowing Decline in Blood Collection and Transfusion in the United States – 2017.” Transfusion 60, no. S2 (2020): S1–9. https://doi.org/10.1111/trf.15604.

[3]Suter, Steven. “Apheresis in Companion Animals.” In *Clinical Small Animal Internal Medicine*, 1369–72. John Wiley & Sons, Ltd, 2020. https://doi.org/10.1002/9781119501237.ch156.

[4] CDC. “Data and Statistics-Inflammatory Bowel Disease Prevalence (IBD) in the United States,” August 18, 2020. https://www.cdc.gov/ibd/data-statistics.htm.

20201127 - Platelet, WEPLEX Physiology (Report Edits)

ADITYA AILIANI - Nov 29, 2020, 4:29 PM CST

Title: Final Report Edits - Platelet, WEPLEX Physiology

Date: 11/27/2020

Content by: Aditya

Present: Aditya

Goals: To conduct background research on platelet physiology and conclusions from the WEPLEX study to add to the final report.

Content:

One of the comments we received was that the introduction did not have enough specific information on the following topics: HOW platelets are involved in healing, conclusions of the WEPLEX study, existing apheresis for animals, and existing devices with different tubing designs.

Platelet physiology [1]:

This source describes platelet function in great detail. Mainly, it seems like activated anucleate platelet cells help make thrombin, which converts fibrinogen into a fibrin network, which forms the clot. The article goes on to describe how platelets secrete different types of growth factors that recruit stem cells for wound healing and both soft and bony tissue regeneration. I don't think it will be necessary to go into so much detail in the report, but it is important to briefly note that platelets have functions outside clotting that promote wound healing, which is why they are so useful in transfusions and injections. One example the article uses is that autologous platelets are used to accelerate bone regeneration.

WEPLEX Study Results, Conclusions [2]:

The study used three experiments to assess the efficacy of WEPLEX: T cell suppression, macrophage polarization, and treatment of mice with experimentally-induced colitis

-T cell: T-cell suppression is a marker of how well the product could reduce an inflammatory response. Cultures of human and equine T cells were treated with varying concentrations of WEPLEX extract, and 1% v/v WEPLEX was shown to substantially decrease T cell proliferation

-Macrophage polarization: Peritoneal macrophages (from the lining of the abdominal cavity) were extracted from mice. The researchers cultured macrophage monolayers with 0.01-0.1 % v/v WEPLEX and measured IL-10 concentrations. The idea was that resting macrophages can be "polarized" to the IL-10+ M2 phenotype, which has an immunosuppressive function. The study found significant concentrations of secreted IL-10 in WEPLEX cultures and none in cultures without WEPLEX.

-DSS-induced colitis in mice (most dramatic): Mice were given water with DSS, a chemical that is toxic to epithelial tissue and results in colon damage and inflammation. Treated mice were given intra-peritoneal injections of WEPLEX (lyophilized and fresh). Treated mice has significantly greater body weight recovery over the duration of the study and greater survival rates than those treated with the control (PBS).

Current state of animal apheresis:

This review from 2020 claims that apheresis is not widely used for animals, but the COBE Spectra has been validated for dogs and horses (it cites the same studies that Prof Brounts showed us) [3]. The only other system validated for use in horses was the Fresenius AS104, and that was for plasmapheresis. This also seems to be an outdated design, and I couldn't find any tubing diagrams or user manuals for this system. According to a 2018 review of apheresis systems, the COBE Spectra was the main workhorse for apheresis before Terumo discontinued service in order to modernize its products [4].

The other major apheresis device is the Baxter-Fenwal CS 3000+, which seems more complicated than the COBE Spectra. It uses two centrifuge chambers and, instead of separating blood within a circular tubing set, it processes blood in a large pouch that fits into the centrifuge [5]. It uses two centrifuge chambers - one two process whole blood and extract platelet-rich plasma, and the other to process the platelet-rich plasma and output platelet-poor plasma to be returned to the donor.This study used a "monitor pack" for patient safety - it is not clear how they did this, but they did not report that they had to change or override any existing sensors in the CS 3000. The study was from 1983, so we probably wouldn't be able to replicate its results with newer devices anyways. Source [5] does have a useful diagram of how computer control with RBC sensors works to reverse the centrifuge.

Conclusions/action items:

Update the report with these findings, except maybe the Baxter-Fenwal design. I'll consult with the team on this, because it might have been useful earlier in the design process, but right now it might not be relevant to have a tubing design with a centrifuge chamber when we know we have to use a centrifuge loop anyways.

Sources:

[1] Nurden, Alan T., Paquita Nurden, Mikel Sanchez, Isabel Andia, and Eduardo Anitua. “Platelets and Wound Healing.” *Frontiers in Bioscience: A Journal and Virtual Library* 13 (May 1, 2008): 3532–48.

[2] Pennati A, Apfelbeck TM, Brounts SH, Galipeau J. Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical [published online ahead of print, 2020 Aug 28]. Tissue Eng Part A. 2020;10.1089/ten.TEA.2020.0160. doi:10.1089/ten.TEA.2020.0160

[3] Suter, Steven. “Apheresis in Companion Animals.” In *Clinical Small Animal Internal Medicine*, 1369–72. John Wiley & Sons, Ltd, 2020. https://doi.org/10.1002/9781119501237.ch156.

[4] Maitta, Robert W. “Current State of Apheresis Technology and Its Applications.” *Transfusion and Apheresis Science* 57, no. 5 (October 1, 2018): 606–13. https://doi.org/10.1016/j.transci.2018.09.009.

[5] Buchholz, D. H., J. H. Porten, J. E. Menitovea, L. Rzad, R. R. Bucheger, R. H. Aster, A. T. Lin, and J. Smith. “Description and Use of the CS-3000 Blood Cell Separator for Single-Donor Platelet Collection.” *Transfusion* 23, no. 3 (1983): 190–96. https://doi.org/10.1046/j.1537-2995.1983.23383224893.x.

ADITYA AILIANI - Nov 27, 2020, 8:22 PM CST

Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical

Andrea Pennati, MS, PhD[1], Taylor Apfelbeck, BS[1,2], Sabrina Brounts, DVM, MS, PhD[2] and Jacques Galipeau, MD[1]

[1]Department of Medicine, University of Wisconsin, School of Medicine and Public Health and University of Wisconsin Carbone Cancer Center, [2]Department of Surgical Sciences, School of Veterinary Medicine, Madison, WI, USA 53706

Corresponding Author: Jacques Galipeau, MD [jgalipeau@wisc.edu]

Mailing address: 1111 Highland Ave, 3009 Wisconsin Institute for Medical Research, Madison, WI 53705

Telephone: (608) 262-6701, Fax: (608) 265-3913

platelet_rich_plasma_1_.pdf(1 MB) - download

20200914 - COBE Disposable tubing

ADITYA AILIANI - Sep 14, 2020, 1:18 PM CDT

Title: COBE Disposable tubing

Date: 9/14/20

Content by: Aditya

Present: Aditya

Goals: To get product specifications on disposable tubing sets used in the COBE Spectra device

Content:

This edition was published in 2005 and is valid for versions 4.7, 5.1–5.9, 6.0–6.9, 7.0–7.9 (not sure what the client's model number is right now) [1]. This entry will focus on the safety information in the Preface (section 1.7 mainly), information on Disposable Tubing Sets (2-21), and sections of Chapter 4 (machine setup) detailing the use of the tubing sets.

Warnings relevant to tubing (Preface):

-Take precautions to avoid air bubbles in access/return lines

-Only qualified individuals should perform equipment modifications and Gambro BCT must approve them in writing (Dr. Pennati is qualified, so maybe not an issue?)

-DO NOT use any disposable tubing sets if access needle is disconnected from tubing set

-Lists a set of indications that a tubing set is not functionally closed (able to be opened/exposed and must be sterilized before use [2])

-Tubing should allow for sample-taking without air embolism (check client's procedure to see if that is something we need to consider). It seems like an embolism due to sample taking would come from user error, but our tubing should reproduce the functionality rather than being a simple length of tube.

-Disposable tubing sets are sterilized by EtO before use (no longer sterile if end caps are not in place) and should not be sterilized again.

-Tubing sets should not be kinked or occluded (clotting is a concern). Interior diameter must be larger than that of the needle.

-Extreme conditions that should not be combined: Room temp > 27.5 C, 2400rpm centrifuge speed, Inlet pump flow rate <= 25 mL/min

-Tubing must be checked before use, as it can fail (leak). Also, don't stretch tubing.

Safety Information:

-Classified as Type B equipment according to EN 60601-1 standards for electrical equipment [3]. Mainly seem to be eletrical hazards (biohazardous info?).

-(pg. 11-4): EtO residuals from sterilization can cause reactions in some donors [5] - there is an alternative single-pass priming procedure to address this.

Disposable Tubing:

-Several types of tubing sets based on use. Based on the Sumner et al. paper that our client said was a proof of concept for plateletpheresis on horses, the tubing we are looking for is Dual-Needle Extended Life Platelet (ELP) disposable tubing set with LRS (trademarked LeukoReduction System) chamber [4]. The tubing sets had to be primed, and Sumner et al. used 0.9% NaCl and 20cc acid-citrate dextrose formula a from Fenwal [4].

-This set can be primed 3x (depends on volume of waste bag [included in the set])

-Apparently the LRS setting eliminates the need for post-collection leukoreduction. Fewer immune cells in the blood is better for plasma-based treatments.

-Based on the instructions alone, it is hard to understand how the tubing connects to various ports and chambers. Some of the tubing is multi-lumen tubing.

Conclusions/action items:

Source:

[1]*COBESpectra Apheresis System Essentials Guide.* Gambro BCT, Inc., Lakewood, CO, USA, 2005.

[2] https://www.biospherix.com/blog/how-closed-systems-is-a-functionally-closed-system#:~:text=Functionally%20closed%3A%20A%20process%20system,step%20prior%20to%20process%20use.

[3] International Electrotechnical Commission. International standard for medical equipment, Part 1: General requirements for safety, IEC 601-1. 2nd ed. Geneva, 1988.

[4] https://onlinelibrary.wiley.com/doi/full/10.1111/trf.14124

[5] Leitman SF et al. New England Journal of Medicine,1986;5(19):1192-6.

20200916 - PDS - Standards and Specifications

ADITYA AILIANI - Sep 16, 2020, 9:34 PM CDT

Title: PDS - Standards and Specifications

Date: 9/16/20

Content by: Aditya

Present: Aditya

Goals: To organize research for the "Standards and Specifications" section of the Product Design Specifications. Likely sections include general device-handling safety (electrical standards), biosafety (for handling blood), and testing/material requirements (for tubing)

Content:

Ctrl+F searched the IEC Standard 60601-1 pdf for the following keywords: "type b", "sterilization", "protective earthing"

From the COBE Spectra Apheresis manual, the device is subject to regulations on electrical safety - it is a Type B device according to IEC Standard 60601-1, which regulates medical electrical equipment [1]. Type B is the least regulated equipment in this standard because the patient connections are not electrical [2]. We will not be changing any electrical components of the device, so electrical safety is a concern only in the general sense and we will not need to include safety measures beyond user expectations. One point of confusion I have is that the IEC standard I'm looking at defines Classes (I and II) for equipment, while individual parts are labelled as Type B, BF, or CF. The Spectra user manual refers to the entire system as Type B, but this may be from using an older edition (2nd edition vs. the 3rd edition I'm looking at).

IEC document cited an ISO 11134 standard for moist heat sterilization and 11135 for ethylene oxide [2]. I can't access the full text of these right now - check library access later.

FDA document describing environmental hazards of EtO sterilization has a helpful overview of sterilization standards [3]. It also cites ISO 11134 - apparently it has a detailed description of how sterilization techniques are validated. One validation method takes the "bioburden" (number of microbes and their resistance) into account, using a biological indicator - this document did not have the details [3]. One of the biggest concerns with EtO, apart from the environmental hazard, is the fact that materials can accumulate EtO residuals, which are hazardous. The FDA document cites ISO/ANSI/AAMI 10993-7, which describes acceptable maximum EtO residual levels [4]. The cited source is just part of the introduction - I couldn't see the full text. As things stand, those seem to have good specific information on how we can test our final system, but actually testing these materials might be a problem.

Searching the FDA CFR21 website, I found that blood collection devices (includes tubing, serum collection vials, collection trays) are considered Class II medical devices, so they need premarket notification but usually not approval [5]. Also, there is a FDA Device lookup tool that showed me the details on regulation for the COBE Spectra device - it was not classified (Pre-Amendment) but was subject to premarket review [6]. Reviewing the COBE Spectra manual for tubing-related specifications, I found a warning against kinking and the upper and lower pump pressure limit: -250 to 450 mmHg [1]. In the "Safety Certifications" section, the tubing inner diameter is written as 0.113 in (larger than the inner diameter of the 17 gauge needle used).

US Patent search "apheresis tubing"

Found a patent for an apheresis tubing set with a cell sampling port and collection bag [7]. This type of tubing is used in the COBE Spectra system. Preferred materials are PVC for the tubing and Teflon for the collection bag.

Conclusions/action items:

Between the US Patent for currently available apheresis tubing, FDA and ISO regulations on sterilization procedures, and the COBE Spectra's user manual, we have a number of quantitative specifications on how the tubing should perform (once we can read the ISO regulations). A few questions remain about material properties - a few of these sources mentioned avoiding certain plasticizers (make this another entry), but I haven't found much information yet on how much of a concern physical reactions are to currently available medical tubing. Also, I haven't found much information yet on mechanical failure of the tubes, aside from the working pump pressures.

I will put this information into the Standards and Specifications section of the PDS. Some of it may overlap with the "Safety" or "Materials" sections, but that can always be repeated. We are the ones using this document to keep track of specifications.

Sources:

[1] *COBESpectra Apheresis System Essentials Guide.* Gambro BCT, Inc., Lakewood, CO, USA, 2005.
Available: http://startrinity3.com/mssn/04/Apheresis%20System%20Essentials%20Guide.pdf

[2] https://www.ele.uri.edu/courses/bme484/iec60601-1ed3.0_parts.pdf

[3] https://www.fda.gov/media/132186/download#:~:text=The%20FDA%20regulation%20of%20EtO,for%20a%20specific%20medical%20device.

[4] https://www.fda.gov/media/132186/download#:~:text=The%20FDA%20regulation%20of%20EtO,for%20a%20specific%20medical%20device.

[5] https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=862.1675

[6] https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfpcd/classification.cfm?id=2243

[7] https://patentimages.storage.googleapis.com/32/12/1d/4328859f8364be/US20080103428A1.pdf

20200930 - Tubing Set Patents

ADITYA AILIANI - Oct 05, 2020, 9:57 AM CDT

Title: Tubing Set Patents

Date: 10/5/2020

Content by: Aditya

Present: Aditya

Goals: To look through Google Patents and identify platelet collection patents that could give us useful diagrams of the tubing.

Content:

We have had difficulty analyzing the actual tubing set because it is so complex, and only one of us can handle it at a time. Therefore, our goal is to find a labelled schematic that we can study individually and just use the physical tubing set for clarification of exactly how each part is put together. I used the search terms "apheresis tubing set", "COBE Spectra disposable tubing set", "platelet collection tubing", and "tubing pump cartridge" on Google Patents and filtered by assignee (Terumo BCT, Gambro BCT, Caridian BCT)

https://patents.google.com/patent/US8123713B2/en?q=COBE+Spectra&assignee=Terumo+Bct%2c+Inc.

This patent was granted in 2012 to Terumo BCT. The body doesn't describe platelet collection in detail, but says it can be done with a similar procedure (it describes plasma protein extraction). However, the included tubing diagram only has one block where the tubing connects to four peristaltic pumps, and from pictures of our tubing set, we know our tubing set has a set of two cartridges that connect to two pumps each.

https://patentimages.storage.googleapis.com/db/b3/05/401a1d9ed3accf/US7780618.pdf

This patent was granted in 2010 to Terumo BCT. It goes into more detail about the feedback loops involved with the pressure sensors - this would be good to review for when we consider the device parameters (our priority now is the actual tubing structure). However, the tubing diagram in this patent also does not include two pump cartridges.

https://patentimages.storage.googleapis.com/3b/d8/93/08f884c3845513/US6730055.pdf

This patent was granted in 2004. It goes into more detail about the structure of the centrifuge, which is not entirely relevant for tubing. It also has a single set of four pump connections, so this design may be too recent for our use.

https://patentimages.storage.googleapis.com/24/79/35/7e4d1b2df2db28/US4824339.pdf

This patent was granted in 1989 to COBE laboratories. It only describes the pump cartridge system and has helpful diagrams for how the tubing actually fits into the cartridge. This will be helpful while we are looking at the physical tubing set because now we can track each tubing line through the pump system.

https://patentimages.storage.googleapis.com/6b/4f/3b/eac079365762ac/US6022306.pdf
I think this is the patent we are looking for - it was granted in 2000 to COBE Laboratories. It includes a numbered diagram of the tubing set that closely matches the physical tubing set we have, with two pump cartridges and 4 attached bags. This is the diagram we can include in our preliminary report, and further research can look into what each part of the tubing system does.

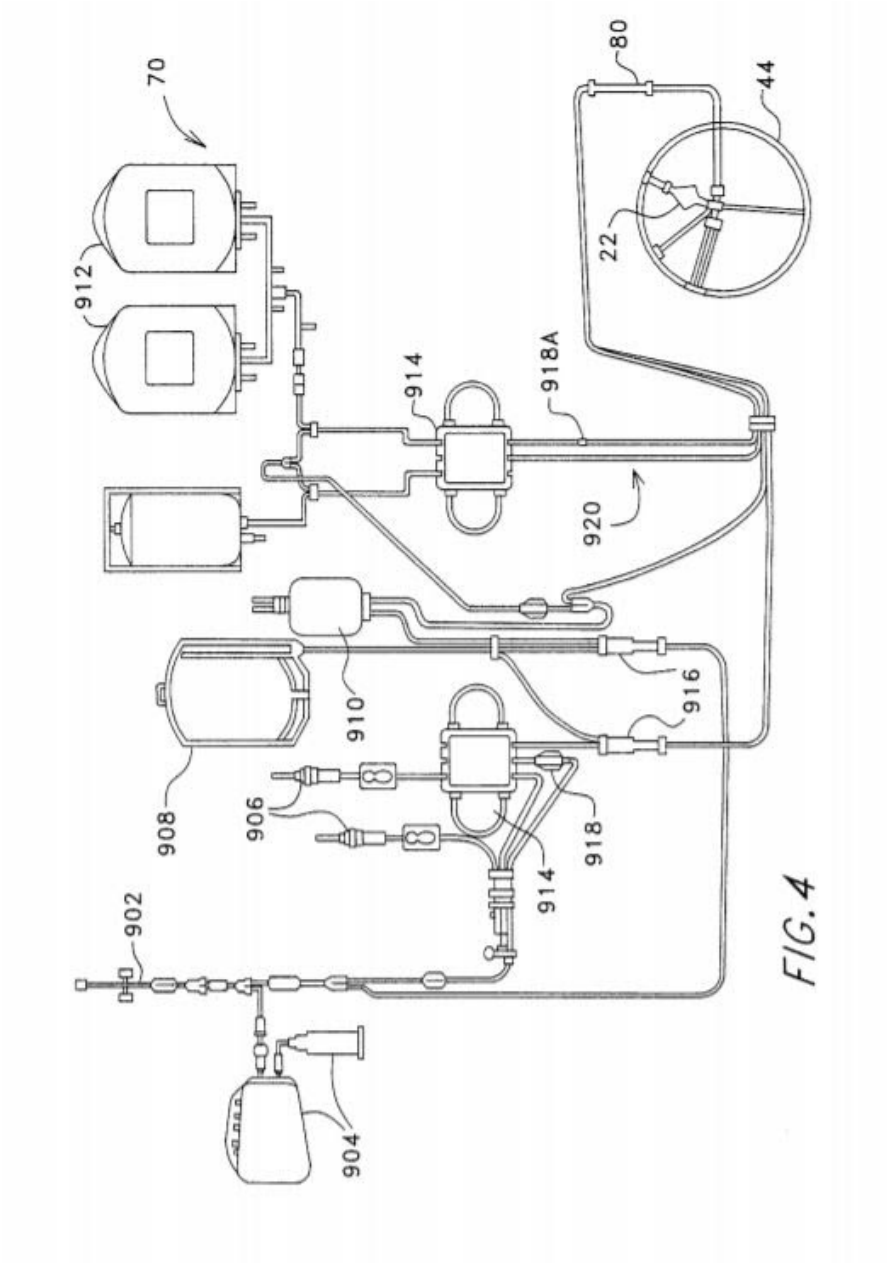


FIG. 4

Conclusions/action items:

Use the 2000 patent to understand how the tubing components fit together. It is unlikely that this will be ready by the preliminary design report, but it will be useful information for the client meeting next week. We can discuss how to simplify the tubing in a meaningful way now that we have information on the structure.

20201005 - ELP Tubing Set company specifications

ADITYA AILIANI - Oct 07, 2020, 11:51 AM CDT

Title: ELP Tubing Set patents, company specifications

Date: 10/5/2020

Content by: Aditya

Present: Aditya

Goals: To look through specifications received from a Terumo representative

Content:

Last week, I made an account with the Terumo BCT website and sent a customer support request for a schematic of the Dual-Needle Extended Life Platelet Set with Leukoreduction chamber. A representative emailed me back with dimensions (weight, wall thickness) and a diagram of the tubing set, which is in the Project Files folder. Here, I will review the diagram and make note of additional parts we will need to purchase or simplify.

General overview:

-The tubing set is first primed with saline from the access saline line and anticoagulant from the anticoagulant line. This flushes air out of the tubing before it is attached to the patient. The tubing set is flushed with saline again in the "Rinseback" stage, when the procedure ends and RBCs are completely returned to the patient. Both the access saline and anticoagulant lines pass through 0.2 micron filters [2].

-During the procedure, whole blood is pumped into the inlet line, where it is mixed with anticoagulant pumped from the anticoagulant line. This section uses two peristaltic pumps: one to pump anticoagulant in, one to pump anticoagulated blood in. The anticoagulated blood passes through a pressure sensor, then through the pump cartridge (here it wraps around the corresponding peristaltic pump) [1].

-From the pump cartridge, the anticoagulated blood moves into a 0.2 micron filtered air chamber. Air and excess saline is diverted upward into a waste bag, while the anticoagulated blood moves into the centrifuge loop [1].

-There are four tubes connected to the centrifuge loop: anticoagulated blood (moving into the centrifuge), leukoreduced platelet concentrate (moving out of the centrifuge), plasma (moving out of the centrifuge), and RBC/WBC concentrate (moving out of the centrifuge). These are held together by a four-tube connector and merged into a large four-lumen tube [1].

-The four-lumen tube is surrounded by a protective mesh and is secured into the centrifuge chamber with two bearings ("upper" and "lower"). The start of the four-lumen tubing is marked by an "upper" collar and ends with a "centrifuge collar", where it splits into four different tubes [1].

-Anticoagulated blood enters the inlet chamber of the centrifuge loop, and the components exit the collection chamber. There is a separate outlet for leukoreduced platelets, which must pass through a leukoreduction chamber [1] <-Need to research how this works

-Platelet concentrate passes through a CCM (collection concentration monitor-checks platelet concentration, RBC contamination), which seems to be an optical sensor in the COBE device - a cuvette that wraps around the tubing must lock into the CCM housing (see COBE Essentials Guide, 4-7) [2]. It then passes through a second pump cartridge, where a collect/replace peristaltic pump is housed. This pumps the platelet concentrate into collection bags (up to 3) [1, 2]. These bags can be closed off with attached clamps. The collect/replace pump can either pump collected cells into bags or into the return line - it seems to me that, for our purposes, we only need the collection bag. However, I'm not sure if this causes problems during the Rinseback stage.

-The plasma line moves from the centrifuge loop, through the four-lumen tubing, over a plasma collect/replace peristaltic pump. It then goes into a valve that can either direct it to a plasma collection bag or to a return line [2]. We might be able to get rid of this part of the tubing - I don't think our client has any need for plasma. The return plasma goes through a pressure sensor before merging with the RBC/WBC line from the centrifuge loop to go into a return air detector.

-The plasma/RBC line valve also has an RBC detector that closes the waste divert valve and opens the return line valve when it detects RBCs [2].

-The return air detector has an input of the combined return RBC/WBC/plasma and outputs air/saline to waste (same waste bag as before) and RBC/WBC/plasma to return to the patient [1].

-The return fluids are mixed with return saline (also has a 0.2 micron filter) before going into the return needle back to the patient [1].

List of parts:

-Plastic casing for pump cartridge (can reuse from old set)

-Bags: access saline, return saline, waste, plasma, collection x 3. Plasma and platelet collection bags are made from "citricized PVC" [1].

-Pressure sensor connectors x 2: access and return lines (look like plastic buttons that click into slots on the front panel) [2]

-Air detector chambers x 2: access, return (there is an anticoagulant ultrasonic air detector, but it seems to detect air in the AC line itself rather than in a bubble chamber - possible route for simplification)

-CCM cuvette: clear plastic casing around platelet collect line that allows it to lock into the CCM housing

- Four-lumen connector: Holds plasma, RBC, AC blood, and platelet collect lines together before entering the protective mesh sleeve

-Collars: 2x, one upper one lower to hold the four-lumen tubing to the centrifuge

-Mesh sleeve, hard protective sleeve, centrifuge channel: protects tubing in the centrifuge loop

-Bearings: 2x, one upper one lower to attach the loop to the centrifuge arms **The size of the bearings and collar may limit our ability to change tubing diameter - we will need to see how these interact with the centrifuge**

https://www.youtube.com/watch?v=DbKd7tkJR_Q&t=139s -> From this video, the hexagonal centrifuge collars fit exactly into two levers inside the device. **We will not be able to change the size of the four-lumen tubing without making modifications to the device itself.**

-Spikes, drip chambers x3: access saline, return saline, anticoagulant

-Flow controller (w/ roller): x1, return saline

-0.2 micron filters x7 (at least): access saline, return saline, anticoagulant, inlet air detector, return air detector, access line, return line

-H connectors: x2 for plasma/RBC return/collect valve, platelet concentrate return/collect valve

-Y connector x1: waste divert valve

-ACCESS LINE equipment

->needle, needle protector (MasterGuard (R) Anti-Stick), clamp, access luer connection

->Plastic connections for "access manifold" (where AC, access saline, and inlet lines meet)

-RETURN LINE equipment

->needle, clamp, return luer connection

->Plastic connections for "return manifold" (where return saline and return lines meet)

-Leukoreduction chamber - ???

Conclusions/action items:

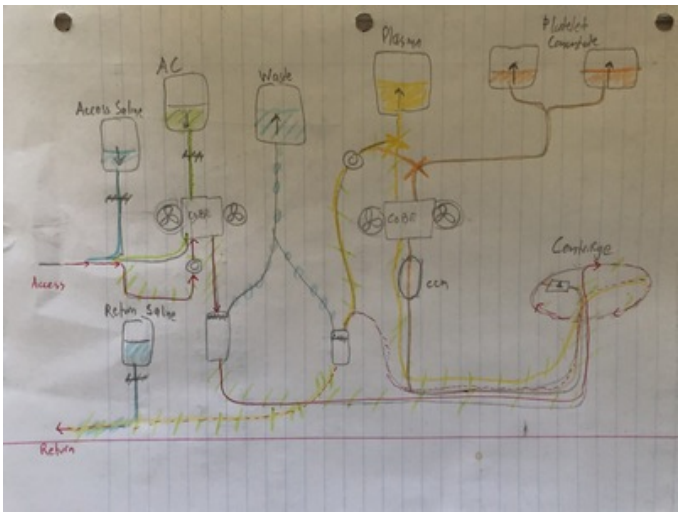
The COBE Essentials Guide refers me to a "Platelet Collection Guide" for more information on the leukoreduction system. If this could be removed from the tubing system somehow, it could help simplify the design so we could fabricate it. I am not hopeful about pursuing this strategy, though, because of the complexity of the system and the number of sensors that must connect to the tubing. One possibility might be cannibalizing this set of tubing for hard plastic parts, but that is very risky. The tubing is expensive, and we are unlikely to get it right the first time.

