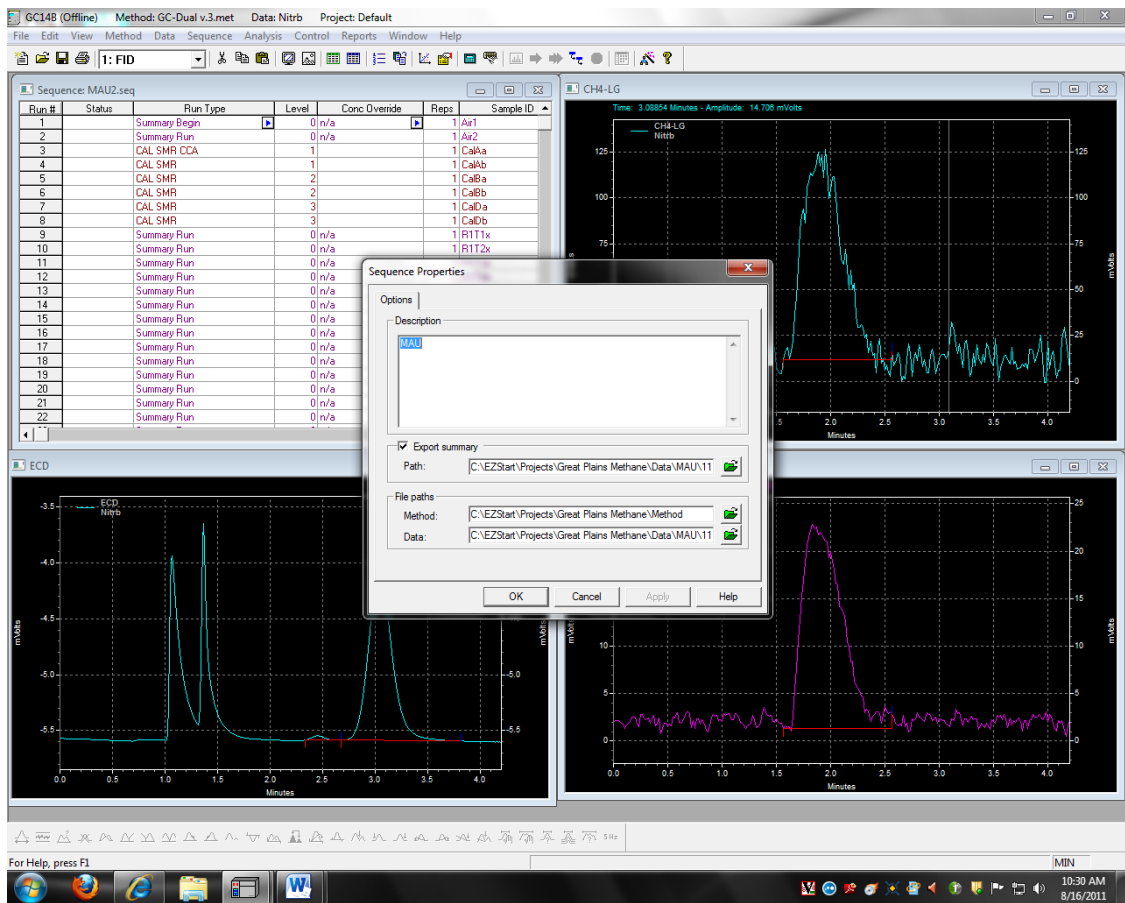


Run Protocol for Great Plains Methane Gas Samples
[CH₄], [CO₂], [SF₆]
Last revised by MS, 8-15-11