Sources:

[1]COBESpectra Apheresis System Essentials Guide. Gambro BCT, Inc., Lakewood, CO, USA, 2005. Accessed September 17, 2020. http://startrinity3.com/mssn/04/Apheresis%20System%20Essentials%20Guide.pdf

[2]Terumo BCT. "Dual-Needle Extended Life Platelet Set with LRS® Chamber
Catalog Numbers 777003-015, 70300" (2007). Available from Terumo, BCT upon request.

ADITYA AILIANI - Oct 05, 2020, 11:24 AM CDT



ApheresisTubingCartoon.jpg(2 MB) - download Sketch of ELP tubing. Red = whole blood, dotted red = RBCs, blue = saline, hashed blue = saline/air, green = anticoagulant, yellow = plasma, orange = platelet concentrate. Filters, centrifuge, pumps, air detectors, pressure detectors, and collect monitor are depicted.

20201006 - Testing Research

ADITYA AILIANI - Oct 07, 2020, 10:27 AM CDT

Title: Testing Research

Date: 10/6/2020

Content by: Aditya

Present: Aditya

Goals: To research options for testing our tubing to present them in the preliminary design report

Content:

The tubing should resist kinking, be easily compressed in the peristaltic pumps, not leak fluid, and be sterilizable (preferably by autoclave)

Material Properties:

[1] This article in Medical Design Briefs has a lot of useful information on testing medical-grade tubing. It describes a simple test to measure a tube's resistance to kinking: the ends are put over each other into a loop, and the loop is pulled tighter until the tubing wall collapses. The reported value is the minimum bend diameter - the final diameter of the loop. The article also describes tests to measure elastic modulus and necking, but because we are not manufacturing the plastic ourselves, these tests are likely unnecessary. Even the bend test is probably unnecessary if we are purchasing medical-grade tubing, but it should be done to double-check.

Performance:

The tubing should not leak saline or horse blood.

Nesya found a paper in which the authors made a substance to mimic horse blood [2]. This was for forensic purposes, and they measured its ability to mimic equine blood by looking at the bloodstain patterns it made. They found that the following formulation (ingredient brands in parentheses - they are British) came closest to mimicking horse blood in stain patterns:

Superfine Plain Flour (Waitrose) 39.6g
D(+) Glucose Anhydrous (GPRTM) 5g
Glycerol (Supercook) 2ml
Sodium Chloride (Sigma) 0.7g
Bovine Serum Albumin (Sigma) 1.5g
Sterile Distilled Water 125ml
Scarlet Food Colouring (Supercook) 2ml

This will have to be verified in a viscometer to see if it actually mimics flow properties of equine blood. The paper did not mention an actual viscosity value. Source [3] describes several whole blood viscosities, depending on shear rate, so I will have to calculate shear rate for this system (I think it's dvz/dr, but I'll check with Prof. Kinney).

Sterilization:

I couldn't find FDA guidelines on apheresis tubing, but there was a set of guidelines for hemodialysis tubing, which should be relevant [4].

These guidelines recommend tests for sterility as well as pyrogenicity. Pyrogens are usually bacterial endotoxins that induce fever and are a concern in any blood handling device. However, they seem to be handled using dry heat, not moist heat as our client recommended. For dry heat, there are LAL assays available, which form a gel clot when endotoxins are detected. They're mostly used for dry heat, but they could be used for moist heat (it's not as effective for endotoxins) [4, 5].

For regular biological indicators, I found this company (Steris Life Sciences) that sells small (0.25 in diameter) spore discs and suspensions to inoculate surfaces with bacterial spores [6]. These could be put into tubing to check that the interior is sterile. However, I'm not sure if any ends of the tubing could be open for us to insert the disc.

Source [7] has good general information on autoclave cycles and developing new protocols. In particular, it mentions that a vacuum cycle (as opposed to a usual gravity cycle) is best for tubing, as it makes sure all air from the tubing product is purged before sterilization.

Conclusions/action items:

Summarize this information in the Testing section of the report. Major questions that arose were: should we check for endotoxins? If so, can we consider a dry heat sterilization protocol? How exactly should the tubing be placed in the device (should it be packaged? sealed off?)?

Sources:

[1]https://www.medicaldesignbriefs.com/component/content/article/mdb/features/applications/14643

[2]http://edge.rit.edu/content/DRIL_Modeling/public/Nicole_Varble/Articles/Viscosity/Millington%20%282000%29%20development_of_synthetic_blood_substitute.pdf

[3]https://physoc.onlinelibrary.wiley.com/doi/epdf/10.1113/eph8802496

[4]https://www.fda.gov/media/71429/download

[5]https://www.fda.gov/inspections-compliance-enforcement-and-criminal-investigations/inspection-technical-guides/pyrogens-still-danger

[6]https://www.sterislifesciences.com/products/biological-and-chemical-indicators/biological-indicators

[7]https://consteril.com/sterilization-cycle-development/

20201009 - Reusable Tubing Sets

ADITYA AILIANI - Dec 09, 2020, 11:14 AM CST

Title: Reusable Tubing Sets

Date: 10/9/2020

Content by: Aditya

Present: Aditya

Goals: To look for patented reusable blood tubing

Content:

https://patentimages.storage.googleapis.com/82/42/0c/7d30b1001d7373/EP2334412B1.pdf

This is a patent for hemodialysis tubing that has some actual specifications for how the tubing should interface with pressure sensors.

https://patentimages.storage.googleapis.com/0f/48/eb/762977f2cc192e/WO2002070042A1.pdf

This is a patent for apheresis tubing. The Ctrl+F function doesn't work on it, so I will have to read through this carefully to see if there are any sterilization protocols. It also has a lot of detailed information on each of the safety features - they might be different from the Terumo set, but the general information should be useful. Update - the safety features seem to be similar to general extracorporeal circuit features.

https://www.smiths-medical.com/-/media/M/Smiths-medical_com/Files/Import-Files/VE195802EN-112017.pdf

This is a catalog for veterinary medical products. Apparently equine uterine flushing tubing and fluid delivery tubing are made of silicone and can be autoclaved and reused.

Conclusions/action items:

There does not seem to be precedent for reusing blood tubing. I have not yet looked into veterinary hemodialysis, so that might yield results, but for now no one on the team has yet found any research to suggest that tubing that carries blood can be cleaned as sterilized for reuse.

20201026 - COBE Spectra associated patents

ADITYA AILIANI - Dec 09, 2020, 11:17 AM CST

Title: COBE Spectra associated patents

Date: 10/26/2020

Content by: Aditya

Present: Aditya

Goals: To categorize the patents listed on the COBE Spectra Essentials Guide

Content:

US 4,468,219: Describes feedback loop for maintaining constant flow rate of biological fluids using a peristaltic pump

https://patents.google.com/patent/US4468219A/en?oq=+4%2c468%2c219

US 4,647,279: Describes centrifugal separator (does not go into detail on tubing within centrifuge loop)

https://patents.google.com/patent/US4647279A/en?oq=US4%2c647%2c279

US 4,674,962: Describes peristaltic pump specifications used in COBE Spectra

https://patentimages.storage.googleapis.com/1d/0a/11/d2596d14efc470/US4674962.pdf

US 4,708,712: Describes a continuous loop centrifugal separator with a dam for separating heavy-phase components

https://patents.google.com/patent/US4708712A/en?oq=US+4%2c708%2c712

US 4,795,314: Describes control system used to obtain a **specified volumetric flow rate** through the pumps

https://patents.google.com/patent/US4795314A/en?oq=US4%2c795%2c314

US 4,810,090: Describes concentration collect monitor

https://patents.google.com/patent/US4810090A/en?oq=US+4%2c810%2c090

US 4,824,339: Describes cartridge for organizing tubing through the peristaltic pumps

https://patents.google.com/patent/US4824339A/en?oq=US+4%2c824%2c339:

US 4,850,995: Describes tubing apparatus for returning RBCs to the donor

https://patents.google.com/patent/US4850995A/en?oq=US+4%2c850%2c995

US 4,861,242:Describes peristaltic pump with an external rotor (front panel of the device)

https://patentimages.storage.googleapis.com/da/27/76/c3596cfa306300/US4861242.pdf

US 4,900,298: Centrifuge drive and support assembly

https://patents.google.com/patent/US4900298A/en?oq=US+4%2c900%2c298

US 4,978,446: Describes the use of filters for sterile blood component collection

https://patents.google.com/patent/US4978446A/en?oq=US+4%2c978%2c446

US 4,991,743: Describes flow control apparatus (external feature described in the COBE Spectra Essentials Guide but not required)

https://patents.google.com/patent/US4991743A/en?oq=US+4%2c991%2c743

US 5,263,831: Another peristaltic pump?

https://patentimages.storage.googleapis.com/55/49/13/8be4781dc2be08/US5263831.pdf

US 5,345,670: Power magnetic devices (like inductors) for circuits

https://patentimages.storage.googleapis.com/8b/82/c2/9cd20000f53dde/US5345670.pdf

US 5,352,371: Describes how fluid flows through tubing multiple times, goes into detail on tubing connectors

https://patentimages.storage.googleapis.com/57/c2/f2/bd0f5238ba65e0/US5352371.pdf

US 5,496,265: Describes how the COBE Spectra optimizes parameters

https://patents.google.com/patent/US5496265A/en?oq=US+5%2c496%2c265

US 5,496,301: Method for sampling blood without opening the extracorporeal circuit

https://patents.google.com/patent/US5496301A/en?oq=US+5%2c496%2c301

US 5,611,997: Describes overall process control method for collecting platelets

https://patents.google.com/patent/US5611997A/en?oq=US+5%2c611%2c997

Conclusions/action items:

Most of these patents are about process control and will be useful in another semester for optimizing blood flow. For now, US 4,708,712 (centrifuge loop) and US 4,674,962 (peristaltic pump) will be most useful because we need to show how complicated the centrifuge loop and potentially go into peristaltic pump specifications if none of our tubing sets can interface with them to find out why. I was initially excited about this source because it listed allowable durometers for the peristaltic pump, but it turns out that those were for the arms of the peristaltic pump itself, not the tubing that wraps around it. Maybe there is a calculation for allowable tubing hardness based on this?

20200910 - Client Meeting 1

ADITYA AILIANI - Sep 14, 2020, 11:30 AM CDT

Title: First Client Meeting

Date: 9/14/2020

Content by: Aditya

Present: Prof. Brounts, Cate, Trevor, Nesya, Lokesh, Aditya

Goals: To understand the client's goals for the project

Content:

Broadly, the client wants to decrease the cost per use of their COBE apheresis device to justify its use for making a new platelet lysate product (patent pending). Disposable tubing accounts for most of the $2000-2600 cost per use, so they would like us to either 1) find an alternative tubing source that is significantly cheaper 2) find/manufacture tubing from a material allowing for repeat use OR 3) Develop an alternate sterilization method that will not degrade the current tubing. A combination of these tasks would be ideal. The client has access to steam and gas (EtO?) sterilization methods, but steam is cheaper.

The dimensions of the current disposable tubing set are acceptable for their use in horses. Prof. Brounts advised those of us who are available (Cate and Lokesh) to visit the clinic and view the machine and tubing set (set up a meeting with Dr. Andrea Pennati). She also said we could use clinic sterilization methods (steam and gas) and the apheresis device for testing, recommending that we run water or horse blood (without connecting a horse to the device) through the machine to test our new tubing. She does not want us to destroy the tubing they have, as it is expensive.

Safety measures that would be taken in humans should be taken in animals, although there is a decreased risk of bloodborne diseases. We should find for ourselves what this will mean specifically for how we should test sterilization techniques. Our current budget is $500 (negotiable if we have to), and our goal is to bring the cost per use down to around $100. 20-25 uses per set of tubing would also be a great improvement (does not to be indefinitely reusable). Prof. Brounts did not know of a way her goals could be met if the campus was shut down and said she would consult with Dr. Pennati to brainstorm virtual possibilities. A user manual will be available at the clinic.

Another problem to consider is that the client is using an old donated model of the COBE device, and disposable tubing sets for that model are being phased out in favor of slightly different tubing for new models.

Conclusions/action items:

It seems like we should primarily look at tubing materials and try to find a 3rd party supplier to purchase tubing that we can test. Trevor has a contact in the tubing business who could help us out.

Revise problem statement with new information and use this information to guide research into safety, PDS.

20201001 - Max blood processing volume calculations

ADITYA AILIANI - Oct 06, 2020, 10:11 PM CDT

Title: Maximum blood processing volume calculations

Date: 10/01/2020

Content by: Aditya

Present: Aditya

Goals: To define the maximum volume of blood processed in the COBE Spectra given the following parameters: time (30min -1hr), catheter diameter (10-14 gauge), equine blood viscosity, and pump pressure in the device.

Content:

Dr. Galipeau informed us that, because we are using a device designed for human apheresis on horses, we will need to change the tubing diameter (and maybe the device's pressure parameters) to allow for large amounts of blood to be processed in a short amount of time. Human apheresis normally takes about 3 hours, while equine apheresis can take a maximum of 1 hour (under sedation). Dr. Galipeau referenced the above parameters in our meeting today and also said there is significant flow resistance in the catheter but negligible resistance in the veins. Prof. Brounts said she pulls blood from the jugular vein of the horse, which is around 1.5mm in diameter.

There are 4 peristaltic pumps on the device: inlet blood flow, anticoagulant inlet, plasma/RBC to collect/waste/return, and cells to collect/return. Based on the client interview, we are primarily concerned with inlet blood flow. The COBE Essentials Guide has an equation for determining anticoagulant flow rate based on total blood volume (default AC Infusion rate is 0.8mL/min/L TBV:

TBV x Configured AC Infusion Rate = AC Returned to the Donor/Patient

Human apheresis parameters - 500lb, 7ft tall female [1] Sumner et al. collected 1 L of platelet concentrate. Hematocrit, platelet count, total blood volume for horses to manually calculate infusion rate, pump rate, inlet flow rate. They reported a mean run time of 163 min and mean total blood processed of 13.1 L, implying a total flow rate of 80.4 mL/min. They used a 12 gauge needle for inlet and 14 gauge for return and ended up infusing about 1.5L ACD-A (anticoagulant - citrate dextrose solution) to each donor.

From the COBE Spectra Essentials guide, inlet pump max flow rate is 150 mL/min. We know that the maximum length of time the apheresis procedure can take place for a horse is 1 hr, which gives a max processing volume of 9L. The tubing set our client uses (Dual-Needle ELP) has a total volume of 260 mL, with 131 mL of that being blood volume. Based on this, and the fact that the inner diameter of 0.113 in, the length of tubing with blood flow is (131mL) / (pi*(0.0565in)^2). Converting mL to in^3 (131mL = 7.99 in^3), the length of tubing with blood flow is 797in.

The inlet pump max flow rate may be 150 mL/min, but the manual lists several factors for calculating inlet pump flow rate:

AC Pump Rate X Inlet:AC Ratio = Inlet Pump Flow Rate (Inlet: AC ratio is the ratio of inlet and AC pump flow rates - should not exceed 15:1 - chosen based on hematocrit)

If the max AC pump rate (12 mL/min) is used with the max inlet flow (150 mL/min), the Inlet:AC Ratio would be 12.5, which is high but technically not impossible. It would depend on the donor's hematocrit (conc. RBC).

I might have enough information to roughly apply the Hagen-Pouiseulle equation to see if I'm understanding the parameters correctly:

-Assumptions: steady-state (probably not), laminar flow (fine - peristaltic pumps should produce laminar flow), inlet pump pressure gradient can be extended to the whole device (very questionable, but just getting a rough idea so far)

Q = [pi * r^4 * delta_P] / [8 * mu * L]

r = 0.113 / 2 in = 0.001435 m (current - will change)

delta P = max inlet pump pressure: -250 mmHg (+400 mmHg outlet pressure) = 33330.6 Pa

mu = 7.1 cp = 7.1 g/ms = 0.0071 kg/ms[3]

L = 797 in = 20.24 m

Qmax = 3.862E-07 m3 / s = 2.32E-06 m3/min = 23.2 mL/min => **assumptions definitely need corrections - max flow rate is 150 mL/min**

Maybe look at general apheresis calculations for humans, see how TBV applies there.

[4] This article does not include tubing radius as part of the calculations, could could be good to look at later for TBV calculations. Recommends extracorporeal volume limited to 15% of TBV, which can be checked after we have an ideal tubing radius.

Simpler idea: Flow rate scales with r^4, so just use a proportion to decide how much radius should change.

1) Human to Equine blood flow

-Volumentric flow through a tube is inversely proportional to fluid viscosity

-Above equation used whole blood viscosity - this is highly variable and depends heavily on RBC levels [5]. If I'm just using a ratio to figure out equine blood flow relative to human blood flow, I can try using the ratio of plasma viscosity. These are the values cited in Sumner et. al (horse plateletpheresis study) from source [5]: 1.24 cp (human plasma), 1.66 cp (equine plasma)

Q_human = 150 mL/min (from COBE Essentials Guide - max inlet flow rate) [2]

Q_human / Q_equine = mu_equine / mu_human = 1.66 / 1.24

Q_equine = 112.05 mL/min

2) Current equine blood flow to target flow

Qtarget = 13.1 L / 60min = 218.3 mL/min

Qtarget / Qcurrent = 218 / 112.05 = 1.945 = r^4 ideal / r^4 current

r target / r current = (1.945) ^ (1/4) = 1.181

r target = (1.181) (0.0565 in) = 0.0667 in

Target inner diameter is 0.133 in from this method - run this by the team to see if it makes sense. Because the tubing has to be larger than the needle, this tubing would likely not be compatible with a 10 gauge needle (0.134 in outer diameter) but would be compatible with a 12 gauge needle (0.109 in outer diameter) [4].

Conclusions/action items:

We have a number for an improved inner diameter. I have a few concerns with this - namely that it doesn't take into account TBV and AC volume, it doesn't take into account the 4 different pumps with different max pressures, and that the Sumner et al. study already seems to have processed a large blood volume. We are also changing very small values in inches, so there may be error from estimating viscosity values from averages. Source [5] has some interesting physiological information on horse blood - I don't know if high levels of red blood cell aggregation is relevant right now, but it might be worth looking at later.

Sources:

[1] https://onlinelibrary.wiley.com/doi/full/10.1111/trf.14124

[2] COBE Essentials Guide

[3] https://onlinelibrary.wiley.com/doi/pdf/10.1111/j.1939-1676.1990.tb00895.x

[4] https://darwin-microfluidics.com/blogs/tools/syringe-needle-gauge-table

[5]https://physoc.onlinelibrary.wiley.com/doi/epdf/10.1113/eph8802496

20201110 - Pressure Sensor Research/Ideas

Title: Pressure Sensor Research/Ideas

Date: 11/10/2020

Content by: Aditya

Present: Aditya

Goals: To brainstorm ideas for manufacturing a cheap pressure sensor

Content:

Motivation: The pressure sensors in the COBE Spectra are small and are made of a nearly uniform material. This means they may be easy to 3D print.

[1]: This website describes a commercially available pressure transducer and was recommended for Show and Tell. It is interesting that pressure transducers can be integrated directly into the referring to are just relaying information to the transducers to keep the blood from getting into the device.

[2]: Spectra Optia Service Manual - describes how pressure sensors are integrated into the device. I could not find an equivalent manual on the Internet for the COBE Spectra. I called Terumo seems that the pressure pods in the tubing actually transmit pressure to the sensors within the device. Apparently they are "magnetically coupled" to the device. I thought this was some kind o locks the pressure pod in because it has a metal component.

[3]: Integrated pressure sensor patent- NOT what COBE Spectra uses. This describes a method for integrating pressure sensors into an extracorporeal tubing set. In this system, the pressure

[4]: Diaphragm pressure pod patent. This patent describes a "diaphragm" pressure pod that has blood running on one side of an impermeable but flexible diaphragm (silicone rubber). The diap and they refer to diaphragms that have strain gauges to directly measure the deflection of the diaphragm. This doesn't seem to use that method.

[5] PVC Sterilization guidelines. This source is from ThermoFischer and confirms that, although PVC should be able to hold up to autoclaving (but our TYGON PVC set has a max temperature for flexibility) changes with after gas sterilization. According to the Grainger site for TYGON PVC [10], our initial measurement should be 7mm.

[6] Makerspace 3D Printing website. Polypropylene seems to be autoclavable and gas sterilizable up to 200 cycles [11], so I chose to use that to describe how to 3D print a pressure pod.

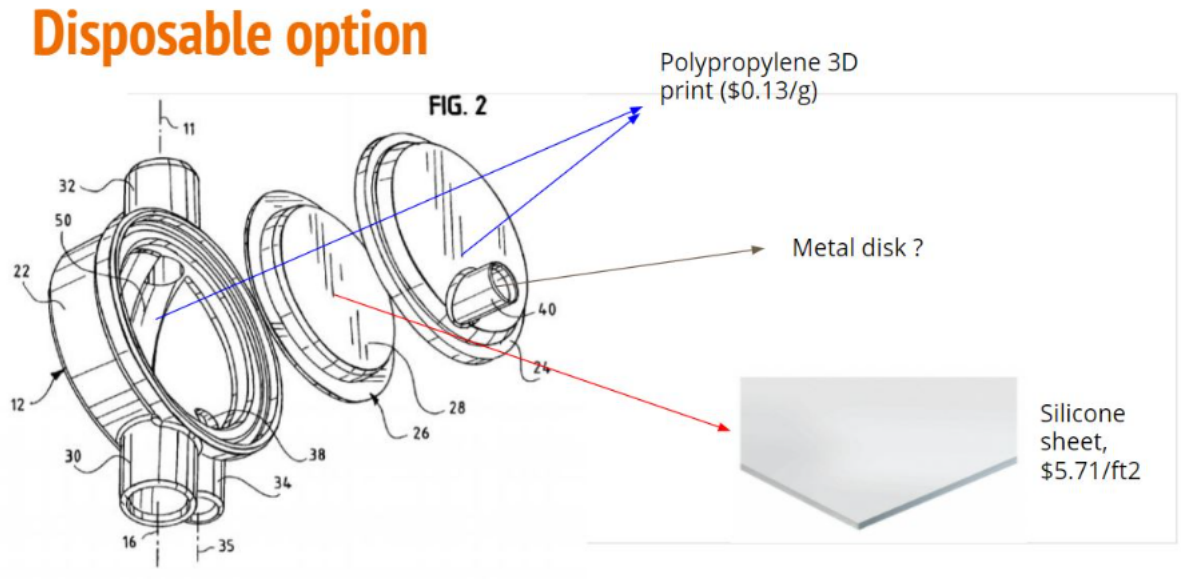
[7] Silicone order site. We will need silicone rubber as the diaphragm if we make a pressure pod - this sheet is slightly thinner than 1mm (1/32 in = 0.8mm), but the patent did not have a dimen

[8] This was a letter from the FDA approving the Spectra Optia for emergency use in treating COVID patients. It includes a table with all the materials in the tubing set. Interestingly, while most phthalate). PET, Polyurethane, and Acetal are all noted to be in indirect contact with blood, while the PVC and Polyetheylen are in direct contact. Regarding the pressure pod, the listed materia in other patents. The steel and nickel could make up the metal disk that Cate and Lokesh noted in their last observation of the tubing set that could be used to "magnetically couple" the tubing and diaphragm cutting (laser?).

[9] *Attached pdf: description of sensors used in hemodialysis lines. This is where I first looked to try to identify how the pressure sensor in the COBE Spectra works. The pressure pod seems

Conclusions/action items:

I can include this image in the next client meeting to summarize the important findings here. The image is from the diaphragm pressure pod patent [4], the polypropylene price is from the UW M per diaphragm [7]. However, making sure each components locks together exactly will be challenging, and I'm not sure how to draw the tiered outer structures depicted below in SolidWorks. I the centrifuge loop including several different plastics, as this could be a cause for concern when trying to test sterilization.



Sources:

[1] https://www.edwards.com/devices/pressure-monitoring/transducer

[2] http://www.frankshospitalworkshop.com/equipment/documents/dialysis_units/service_manuals/Caridian%20BCT%20Spectra%20Optia%20Apheresis%20System%20-%20Service%20man

[3] https://patentimages.storage.googleapis.com/39/db/49/07609d9c57a551/US6887214.pdf

[4] https://patentimages.storage.googleapis.com/97/c0/ed/12afc5fb3456d0/US8092414.pdf

[5] https://www.thermofisher.com/us/en/home/life-science/lab-plasticware-supplies/plastic-material-selection/polyvinyl-chloride-pvc-labware.html#:~:text=Nalgene%20180%20PVC%20tubing%

[6] https://making.engr.wisc.edu/3d-printers-2/

[7] https://www.grainger.com/product/56GT73?gclid=CjwKCAiAtK79BRAIEiwA4OskBsyLAZKck8VugwCoojDsWc_uY3YE3YaVamtijseWSlYOKh_lCqydKRoCNz0QAvD_BwE&cm_mmc=PPC:+Google+PLA&ef_id=CjwKCAiAtK79BRAIEiwA2295:4P7A1P:20501231

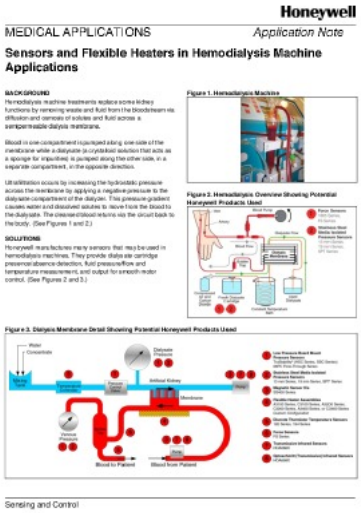
[8] https://www.fda.gov/media/136834/download

[9] Attached pdf - could not find a URL because it downloaded directly from Google

[10] https://www.grainger.com/product/TYGON-PVC-Tubing-22XH86?breadcrumbCatId=CC_1041_1001014_18429

[11] https://www.ensingerplastics.com/en-us/shapes/plastic-material-selection/sterilisable-autoclavable

ADITYA AILIANI - Nov 12, 2020, 1:01 PM CST



Honeywell

MEDICAL APPLICATIONS *Application Note*

Sensors and Flexible Heaters in Hemodialysis Machine Applications

BACKGROUND

Hemodialysis machine treatments replace some kidney functions by removing waste and fluid from the bloodstream via diffusion and osmosis of solutes and fluid across a semipermeable dialysis membrane.

Blood in one compartment is pumped along one side of the membrane while a dialysate (a crystalloid solution that acts as a sponge for impurities) is pumped along the other side, in a separate compartment, in the opposite direction.

Ultrafiltration occurs by increasing the hydrostatic pressure across the membrane by applying a negative pressure to the dialysate compartment of the dialyzer. This pressure gradient causes water and dissolved solutes to move from the blood to the dialysate. The cleansed blood returns via the circuit back to the body. (See Figures 1 and 2.)

SOLUTIONS

Honeywell manufactures many sensors that may be used in hemodialysis machines. They provide static-use cartridge pressure/absence-detection, fluid pressure/flow and temperature measurements, and output for smooth motor control. (See Figures 2 and 3.)

Figure 1. Hemodialysis Machine

Figure 2. Hemodialysis Overview Showing Potential Honeywell Products Used

Figure e 3. Dialysis Membrane Detail Showing Potential Honeywell Products Used

Sensing and Control

Application_Note_Hemodialysis_Machines_008156-4-EN.pdf(394.6 KB) - download

Project Description

TREVOR SILBER - Oct 07, 2020, 10:10 AM CDT

Title: VetMed: Conversion of Human COBE Plateletpheresis Machine for Large Animal Use

Date: 9/7/2020

Content by: Trevor

Present: N/A

Goals: Learn about what the client wants from the project

Content:

From the design website.