- **PLEASE READ ALL OF THIS PAPER BEFORE STARTING**
1. Once the GC (Shimadzu GC-14B) and LG (Los Gatos DLT-100) are turned on (GC should not be off), you will need to check the LG's conversion setup. This feature tells the LG what amount of voltage to send to computer (technically it sends it to the SS420 Analog to Digital Converter then the computer) for each ppm of gas it registers. To modify this feature go to the LG display computer. Using its built in mouse, from the main screen go to "Setup", Ok, and then "Analog Out". Then set the numbers with the keyboard to 1 ppm CH₄ at 5 volts and 1000 ppm CO₂ at 5 volts. Hit Ok and then Return to get back to the main screen. These numbers are saved, but different experiments may change them so keep track of it.
 2. Next you need to set up the Zero Air. The Zero Air tank is connected to a Flow Meter (which is located next to the desk) and then the Flow Meter is connected to the LG. Turn the Zero Air tank on and set its outward pressure (left hand gauge on Regulator) to 75 kPa. Then look at the Flow Meter (which has a valve system also). You want to turn the knob labeled L until the Flow Meter is set at 12 to 13 units. Now check the LG to make sure it has a low, stable baseline (minimal baseline fluctuations for at least 2 minutes on both CO₂ and CH₄ channels). Now these machines are ready to run.
 3. On the computer, create folders for storing the **Sequence** and **Data** files that will be generated by the run. All gas sampling data is being stored in C: *EZStart: Projects: Great Plains Methane*. There should already be site files named with the 3-4 character code on the data sheet included in the sample bag within "Sequence" and "Data" that contain multiple folders, one for each sampling date with the format: YYMMDD (i.e 110601). Add a folder for the date that the samples were collected in the field as written on the data sheet included with the samples.
 4. Using the EZStart 7.2.1 SP1 software, to set up a new run you will need to make a new sequence file. To use a previous file as a template for your new sequence, go to *File: Sequence: Open* and select a previous sequence file from the Great Plains Methane Sequence file section. In the Help folder within Great Plains Methane, there is a file called "**Template for double sample GC Run**", which is useful for running two plots at once (or could be reduced down to just one plot easily). Click *File: Sequence: Save As* to save the file as a new name in a different sequence folder so you can make changes to it.
 5. Under the "Sequence" pull down menu in EZstart, choose "**Properties**". Your screen should now look like the picture below.



- a. This is where you will change the file paths for the data that will be generated during the run. Change the file path so that its terminus is the folder you just created under the “Data” folder. You want to make sure to change both the “path:” and the “data:” options to your current Data folder for this plot.
6. Now modify the sequence by adding or removing samples as needed, each sample will take ~ 4.5 minutes to run. The sequence should be mostly assembled already.
 - a. Make sure to include two Tank B standards in the middle and at the end for check standards.
 7. Now it is time to set up the Auto Sampler. Each tray holds 32 samples. The tray nearest the computer is Tray 1, the one to its right is Tray 2, and then Tray 3 is on the far side. On the Auto-Sampler control pad, add or modify a “job” in the job queue screen on the main menu (called also HOME). Your options in the Job Menu (click the center button of the rotating wheel over a job) are tray, first, last, increment, count, and method. Increment and count

- you will likely not use. The first should be 1 and the last depends on the amount of samples you are running. The method should be **Gcdual**.
- a. For example, if you want to run 84 samples (which is the set-up of the **“Template for double sample GC Run”**) then you will need three jobs. The first job (from top to bottom on the list) should be Tray 1, 1, 32, 1, 1, and then Gcdual respectively in the options for the job. The second job in the list should be Tray 2, 1, 32, 1, 1, and then Gcdual respectively. And the third job should say Tray 3, 1, 20, 1, 1, and then Gcdual respectively.
8. Once you are confident in how you entered the sequence information, go to the desktop computer and press the green button on the menu bar that says “Sequence Run” when highlighted. After you hit this button and click Start on the next screen, the control panel on the GC will display flashing codes (this shows communication is working between the GC and desktop) and it will say “Acquiring” in the sequence edit window (if sequence edit window is not open go to *Sequence: Edit*), this means that the computer is ready to receive information from the GC (SF6) and the LG (CO2, CH4).
 9. Before you start running the samples, you need to wrap (or secure in some way) the vials to the tray. Before you wrap the aluminum foil around the tray, you need to push down any of the protrusions on the vial caps that could cut the foil. Then, I used aluminum foil wrapped underneath the tray holder itself with tape around the outside of the foil for a support system to any shifting. Make sure that the foil is very secure and tight along the tray.
 10. To actually start the run, from the job queue menu (or HOME) press “F4” on the Auto Sampler control pad, select “All”, and click Okay on the needle warning (unless, of course, there is no needle present). This will actually start the process of running these samples.

Retrieving Data after a Completed Run
Last revised by MS, 8-15-2011

11. After the sampling has finished (in the sequence edit window all the runs will have a complete next to them), it is time to process and retrieve the data. Make sure the Sequence you ran is still loaded (i.e. the sequence edit window has the right name shown).
12. If you used the **“Template for double sample GC Run”** without erasing any runs (except maybe the HS and LS if not provided in the bag), then you were able to run two different plots on one sequence. Now you need to separate these two plots so they can have their own individual reports which you can open in Excel. Even if you didn’t use the **“Template for double sample GC Run”**, following these following steps (when applicable) will help you retrieve your data after a run.

13. To separate the two plots into two different sequences, you need to make sure you create a sequence and data folder for the plot that ran at the end of the sequence run (the initial plot should be the first 42 runs and its name should be the current saved data and sequence folders). For example, you ran SVS and SVC in this sequence. SVS was the first plot you put in the trays, so you named the previous data and sequence folders this plot name. Now you need to make folders for SVC, so we can store this plot in a different spot.
 - a. Each plot's sequence should have two airs (just for initial checks of the machine), 6 calibrations, the first three rings with 4 times, two high standards, two check standards, the last three rings with 4 times, two low standards, two check standards, and then two nitrogen vials (N₂). There will be some variation sometimes, but this is the rough outline of the 42 possible samples for each plot.
14. After creating the folders, you will first create and process the initial plot. Go to *File: Sequence: Save As* and save the sequence file in the same place as the previous sequence but with a slightly different name (I used 1 and 2 as labels at the end of the plot name). Once saved (in the sequence edit window you now should see the new name at the top) you can now delete the entire second plot leaving just the 42 runs you used for your initial plot. To delete runs in the sequence edit window, highlight the rows and push the delete button on your keyboard. Below is an example of an ideal sequence edit window of just the initial plot (enlarged).