- Design materials that are low cost and or reusable for a COBE plateletpheresis machine so it can be used for large animals.
- Plateletpheresis is the collection of platelets from blood. This is done by using a centrifuge to separate blood into its components, Then plateletpheresis is performed, and the rest of the blood is returned to the patients.
- The platelets can then be used in regenerative medicine to create platelet lysate.
- The process can also be done in dogs and horses.
- It can cost anywhere from 2000-2600 dollars every time the machine is used for just the tubing alone.
- Our job is to create parts and/or tubing for the machine to make this process more cost effective to use on large animals.
- The parts should be multipurpose and easy to sterilize.

Conclusions/action items:

I now have a better understanding on what we are going to be doing. Moving forward it would be a good idea to start researching about the process of platelet lysate to better understand exactly what our project will be used for. Competing designs would be a great starting point to understand some of the machinery of the centrifuge device.

Platelet Lysate

TREVOR SILBER - Oct 07, 2020, 11:00 AM CDT

Title: What is Platelet Lysate?

Date: 9/7/2020

Content by: Trevor

Present: N/A

Goals: Understand how platelet lysate is

Content:

“Platelet Lysate,” Ortho Regenerative, 06-Jun-2017. [Online]. Available: https://orthoregenerative.com/platelet-lysate/. [Accessed: 06-Oct-2020].

- Platelets release growth factors slowly. It can be beneficial in the healing process to speed this process along.
- A platelet rich plasma (PRP) can be used for this process. PRP is created by collecting a highly concentrated sample of platelets. PRP can then be injected into a patient to speed up the healing process. However, using PRP can cause inflation in certain parts of the body.
- Platelet lysate (PL) is the answer to this problem. It is very anti-inflammatory, so it can be used around more sensitive areas, i.e. places with high nerve concentration.
- Platelet lysate can be used to break apart the platelets to release the growth factors to the inflamed areas.
- Regenexx created the first PL by freezing a sample of PRP. The ice crystals formed broke open some of the platelets, releasing the growth factors, and creating a great healing tool.
- PL's can also be used to help culture mesenchymal stem cells (MSCs). It has also been found to help replace toxic, high dose steroids in epidural injections and help in nerve hydro-dissection.
- The process can be used to treat lower back pain, butt and leg pain, or numbness and tingling in the legs or feet.

Conclusions/action items:

The isolation of platelets to create platelet lysate seems like it can be a great tool to for doctors to help heal patients. If cost is decreased, it can be more widely used and hopefully be found helpful in other situations.

Platelets and Wound Healing

TREVOR SILBER - Oct 07, 2020, 11:00 AM CDT

Title: Platelets and Wound Healing

Date: 10/6/2020

Content by: Trevor Silber

Present: NA

Goals: Learn how growth factors work inside platelets.

Content:

A. T. Nurden, P. Nurden, M. Sanchez, I. Andia, and E. Anitua, “Platelets and Wound Healing,” Pubmed.gov, 01-May-2008. [Online]. Available: https://pubmed.ncbi.nlm.nih.gov/18508453/. [Accessed: 06-Oct-2020].

If an injury occurs that draws blood, platelets are crucial in the healing process.

Platelets release substances called growth hormones. GH's promote tissue repair and influence other processes such as angiogenesis, inflammation and the immune response.

The growth factors that are released from the platelets bind to the healing site and create a gradient that favors the recruitment of stem cells to the site.

Conclusions/action items:

Having a good understanding of how growth hormones is crucial when talking about platelet lysate and other blood-derived biological products.

WEPLEX

TREVOR SILBER - Oct 07, 2020, 10:59 AM CDT

Title: Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical,

Date: 10/6/2020

Content by: Trevor

Present: NA

Goals: Learn about out clients technique

Content:

A. Pennati, T. Apfelbeck, S. Brounts, and J. Galipeau, “Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical,” Weplex Paper, 14-Sep-2020. .

Washed equine platelet extract, or WEPLEX, can be created by taking concentrated equine platelets, washing, and fysing the solution by a detergent, Triton X-114.

This process completely removes platelet materials, such as albumin, fibrinogen and immunoglobulins, and is 266 times more enriched in platelet derived growth factors than PRP.

When WEPLEX was injected into mice with acute tissue injuries, WEPLEX was found to have protective effects against the tissue injuries.

Conclusions/action items:

Attached below is the WEPLEX paper written by our clients. This paper can be used to help understand exactly what they are doing in the lab. It's interesting to know that our research project can help them do more research on this new method of creating a blood-derived biological product. Their research may be used to help millions of people in the future heal better and faster.

TREVOR SILBER - Oct 07, 2020, 10:58 AM CDT

Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical

Andrea Pennati, MS, PhD[1], Taylor Apfelbeck, BS[1,2], Sabrina Brounts, DVM, MS, PhD[2] and Jacques Galipeau, MD[1]

[1]Department of Medicine, University of Wisconsin, School of Medicine and Public Health and University of Wisconsin Carbone Cancer Center, [2]Department of Surgical Sciences, School of Veterinary Medicine, Madison, WI, USA 53706

Corresponding Author: Jacques Galipeau, MD [jgalipeau@wisc.edu]

Mailing address: 1111 Highland Ave, 3009 Wisconsin Institute for Medical Research, Madison, WI 53705

Telephone: (608) 262-6701, Fax: (608) 265-3913

Weplex.pdf(1 MB) - download

COBE Spectra Apheresis Operator’s Manual

TREVOR SILBER - Oct 07, 2020, 11:09 AM CDT

Title: COBE Spectra Apheresis Operator’s Manual

Date: 10/6

Content by: Trevor

Present: Aditya

Goals: Get tubing parameters for machine

Content:

TerumoBCT, “Section 10,” in COBE Spectra Apheresis Operator’s Manual, Terumo.

Aditya reached out to Terumo and got the tubing specifications. Here is an email from Angela.Richardson@terumobct.com. She informed us that it was from the owner's manual that we can't access.

COBE BCT PLATELET AND PLASMA COLLECT BAG TUBING SPECIFICATIONS

OUTSIDE TUBING DIAMETER		WALL THICKNESS	
MINIMUM	**MAXIMUM**	**MINIMUM**	**MAXIMUM**
0.157 inches	0.163 inches	0.020 inches	0.024 inches
3.99 mm	4.14 mm	0.51 mm	0.61 mm

CLEARED TUBING SPECIFICATION FOR

USE WITH THE SCD 312 STERILE TUBING WELDER

OUTSIDE TUBING DIAMETER		WALL THICKNESS	
MINIMUM	**MAXIMUM**	**MINIMUM**	**MAXIMUM**
0.152 inches	0.220 inches	0.020 inches	0.043 inches
3.86 mm	5.6 mm	0.508 mm	1.1 mm

PLATELET COLLECT BAG TARE WEIGHT:

Section 10, the “Helpful Hints” section, of the COBE Spectra Apheresis Operator’s Manual, provides instructions for “How to Calculate Collect/Plasma Bag Tare Weights”. The weights listed in the Operator’s manual are accurate for the slide clamp and tubing which will be included in disposable sets beginning in August (lot numbers beginning approximately 07D). The table below summarizes the component weights (grams):

COMPONENT	OPERATORS MANUAL	01D-07D lots (approximately)	after 07D lots (approximately)
Platelet Collect Bag	36	33	33
Slide Clamp	1.4 (Blue)	1.5 (White)	1.4 (Blue)
Weight of tubing (per inch)	0.20 (thin wall)	0.35 (thicker wall)	0.20 (thin wall)

Please note that the average Collect Bag weight is 33 grams, instead of 36 grams, due to the recent elimination of one of the receptors.

The Operator's Manual will be updated at the next scheduled revision.

Conclusions/action items:

These tubing dimensions can be provided to companies to get quotes on tubing fabrication costs. Since the outer diameter was provided, this means it is a more critical measurement than the inner diameter. This information has been provided to Cole Parmer.

How to make adhesives work

TREVOR SILBER - Nov 04, 2020, 11:37 AM CST

Title: How to make adhesives work for your medical tubing

Date: 11/4

Content by: Trevor

Present: None

Goals: Learn about different adhesive methods

Content:

https://www.medicaltubingandextrusion.com/how-to-make-adhesives-work-for-your-medical-tubing/

Nylon is a difficult-to-bond thermoplastic because of its low surface energy and texture of its surface.

Adhesives can either spread out, in a process called “wetting,” or bead up on the surface of a plastic substrate. This can be reduced by grit blasting, micro blasting and sanding. They will scratch the surface to expose crystalline microstructures within the substrate with different wetting characteristics.

UV/Visible light-curable adhesives can fully cure in seconds into thermoset polymers. These form lasting bonds to different difficult-to-bond types of plastics. Adhesives that fluoresce red work well with substrates that naturally fluoresce blue, such as PVC and PET, and allow for easy inspection of bond lines.

Conclusions/action items:

Since we are testing a nylon tubing we may have to try the abrasive techniques to make sure the glue sticks. A UV cured glue may be used when testing the PVC tubing.

Tubing Research

TREVOR SILBER - Dec 09, 2020, 12:09 PM CST

Title: Tubing Prices

Date: 10/21/2020

Content by: Trevor Silber

Present: N/A

Goals: Get base set tubing prices

Content:

Since Cole-Parmer has not responded about custom tubing, I looked into premade tubing that would fit into our device. I looked into McMaster - Carr, Grainger Industrial Supplies, MSC Industrial Supply, Sigma Aldrich, Unisource, and VWR. We want to test the tubing for its inner diameter/flow rate and its material/serializability.

Company	OD (in)	ID (in)	Length (ft)	Material	Cost	Link
Current	5/32	1/8	63	PVC w DEHP	2300	
Grainger	5/32	1/4	50	Fluoropolymer	144.5	https://www.g
Grainger	5/32	7/64	100	Nylon	17.59	https://www.g
Grainger	5/32	3/32	50	Tygon(R) PVC	16.05	https://www.g
Grainger	5/32	3/32	100	Nylon	29.79	https://www.g
MSC	5/32	3/32	100	Silicone	87	https://www.n
Grainger	0.15748 ~ 5/32	.0787402 ~ 5/64	49	7	23.5	https://www.g
MSC	5/32	1/16	100	Polyurethane	30.05	https://www.n
Grainger	5/32	1/32	100	Silicone	232	https://www.g
Grainger	3/16	1/8	50	Fluoropolymer	79.3	https://www.g
Fischer Scientific		0.125-0.156	00 Y connectors	Polypropylene	104	https://www.fi

Conclusions/action items:

We decided to purchase 3 of the tubing options. All made of different materials and have different diameters. This tubing will be used to fabricate a prototype.

Grainger	5/32	7/64	100	Nylon	17.59	https://www.grai
Grainger	0.15748 ~ 5/32	.0787402 ~ 5/64	49	PVC	23.5	https://www.grai
Grainger	3/16	1/8	50	Fluoropolymer	79.3	https://www.grai

Transfusion Medicine Apheresis Plateletpheresis

TREVOR SILBER - Oct 07, 2020, 10:06 AM CDT

Title: Transfusion medicine Apheresis Plateletpheresis

Date: 9/07/2020

Content by: Trevor Silber

Present: NA

Goals: Learn about apheresis and plateletpheresis

Content:

http://www.pathologyoutlines.com/topic/transfusionmedplateletpheresis.ht

Plateletpheresis is the process of removing platelets using an automated machine

Currently available instrumentations for plateletpheresis procedures are COBE Spectra and Trima Accel from Terumo and Fenwal Amicus from Fresenius Kabi (competing designs)

Can be used both as a platelet collection procedure from a donor and as a therapeutic modality to quickly decrease the platelet count in a patient with thrombocytosis

Apheresis is the removal of blood plasma from the body by the withdrawal of blood, its separation into plasma and cells, and the reintroduction of the cells, used especially to remove antibodies in treating autoimmune diseases.

Conclusions/action items:

Now that I have a better understanding of what our client is doing and using the machine for I feel more comfortable moving forward. There are a couple competing designs from this website that may be helpful to investigate.

COBE Spectra Apheresis System

TREVOR SILBER - Sep 07, 2020, 12:51 PM CDT

Title: COBE Spectra Apheresis System

Date: 9/7/2020

Content by: Trevor

Present: N/A

Goals: Learn about Terumo's apheresis machine

Content:

https://www.terumobct.com/cobe-spectra

- The COBE Spectra machine is widely customized to help doctors get the desired outcome for the multitude of procedures that it can be used on.
- The device uses continuous flow centrifugal technology.
- Can also be used for white blood cell procedures and bone marrow processing.
- The COBE takes in whole blood from the patient, adds an anticoagulant, separates the blood components using a centrifuge, collects or removes specific components and returns uncollected components to the donor or patient.

Conclusions/action items:

This seems to be the device we are going to be making tubing for. If not this device, then something similar.

Tubing Prices and Comparisions

TREVOR SILBER - Oct 21, 2020, 1:08 PM CDT

Title: Tubing Prices

Date: 10/21

Content by: Trevor Silber

Present: N/A

Goals: Find different prices for tubing

Content:

Company	OD (in)	ID (in)	Length (ft)	Material	Cost
Current	5/32	1/8	63	PVC w DEHP	2300
Grainger	5/32	1/4	50	Fluoropolymer	144.5
Grainger	5/32	7/64	100	Nylon	17.59
Grainger	5/32	3/32	50	Tygon(R) PVC	16.05
Grainger	5/32	3/32	100	Nylon	29.79
MSC	5/32	3/32	100	Silicone	87
Grainger	0.15748 ~ 5/32	.0787402 ~ 5/64	49	PVC	23.5
MSC	5/32	1/16	100	Polyurethane	30.05
Grainger	5/32	1/32	100	Silicone	232

Conclusions/action items:

These different sizes of tubing can be used in our testing protocol. In order to see how larger inner diameters will change the flow rate, a large cost hike will be needed.

Tubing Diagram

TREVOR SILBER - Oct 29, 2020, 5:42 PM CDT

Title: Tubing Diagram

Date: 10/29/2020

Content by: Trevor Silber

Present: N/A

Goals: Create a simplified tubing diagram

Content:

See file attached.

Conclusions/action items:

This tubing diagram can be used in our Show and Tell upload to canvas. Also, it can be used when we are fabricating our tubing to know exactly what goes where.

TREVOR SILBER - Oct 29, 2020, 5:41 PM CDT

Tubing_Diagram(5.4 KB) - download

Tubing Pressure Sensor

TREVOR SILBER - Nov 11, 2020, 11:48 AM CST

Title: EXTRACORPOREAL BLOOD PROCESSING APPARATUS AND METHODS WITH PRESSURE SENSING US 7,780,618B2

Date: 11/11/2020

Content by: Trevor Silber

Present: N/A

Goals: Learn is the pressure sensors can be autoclaved

Content:

The main source of contention is a thin circular diaphragm, 134A/138A, in the center of the pressure sensor 134/138. A tight seal is formed with cylindrical components 134B/138B. This tight interaction will allow the pressure to be read using the rest of the pressure sensing component. It said US 5,795,317 would provide more information on the pressure sensor but I didn't find anything about what material the diaphragm is made out of. A max pressure was provided of 1350 mmHg but this corelates to when the machine will turn off. We know that the diaphragm can withstand this pressure, so the pressure inside the autoclave (about 775 mmHg) should not be a problem. I am mostly concerned about the material being so thin that it will easily warp from the heat of the autoclave and not be able to give accurate readings.

Conclusions/action items:

Need to do more research into autoclaving or reach out and call Terumo to see if they can provide more information. UPDATE: After calling they told me they could not give out that information and since the tubing has been taken out of production they don't have access to it.

Tubing Fabrication Idea

TREVOR SILBER - Dec 09, 2020, 12:23 PM CST

Title: Tubing Fabrication Idea

Date: 11/16/2020

Content by: Trevor

Present: BME Group

Goals: Help Cate fabricate tubing with connectors that are too large.

Content:

After our tubing and connectors came in, Cate was tasked with fabricating a new tubing set prototype. The connectors had a larger OD than the ID of our tubing, Cate has mentioned that it was impossible to put the connector in the tube. I thought it would be a good idea to heat up the end of the tubing with a hair dryer and then put the connector inside of the tubing,

Conclusions/action items:

Cate tried it and it worked. It also, unintentionally, made a really tight seal around the connectors. The tight connection will be crucial when it comes to actually running liquid inside the machine to prevent leaks.

Client Meeting

TREVOR SILBER - Sep 10, 2020, 3:44 PM CDT

Title: Client Meeting

Date: 9/10/2020

Content by: Trevor Silber

Present: BME Team

Goals: Learn about project details

Content:

Dr. Brounts is a large animal and sports medicine specialist. Her and two other doctors have created a new product utilized from blood platelets.

Is there a way we can create a new tubing, easy to fabricate, possibly sanitized via gas or steam.

Heat sterilization via heat and gas sterilization.

Good with 20-25 uses on a tube. Tubing is the main focus.

Tubing in the machine, from catheter to machine, in machine, and from machine to patient.

Plastic silicon type of tubing.

Stick with tubing dimensions currently.

Fabrication on campus or third party system.

Testing would be done with water, and then possibly on a horse.

Their product "PRP on steroids" can be used across species, as well as turned into a powder and hydrated to be used again.

Conclusions/action items:

We are going to need to get licensed to work with animal blood.

Project Selection Link

TREVOR SILBER - Sep 17, 2020, 12:21 PM CDT

Title: VETMED: CONVERSION OF HUMAN COBE PLATELETPHERESIS MACHINE FOR LARGE ANIMAL USE

Date: 9/17

Content by: Trevor

Present: N/A

Goals: Get link to project page

Content:

https://bmedesign.engr.wisc.edu/selection/projects/25de148e-1405-47a7-9894-c76d2366793d

Information provide from client.

Conclusions/action items:

I hated having to look through my email for this link and could not find a way to access it through the website, so I put it here for easy access.

Client Meeting 2

TREVOR SILBER - Sep 23, 2020, 2:46 PM CDT

Title: Client Meeting 2

Date: 9/23/2020

Content by: Trevor Silber

Present: BME Group, Sabrina Brounts, Jaques Galipeau, Andrea

Goals: Understand more about

Content:

14 gage needle is currently being used

Would like to be 30-40 minutes long at most with no sedation

Need to calculate maximum volume that can be processed

input catheter will be the limit on flow rate

Could be looking to use a 10/12 gage needle instead of a 14 gage

Want to rearrange parameters on device to pull in the largest volume possible of device

using the jugular vein

input has viscosity of whole blood

can the machine cope with the increase in speed

have means of completely taking apart the machine and reassembling it

120 C for autoclaving

autoclaving is the cheapest option for sterilization

keep the machine where it is, risk of covid closing the building and machine is locked where it is would be an issue

horse has 50/60 L of blood

While lock down is in effect it would be smarter to just start working on tubing fabrication and programming the machine

Conclusions/action items:

From this information, it would be a good idea to dig info packet from the company and start searching through the specific machine details. Must reach out to more tubing companies to see what their costs of tubing are. Create powerpoint for the time frame of the project. Include a gantt chart.

PACT

TREVOR SILBER - Oct 07, 2020, 10:50 AM CDT

Title: PROGRAM FOR ADVANCED CELL THERAPY

Date: 10/1/2020

Content by: Trevor Silber

Present: NA

Goals: Learn more about or clients

Content:

Program for Advanced Cell Therapy. [Online]. Available: https://pact.wisc.edu/our-team/. [Accessed: 06-Oct-2020].

Jacques Galipeau, MD, is the director for PACT. Works for the Department of Medicine and UW Carbone Comprehensive Cancer Center at the University of Wisconsin in Madison, and is the inaugural Associate Dean for Therapeutics Development at the University of Wisconsin School of Medicine & Public Health.

Andrea Pennati, PHD, is the associate director for research and development. He leads translating cell-based research projects into clinical investigations by developing, standardized and establishing cellular processing protocols for use in cellular therapies.

Conclusions/action items.

This information can be used in our video presentation and in our preliminary presentation. Dr. Brounts info has all been provided by her in our first client meeting.

Cole-Parmer Email

TREVOR SILBER - Dec 09, 2020, 11:55 AM CST

Title: Cole-Parmer Emails

Date: 10/6/2020

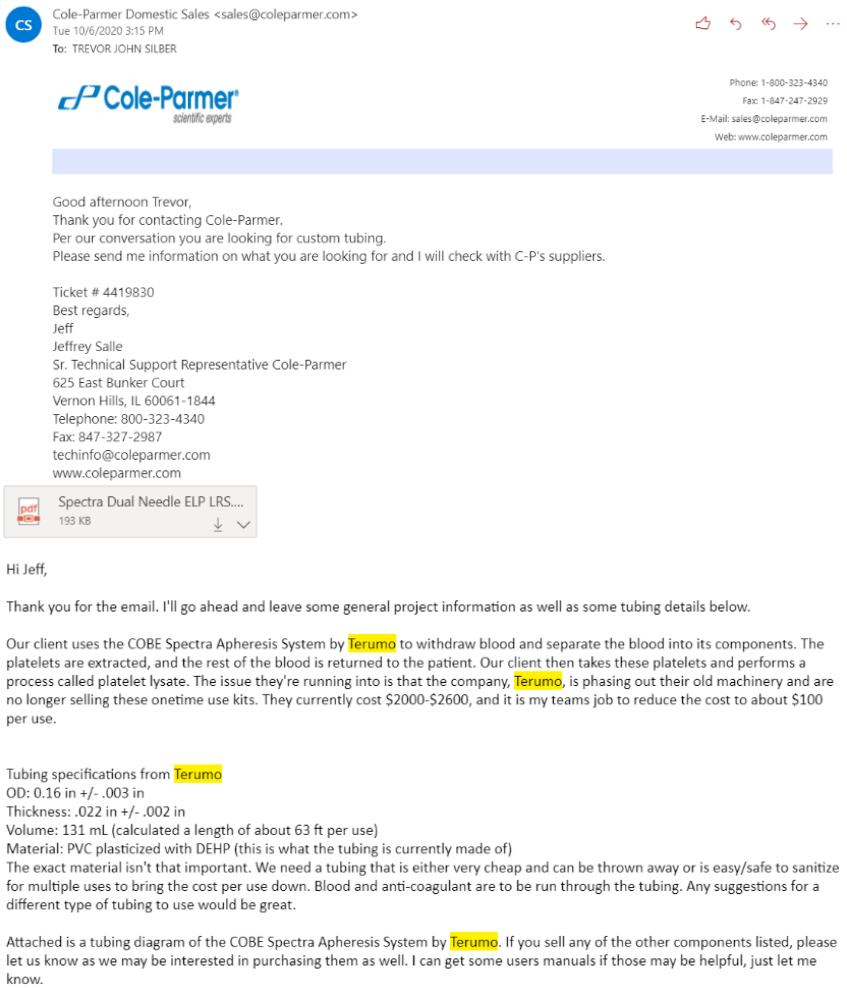
Content by: Trevor Silber

Present: N/A

Goals: Get a tubing price quote

Content:

I did a majority of my tubing searching over the phone. I called Terumo multiple times asking questions on materials and dimensions. I also called Cole-Parmer, my first phone called I talked with a guy named Jim and then later Jeff. I earned that we would need to order 10,000 ft at the minimum if we were going to do a special order. There would be a $250-500 set up cost for making the molds. Once the tubing is produced they can do custom cutting of the tubing sections, have it spooled to different sizes for delivery or have it come sterilized and bagged. I was also informed that due to COVID restrictions they were not taking any in person meetings. This email transpired after our phone call.



Cole-Parmer Domestic Sales <sales@coleparmer.com>
Tue 10/6/2020 3:15 PM
To: TREVOR JOHN SILBER

Cole-Parmer
scientific experts

Phone: 1-800-323-4340
Fax: 1-847-247-2929
E-Mail: sales@coleparmer.com
Web: www.coleparmer.com

Good afternoon Trevor,
Thank you for contacting Cole-Parmer.
Per our conversation you are looking for custom tubing.
Please send me information on what you are looking for and I will check with C-P's suppliers.

Ticket # 4419830
Best regards,
Jeff
Jeffrey Salle
Sr. Technical Support Representative Cole-Parmer
625 East Bunker Court
Vernon Hills, IL 60061-1844
Telephone: 800-323-4340
Fax: 847-327-2987
techinfo@coleparmer.com
www.coleparmer.com

Spectra Dual Needle ELP LRS....
193 KB

Hi Jeff,

Thank you for the email. I'll go ahead and leave some general project information as well as some tubing details below.

Our client uses the COBE Spectra Apheresis System by Terumo to withdraw blood and separate the blood into its components. The platelets are extracted, and the rest of the blood is returned to the patient. Our client then takes these platelets and performs a process called platelet lysate. The issue they're running into is that the company, Terumo, is phasing out their old machinery and are no longer selling these onetime use kits. They currently cost $2000-$2600, and it is my teams job to reduce the cost to about $100 per use.

Tubing specifications from Terumo
OD: 0.16 in +/- .003 in
Thickness: .022 in +/- .002 in
Volume: 131 mL (calculated a length of about 63 ft per use)
Material: PVC plasticized with DEHP (this is what the tubing is currently made of)
The exact material isn't that important. We need a tubing that is either very cheap and can be thrown away or is easy/safe to sanitize for multiple uses to bring the cost per use down. Blood and anti-coagulant are to be run through the tubing. Any suggestions for a different type of tubing to use would be great.

Attached is a tubing diagram of the COBE Spectra Apheresis System by Terumo. If you sell any of the other components listed, please let us know as we may be interested in purchasing them as well. I can get some users manuals if those may be helpful, just let me know.

He then hadn't responded until 11/16 and by that time we had already purchased our other tubing sets, so there was no reason for any further communication.

Good morning Trevor,

Thank you for contacting Cole-Parmer.

Per the manufacturer, "We do not as standard do a PVC 4.0 x 0.6mm wall. We do have in stock a 4.0 x 0.8mm wall. Would this be an option to try?

If not, it may well be possible but we would have to do a trial run to see if it is possible to obtain with the current tooling. Let me know what you'd like to try.

Also, do you know if this will be used on a pump?

Conclusions/action items:

If this tubing project is carried out in future semesters it may be a good idea for them to used Cole-Parmer as a resource. If the tubing set is meant to be mass produced, a large amount of tubing will be required. I was thoroughly disappointed by the long response time so maybe it will be a better idea to go through someone else.

09/14/2020 Platelet Extract as an Anti-Inflammatory

Cate FLYNN - Oct 07, 2020, 9:37 AM CDT

Title: Platelet Extract as an Anti-Inflammatory

Date: 09/14/2020

Content by: Cate Flynn

Goals: To learn about the client's research

Content:

The clients have developed a new platelet extract where equine blood platelets are concentrated, washed and thereafter lysed. Washed equine platelet extract (WEPLEX) is amenable to lyophilization without loss of biological activity

The client has observed success with decreasing inflammation markers in mice with a condition similar to Crohn's disease. This is a unique breakthrough because it is exceedingly difficult to have success with gene therapy across species.

Broader Impact on Society: This therapy may have broad impact in both veterinary medicine and human medical field, on acute injury of tissue, regenerative medicine, but also in other medical fields were platelet derived products have been used such as dermatology, cardiology and sports medicine. Approximately 1.6 million million people in America have Crohn's disease or Ulcerative Colitis and existing treatment options are modeled for life-long use and often function as immunosuppressants. This gene therapy could help 1.6 million people with this one autoimmune condition classification, so the impact of the project and the client's research cannot be understated.

Source: “Washed Equine Platelet Extract (WEPLEX) as an Anti-Inflammatory Biologic Pharmaceutical,” n.d. Washed equine platelet extract (WEPLEX) as an anti-inflammatory biologic pharmaceutical.

Conclusions/action items:

The lysate therapy the client is working on has broad societal impacts on health for both veterinary and human medicine.

09/17/2020 Existing Tubing Conditions

Cate FLYNN - Oct 07, 2020, 8:58 AM CDT

Title: Existing Tubing Conditions

Date: 09/17/2020

Content by: Cate Flynn

Goals: To going information on the existing conditions and regulations the current tubing set operates under.

Content:

The tubing and machine cannot be exposed to temperatures above 81° F.
The inlet flow rate cannot be 25 mL/min or less.
The tubing cannot be occluded or bent before use, this will damage the machine.
Any stretching of the tubing will result in damage to the tubing itself or the machine if it has already been connected.
Tubing must be stored in a sterile environment: outside contaminants such as hair or dirt will damage the machine or contaminate the extracted platelets.
The tubing must be dry upon use: any excess fluid can cause an unwanted electrical response.
The tubing should only be handled by this team in testing or the client and their approved partners when finished.

The inlet pump rate will have a maximum restriction of 65 mL/min for the Dual-Needle procedure

There is a lower concern for bloodborne pathogens in horses, but sterilization between uses will still be required. Ethylene oxide is currently used as a sterilization agent.

Source: “Apheresis System Essentials Guide.Pdf.” Accessed September 17, 2020. http://startrinity3.com/mssn/04/Apheresis%20System%20Essentials%20Guide.pdf.

Conclusions/action items:

We need to be especially mindful of any parameters that impact the quality and operation of the machine; if our tubing damages the machine, the client will lose an important tool in their research.

09/22/2020 WIMR Visit #1

Cate FLYNN - Dec 07, 2020, 12:05 AM CST

Title: WIMR Visit #1

Date: 09/22/2020

Content by: Cate Flynn

Present: Cate, Lokesh

Goals: Gain a greater understanding of the machine, collect tubing set from client

Content:

Seeing the machine in person has increased my understanding of the complexity of this project. I am concerned about the pressure sensors and making sure the tubing we use fits into the front panel of the machine because the pumps and secures on the machine have a fixed size. We will need to seriously consider the rigid nature of where the tubing fits into when selecting our material and size of tubing because too much deviation from the current size and flexibility of the tubing will make it difficult or impossible to use with the machine. A rigid material would not be ideal because of the relatively congested spacing of the panel: the tubing must be flexible to feed into all of the fixtures. Lokesh is holding on to the tubing for now and we will begin to look at simplifying the set moving forward.

Below is an image of the machine (specifically the machine's front panel)

Conclusions/action items:

Given the fixed nature of the front panel of the machine, we will not be able to deviate too greatly from the current size and flexibility of the existing tubing set. We are going to look at how we can simplify the set now that we have a physical set.

10/02/2020 Autoclave Impact on Silicone Stability

Cate FLYNN - Oct 07, 2020, 2:51 AM CDT

Title: Autoclave Impact on Silicone Stability

Date: 10/02/2020

Content by: Cate Flynn

Goals: To learn more about the usability of silicone as a fabrication material

Content:

This source follows a sterilization study conducted by dentists to investigate the sterility and structural integrity of silicone based impressions after autoclave sterilization.

The study shows the ability of cured silicone to remain dimensionally stable through disinfection procedures.

One thing to keep in mind is that hydrophilic silicones may absorb water during the immersion disinfection procedure. This effect will vary material to material based on the quantity of intrinsic surfactant present. It has been shown that tear strength is directly related to wettability due to the level of surfactant incorporated in the material in question.

The ultimate conclusion of this article was that autoclaving is an effective sterilization technique for silicone and silicone based materials. The autoclaving also doesn't damage the structural integrity of the material and the existing shape should not vary in any discernible quantity either.

The implications of this article on this project are as follows:

Autoclaving is an acceptable form of sterilization for silicone material.

This may be the ideal material to pursue considering that the client already has access to an autoclave.

If the current sterilization technique, the use of ethylene oxide, needs to be maintained then silicone won't be an ideal material for fabrication as PVC is more suitable for chemical sterilization.

Source: Millar, Brian J., and Sanjukta Deb. “Effect of Autoclave Sterilisation on the Dimensional Stability and Tear Strength of Three Silicone Impression Materials.” *Open Journal of Stomatology* 04, no. 12 (December 25, 2014): 518. https://doi.org/10.4236/ojst.2014.412069.

Conclusions/action items:

Silicone is an acceptable material to use with autoclaving sterilization.

This may be the most convenient material to use considering the team already has access to an autoclave.

Material decisions are still heavily dependent on contractor material availability which is not yet known.

10/02/2020 Use of Ethylene Oxide for Sterilization

Cate FLYNN - Oct 07, 2020, 3:09 AM CDT

Title: Use of Ethylene Oxide for Sterilization

Date: 10/02/2020

Content by: Cate Flynn

Goals: To consider the use of ethylene oxide as a chemical sterilization technique

Content:

Ethylene oxide is currently used as a sterilization agent for long tubing sets in hospitals.

It is capable of penetrating deep into long tubing routes via hospital ethylene oxide steriliser, but there can be residual quantities of ethylene oxide in plastic tubing.

These residual values were initially high, but a storage period of approximately four days at room temperature had reduced them to an acceptable level.

The paper concludes that if there are adequate controls on the sterilising process and storage practices, then sterilization via ethylene oxide is considered to be safe.

Initial questions to consider:

Does the rest period for the residual levels to drop depend on the type of plastic material, or is it relatively consistent?

How would we test the residual quantity remaining in the tubing assuming we pursue this sterilization technique?

Does the acceptable quantity of residual ethylene oxide vary by species (for example humans versus horses) or is it consistent across life forms?

Source: Gillespie, E H, J M Jackson, and G R Owen. “Ethylene Oxide Sterilisation--Is It Safe?” *Journal of Clinical Pathology* 32, no. 11 (November 1979): 1184–87.

Conclusions/action items:

If ethylene oxide is chosen as the sterilization technique, we need to develop a testing plan and specific parameters to demonstrate irs effectiveness in between uses.

10/18/2020 Autoclave Sterilization Methods

Cate FLYNN - Nov 28, 2020, 12:10 PM CST

Title: Different Autoclave Techniques

Date: 10/18/2020

Content by: Cate Flynn

Present: NA

Goals: To outline some of the possible sterilization techniques that could be used to clean the tubing set in between uses.

Content:

http://stmichaelshospitalresearch.ca/wp-content/uploads/2015/09/Autoclave-3850.pdf

This document is a user manual that includes multiple sterilization techniques on pages 16-20.

There are five sterilization programs with and without drying stages. Each program lists the temperature, time and operation sequence. One of the materials being considered for fabrication, PVC, has a temperature limit of 160 ° F, so that should be kept in mind when choosing a sterilization program assuming we can get access to the autoclave later on in the semester. It appears that the autoclave will be far too hot if we use this material for fabrication and that we will have to consider other sterilization methods if we pursue this material.

Conclusions/action items:

This source has five good examples of sterilization sequences to consider assuming we use a material that can tolerate the heat, if not we will have to consider alternative means of sterilization.

10/19/2020 Hybrid Sterilization Technique

Cate FLYNN - Nov 28, 2020, 12:41 PM CST

Title: Hybrid Sterilization Technique

Date: 10/19/2020

Content by: Cate Flynn

Present: NA

Goals: To outline an alternative sterilization method or combination of methods to avoid relying fully on the autoclave.

Content:

https://pdfs.semanticscholar.org/0a4a/677d8a66923b6d63c3e3601e1505c1208fba.pdf

There is a five step sterilization protocol on page 2 of this document.

It still uses autoclave, but it also uses ethanol, a drying solution and UV light. I don't think that this specific protocol will work for our purposes, but I think it is a good example of how multiple methods in combination can be an effective tool. I think we should reconsider the ethylene oxide solution for sterilization if we use a material like PVC for fabrication that can't be put in the autoclave without being damaged.

Conclusions/action items:

If we have time for sterilization testing, we need to consider using a chemical mixture of some sort to clean or need to find a different alternative to autoclaving for the tubing if it is fabricated from a low temperature tolerant material. Autoclave should still work well for hard plastic parts.

10/29/2020 WIMR Visit #2

Cate FLYNN - Dec 07, 2020, 12:35 AM CST

Title: WIMR Visit #2

Date: 10/29/2020

Content by: Cate Flynn

Present: Cate/ Lokesh

Goals: Fit the tubing into the machine to gain greater understanding for fabrication

Content:

We went to WIMR for the second time to fit the tubing into the machine itself. This process took about 50 minutes and was a bit troublesome due to the material's inclination to tangle. Once the tubing was set in the machine, I saw a better picture of what we're trying to create. It was hard to see the goal when the tubing was in its packaging or laid across the floor. We did run into one unforeseen challenge that we will have to ask the client about. The centrifuge loop doesn't fit into the current centrifuge cap, the loop is much too small. Upon further research, Aditya discovered that there are actually four centrifuge caps that come with the machine, depending on what procedure you wish the run (image inserted below). The current cap with the machine is the single-stage cap which is significantly larger than the other three caps. I am going to reach out to the clients to see if they have any of the other caps in storage or if the previous owner has them. I plan on doing more research into the LRS chamber to see which cap we should purchase. If the client doesn't have access to the other caps, we will need to look into different means of acquiring it. Since the tubing for this machine is no longer sold, I anticipate that we might have a difficult time getting a separate cap from Terumo, but we are in contact about getting extra tubing sets so it's possible they'll consider it. If we cant get the cap from them, we might want to consider a third party vendor like eBay or craigslist. Lokesh has already found the dual-stage cap for about $100 on eBay which would be a viable option. We also turned on the machine and tried to run it, but there needs to be fluid present (anticoagulant and saline) to get the machine to run any of its protocols. Without fluid, the pressure readings don't work and we can't get around the error messages. I will reach out to the client to ask if we could have access to some saline bags for the next round of testing. Based off of the users manual, I suspect that we can use the saline for the anticoagulant as well, especially since we only need to run the saline rinse to test the tubing. I have also attached an image of the tubing in the machine and a closer image of the size issue with the current centrifuge loop/cap.

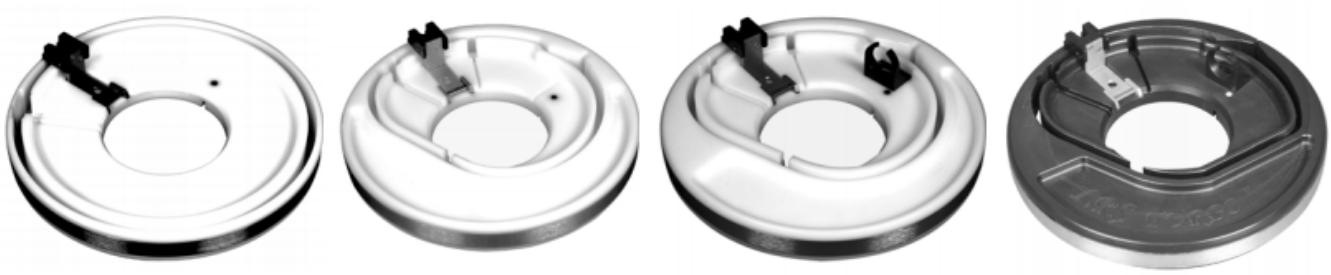
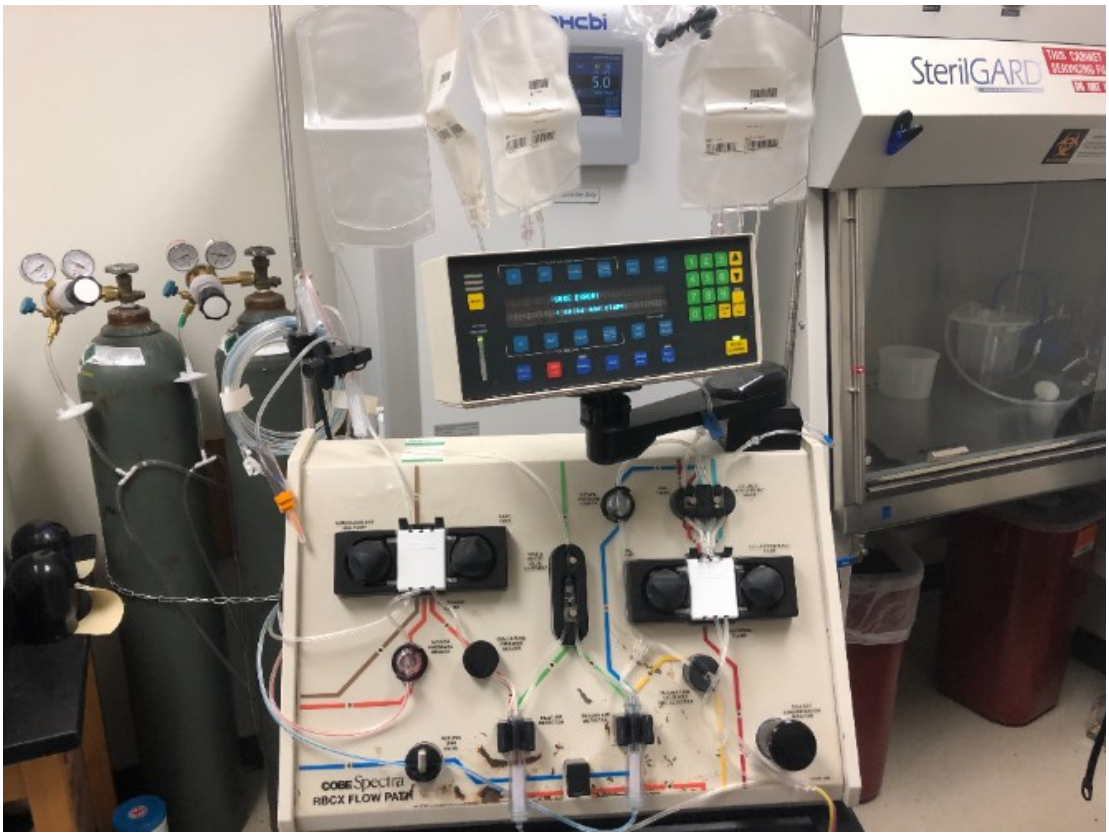


Figure 2-6: Fillers from left to right: single-stage, dual-stage, dual-stage with LRS bracket, and dual-stage LRS Turbo



Conclusions/action items:

I will reach out to the clients to see if they have knowledge of where the other centrifuge caps are. If they do not, I will present other means of getting the cap. I will also do research in order to determine which cap we will need and I will reach out to the client to ask about saline bags for the next round of testing.

11/09/2020 Selecting Tubing Material

Cate FLYNN - Dec 09, 2020, 2:28 AM CST

Title: Selecting Tubing Material (for Prototype Fabrication)

Date: 11/09/2020

Content by: Cate Flynn

Present: Cate, Aditya, Lokesh, Nesya, Trevor

Goals: Select one of three available tubing materials to fabricate our prototype from

Content:

I picked up our first round of materials from the client on the 6th and we decided on what material to use for the prototype during our team meeting on the 9th. I described the physical characteristics of all three materials to the team and recommended we use the PVC tubing because it will be flexible enough to feed into the machine, is clear which will be convenient for testing and is most similar to the existing tubing material. The team concurred and decided on PVC for fabrication because of those reasons. The other two materials ,nylon and fluoropolymer, were much more rigid and would not fit into the machine and the nylon is also black in color which will make observing testing difficult.

Conclusions/action items:

Use the selected material to fabricate the prototype in the future.

11/09/2020 WIMR Visit #3

Title: WIMR Visit #3

Date: 11/09/2020

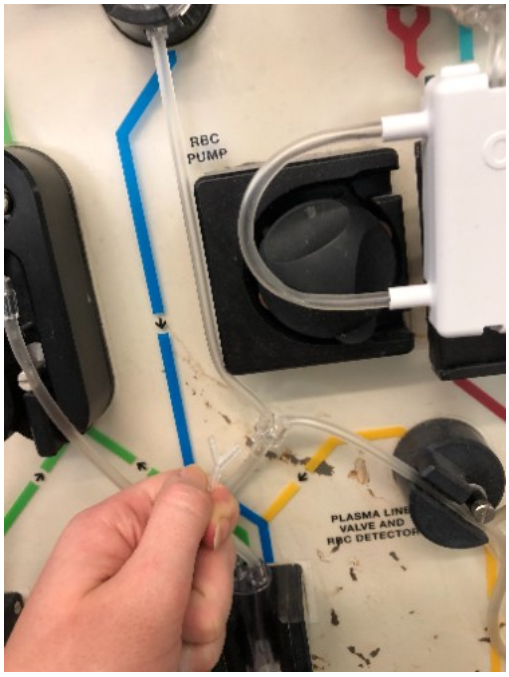
Content by: Cate Flynn

Present: Cate, Lokesh

Goals: Run fluid through the existing machine to gain a greater understanding of the pressure sensors for simplification of the tubing set

Content:

We acquired three bags of saline from the client and connected them to the machine in the correct locations. The machine produces prompts for which lines need to be clamped at what time d bag following these prompts. We are still unsure of how to drain the tubing after the saline rinse. Unfortunately for simplification purposes, the pressure detectors are very sensitive. We tried to could bypass them and we could not. This means that we need to maintain the current pressure structures. To me, the easiest solution for this would be to fabricate around the necessary parts like to work on creating our own pressure sensors, as some of them seem like they could be 3D printed, but I think our timeline might be a bit strained to replace all essential parts with parts of machine's Y Connector. The idea would be to go through and replace as much of the existing tubing and connectors with our own. I have attached our ordering form for the proposed prototype



For Order:							
Company	**Outer Diameter (in)**	**Inner Diameter (in)**	**Length (ft)**	**Material**	**Cost**	**Link**	**Part Numbe**
Grainger	5/32	7/64	100	Nylon	17.59	https://www.grainger.com/pro	
Grainger	0.15748 ~ 5/32	0.0787402 ~ 5/64	49	PVC	23.5	https://www.grainger.com/pro	
Grainger	3/16	1/8	50	Fluoropolymer	79.3	https://www.grainger.com/pro	
Fischer Scientific		0.125-0.156	20 Y connectors	Polypropylene	30.29	https://www.fishe	S50701A
Fischer Scientific		1/8	100 straight connectors	Kynar (PVDF)	69.3	https://www.fishe	01-000-510

Conclusions/action items:

We need to generate a labeled graphic with all of the lengths of tubing we wish to replace labeled. We need to come up with a way to drain the tubing set following the saline rinse in preparatio

11/19/2020 WIMR Visit #4

Cate FLYNN - Dec 08, 2020, 10:13 PM CST

Title: WIMR Visit #4

Date: 11/19/2020

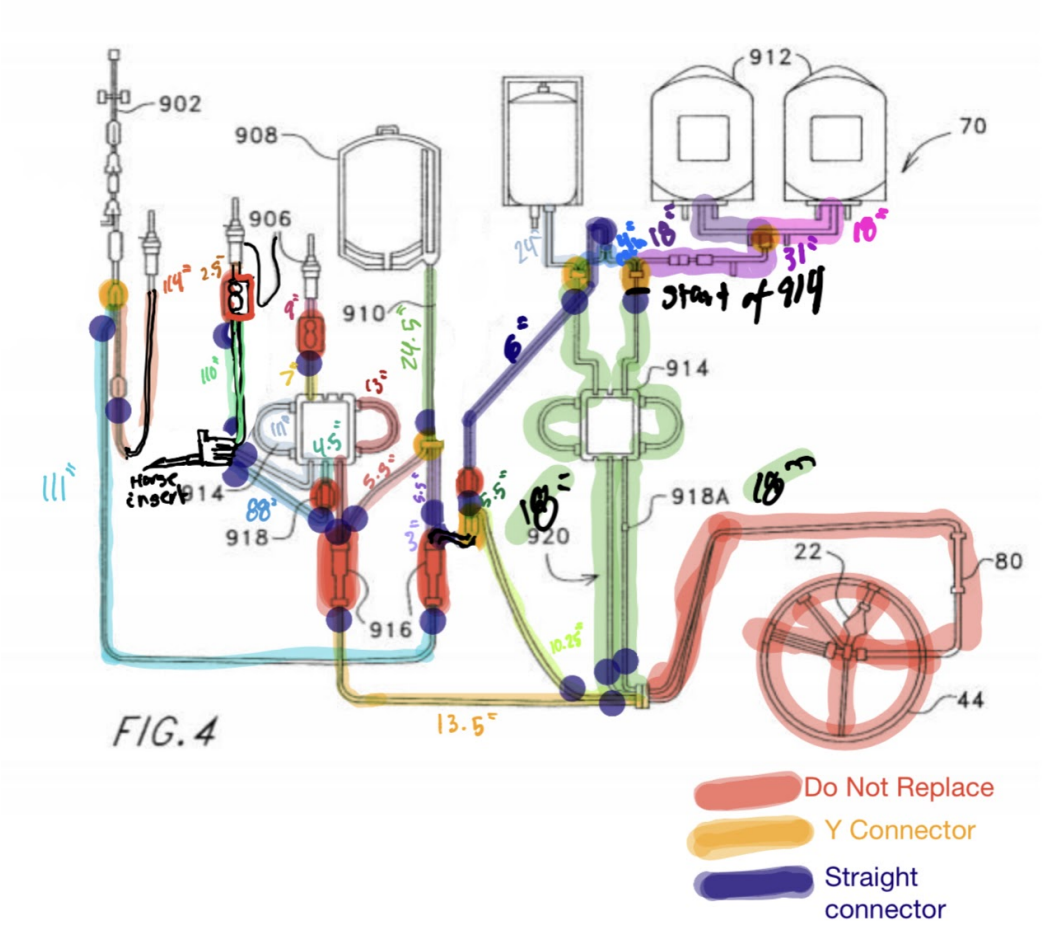
Content by: Cate Flynn

Present: Cate, Lokesh

Goals: Create a graphic with measurements to be used to fabricate the prototype

Content:

We went to WIMR where we had left one of the previous tubing sets loaded into the machine which made taking measurement easier as the tubing is long enough that it is difficult to decipher if it isn't in the machine. Lokesh brought his tape measure and I brought up a graphic of the Terumo tubing set on my iPad. We first made sure we understood which part of the graphic aligned with which parts of the tubing. Once we were confident in the diagram, we went through and measured the tubing set, Lokesh taking the measurements and me highlighting and recording on the graphic and modifying the image for any discrepancies (see portion labeled "Horse Insert"). I also highlighted the graphic for Y connectors and straight connectors and included a key with that information. I highlighted parts that we cannot fabricate during this semester and that therefore cannot be replaced in red and included this in the key as well. From here, I will fabricate the prototype, replacing the highlighted sections of tubing with the appropriate lengths and connectors. I have attached the graphic below.



Conclusions/action items:

Fabricate the prototype from the generated graphic.

11/20/2020 Testing Brainstorming

Cate FLYNN - Dec 09, 2020, 2:16 AM CST

Title: Testing Brainstorming

Date: 11/22/2020

Content by: Cate

Present: Cate

Goals: Outline a possible testing protocol for the prototype

Content:

Once I have fabricated the prototype, we will need a way to test it with the machine. Lokesh and I already know that we can run the saline rinse with the tubing set and have enough leftover saline bags to run the sterilization protocol (we have three saline bags at WIMR). Running the machine will be a good indicator of the quality of the seals and will tell us if there are any pressure issues with the prototype. My only concern is our ability to get the remaining water out of the tubing set. Because we don't have the correct centrifuge cap for our tubing set, we can't run the centrifuge which is the only way to drain all of the water from the centrifuge loop. I'm thinking that we can clamp the sections that lead into the loop off and avoid accumulating water there in the first place, but we will have to test that out and see if the machine will still run with unintended sections clamped off.

Conclusions/action items:

Test the prototype with the apheresis machine's saline rinse protocol at WIMR.

11/23/2020 Fabrication of Prototype

Cate FLYNN - Dec 09, 2020, 2:29 AM CST

Title: Fabrication of Prototype

Date: 11/23/2020

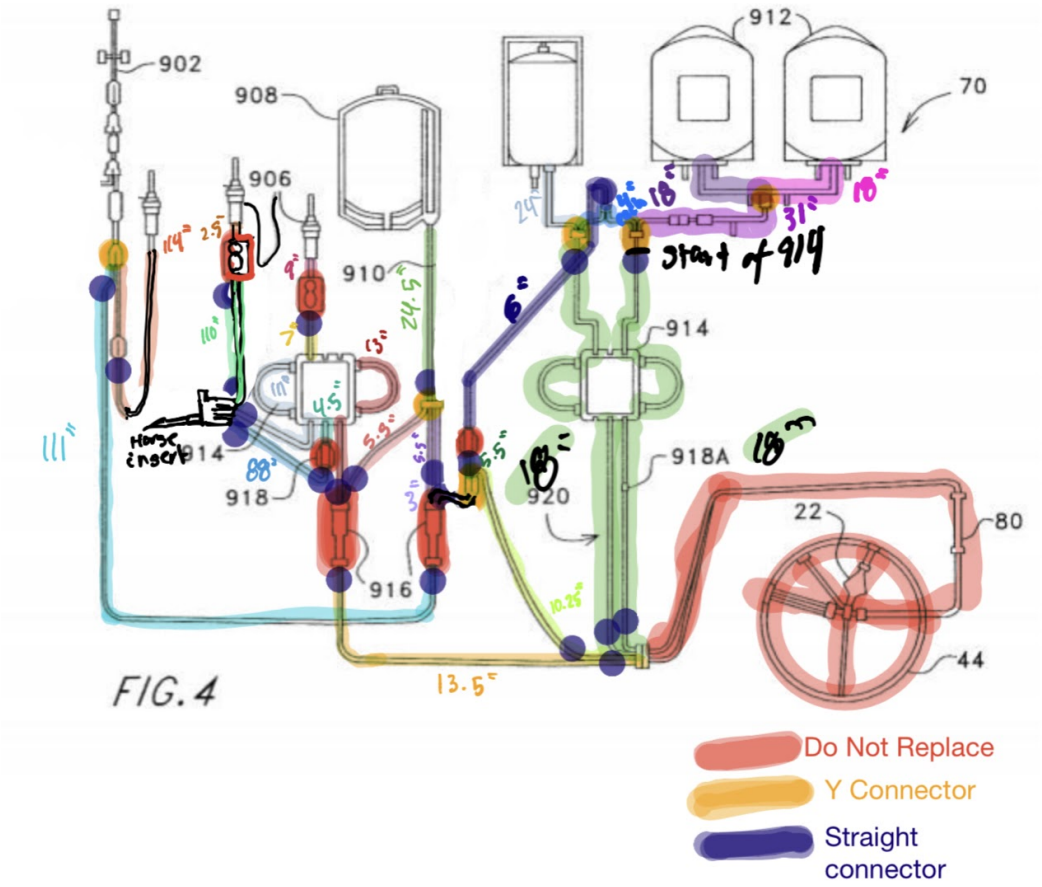
Content by: Cate Flynn

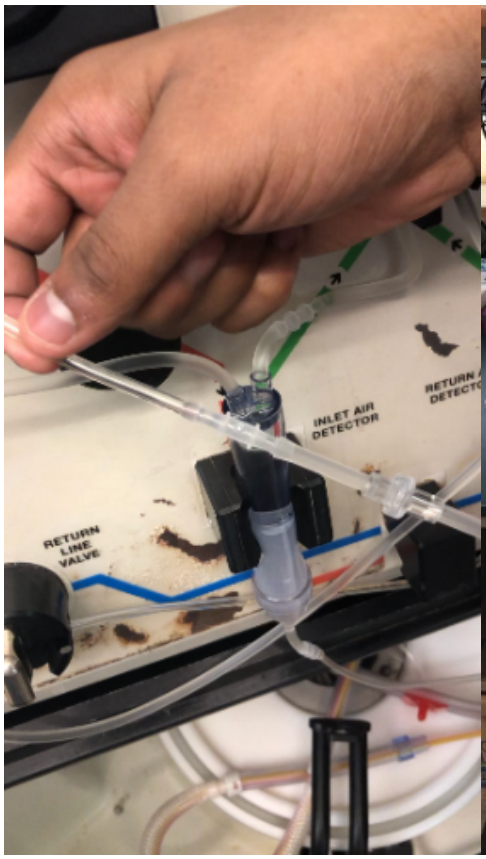
Present: Cate

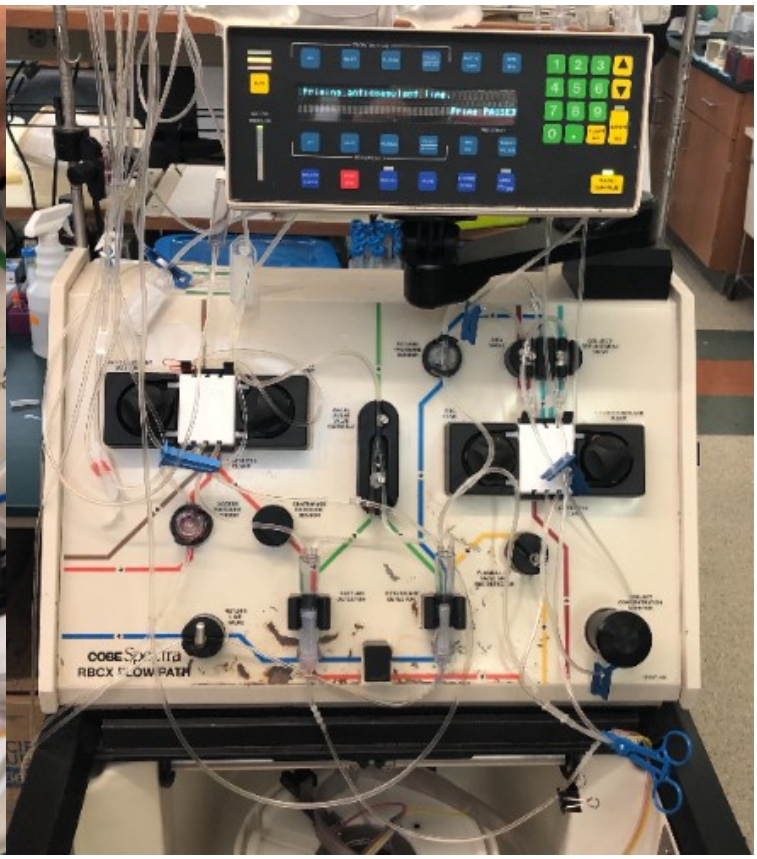
Goals: Fabricate the prototype as described in WIMR visit #4

Content:

In fabricating this prototype, I measured out the tubing segments to cut from our PVC tubing as shown in the measurement document created during our fourth trip to Wisconsin Institutes of Medical Research (WIMR). I have attached this image below for reference. Once I had measured out the indicated lengths of tubing, I used a hairdryer on medium heat to warm the PVC tubing. I held each section of tubing under the hair dryer for about 5 seconds before pushing the 1/8" straight connectors into the warmed section of tubing as indicated on the measurement graphic. Once the tubing had cooled, a strong seal formed. I continued this process, leaving the parts that we could not replace as they were. I attempted this same procedure with the 1/8" Y connectors, but was unable to get the connector to fit into the tubing regardless of how long the tubing was heated for. This will be something to consider moving forward, as smaller Y connectors are likely necessary. Due to this complication, I left the original Y connectors intact. I have attached a close up image of one of the seals below as well as an image of our prototype in the apheresis machine itself. I did run into one tubing complication with the existing set that I have described in more detail in the next entry.