Run #	Status	Run Type	Level	Conc Override	Repts	Sample ID	Method	Filename	Sample Amt	ISTD Amt	Multiplier 1	Multiplier 2
1		Summary Begin	0 n/a		1	Lab Air1	sts\Default\Method\GC-Dual v.3.met	Lab Air1	1	1	1	1
2		Summary Run	0 n/a		1	Lab Air2	jects\Default\Method\GC-Dual v.3.met	Lab Air2	1	1	1	1
3		CAL SMR CA	1		1	C1a	jects\Default\Method\GC-Dual v.3.met	C1a	1	1	1	1
4		CAL SMR	1		1	C1b	jects\Default\Method\GC-Dual v.3.met	C1b	1	1	1	1
5		CAL SMR	2		1	C2a	jects\Default\Method\GC-Dual v.3.met	C2a	1	1	1	1
6		CAL SMR	2		1	C2b	jects\Default\Method\GC-Dual v.3.met	C2b	1	1	1	1
7		CAL SMR	3		1	C3a	jects\Default\Method\GC-Dual v.3.met	C3a	1	1	1	1
8		CAL SMR	3		1	C3b	jects\Default\Method\GC-Dual v.3.met	C3b	1	1	1	1
9		Summary Run	0 n/a		1	R1T1	jects\Default\Method\GC-Dual v.3.met	R1T1	1	1	1	1
10		Summary Run	0 n/a		1	R1T2	jects\Default\Method\GC-Dual v.3.met	R1T2	1	1	1	1
11		Summary Run	0 n/a		1	R1T3	jects\Default\Method\GC-Dual v.3.met	R1T3	1	1	1	1
12		Summary Run	0 n/a		1	R1T4	jects\Default\Method\GC-Dual v.3.met	R1T4	1	1	1	1
13		Summary Run	0 n/a		1	R2T1	jects\Default\Method\GC-Dual v.3.met	R2T1	1	1	1	1
14		Summary Run	0 n/a		1	R2T2	jects\Default\Method\GC-Dual v.3.met	R2T2	1	1	1	1
15		Summary Run	0 n/a		1	R2T3	jects\Default\Method\GC-Dual v.3.met	R2T3	1	1	1	1
16		Summary Run	0 n/a		1	R2T4	jects\Default\Method\GC-Dual v.3.met	R2T4	1	1	1	1
17		Summary Run	0 n/a		1	R3T1	jects\Default\Method\GC-Dual v.3.met	R3T1	1	1	1	1
18		Summary Run	0 n/a		1	R3T2	jects\Default\Method\GC-Dual v.3.met	R3T2	1	1	1	1
19		Summary Run	0 n/a		1	R3T3	jects\Default\Method\GC-Dual v.3.met	R3T3	1	1	1	1
20		Summary Run	0 n/a		1	R3T4	jects\Default\Method\GC-Dual v.3.met	R3T4	1	1	1	1
21		Summary Run	0 n/a		1	HS1	jects\Default\Method\GC-Dual v.3.met	HS1	1	1	1	1
22		Summary Run	0 n/a		1	HS2	jects\Default\Method\GC-Dual v.3.met	HS2	1	1	1	1
23		Summary Run	0 n/a		1	C2d	jects\Default\Method\GC-Dual v.3.met	C2d	1	1	1	1
24		Summary Run	0 n/a		1	R4T1	jects\Default\Method\GC-Dual v.3.met	R4T1	1	1	1	1
25		Summary Run	0 n/a		1	R4T1	jects\Default\Method\GC-Dual v.3.met	R4T1	1	1	1	1
26		Summary Run	0 n/a		1	R4T2	jects\Default\Method\GC-Dual v.3.met	R4T2	1	1	1	1
27		Summary Run	0 n/a		1	R4T3	jects\Default\Method\GC-Dual v.3.met	R4T3	1	1	1	1
28		Summary Run	0 n/a		1	R4T4	jects\Default\Method\GC-Dual v.3.met	R4T4	1	1	1	1
29		Summary Run	0 n/a		1	R5T1	jects\Default\Method\GC-Dual v.3.met	R5T1	1	1	1	1
30		Summary Run	0 n/a		1	R5T2	jects\Default\Method\GC-Dual v.3.met	R5T2	1	1	1	1
31		Summary Run	0 n/a		1	R5T3	jects\Default\Method\GC-Dual v.3.met	R5T3	1	1	1	1
32		Summary Run	0 n/a		1	R5T4	jects\Default\Method\GC-Dual v.3.met	R5T4	1	1	1	1
33		Summary Run	0 n/a		1	R6T1	jects\Default\Method\GC-Dual v.3.met	R6T1	1	1	1	1
34		Summary Run	0 n/a		1	R6T2	jects\Default\Method\GC-Dual v.3.met	R6T2	1	1	1	1
35		Summary Run	0 n/a		1	R6T3	jects\Default\Method\GC-Dual v.3.met	R6T3	1	1	1	1
36		Summary Run	0 n/a		1	R6T4	jects\Default\Method\GC-Dual v.3.met	R6T4	1	1	1	1
37		Summary Run	0 n/a		1	LS1	jects\Default\Method\GC-Dual v.3.met	LS1	1	1	1	1
38		Summary Run	0 n/a		1	LS2	jects\Default\Method\GC-Dual v.3.met	LS2	1	1	1	1
39		Summary Run	0 n/a		1	C2e	jects\Default\Method\GC-Dual v.3.met	C2e	1	1	1	1
40		Summary Run	0 n/a		1	C2f	jects\Default\Method\GC-Dual v.3.met	C2f	1	1	1	1
41		Summary Run	0 n/a		1	N2a	jects\Default\Method\GC-Dual v.3.met	N2a	1	1	1	1
42		Summary End	0 n/a		1	N2b	jects\Default\Method\GC-Dual v.3.met	N2b	1	1	1	1
43												

15. Go to *Sequence: Process*, check the Review tab, and hit Start. The Sequence will now be processed one by one. Processing enables you to create (or save over a previous) an advanced report in the folder you specified in the *Sequence: Properties: Path* from earlier. Hit the green down arrow at the bottom of your screen to move down through the list of graphs. Please note any issues or problems you see with the graphs (I used “**Quality Analysis of GPM**” in *Great Plains Methane: Analysis* to record my stats and notes on the sequence runs). Make sure to hit the green arrow until you have gone through all of runs.