Conclusions/action items:

Test the prototype with the apheresis machine as described in Testing Brainstorming.

11/23/2020 Unforeseen Inner Diameter Issue

Cate FLYNN - Dec 09, 2020, 2:40 AM CST

Title: Unforeseen Inner Diameter Issue

Date: 11/23/2020

Content by: Cate Flynn

Present: NA

Goals: Make note of the small inner diameter sections of the tubing for future considerations

Content:

While I was fabricating the prototype, I can across two sections of tubing that had an exponentially smaller inner diameter than the rest of the tubing set. These two sections went through a conversion piece that lead back into regularly sized tubing sections. I fabricated from the converter and left the smaller inner diameter sections of tubing intact in the prototype. This is an important note because we had not found evidence of it in any of our documents and had not anticipated it. If the smaller inner diameter is a necessity in order to maintain the correct pressure throughout the set, than future semester's teams will need to consider ordering custom tubing or sterilizing the existing tubing. If the smaller inner diameter isn't necessary, they can just replace those sections with the same tubing as the rest of the set.

Conclusions/action items:

The tubing set has two sections of tubing with significantly smaller inner diameters than the rest of the set. It is unknown if this is necessary to maintain the correct pressure or if these lengths can be replaced by standard tubing at this time.

11/24/2020 WIMR Visit #5

Cate FLYNN - Dec 09, 2020, 9:07 AM CST

Title: WIMR Visit #5

Date: 11/24/2020

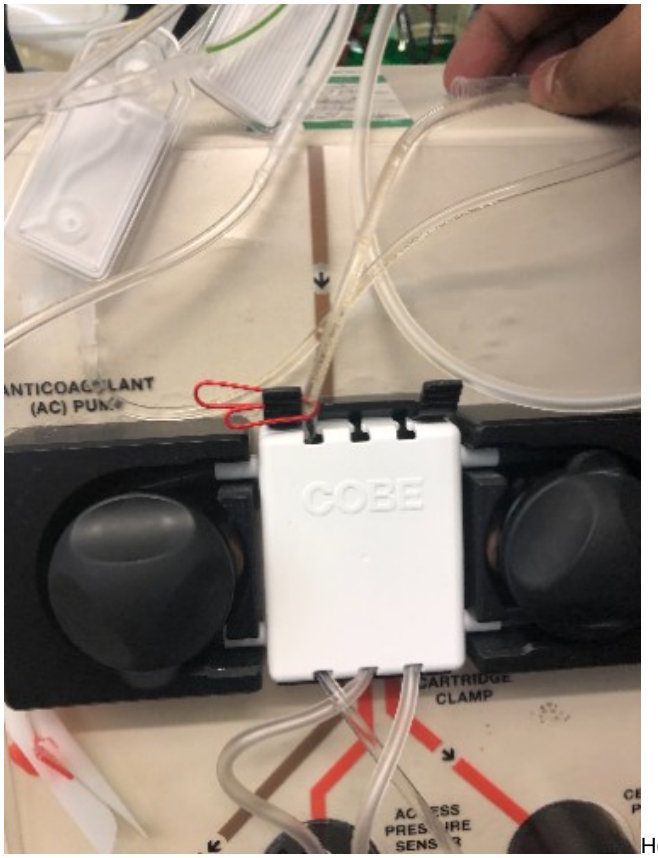
Content by: Cate Flynn

Present: Cate, Lokesh

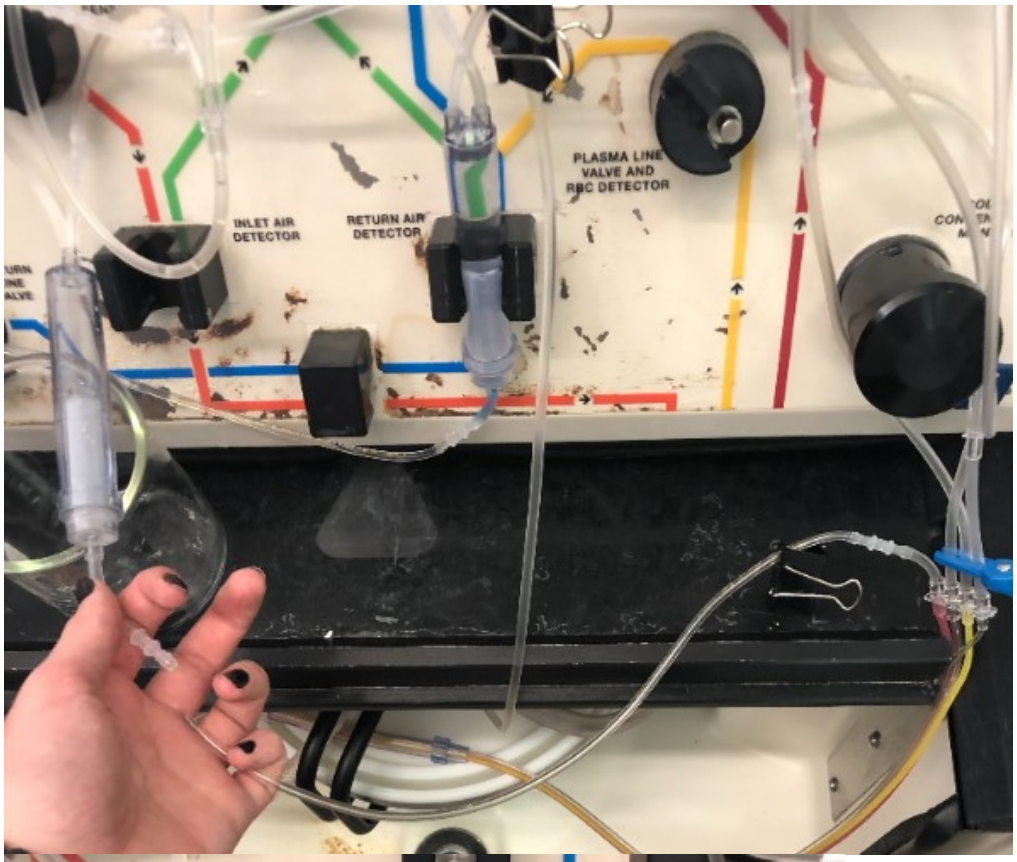
Goals: Test the prototype with the apheresis machine at WIMR

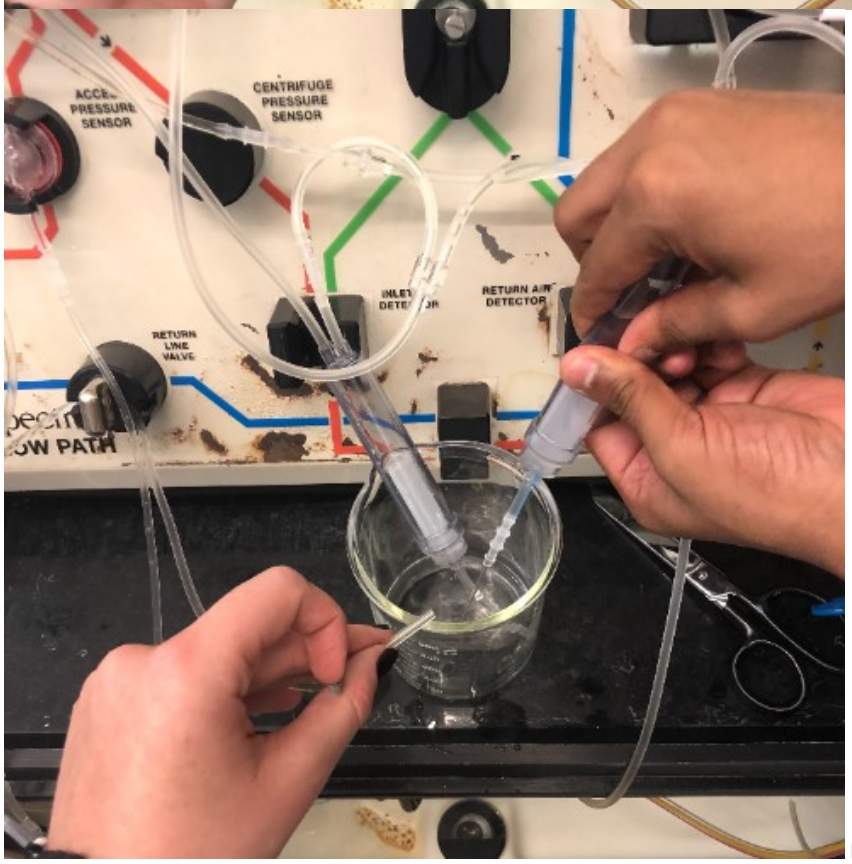
Content:

We went to WIMR to run the machine with the prototype. We ran the saline rinse cycle as planned and had success with this test. We did clamp off the centrifuge loop and there were no issues in doing this and we prevented water from getting in to this section of the tubing that we wouldn't be able to remove. There were no visible leaks in the tubing and no pressure issues as indicated by the lack of error messages. We now know that it is a viable option to replace the existing tubing with commercially available tubing and that strong seals can be formed and that custom tubing shouldn't be necessary as was initially thought in the semester. There were a couple of things that will be important to note moving forward. The white plates that connect to the machine to feed the pumps need to have a adhesive between them and the tubing or the tubing will be pulled by the pump. Without an adhesive, no tension is created and there is slack around the pump at the end of the loading. We solved this by holding the tubing in place and then putting a paperclip in, but this is a good consideration for the next prototype. We also didn't have a regulated way to drain the saline because we couldn't run the centrifuge portion, so we cut the tubing below both of the air connectors and drained the saline into beakers. We cut close to the connector, so this portion of tubing can just be reheated and reattached.

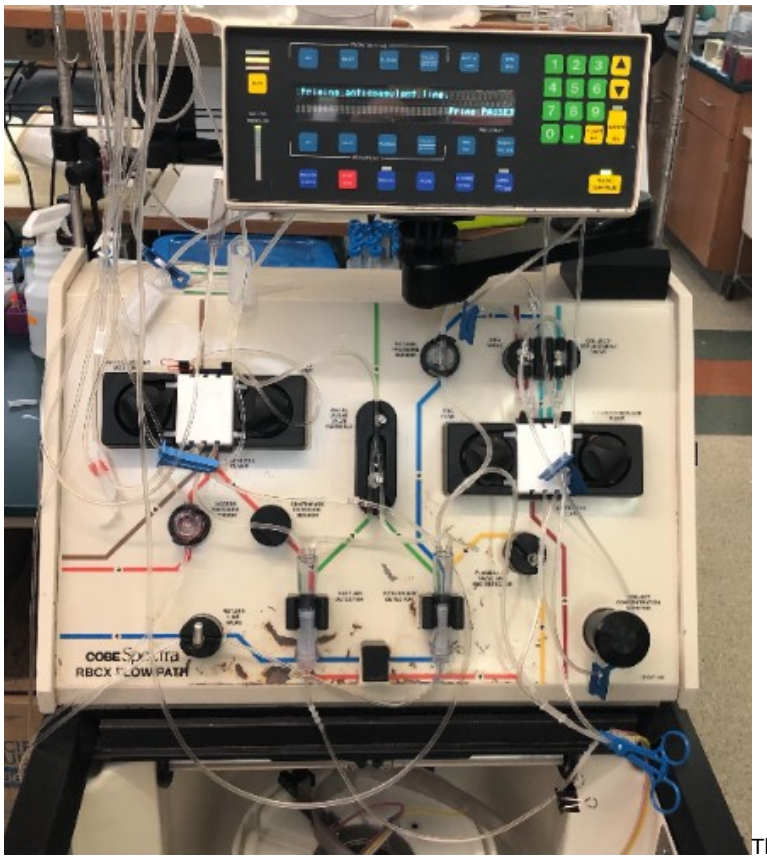


Here is an image of the paperclip we placed near the pump on the white plate to maintain tension.





Here is an example of how we cut the tubing, we did this below each air pump and drained the saline into a beaker. In the lower right, you can also see where we clamped off the centrifuge loop.



This is an image of the entire prototype in the apheresis machine.

Conclusions/action items:

This test proves that the existing tubing and connections can be replaced. In the future, it would be informational to be able to run the centrifuge and run the apheresis procedure in its entirety to gain a greater understanding of the machine.

11/30/2020 Large Connectors vs. Small Connectors Testing Ideas

Cate FLYNN - Dec 09, 2020, 9:14 AM CST

Title: Large Connector vs. Small Connector Testing Ideas

Date: 11/30/2020

Content by: Cate Flynn

Present: Cate, Aditya

Goals: Create a testing plan for comparing the 1/8" inner diameter (ID) connectors and the 1/16" ID connectors

Content:

We are creating a plan to compare the relative merits of the two connectors that will also produce data that can undergo statistical analysis. I have access to 2.5 mL syringes that I can use to inject water into tubing from home in a fairly standardized time. I think that 3 ft of tubing is an appropriate length to use for this quantity of water. I am going to construct two tubing sets, each with 3 of either the larger or smaller connectors. I am going to record the time to load each set from the syringe and use this value to calculate the flow rate to see if there's an advantage for the larger connectors in this aspect. I am also going to measure the water regained after the water has been ran through the set to test the quality of the seal. It was Aditya's idea to create a control tubing length with not connectors and to do the testing for each set in a trial of three for consistency. It was also his idea to vary which set I'm testing randomly in order to minimize the human bias that a manually powered syringe introduces.

Conclusions/action items:

Follow out with this testing plan to conclude on which connectors are favored. Run statistics on the results.

12/03/2020 Connector Comparison Test Results

Cate FLYNN - Dec 09, 2020, 9:32 AM CST

Title: Connector Comparison Test Results

Date: 12/03/2020

Content by: Cate Flynn

Present: NA

Goals: Recap the findings of the connector test.

Content:

After conducting this test, I have concluded that the 1/8" connectors are better than the 1/16” connectors. The fabrication for the 1/8" is a bit more difficult as the tubing needs to be heated to place the connectors, but once the tubing cools there is a very strong seal whereas the 1/16” connectors can just be slid in during fabrication, but also can be displaced from only a moderate amount of force.

I used the times to load the 3 ft of tubing with the 2.5 mL syringe to calculate the flow rates for the two sets. The flow rate for the 1/8" connectors is 16.57 mL/min and the flow rate of the 1/16” connectors is 16.53 mL/min, so the larger connectors produce a marginally greater flow rate. (Calculations included below)

2.5 mL of water was used to see how much water was lost after moving through each set. No tubing set, including the control tubing that had no connectors, retained all 2.5 mL This is not due to an issue with the seals, but rather the droplets that accumulate on the inside of the tubing that are difficult to remove. This will be an important consideration for possible sterilization efforts as this tubing has always been thrown away after one use and has an inclination to hold on to particles and will likely do the same with blood.

I generated a MATLAB script where I entered the times for loading each set and the water regained data for the 1/16" connector set, the 1/8" connector set and the control set. Using MATLAB, I found the average time for the 1/16" and 1/8" sets (10.43 s and 10.403 s), the standard deviation of the times (0.0100 and 0.0058) and the standard error of the times (0.0058 and 0.0033). These figures displayed that the times were fairly consistent between the two sets.

I sent my script to Aditya for him to run an ANOVA test with the water regained data to guarantee that there is no significant correlation and that the water regained was uniform across sets as we suspect.

I generated a graph in Excel with the water regained data as well, showing mean and error bars (also attached below).

Connector testing
Dec 2, 2020 at 8:44 AM

ID	1/16" (s)	1/8" (s)
Trial 1	10.42	10.40
trial 2	10.43	10.41
trial 3	10.44	10.40
avg	10.43	10.403

Time trials
std 1/16 = 0.0100
std 1/8 = 0.0058
Std. error 1/16 = 0.0058
Std. error 1/8 = 0.003

$3\,ft \cdot \frac{1\,m}{3.2808\,ft} = 0.9144\,m$

velocity (avg)

$1/16": \frac{0.9144\,m}{10.43\,s} = 0.0877\,m/s$

$1/8": \frac{0.9144\,m}{10.403\,s} = 0.0879\,m/s$

Flow rate $Q = AV$

$A = \frac{\pi}{4}(d^2) = \frac{\pi}{4}(2\,mm)^2 = \pi\,mm^2$

$1/16": 0.0877\,\frac{m}{s}\left(\frac{1000\,mm}{1\,m}\right)\left(\frac{60\,s}{1\,min}\right) \times \pi\,mm^2 =$

$1653.06\,\frac{mm^3}{min} \times \left(\frac{0.1\,cm}{1\,mm}\right)^3 = 16.53\,mL/min$

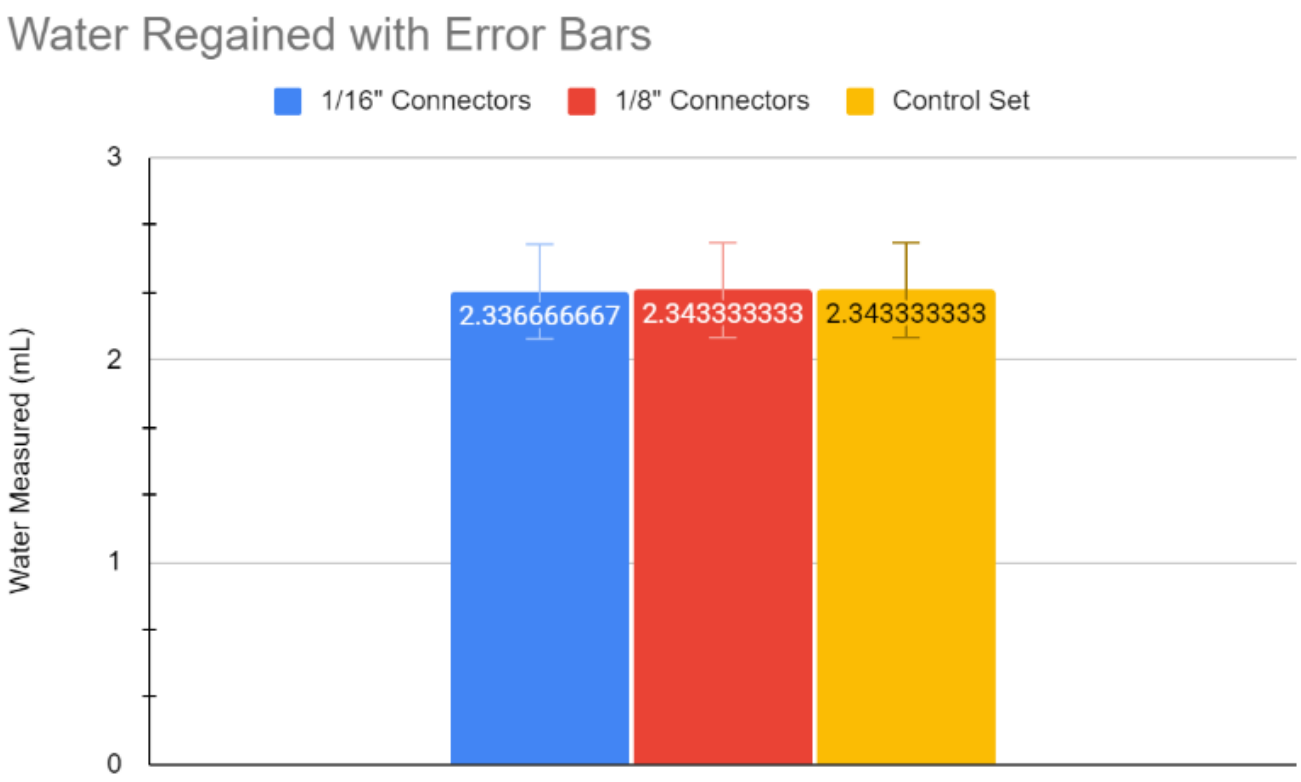
for 1/8": $16.57\,\frac{mL}{min}$

Starting volume for all tests:

2.5 mL

water reaquired:

ID	1/16" (mL)	1/8" (mL)	control
trial 1	2.32	2.33	2.37
trial 2	2.35	2.36	2.34
trial 3	2.34	2.34	2.32
avg	2.337	2.343	2.343



Conclusions/action items:

This test has helped us to determine that the 1/8" inner diameter connectors are favorable for fabrication over the 1/16" inner diameter connectors.

20200906 Initial Research

LOKESH KUMARAVEL - Oct 05, 2020, 8:02 PM CDT

Title: Initial Research

Date: 09/6/2020

Content by: Lokesh Kumaravel

Goals: To familiarize myself with the project, and to get an idea of what the semester will look like in terms of a deliverable

Content:

Project Description (*As described on BME Design Website)*[1]

Apheresis equipment uses centrifugal technology to separate whole blood into its components, collecting platelets (plateletpheresis) and returning the uncollected components to the donor. Unlike whole blood collection, apheresis is a technique that does not deprive the donor of blood components, other than platelets, and thus is a potentially safer and more effective method of platelet collection. These collected platelets can then be used in regenerative medicine to create platelet lysate. This employed method of plateletpheresis to produce platelet lysate in human medicine has also been proven to be feasible and safe in dogs and has been demonstrated to be feasible in horses. The cost for tubing used in human medicine runs $2000-2600 per time the machine is used. Plus in human medicine tubing is only allowed to be used once for each patient. For the use in dogs and horses up till now, the materials used such as tubing were thrones used in humans. This makes it cost prohibitive to use this machine in veterinary medicine more frequently. We would like the students to come up with ideas to create parts of this machine such as tubing and such to make it more economically and cost effective to be used in veterinary medicine. That way this procedure could collect platelets from large animals such as horses and cows.

Initial Questions:

What other parts of the machine could be made more economically efficient?

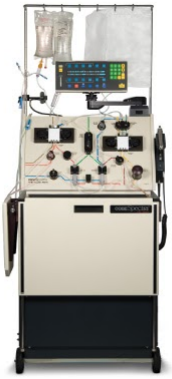
What causes the tubing to cost so much?

Why can’t we sterilize the current tubing?

How essential are the parts of the tubing? Can they be removed, but still preserve functionality?

COBE Spectra Apheresis System [2]

- Perform tailored procedures for a wide variety of patients using continuous flow centrifugal technology
- Customize procedures to fit operator needs, providing flexibility in therapeutic apheresis and transfusion medicine
- Utilize a wide array of therapeutic apheresis procedures all on one platform
- System includes
    - Four pumps, Five valves, Optical sensors, Display Panel, Centrifuge Container, Return flow controller
- The protocol to focus on is the Therapeutic Plasma Exchange
- Brochure Includes basic information for the consumer
    - https://www.terumobct.com/Public/306670810.pdf
- How it works
    - The system draws whole blood from a donor or patient, adds anticoagulant, separates the blood components, collects or removes specific components and returns uncollected components to the donor or patient. In therapeutic plasma exchange and red blood cell exchange procedures, appropriate replacement fluid is continuously returned.



- Company is making a new machine, this one will not longer be on the market soon
- Full Manual (Contains information about various parts their functions listed in detail) Technical specs also listed
    - http://startrinity3.com/mssn/04/Apheresis%20System%20Essentials%20Guide.pdf

Conclusions/Action Items: Meet with group and discuss basic plans for the project

Sources:

[1] BME Design Website https://bmedesign.engr.wisc.edu/projects/f20/plateletpheresis
[2] COBE Spectra Apheresis System Website https://www.terumobct.com/cobe-spectra

20200908 Platelet Lysate

LOKESH KUMARAVEL - Oct 05, 2020, 9:49 PM CDT

Title: Platelet Lysate Research

Date: 09/08/2020

Content by: Lokesh Kumaravel

Goals: To research platelet lysate, the reason the client needs the device

Content:

What is Platelet Lysate? (National Spine and Pain Center) [1]:

- Platelets release into the body in a timed manner to promote healing within the body
- Physicians often need access to all of the growth factors in platelets aid in faster healing in the body
- Platelet lysate breaks open platelets to release all growth factors into body at once

Platelet Lysate (Ortho Regenerative 2020) [2]:

- Blood is extracted from a donor via blood draw and then put into a centrifuge to spin the blood into its different parts
    - The COBE apheresis system does this and then returns red blood cells into the donor
- The platelets are then frozen
    - The ice crystals formed when freezing and thawing is enough to break the platelets allowing for access to all the growth factors

Simple tube centrifugation for processing platelet-rich plasma in the horse (Fontenot et al., 2012) [3]:

- Blood withdrawn from the horse's jugular vein
- This study used a separate blood centrifuge to extract plasma
    - Successfully was able to extract plasma and create platelet lysate in a horse

Platelet lysate obtained via plateletpheresis performed in standing and awake equine donors (Sumner et al., 2017) [4]

- The study used the COBE Spectra machine to extract plasma
- Concluded that it is feasible to do
- They used six female horses that were chemically restrained
- Plateletpheresis Procedure
    - Followed the instructions as per COBE Apheresis device
    - Used dual-needle procedure option of the device
    - disposable tubing set was primed with 0.9% sodium chloride plus 20 cc acid-citrate-dextrose formula A (Fenwal) in the inlet line before attachment to the horse.
    - Machine parameters set for 7ft 500lb female human
- 1L of platelets collected after 3 hours of extraction

Conclusions/ Action Items: Platelet lysate can be performed on large animals like horses. The platelet can be used similarly to that of human platelets where they can provide growth factors to horses in need. Apheresis via the COBE device is feasible.

Sources:

[1]: National Spine & Pain Centers. (n.d.). Retrieved September 08, 2020, from https://www.treatingpain.com/treatments/platelet-lysate/

[2]: Platelet Lysate. (2017, June 06). Retrieved September 08, 2020, from https://orthoregenerative.com/platelet-lysate/

[3]: Fontenot, R., Sink, C., Werre, S., Weinstein, N., & Dahlgren, L. (2012, December). Simple tube centrifugation for processing platelet-rich plasma in the horse. Retrieved September 08, 2020, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3500116/

[4]: Sumner, S., Naskou, M., Thoresen, M., Copland, I., & Peroni, J. (2017, April 25). Platelet lysate obtained via plateletpheresis performed in standing and awake equine donors. Retrieved September 08, 2020, from https://onlinelibrary.wiley.com/doi/full/10.1111/trf.14124

20201015 Real World Applications

LOKESH KUMARAVEL - Dec 09, 2020, 12:19 AM CST

Title: Real World Need For Platelet Lysates

Date: 10/15/2020

Content by: Lokesh Kumaravel

Goals: To understand the problem that this machine/tubing set could potentially solve

Content

Platelet lysates are being used has a novel method to provide supplements to an equine bone marrow derived mesenchymal stem cell. The growth factor rich lysates provide nourishment for the cell cultures as they grow in vitro. [1] Without a supplement mesenchymal stem cells have a hard time growing. Ordinarily a fetal bovine serum is used to supplement the cell cultures. FBS is acquired from the fetuses of pregnant cows during slaughter. Fetuses are most likely exposed to pain or at least discomfort during the procedure making the harvest of FBS an inhumane procedure. It has been recommended that FBS is not used as supplements and we should move towards a more humane or synthetic alternative. [2] In addition, there is evidence showing that FBS contain endotoxins and xenogeneic antigens that may alter the phenotype of the MSCs. This would make the body reject the stem cells. [1]

Platelet lysate extracted from equines may be an alternative. The procedure is conducted on fully grown horses and has been tested to be proved functional. The results of an experiment conducted by Naskou, M.C., Sumner, S.M., Chocallo, A. *et al. ,* indicate that equine platelet lysate actually produced more viable stem cell cultures without inhibiting growth.

The tubing set paired with the machine could provide a fast and reliable way of harvesting large amounts of equine platelets to be then turned into lysate. The process would be cheap and could help anyone that would benefit from cell therapy.

Dr. Brounts, Dr. Galipeau et al, have used platelets extracted from equines to create WEPLEX an enhanced version of lysate. Lysate is acquired from an equine, concentrated and washed. If is later lysed with Triton X-114. This newly developed lysate contains 266 fold the platelet-derived growth factor content relative to normal platelet rich plasma. The researchers purpose that WEPLEX can serve as an anti-inflammatory biological therapy across mammalian species. WEPLEX was administered to mice with colon inflammation. The mice were able to recover and maintain a healthy body weight.[3] Meaning WEPLEX has cross species compatibility and could potentially be used in humans as well. Currently, the team has to extract whole blood from a horse and manually separate it out into its components. Our tubing set would streamline the extraction processes allowing for the researchers to have more time and lysate to study.

Conclusions:

Platelet lysates have broad applications in the world. People across the globe suffer from muscular degeneration, chronic inflammatory diseases, and other illnesses that can be treated via the growth factors of lysate. Research needs to be continued before WEPLEX can be used in humans, however our tubing set will aid in the efforts of Dr. Brounts and Dr. Galipeau potentially having positive effects across the entire medical field.

Sources:

[1]: Naskou, M.C., Sumner, S.M., Chocallo, A. *et al.* Platelet lysate as a novel serum-free media supplement for the culture of equine bone marrow-derived mesenchymal stem cells. *Stem Cell Res Ther* 9, 75 (2018). https://doi.org/10.1186/s13287-018-0823-3

[2]: Jochems CE, van der Valk JB, Stafleu FR, Baumans V. The use of fetal bovine serum: ethical or scientific problem? Altern Lab Anim. 2002 Mar-Apr;30(2):219-27. doi: 10.1177/026119290203000208. PMID: 11971757.

[3]: Pennati A, Apfelbeck T, Brounts S, Galipeau J. Washed Equine Platelet Extract as an Anti-Inflammatory Biologic Pharmaceutical. Tissue Eng Part A. 2020 Sep 30. doi: 10.1089/ten.TEA.2020.0160. Epub ahead of print. PMID: 32854583.

20201017 Safety and Standards

LOKESH KUMARAVEL - Dec 09, 2020, 12:19 AM CST

Title: Safety and Standards

Date: 10/17/2020

Content by: Lokesh Kumaravel

Goals: To understand any safety and standard requirements

Content

When working with any biomaterials, safety is number one priority. The use of our design will require handling of blood components so standard blood drawing and transporting safety apply. The FDA does not have strict or explicit blood safety regarding animal blood, however several Universities have provided a safety guidelines for processes animal blood.

University of Kentucky [1]

- Highlights safe blood drawing amounts from different types of animals including horses
- Explain common areas of extraction for blood from animals

OSHA Guidelines [2]

- Highlights common safety and standard when handling blood
- Provides guidelines for save blood practices in a laboratory

When it comes to the tubing set, the main concern of safety is leaking of blood components. A tubing material that is safe to transport blood is required. This will protect both the operator of the machine and the animal whose donating. An aspect that needs to be considered for the materials is biocompatibility. This is the only standard and regulation in effect for our device.

The FDA defines biocompatibility as the ability of a device material to perform with an appropriate host response in a specific situation. Our specific situation is direct contact of living biocomponents (blood) with the tubing material. According to the FDA, a new material that is used in situations where biocompatibility is a concern requires a biocompatibility test. The material must follow all standards for biocompatibility. The FDA will test the material for cytotoxicity, irritation, or any other issues that may arise. There are exceptions for when this test is not needed. If a material has extensive literature about its chemical and physical abilities then no test is needed. Also if a material has a long history of safe use then a new test is not needed. [3]

Conclusions/Action Items:

It its unlikely that we actually handle blood and run the tubing set with an actual horse this semester, so blood handling and safety is not a large concern. If we do run blood components through the tubing as a test then we will have our client, Dr. Brounts, help us with that. The main safety and standard issue is biocompatibility. It will be easiest for our case to choose materials that already have been approved by the FDA and are in use in the field. PVC tubing is a common material used for blood tubing.

Sources:

[1]: https://www.research.uky.edu/division-laboratory-animal-resources/guidelines-blood-collection-laboratory-animals

[2]: https://www.osha.gov/Publications/laboratory/OSHA3404laboratory-safety-guidance.pdf

[3]: https://www.fda.gov/media/85865/download

20200922 Initial Impression of Apheresis Machine

LOKESH KUMARAVEL - Oct 05, 2020, 11:11 PM CDT

Title: Initial Impressions of the COBE Spectra Machine

Date: 09/22/2020

Content by: Lokesh Kumaravel

Present: Lokesh Kumaravel and Cate Flynn

Goals: To meet with Dr. Pinnati and get a look at the COBE Spectra Machine

Content:

The machine was in very good condition despite some rust spots here and there. One thing that stood out was the weight of the machine. Although small in size the machine is extremely heavy. I'd say similar to that of a couch. We discussed with Dr. Pinnati about moving the machine. We are free to do so however it cannot be transported via car. It simply weighs too much. Dr. Pinnati gave us permission to even wheel the machine to one of our apartments. We also acquired a set of the current tubing for studying purposes.

Design Notes:

- The dimensions of new improved tubing cannot change too much. The machine has set size parameters that need to be kept otherwise the tubing won't fit.
- We may want to consider different colored tubing since the machine uses different tubing for different purposes
- We may want to consider using the machine with water just to understand the flow of liquids.

Conclusions/Action Items: Meet with team and clients during client meeting and understand the final deliverable the client is looking for. Ask advisor about moving the device to a more accessible location

20200930 Hands on Investigation of Current Tubing

LOKESH KUMARAVEL - Oct 06, 2020, 12:30 AM CDT

Embedded private docs from Google Drive cannot be displayed in PDF

LOKESH KUMARAVEL - Dec 07, 2020, 2:09 PM CST

Title: Hands on Investigation of Current Tubing

Date: 09/30/2020

Content by: Lokesh Kumaravel

Goals: To further understand the intricacies of the tubing that needs to be replicated

Content:

Procedure:

1. I opened the sealed sterile tubing package. The opened package is shown in Fig. 1
2. I removed all the said contents.
    1. Note: The whole tubing set including bags and needles came out as one long set. They are all interconnected.
3. I cross referenced the tubing with the tubing schematic shown in Fig. 12
4. Ran into some difficulties with arranging the tubing according to the image above. The tubing was heavily tangled. The best possible recreation I could make is shown below in Fig. 11
5. After taking detailed pictures of the different parts of the tubing I placed the tubing back into it’s package.

The tubing is much more complex than first aniticapted. There are none tubing parts of the tubing that need to be considered. Mesh supports (Fig. 2), plastic joints (Fig. 3and Fig. 4), filters (Fig. 5) , plastic feeders (Fig. 6) and needles (Fig. 7) all are a part of the tubing. There is also a large non-tubing circle that connects to the centrifuge (Fig. 8) along with miscellaneous plastic parts whose function is unclear (Fig. 9 and Fig. 10). The tubing cannot be separated. The joints seem to be joined via adhesive. Accurate measurements could not be obtained.

Design Ideas: We may have to consider sterilization as the only option. However, a simplified tubing set may be produced. As per my observations, a lot of the parts on the device seem to serve as safety/regulatory measures. Considering the device is being used on a horse with less standards and requirements we may be able to remove some parts. I think it's possible to cut different diameters of tubing and join them together to resemble the current tubing.

Design Problems: The tubing is unable to disconnect. Needles and bags are connected to the tubing,and non-removable. Unable to accurately create flow of liquids without the actual machine to guide the tubing.

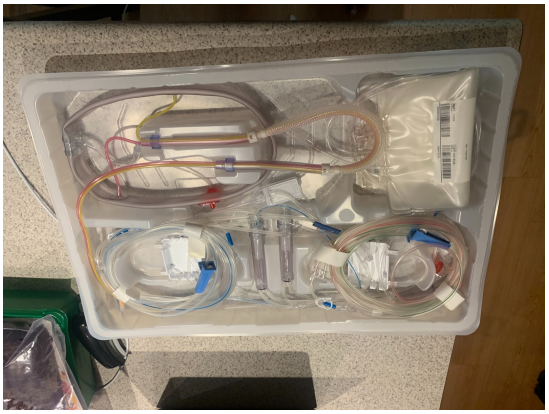


Figure 1: Tubing set in its package

Figure 2: Mesh coverings

Figure 3: Plastic Guide for Tubing

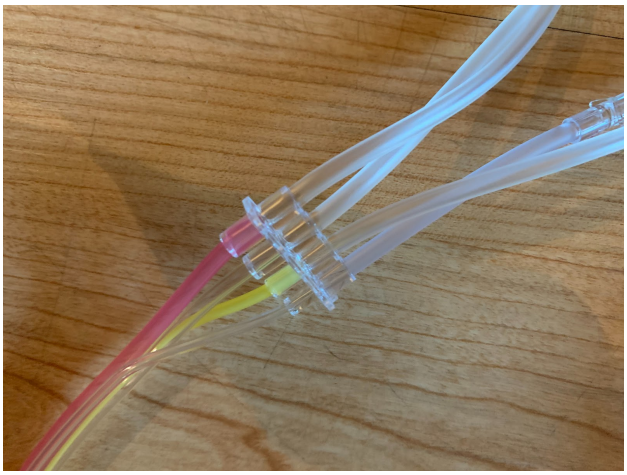


Figure 4: Plastic Joint connecting different types of tubing

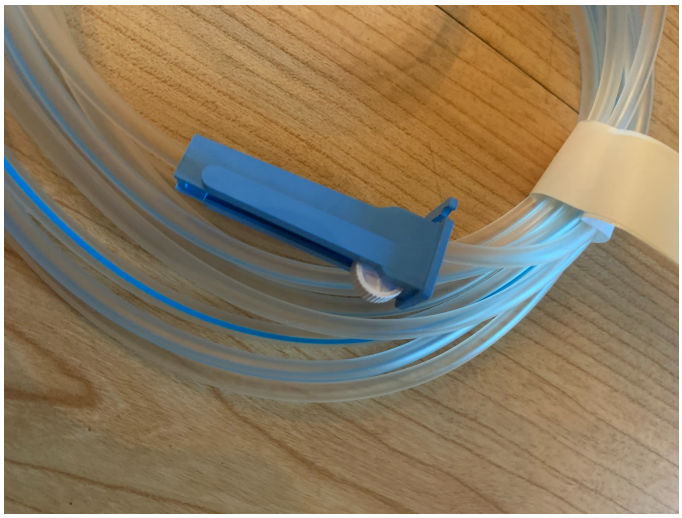


Figure 5: Plastic Feeder/ Clamp

Figure 6: Filter housing case

Figure 7: Needle

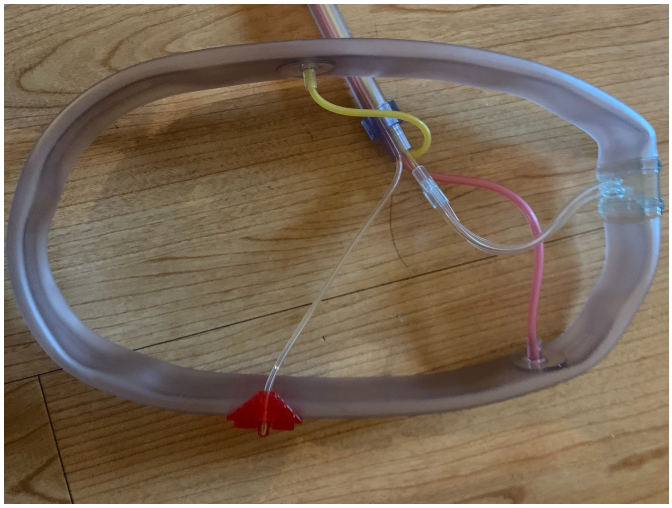


Figure 8: Centrifuge Loop

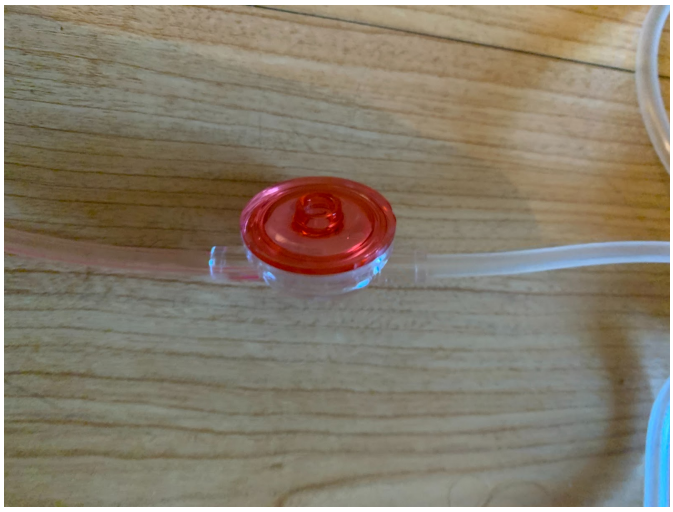


Figure 9: Plastic circle? (Pressor Sensor)

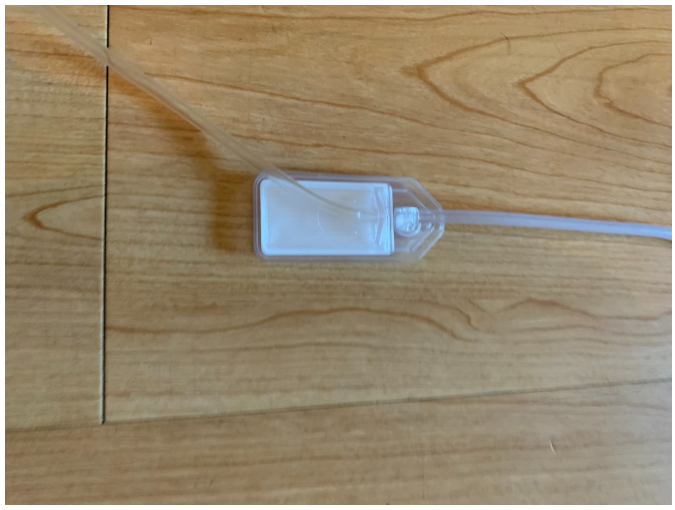


Figure 10: Plastic piece, unkown function

Figure 11: Full Tubing Set

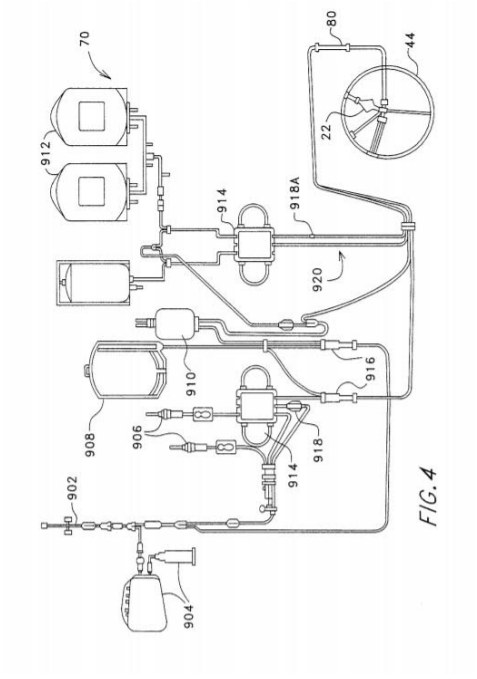


Figure 12: Schematic of Tubing Set

Conclusions/Action Items: Inform team of findings and continue to research the function of each individual part. Contact manufacturer to find specific measurements and ask client if they have information or suggested on how to go about simplifying the design

20201004 Materials Research

LOKESH KUMARAVEL - Dec 09, 2020, 12:21 AM CST

Title: Materials Research

Date: 10/04/2020

Content by: Lokesh Kumaravel

Goals: To determine different specifications materials have and to have an idea of an ideal material to use when deciding the tubing material

Content:

Material technical specifications that need to be meet

- Flexible, Leak proof
- If needed to be sterilized via autoclave then
    - Withstand temps of 250 F
    - Withstand pressure of 15psi
- If needed to be sterilized via chemicals
    - Have some level of chemical resistance

Current tubing is comprised of PVC (Polyvinyl Chloride) medical grade tubing

- Grainger Technical Specs [1]

    - Tensile Strength 2,200 psi
    - Max psi of 60 at 70 degrees
    - Temp range: 25-150
    - Length 100ft
    - Flexibility: Flexible
- PVC Chemical Compatibility [2]
    - Rigid PVC is chemically resistant to many acids, salts, corrosives, bases, fats, and alcohols
    - The melting point of PVC is low, around 100°C / 212°F)
    - Maximum operating temperature is around 60°C / 140°F
    - NOT compatible with tetrahydrofuran or acetone, often incompatible with solvents

Ethyl Vinyl Acetate Tubing (EVA)

- Grainger Specs [3]

- Tensile Strength: 150 psi
- Max psi: 150 at 70 degrees
- Temp Range -30-150 degrees
- Length 200ft
- Flexibility: Flexible, but reinforced

- Plastic Tubing Specifying Guide / Chemical Resistance Properties [4]
  - Weak Acids, Bases, Strong Alkalies, Weak Alkalis: Excellent
  - Strong Acid: Poor
- United States Plastics Corp.[5]
  - Claims this is excellent for surgical, hospital and pharmaceutical alternatives

Silicone Tubing

- Grainger Spec [6]

  - Tensile Strength: 1,200 psi
  - Max Psi 40
  - Temp Range: -100 to 400 degrees F
  - Length 100ft
  - Flexibility: Not an issue
- United States Plastics Corp. [7]
  - Designed for high-purity applications, Tygon® Sanitary Silicone Tubing's ultra-smooth inner bore can reduce the risk of particle entrapment and microscopic buildup during sensitive fluid transfers. In-house analysis of the inner surface of Tygon® Sanitary Silicone Tubing compared to other silicone tubing shows that it is up to three times smoother. Additionally, this smoother fluid path facilitates **complete system cleaning and sterilization.**

Conclusions/Action Items: Depending on the design route we take each material has its advantages and disadvantages, silicone however seems like the best option due to its ability to be sterilized via chemical and autoclave methods

Sources:

[1] https://www.grainger.com/product/4EGU8?gclid=CjwKCAjwiOv7BRBREiwAXHbv3Oj7t0aplYNyd3TOH-h_oTnn3yccuoHrlJ0fr-CV6DghPWSMkYWRtRoCSQ8QAvD_BwE&cm_mmc=PPC:+Google+PLA&ef_id=CjwKCAjwiOv7BRBREiwAXHbv3Oj7t0aplYNyd3TOH-h_oTnn3yccuoHrlJ0fr-CV6DghPWSMkYWRtRoCSQ8QAvD_BwE:G:s&s_kwcid=AL!2966!3!281733298256!!!g!663156281672!&gucid=N:N:PS:Paid:GGL:CSM-2293:99F1R6:20501231

[2] https://www.calpaclab.com/pvc-polyvinyl-chloride-chemical-compatibility-chart/

[3] https://www.grainger.com/product/GRAINGER-APPROVED-200-ft-Flexible-4EGU3

[4] https://www.hudsonextrusions.com/specifying-guide/

[5] https://www.usplastic.com/catalog/item.aspx?itemid=83907

[6] https://www.grainger.com/product/E-JAMES-100-ft-Silicone-Tubing-4CHN7

[7] https://www.usplastic.com/catalog/item.aspx?itemid=23278&catid=864

20201011 Material Cost Research

LOKESH KUMARAVEL - Dec 05, 2020, 9:07 PM CST

Title: Materials Research Regarding Price Points

Date: October 11, 2020

Content by: Lokesh Kumaravel

Present: Lokesh

Goals: To gauge price points of different tubing materials among UW Vendors

Content:

Estimated Length of Tubing for a Single Set: 60ft

PVC Tubing 1/8 “ ID, 3/16” OD (Current Material Used)

- Grainger: $0.49/ft ($29.40 per set) [1]
    - Currently, the best option. It is quite cheap so the materials inability to be autoclaved is not a worry.
- Fisher Scientific: $0.97/ft ($58.20 per set) [2]

Ethyl Vinyl Acetate Tubing (EVA) (Suitable size for the apheresis device is not readily available)

Silicon Tubing 1/8” ID, 3/16” OD

- Grainger: $0.95/ft ($50.00 per set) [3]
    - This silicon tubing is not cured by another substance, hence making it substantially cheaper. However, you lose the properties, mainly it can only be autoclaved a few times before turning gummy.
- Fisher Scientific: $6.40/ft (2mm inner diameter) ($384.00) [4]
    - Not recommended, silicone tubing, although perfect for our use, tends to be cured with other materials such as platinum or peroxide. The addition of these materials adds to its structural integrity and also reduces risks for particle entrapment inside the tubing. These sets are used in laboratories and can be easily sterilized. All appealing, however the cost is expensive.

Miscellaneous Parts that cannot be fabricated and need to be purchased and added on separately (non reusable)

- Collection Bags
    - The tubing set needs 3 per use
    - 10L EVA collection bags: $86.00/ bag (Fisher Scientific) [5]
        - Wasn’t anticipating the cost of these collection bags to be so high, the client may have cheaper way of accessing them
- Needles and Catheters
    - Spoke with client prior, she has multiple 10,12,14 gauge needles and catheters at her disposal
- Saline and Anticoagulant Bags
    - Client may have access

Conclusions/action items:

Of the materials researched so far, PVC seems to be the only viable material in terms of expense. However, a discussion with our advisor and client may lead us in the right direction to try another more effective material. The cost of materials that cannot be reused no matter what like the collection bags and the needles and solution bags might be quite high. In the next client meeting, ask whether or not those components need to be included in the $100 per use goal.

Sources:

[1]: https://www.grainger.com/product/TYGON-PVC-Tubing-22XH86

[2]: https://www.fishersci.com/shop/products/nalgene-non-phthalate-pvc-tubing-25/1371276?keyword=true

[3]:https://www.grainger.com/product/USA-SEALING-Tubing-55YG26

[4]:https://www.fishersci.com/shop/products/wheaton-peroxide-cured-silicone-tubing-for-general-lab-use-8/02928101#?keyword=silicon+tubing

[5]:https://www.fishersci.com/shop/products/corning-flexible-packaging-systems-eva-collection-bags-7/mt9120036?keyword=true

20201019 Microfluidic Tubing Research

LOKESH KUMARAVEL - Dec 09, 2020, 12:21 AM CST

Title: Microfluidic Tubing Options

Date: October 19, 2020

Content by: Lokesh Kumaravel

Present: Lokesh

Goals: To research tubing used in microfluidics as advised by Dr. Kinney

Content:

Microfluidic Tubing and Sleeves [1]

- tubing and sleeves, which are generally used to transport small volumes of liquids to your chip
- unions and adapters, which are used to connect fittings of the same or different threading
- fittings and connectors, which are used to connect, either with rigid or soft tubing, your microfluidic device or your Lab-on-a-chip to external elements such as pumps and reservoirs

Commonly used tubing for microfluidics [1]

- **Microfluidic PEEK tubing (Polyetheretherketone)**
  - Highly biocompatible material paired with chemical compatibility
  - Often described as highest performing thermoplastic tubing due to high chemical resistance
  - Swelling effect can occur on tubing when used in methylene chloride, THF, and DMSO
  - Can be used in high and low pressure applications
- **Microfluidic PTFE Teflon tubing (Polytetrafluoroethylene)**
  - Transparent, chemically inert, nontoxic
  - Has high flexibility so mainly used in low pressure environments
- **Microfluidic FEP tubing (Fluorinated ethylene-propylene)**
  - Good substitute for PTFE, used in low pressure environments
  - High chemical resistance
- **Microfluidic ETFE tubing (Ethylene tetrafluoroethylene)**
  - The highest level of chemical resistance
  - Rigid tubing used in medium pressure environments

Variations of Fluoropolymer tubing seem to be the go to in microfluidics. All of the tubing have strong resistance to chemicals and have high max temperatures so they can be easily sterilized.