16. In Excel, go to *File: Open* and go to the current Data folder. Click the option in the window to view All Files and not just Excel Files. Now open the file called “**Sequence Summary Report – 00005000**” (will always be this same name) and click Finish on the next window. The Report will now be opened in Excel. There are three different tables (ignore the last, fourth table) for each of the three gasses you will be analyzing (SF6, CH4, and CO2). The tables will tell you the Area (which is the integration of the voltage signals received) and ESTD Concentration (which is the concentration of the gas based on the

calibration curve provided by the sample). CH₄ and CO₂ have ESTD Concentrations in ppm and SF₆ has ESTD Concentration listed in ppb.

17. For Quality Analysis, I would suggest using the CV (coefficient of variation) which is equal to the standard deviation over the average. You want to make sure that your check standards and calibrations don't vary much over time, and one way to verify this is by seeing if the CV of the 6 middle standards (C2 or CalB) is below 5 percent. So calculate the CV of the Areas of these 6 reps for all three gases. There are lots of Excel file examples in the 2011 Data folders.
18. Once you are done with your analysis, make sure to save the Report file in the Data folder with everything else as an Excel file. To save the file go to *File: Save As* and then select the "save as type" to be an excel workbook. Change the name to the plot ID (or something appropriate) and hit save.
19. Now you still need to create and process the second plot in the original sequence now. Go to the online or offline version of the EZStart software, and open the original sequence file (the one that still has two plots on it) and then *File: Sequence: Save As* with a new name to the correct folder for the second (ending) plot of the sequence. Using the previous example, this would be the SVC folders and you could just name the sequence file like SVC1 in this correct sequence folder. Once you have saved properly again in the sequence edit window the name should change (Example- Sequence: SVC1.seq). Delete the first plot in this sequence leaving just the second, ending plot. Below is an example of the ideal second plot in the sequence edit window (note the difference in Sample ID and Filename between the second and first plot).

The screenshot shows the GC14B (Offline) software interface. The title bar indicates the method is 'GC-Dual v.3.met', data is 'NitrB', and the project is 'Default - [Sequence: NMu2.seq]'. The menu bar includes File, Edit, View, Method, Data, Sequence, Analysis, Control, Reports, Window, and Help. The toolbar contains various icons for file operations and analysis. The main window displays a table with the following columns: Run #, Status, Run Type, Level, Conc Override, Repts, Sample ID, Method, Filename, Sample Amt, ISTD Amt, Multiplier 1, and Multiplier 2. The table lists 43 runs, starting with 'Summary Begin' and ending with 'Summary End'. Each run is associated with a specific sample ID (e.g., Air1, CaAa, CaAb, etc.) and a method path (e.g., c:\s\Default\Method\GC-Dual v.3.met). The status of each run is 'Summary Run' or 'Summary End', and the level is '0 n/a'. The sample amount is '1' and the ISTD amount is '1'. The multipliers are '1'.

Run #	Status	Run Type	Level	Conc Override	Repts	Sample ID	Method	Filename	Sample Amt	ISTD Amt	Multiplier 1	Multiplier 2
1		Summary Begin	0 n/a		1	Air1	c:\s\Default\Method\GC-Dual v.3.met	Air1	1	1	1	1
2		Summary Run	0 n/a		1	Air2	ojects\Default\Method\GC-Dual v.3.met	Air2	1	1	1	1
3		CAL SMR CCA	1		1	CaAa	ojects\Default\Method\GC-Dual v.3.met	CaAa	1	1	1	1
4		CAL SMR	1		1