Fluoropolymers Available for Purchase (Grainger)

PEEK Tubing: (Sizes are quite small)

- Grainger Specs [2]
  - Cost: Currently unavailable on Grainger
  - Mas Pressure: 5000 psi
  - Temp Range: -320F- 500F

PTFE Teflon Tubing

- Grainger Specs [3]
    - Cost: $18.36 for 10ft
    - Max Pressure: 190 psi
    - Temp Range: -150F - 500F

FEP Tubing

- Grainger Specs [4]
    - Cost: $68.38 for 50ft
    - Max Pressure: 360 psi
    - Temp Range: -100F - 400F

ETFE Tubing (Couldn't find on Grainger)

- Specs as per Polyfluor [5]
- Strong mechanical properties
- Excellent impact resistance
- Good resistance to stress cracking
- Working temperature from -200°C to 150°C
- Good permeability

PFA Fluoropolymer Tubing

- Grainger Specs [6]
- This tubing is one I found by just typing in fluoropolymer. It tends to have a better chemical resistance than FEP, and is used in pharmaceutical, laboratory, and sampling applications. [7]
    - Cost: $78.39 for 50ft
    - Max Pressure: 320 psi
    - Temp Range: -450F - 500F

Figure 1: PEEK Tubing

Figure 2: PTFE Teflon Tubing

Figure 3: FEP Tubing

Figure 4: PFA Fluoropolymer Tubing

Conclusion: Microfluidic and fluoropolymer tubing seem to be good route for tubing in our project. The tubing satisfies all criteria and is widely used in the industry of medicine, so it should be a perfect fit. However, one observation I made is that the tubing seems to be more rigid and hard than the others. The images make it seem like they are a hard plastic type tubing. More research will need to be conducted for hardness.

Sources:

[1]: https://www.elveflow.com/microfluidic-reviews/general-microfluidics/the-basics-of-microfluidic-tubing-sleeves/

[2]: https://www.grainger.com/product/ZEUS-MANUFACTURING-50-ft-PEEK-Tubing-4KLC1

[3]:https://www.grainger.com/product/USA-SEALING-Tubing-742U96

[4]: https://www.grainger.com/product/GRAINGER-APPROVED-Tubing-53XL31

[5]: https://www.polyfluor.nl/en/products/-fluoroplastic--tubing/etfe-tubing/

[6]: https://www.grainger.com/product/GRAINGER-APPROVED-50-ft-PFA-Fluoropolymer-Tubing-2VLU5

[7]: https://www.fluorotherm.com/products/fluoropolymer-tubing/pfa-tubing/

20201024 Hardness of Tubing Research

LOKESH KUMARAVEL - Dec 05, 2020, 9:48 PM CST

Title: Hardness of Tubing

Date: October 24, 2020

Content by: Lokesh Kumaravel

Present: Lokesh

Goals: To understand if hardness of tubing is something that we need to consider

Content:

On Grainger, each tube has its own hardness rating. These ratings are based on the Durometer Shore Hardness Scale. The scale measures the ability for a material to resist indentation. In tubing terms it can affect things such as bend radius.

Figure 1: Shore Hardness Scale [1]

To elaborate on these universal scales:

- Shore A Hardness Scale is used for measuring the hardness of flexible mold rubbers. These can range in hardness from very soft and flexible, to medium and somewhat flexible, to hard with almost no flexibility at all.
- Shore D Hardness Scale is reserved for measuring the hardness of hard rubbers, semi-rigid plastics and hard plastics.

The original tubing set is made from PVC (polyvinyl chloride). According to Grainger [2], PVC is rated with a hardness of Shore A: 73. This puts the tubing somewhere near the pink eraser mark for hardness. Hardness also affects the flexibility of the tubing. The apheresis machine has tight fights and sharp corners where the tubing runs through. The peristaltic pumps also have a sharp radius.

Tubing types that we are considering and their corresponding hardness:

PVC Tubing: Shore A: 73

- Same as Current Tubing

Silicone Tubing: Shore A 65

Microfluidic Tubing (Fluoropolymers): Shore D: 65-85

The microfluidic tubing is actually quite hard similar to that of a rubber wheel or even a hard hat. This will prove to difficult to bend and impossible to hook up to the machine.

Conclusions:

The final tubing set must have a Shore hardness rating in the A category within the 50-80 range.

Sources:

[1]: https://www.artmolds.com/shore-hardness

[2]: https://www.grainger.com

20201029 Initial Loading of Device

LOKESH KUMARAVEL - Dec 09, 2020, 12:24 AM CST

Title: Loading the Original Tubing Set into the COBE Apheresis Device

Date: October 29, 2020

Content by: Lokesh Kumaravel

Present: Lokesh, Cate

Goals: To understand how the tubing set integrates within the system

Content:

Cate and I went to WIMR to attempt to load the tubing set and run water through to understand the flow of liquid of the system. This is the first time he have had any major interactions with the machine.

Initial Observations:

- The overlay guide for the flow of the tubing set it not the correct procedure.
    - This presents no issues as the machine can perform different procedures, and the overlay is only a guide for a technician.
- The device seems like it has not been moved or used in quite sometime.

Procedure:

- Cate and I removed the original tubing set from its packaging and spend some time untangling it.
- We then loaded the the set onto the machine

Problems that occured

- The centrifuge loop does not fit onto the centrifuge cap. At the time we suspected that the tubing set was meant for a different machine, but after further research it was concluded that the machine had the ability to interchange caps. The tubing set we were given did not match the filler cap. A new one needs to be found, or a whole new centrifuge loop needs to be constructed.
- The device will not run unless fluid are present. When attempting to move past the loading phase error signals occurred indicating there was an issue with the access pressure sensor. There was not adequate pressure because there was no fluid. We attempted to bypass this error, but failed.

Figure 1: The centrifuge loop laid on top of the filler cap. (The diameter is much too small and the four blood component tubing sets are attached to different parts of the loop. This presents a problem since the current cap has all of the component tubings laying side by side.)

Conclusion/Action Items:

We need to acquire the proper centrifuge cap otherwise device may not run any fluids at all. We also need a saline bag and and anticoagulant bag to run the priming function. This way we will see how fluids flow through the tubing set.

20201109 Running Fluids Through the Tubing Set

LOKESH KUMARAVEL - Dec 09, 2020, 12:25 AM CST

Title: Saline Test of the Original Tubing

Date: November 9, 2020

Content by: Lokesh Kumaravel

Present: Lokesh and Cate

Goals: To run fluids through the tubing set to further our knowledge

Content:

Cate and I went to WIMR to meet with Andrea who had acquired 3 saline bags for us to use. The team had decided an anticoagulant bag was not required for running this test since the machine could be tricked into thinking saline is anticoagulant. According to Dr. Galipeau, saline and anticoagulant have relatively the same viscosity and the machine shouldn't be able to detect the difference. After loading the tubing set, we hooked up the saline bags into the saline and anticoagulant access ports. After starting the system, we were able to successfully run saline through the entire tubing set. The machine was fully functional.

Observations: The saline ran through the entire tubing set, and eventually drained out into the waste collection bag. However, an issue arises when trying to drain the tubing set of saline. There is a rinseback feature on the device, but it requires for the centrifuge loop to spin. We did not acquire the filler cap yet, so the loop could not attach to the device and spin. We were forced to let the saline sit in the tubing. A possible concern is if exposed to air the NaCl may crystallize clogging some of the tubing.

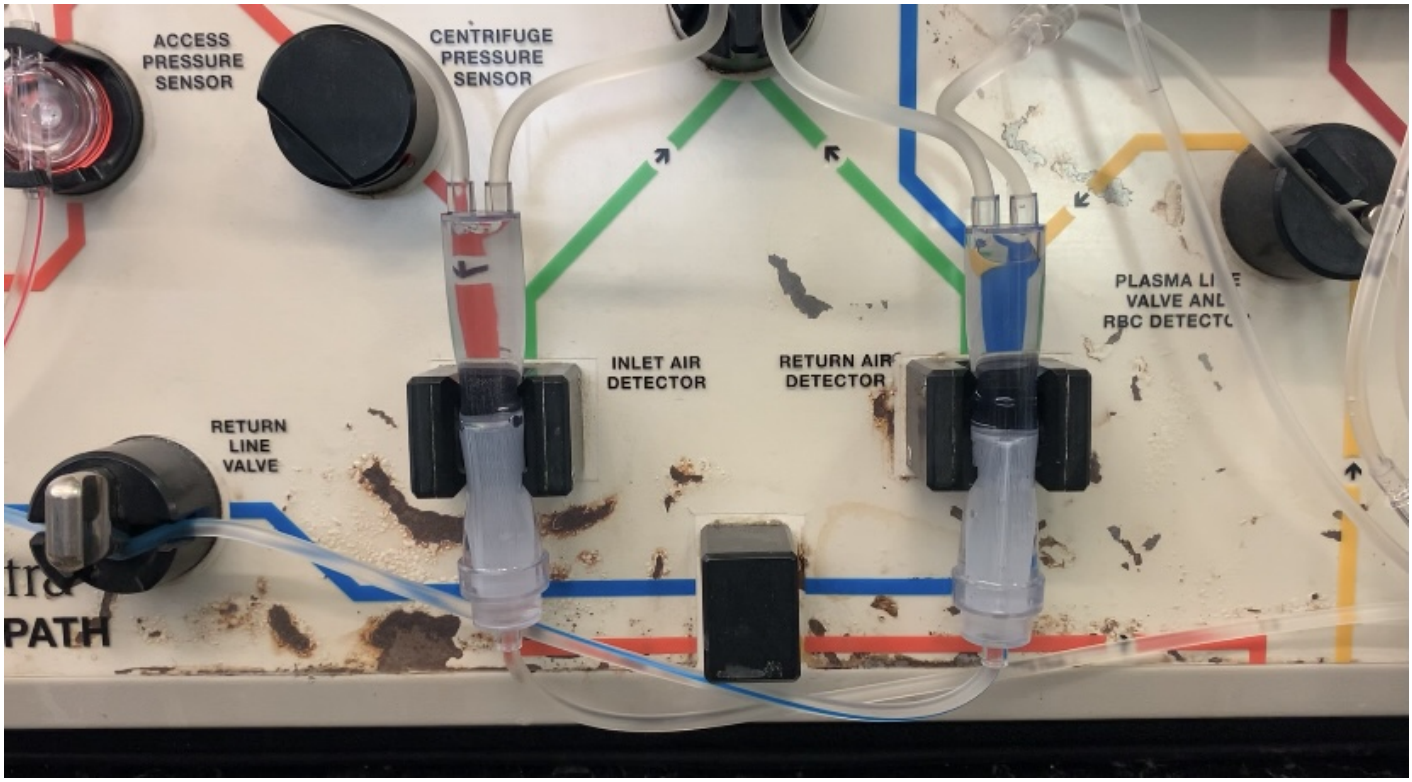


Figure 1: Air Chambers filled with saline

Figure 2: Saline and Anticoagulant Access ports filling with saline

Figure 3: Waste Collection Bag filled with saline

Conclusions/Action Items:

The machine is in fully functioning condition and a proper centrifuge loop is not required to run the priming portion of the procedure. We will need to go back to WIMR soon and find get measurements and also figure out a way to drain the saline before Thanksgiving break.

20201119 Measurements of the Tubing Segments

LOKESH KUMARAVEL - Dec 09, 2020, 12:25 AM CST

Title: Measurements of Individual Tubing Segments

Date: November 19, 2020

Content by: Lokesh Kumaravel

Present: Lokesh and Cate

Goals: To obtain measurements of the tubing for fabrication purposes. Also access where connectors should be placed

Content:

Cate and I went to WIMR to measure the original tubing set so we could fabricate our own set with identical measurements. We also decided where to incorporate connectors and where to cut components that needed to be transferred into our tubing set.

We used a schematic of the prior tubing, however some of the lengths and components didn't line up. The schematic is for a different tubing set than our own, however it is quite similar. We decided to use this schematic as reference for the measurements, and edited some of the excess components out and added components that were missing. We were able to produce the diagram shown below. It is not to scale since the schematic isn't the one for the original tubing set.

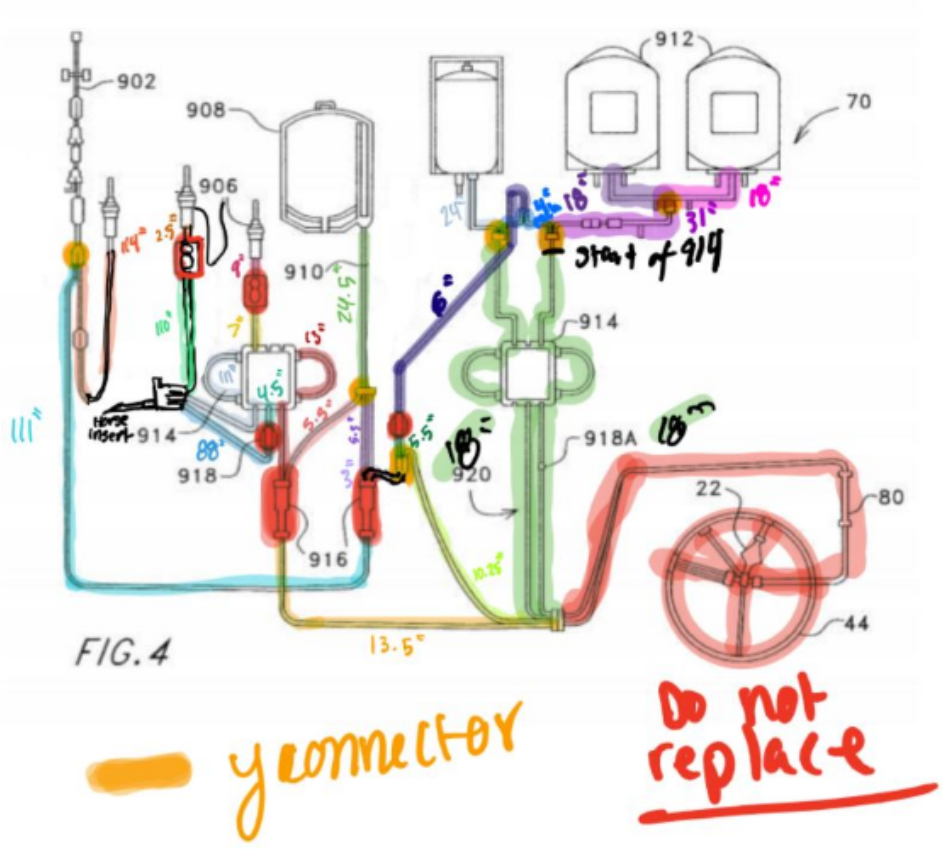


Figure 1: Tubing Schematic with measurements, connectors and components that need to be transferred

Conclusion/Action Items:

We were able to successfully obtain measurements for different tubing segments and got a good idea for how to fabricate a prototype. The next step is to use this diagram as a guide and fabricate our prototype.

20201124 Testing of Prototype

LOKESH KUMARAVEL - Dec 09, 2020, 12:26 AM CST

Title: Testing Prototype

Date: November 24, 2020

Content by: Lokesh Kumaravel

Present: Lokesh and Cate

Goals: To test and see if our prototype integrates with the machine and retains water

Content:

Cate fabricated a final prototype and brought it to WIMR. We loaded the prototype into the machine with little issues. We did run into a problem getting the tubing to lock into the peristaltic pumps. Our tubing is a little smaller than the original tubing set. When loading into the pumps the smaller tubing slides creating slack. This makes it impossible for the tubing to lock in correctly. The solution was simple however, we place a paperclip on the tubing and attached it to the pump cartridge. This prevented sliding so the machine could be loaded properly. We clamped off the return part of the tubing, because we didn't want to have saline run there. We didn't have an effective way to drain the saline, so we wanted to minimize the amount of saline in the tubing. The saline ran without and leaks through our prototype. When unloading we had to cut below the air chamber to drain the saline from the tubing.

Conclusions/Action Items:

Future teams should look into draining the tubing set after running saline through. They should also look into some sort of adhesive to lock the tubing into place to prevent sliding. Overall, the test of a success, because the machine did not detect any pressure issue, and the tubing set was able to retain fluids without any leaks.

20201202 Fabricating the Bubble Trap

LOKESH KUMARAVEL - Dec 09, 2020, 12:26 AM CST

Title: Fabrication Notes of the Bubble Trap

Date: December 2, 2020

Content by: Lokesh Kumaravel

Goals: To fabricate a model for the bubble trap

Content:

The bubble trap is required to remove air bubbles from the blood. This will protect the donor from an embolism. The original tubing set contains a complex bubble trap and due to its skinny nature it may proof to be difficult to fabricate. Nesya was able to design a simpler bubble trap using 3D printed material. Unfortunately all of the parts did not arrive on time to fabricate a fully functional prototype. The PFTE membrane which filters the bubbles did not arrive, so the prototype is only a structural model. I used a thin piece of foam to replicate the membrane for visual purposes.

Procedure:

- I began by inserting the flangeless fittings into the tubing, and then I attached the ferrule onto the tubing end.
- The fittings are supposed to be screwed onto the housing blocks, but the holes were not tapered, so I twisted them on as best as I could
- I then clamped the two housing blocks together
- The C-clamp used to hold the bubble trap together made it hard to handle so I placed tape on the inside of the housing blocks to hold them together.
    - This worked nicely for picture purposes.

Fabrication of the model was simple, however testing could not be done as the main component of the filter was not available.

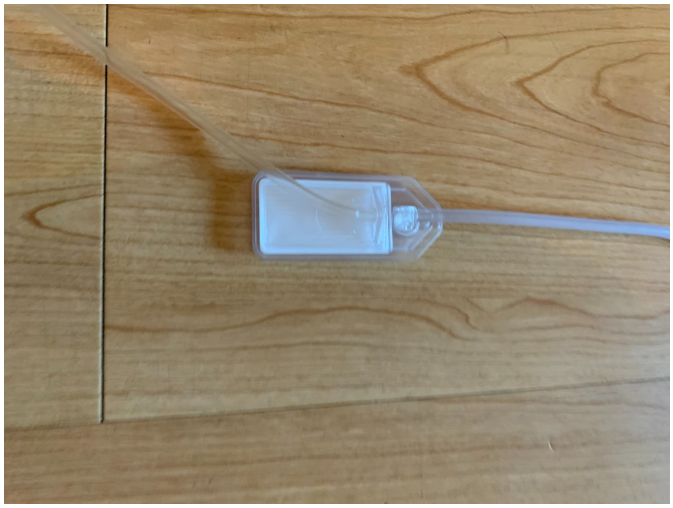


Figure 1: The Original Bubble Trap

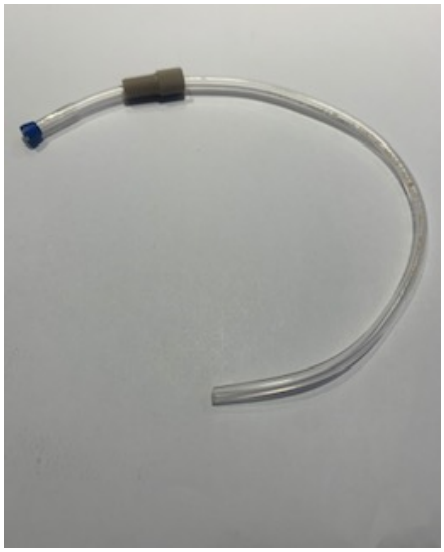


Figure 2: Tubing with flangeless fitting attached (Blue component is ferrule, darker component is the nut)

Figure 3: The nuts twisted on to the top housing block

Figure 4: The bottom housing block with foam membrane substitute

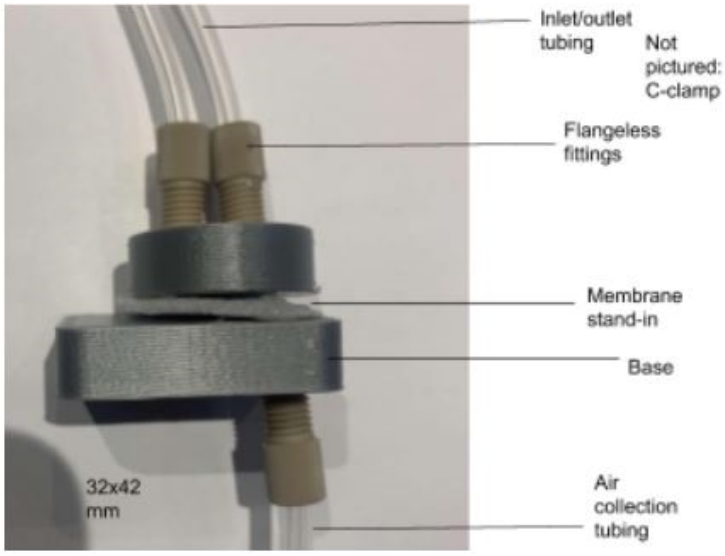


Figure 5: Completed Bubble Trap Model

Conclusions:

The bubble trap is simple and cheap to fabricate. It is hard to know how effective it will be without the membrane, but a future semester's teams should be able to test how well it works and incorporate it into a new tubing prototype.

Initial Startup of the COBE Apheresis Machine

LOKESH KUMARAVEL - Dec 09, 2020, 12:27 AM CST

Title: Initial Start-up of the COBE Apheresis Machine

Date: November 4, 2020

Content by: Lokesh Kumaravel

Present: Lokesh

Goals: To create an easy set-up guide to start and run the machine

Content:

Rough Outline Procedure:

1. Plug the machine in to a wall socket
2. Turn on the machine by flipping the switch located on the right side of the machine
    1. The machine will begin to run some basic tests to make sure start-up is all good (Few seconds of random lights flashing)
3. Follow instructions on the interface of the machine
    1. Machine will ask for what tubing set (Choose ELP)
4. Load the tubing set according to video 2
5. Attach saline and anticoagulant bags
6. Start the process
    1. The machine will begin to prime the whole tubing set, however it will get confused as to when to stop unless real anticoagulant is used.
7. Unload the tubing set

Above is a basic outline of how to get the machine up and running.

There are detailed step by step instruction videos that can be found on youtube.

1. https://www.youtube.com/watch?v=FPm5kuVauXs&t
2. https://www.youtube.com/watch?v=DbKd7tkJR_Q&t=
3. https://www.youtube.com/watch?v=dZupSOWwQaY
4. https://www.youtube.com/watch?v=NFRaIxgqb5o
5. https://www.youtube.com/watch?v=Va1Zym9xMkM

Conclusions:

These videos provide critical and highly useful demonstrations for how to start the and begin using the machine. It is fairly intuitive, however, the machine is smart and will not allow for minor errors. Pressure needs to be within the machines tolerances and there needs to be sufficient fluids (Saline and Anticoagulant) for the machine to draw from.

09/06/20 Research Notes

NESYA GRAUPE - Sep 11, 2020, 2:24 PM CDT

Title: Research Notes

Date: 09/06/2020, Edited on 09/11/2020

Content by: Nesya Graupe

Present: Nesya Graupe

Goals: To better understand platletpheresis and to learn about the project

Content:

Project Objective/Description

-Plateletpheresis uses centrifugal (moving away from the center) technology to separate blood into components, collect platelets, and return uncollected blood components to donor. Platelets form blood clots and can be turned into platelet lysate and used in regenerative medicine

-Apheresis (collecting the platelets) is very expensive. $2,000- $2,600 per time machine is used, tubing can only be used once in human medicine. What is preventing the sterilization of tubing for human medicine?

-Project goal: Create parts (such as tubing) for the apheresis machine that make it more economically effective to be used in veterinary medicine, make realistic to use on large animals such as horses and cows. Should be able to use multiple times and should be easy to sterilize. What will be done with the platelets from large animals? What will the platelet lysate be used for?

COBE Spectra Apheresis System, Terumo Blood And Cell Technologies, www.terumobct.com/cobe-spectra.

-the COBE Spectra Apheresis System

-draws whole blood from a donor or patient, adds anticoagulant, separates blood components, collects specific components, returns uncollected components to the donor

-Can remove plasma, red and white blood cells, stem cells, and platelets

Sumner, Scarlett M., et al. “Platelet Lysate Obtained via Plateletpheresis Performed in Standing and Awake Equine Donors.” *Transfusion*, vol. 57, no. 7, 2017, pp. 1755–1762., doi:10.1111/trf.14124.

-Plateletpheresis using commercially available equipment is feasible in horses

-Platelet lysate is a possible replacement for fetal bovine serum to use in cell cultures. How and why is fetal bovine serum used in cell cultures now?

-Apheresis equipment was adapted for use in horses. Set to largest body parameter (7-foot-tall, 500-pound female). Preliminary bloodwork from each horse was used to manually input baseline platelet count and total volume so that infusion rate, pump rate, and inlet flow rate would be more accurate.

-Donor platelet counts were lower than expected

Rock, G, and Dm Sutton. “Apheresis: Man versus Machine.” *Transfusion*, vol. 37, no. 10, 1997, pp. 993–995., doi:10.1046/j.1537-2995.1997.371098016435.x.

-Since the introduction of the apheresis machine in 1988, 300 million procedures have been carried out on 3000 machines. Apheresis is generally very safe.

Clinical indications and adverse reactions of platelet apheresis by Amanat ST et al in J Coll Physicians Surg Pak 2015;25:403-6.

-Plateletpheresis is generally very safe and effective.

-Anticoagulant (ACD) intoxication, which can cause hypocalcemia (low calcium levels in blood serum), may occur

-Vasovagal reaction (sudden drop in heart rate and blood pressure) is a possible complication

Clinical and clini-copathologic effects of plateletpheresis on healthy donor dogs by Callan MB et al in Transfusion 2008;48:2214-21.

-14 dogs underwent platletpheresis. The procedure was generally well tolerated. All dogs returned to their baseline platelet count by day 16

Conclusions/action items: Meet with group tomorrow (09/07/2020) to talk about research and come up with questions for client.

09/14/2020 Research Notes

NESYA GRAUPE - Sep 14, 2020, 10:57 AM CDT

Title: Research Notes

Date: 09/14/2020

Content by: Nesya Graupe

Present: Nesya Graupe

Goals: Research the COBE Spectra Apheresis System

Content:

“COBE Spectra Apheresis Essentials Guide.” Gambro BCT, Inc. http://startrinity3.com/mssn/04/Apheresis%20System%20Essentials%20Guide.pdf

-Tubing includes a preconnected separation channel
-Dual needle vs single needle extended platelet disposable tubing sets
-Dual needle vs single needle extended life disposable tubing set with LRS (leukoreduction system) chamber
-Tubing that is occluded (closed) or partially occluded can lead to malfunctions of machine

“Spectra Optia Apheresis System Owner's Manual.” Gambro BCT, Inc. https://www.terumobct.com/Public/777379199.pdf

-Extracorporeal volume- volume of blood outside the body

-Dual needle access: typical ECV: 141 mL, maximum ECV: 185 mL

- Single needle access: typical ECV: 185 mL, maximum ECV: 185 mL

-Blood and fluid flow pathways of the tubing are sterilized with ethylene oxide

-Must comply with Class II Type BF electrical safety requirements of IEC 60601-1.

Conclusions/action items: Meet with group later today, work on first version of project design specifications

09/23/2020 Research Notes

NESYA GRAUPE - Oct 06, 2020, 7:27 PM CDT

Title: Research Notes

Date: 09/23/2020

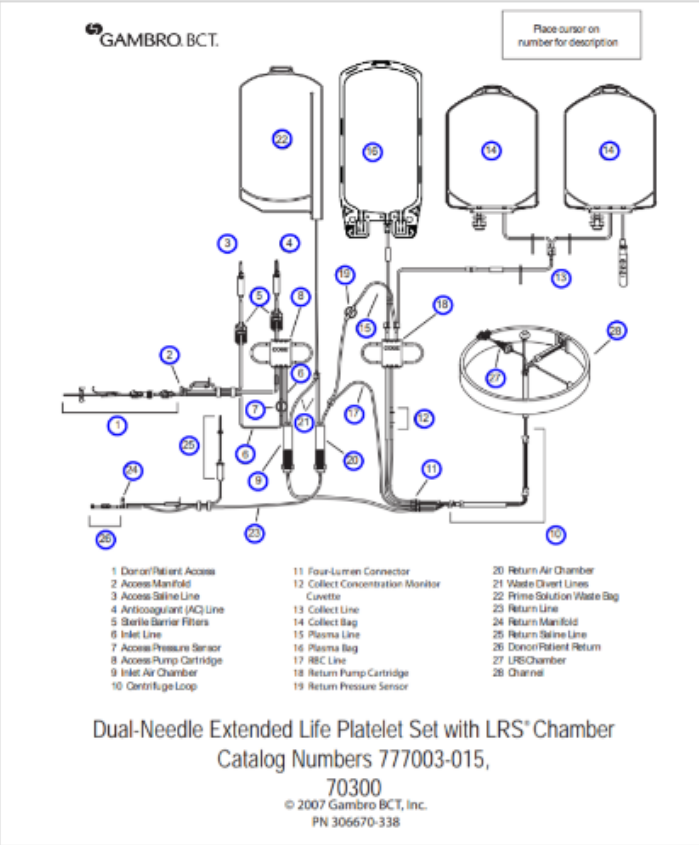
Content by: Nesya Graupe

Present: Nesya Graupe

Goals: Understand the COBE spectra apheresis machine better

Content:

This is a diagram of the Dual-Needle Extended Life Platelet (ELP) disposable tubing set with LRS Chamber. I believe that we do not need the LRS chamber but I could not find a diagram without one.



https://www.ralfstoll.de/szs/306670338-Spectra_Dgrm.pdf

Parts we need on our tubing: Platelet collection bags, collect line and return line tubes, 10 gauge or 14 gauge catheter, clamps to control blood flow

How will we add anticoagulant?

How will we connect the tubing parts together?

What parts are not necessary/ can be simplified?

What to do with saline?

Conclusions/action items: Group meeting on Friday

10/04/20 Research Notes

NESYA GRAUPE - Oct 04, 2020, 5:44 PM CDT

Title: Research notes (Human impacts)

Date: 10/04/20

Content by: Nesya Graupe

Present: Nesya Graupe

Goals: Determine what the global implications/ human impacts of this project will be

Content:

Implications

“14 May Platelet Lysate: The Potential Alternative to the Classic Epidural Steroid Injection.” Cleveland Pain Care, 14 May 1970, Available: www.clevelandpaincare.com/platelet-lysate-the-potential-alternative-to-the-classic-epidural-steroid-injection/.

-Platelets are cells that exist in blood that have healing and regenerative properties.

-Platelet rich plasma is frozen and then thawed to break the platelets and liberate enzymes and growth factors

-Lysate can be injected into the epidural space (part of the spine)

Pennati A, Apfelbeck T, Brounts S, Galipeau J. Washed Equine Platelet Extract as an Anti-Inflammatory Biologic Pharmaceutical. Tissue Eng Part A. 2020 Sep 30

-Through current methods not all growth factors are released from platelets which means that their use is not maximized

-Final product contains impurities which reduce risk of serum sickness or allergic reactions

-Clients developed a new method to collect platelet lysate where platelets are concentrated, washed, and then lysed with Triton X-114, a detergent.

-New method is devoid of impurities and increases the platelet derived growth factor by 266 times relative to PRP

-Will be used in anti-inflammatory therapy

-Ethical consideration in taking blood from horses who can’t consent to experimentation. However, the research is for the benefit of humanity

Conclusions/action items: Meet with group tomorrow, create design idea for simplified tubing.

10/16/20 Research Notes

NESYA GRAUPE - Nov 29, 2020, 5:53 PM CST

Title: Tubing Notes

Date: 10/16/2020

Content by: Nesya

Present: Nesya

Goals: To better understand the different parts of the tubing

Content:

System and method for collecting plasma protein fractions from separated blood components:

https://patents.google.com/patent/US8123713B2/en?q=COBE+Spectra&assignee=Terumo+Bct%2c+Inc

- Apheresis device separates whole blood from donor, connected by tubes to machine, into components automatically and returns uncollected/unneeded components back to donor
- Preconnected disposable tubing set includes extracorporeal (outside body) tubing circuit, blood processing vessel, and plasma separator/concentrator. Set is mounted on blood component separation device (clients have this) which has pump/valve/sensor assembly (need to override sensors) and channel assembly (???) that interfaces with disposable blood processing vessel

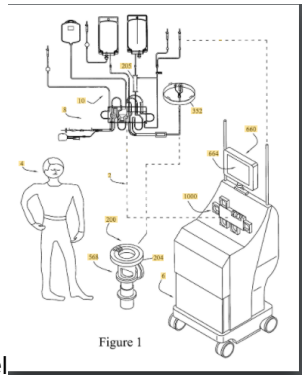


Figure 1

- Are the channel assembly and blood processing vessel things that the client needs also?
- Blood flows from donor/patient, through extracorporeal tubing, into rotating blood processing vessel
    - Centrifuge rotor assembly provides necessary centrifugal forces to separate the different blood components by centrifusion
- Blood components not retained are returned to the body via extracorporeal tubing set
- Tubing assemblies, all parts preconnected for disposable, single use
    - Cassette assembly

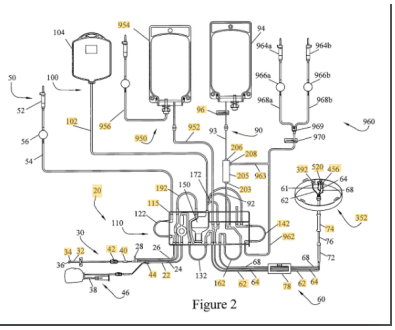


Figure 2

- Anticoagulant tubing assembly
- Plasma or protein collection tubing assembly (needed?)
- Red blood cell collection assembly (needed?)
- Vent bag tubing subassembly
- Platelet collection tubing assembly
- Replacement fluid subassembly (optional)
- Blood removal/return assembly (contains needle subassembly)

Conclusions/action items: Determine which of these tubing parts our client has, which parts we can harvest and sterilize from the original tubing, and which parts we must order online.

10/19/20 Research Notes

NESYA GRAUPE - Nov 29, 2020, 6:24 PM CST

Title: Centrifuge Research Notes

Date: 10/29/2020

Content by: Nesya

Present: Nesya

Goals: Look at patents to better understand how to centrifuge loop works and how we can incorporate a centrifuge loop into our tubing

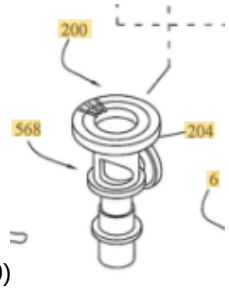
Content:

System and method for collecting plasma protein fractions from separated blood components:

https://patents.google.com/patent/US8123713B2/en?q=COBE+Spectra&assignee=Terumo+Bct%2c+Inc

Centrifuge Loop

- Centrifuge rotor assembly (568) provides centrifugal forces required to separate the blood into different components by weight, interconnected with the channel assembly (200)



- It seems as though the channel assembly and centrifuge rotor assembly are separate from tubing, but connected to the blood processing assembly which is part of tubing
- Blood is continuously removed from the blood processing vessel

Routine Maintenance of Centrifuges Cleaning, Maintenance and Disinfection of Centrifuges, Rotors and Adapters

https://www.eppendorf.com/product-media/doc/en/64426/Eppendorf_Centrifugation_White-Paper_014_Centrifuges_Routine-Maintenance-Centrifuges.pdf

- Rotor (the rotating part), rotor lids, and buckets can be autoclaved
- Typically autoclaved at 121C and 2 bar pressure for 15-20 minutes
- Get centrifuge lubricant?

Conclusions/action items: We will likely decide to reuse the centrifuge loop from the original tubing because it seems complicated to design/fabricate and perfectly sterilizable.

11/05/2020 Research Notes

NESYA GRAUPE - Nov 29, 2020, 10:10 PM CST

Title: Bubble chamber research

Date: 11/05/2020

Content by: Nesya

Present: Nesya

Goals: Learn how a microfluidics bubble chamber works

Content:

Bubble Chambers

-usually have a passive mode or active mode that uses a vacuum. I believe that we would use passive mode for our design

-cost ~$100-$200

-not autoclavable

-reusable: clean with distilled water or 70% ethanol, remove and clean membrane

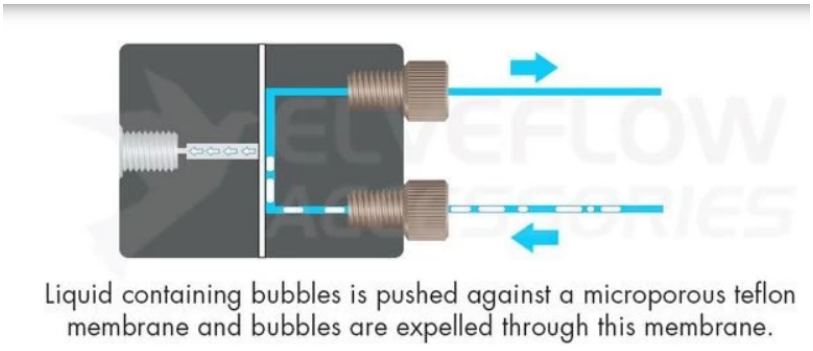
-Fisher Scientific: https://www.fishersci.com/shop/products/NC1102537/nc1102537#?keyword=bubble+trap

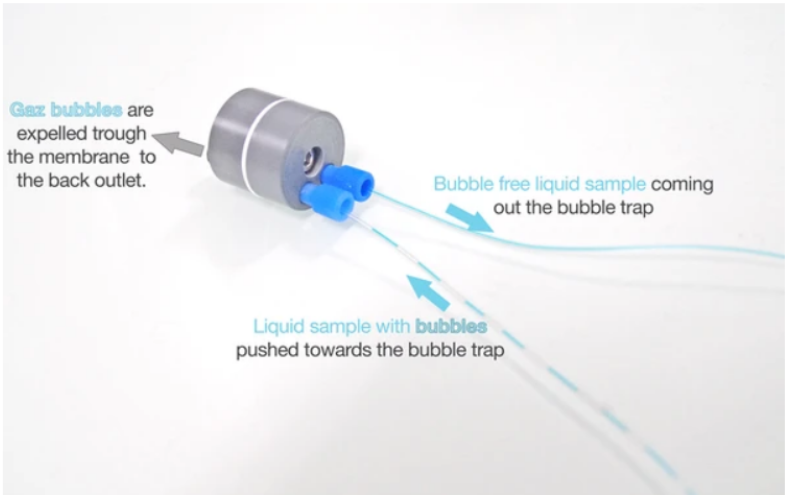
-can handle 0.5-2mL per minute without vacuum, 60 mL per min with vacuum

-work because membrane is permeable to air, not blood. Bubbles are expelled through membrane to collection tube while blood (or other liquid) leaves bubble trap through the outlet port

https://www.precigenome.com/bubble-trap

https://darwin-microfluidics.com/products/microfluidic-bubble-trap





Conclusions/action items: Talk with team and decide whether to purchase a bubble chamber, use preexisting bubble chamber, or fabricate a new bubble chamber

10/4/2020 Design Notes

NESYA GRAUPE - Oct 06, 2020, 8:25 PM CDT

Title: Design Notes

Date: 10/4/2020

Content by: Nesya Graupe

Present: Nesya Graupe

Goals: Create design ideas

Content:

Sterilization

Sterilization

-Client has access to autoclaves in the veterinary school and gas sterilization in the medical school

“Steam Sterilization.” Centers for Disease Control and Prevention, Centers for Disease Control and Prevention, 18 Sept. 2016, Available: www.cdc.gov/infectioncontrol/guidelines/disinfection/sterilization/steam.html.

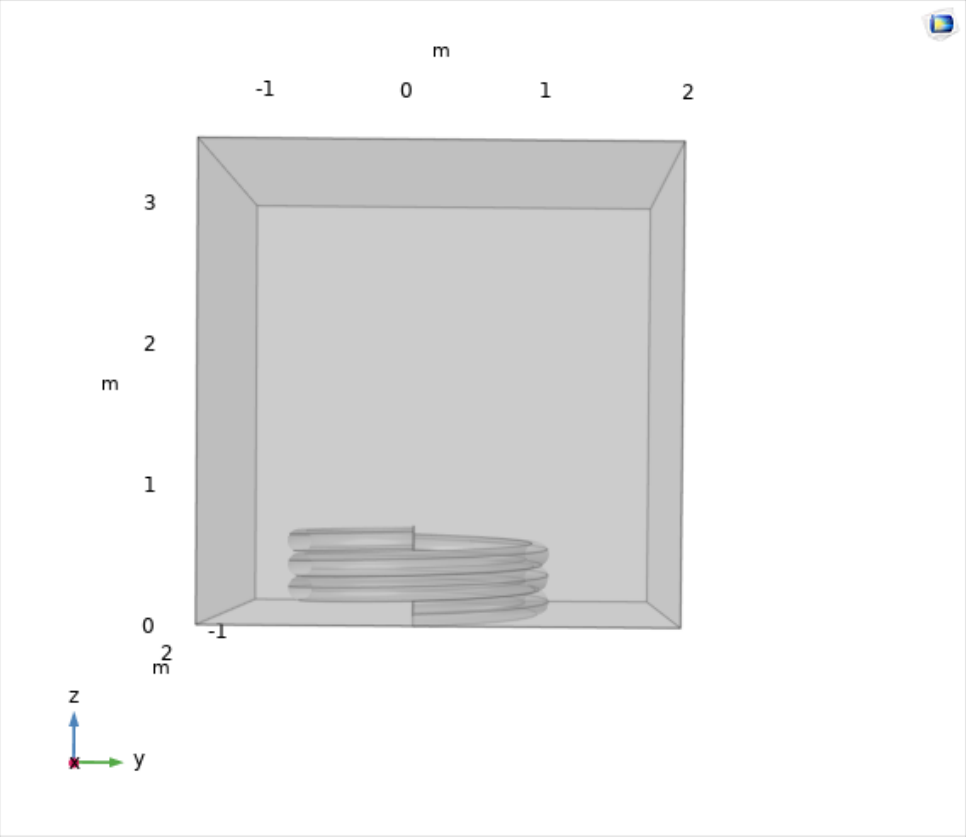
-Autoclave uses moist heat

-Steam sterilization is the most widely used and dependable method of sterilization

-Exposes each item to to direct steam at a certain temperature and pressure for a certain amount of time

-Common temperatures are 121C and 132C

-Minimum of 30 minutes in gravity displacement sterilizer or 4 minutes in prevacuum sterilizer



Representation of what the tubing would look like in an autoclave.

“Ethylene Oxide Sterilization.” Centers for Disease Control and Prevention, Centers for Disease Control and Prevention, 18 Sept. 2016, Available: www.cdc.gov/infectioncontrol/guidelines/disinfection/sterilization/ethylene-oxide.html.

-Ethylene Oxide (ETO)

-Parameters: gas concentration (450- 1200 mg/L), temperature (37- 63C), relative humidity, exposure time

-Takes 1-6 hours

-ETO has been shown to be toxic for animals

-Sterilization cycle has 5 stages: preconditioning and humidification, gas introduction, exposure, evacuation, and air washes

-Used for items that are moisture or heat sensitive

-Using steam sterilization is better because it is cheaper, more accessible to clients, and faster. Also, ETO sterilization is toxic to animals. The gas is aerated but there might be some residual gas, especially with long tubing.

Design

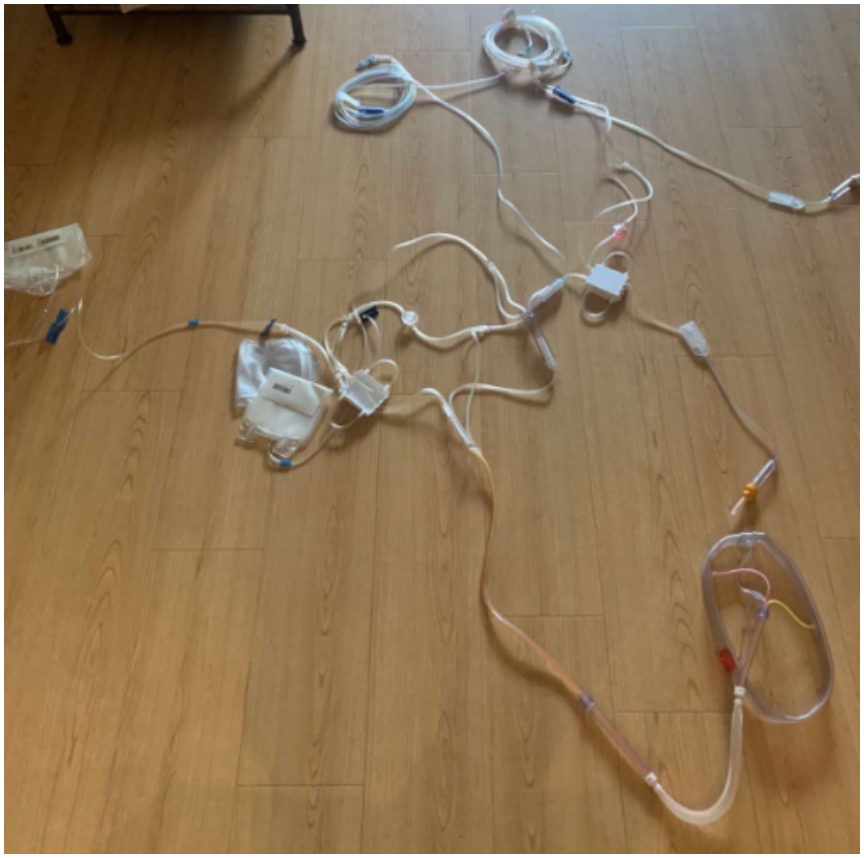
Felt, Thomas, et al. System and Method for Collecting Plasma Protein Fractions from Separated Blood Components. 28 Feb. 2012.

-The preconnected disposable set includes an extracorporeal (outside of body) tubing set, a blood processing vessel, and a plasma separator concentrator

-Sensor assembly on the machine (what sensors does the tubing need to work with?)

-Channel assembly interfaces with blood processing vessel

-Needle assembly -> Blood removal tubing -> Cassette assembly -> blood inlet tubing -> processing vessel -> Return



The tubing from our client

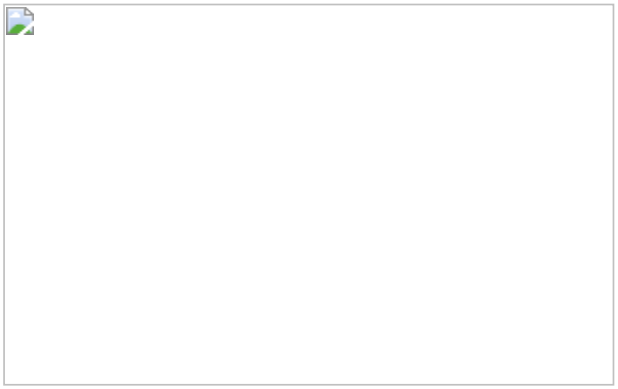


Diagram of simplified tubing

Conclusions/action items:

Meet with the group tomorrow, establish a simplified tubing design

10/30/2020 Call to Action

NESYA GRAUPE - Nov 29, 2020, 5:59 PM CST

Title: Call to Action

Date: 10/30/2020

Content by: Nesya

Present: Nesya

Goals: To clearly and concisely explain roadblocks in our design so that we can receive help from other groups

Content:

Plateletpheresis Call To Action

If you have ever had a medical procedure where you received an IV then you might be familiar with the drip chamber, a device that is used to control the rate that fluid/medicine is administered, and equally importantly, prevent air from entering the bloodstream and blocking blood vessels.

While our project is not with humans and we are not exactly using an IV, our desire for safety by using a drip chamber remains the same. We are developing tubing that will allow for plateletpheresis, the collection of platelets from whole blood, to be performed on horses. The tubing that we are designing will work in conjunction with the COBE Spectra Apheresis machine to pass blood into a centrifuge, separate out platelets, and return uncollected blood components back to the horse. The platelets will then be washed and concentrated into platelet extract that can then be used in anti-inflammatory therapies. We have ordered tubing and Y and T connectors and are planning to autoclave and reuse the centrifuge from existing tubing.

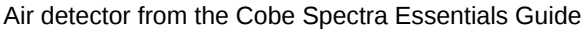
We are requesting help with ideas on how to fabricate drip chambers, and if possible pressure sensors, for our tubing. We have only seen drip chambers connected to IVs and would like ideas to fabricate them as individual pieces, or at the very least separate them from already existing IVs. In addition, we would like help creating and incorporating a pressure sensor into the tubing to monitor the pressure of blood in the tubing. We would like to know the pressure in the tubing so that the machine can potentially turn off if the blood pressure reaches a dangerous level due to occlusion of the tubing.

We realize that since plateletpheresis involves large amounts of blood it can potentially be a dangerous procedure, and therefore are doing everything possible to ensure the safety of our horses. Thank you for your time and ideas!

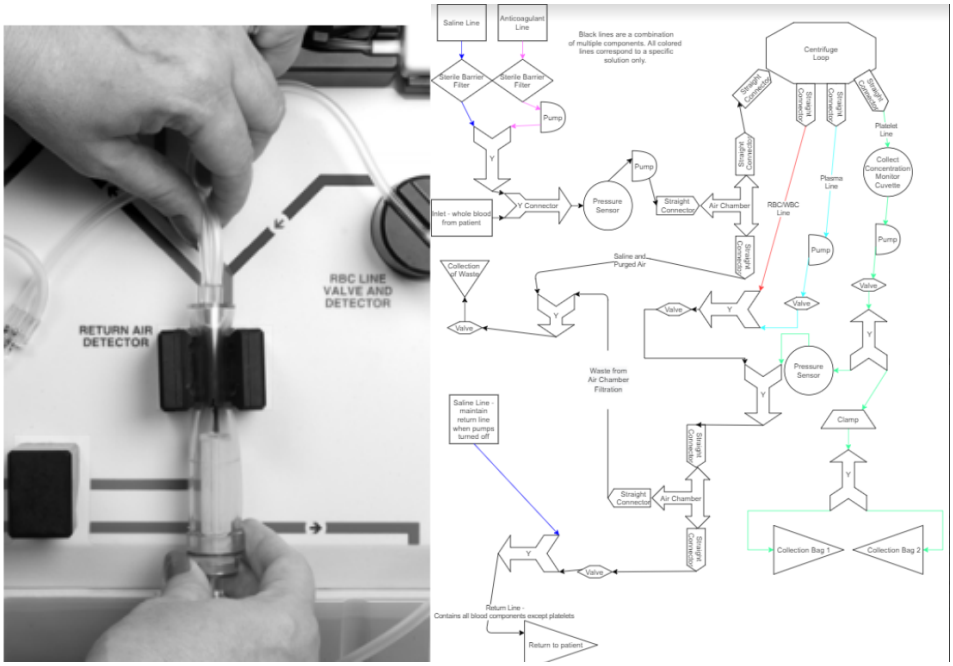
Website: https://bmedesign.engr.wisc.edu/projects/f20/plateletpheresis

Images:

Tubing diagram



Air detector from the Cobe Spectra Essentials Guide



Conclusions/action items: Post the call to action on Piazza so that our group can receive help fabricating drip chamber and pressure sensors, read about other groups' projects and respond to another group's call to action.

11/10/2020 Bubble trap

NESYA GRAUPE - Nov 29, 2020, 9:56 PM CST

John Puccinelli-Nov 03, 2014, 3:20 PM CST

Title: Bubble Trap CAD drawing

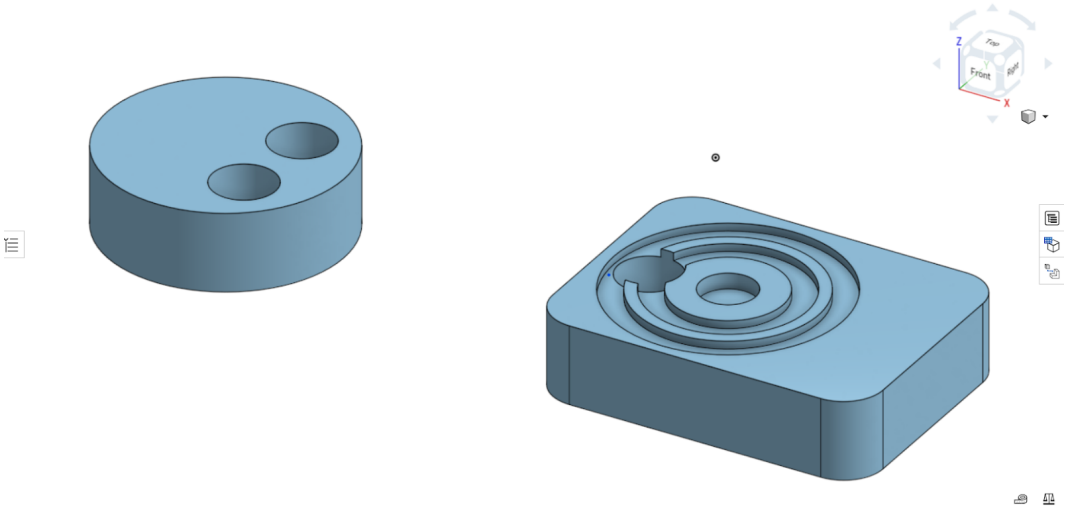
Date: 11/10/2020

Content by: Nesya

Present: Nesya

Goals: To create a CAD drawing of the base for the bubble trap

Content:



Conclusions/action items: This part will be used to remove bubbles from the tubing. I will need to set up an appointment with the Makerspace to 3D print it.

11/17/2020 Bubble trap

NESYA GRAUPE - Nov 29, 2020, 10:01 PM CST

Title: Bubble trap explanation

Date: 11/17/2020

Content by: Nesya

Present: Nesya

Goals: To explain how the bubble trap works

Content:

Bubble trap

PTFE Membrane	$53.29	https://www.fishersci.com/shop/products/m-filter-for-stand-f-bubb-trap/501953960#?keyword=bubble+trap
C-clamp	4.23	https://www.fishersci.com/shop/products/cast-iron-c-clamp-3-in-jaw-opening/s81060#?keyword=clamp
Flangeless Fitting 4mm OD	6.29	https://www.idex-hs.com/store/fluidics/fluidic-connections/fittings/flat-bottom-fittings/flangeless-fittings/flangeless-fitting-peek-5-16-24-flat-bottom-for-4mm-od.html

The membrane will rest in the grooved area. A clamp will sandwich the membrane and the two parts of the body together. On the top cylinder piece of the base, one of the holes is an inlet port and the other hole is an outlet port. The flangeless fittings with a 4mm outer diameter will screw the tubing into the top piece of the base. Air will pass through the membrane to the hole in the bottom of the base, but blood cannot pass through the membrane. It will flow from the inlet port to the outlet port. Can use ethylene oxide to sterilize base, fittings. Possibly autoclave c clamp. Use ethanol to clean the filter. All parts are reusable.

Conclusions/action items: Order the PTFE membrane, C-clamp, and flangeless fittings. Assemble the bubble trap. Come up with a way to test the bubble trap's accuracy.

2014/11/03-Entry guidelines

John Puccinelli - Sep 05, 2016, 1:18 PM CDT

Use this as a guide for every entry

- Every text entry of your notebook should have the **bold titles** below.
- Every page/entry should be **named starting with the date** of the entry's first creation/activity, subsequent material from future dates can be added later.

You can create a copy of the blank template by first opening the desired folder, clicking on "New", selecting "Copy Existing Page...", and then select "2014/11/03-Template")

Title: Descriptive title (i.e. Client Meeting)

Date: 9/5/2016

Content by: The one person who wrote the content

Present: Names of those present if more than just you (not necessary for individual work)

Goals: Establish clear goals for all text entries (meetings, individual work, etc.).

Content:

Contains clear and organized notes (also includes any references used)

Conclusions/action items:

Recap only the most significant findings and/or action items resulting from the entry.

2014/11/03-Template

John Puccinelli - Nov 03, 2014, 3:20 PM CST

Title:

Date:

Content by:

Present:

Goals:

Content:

Conclusions/action items: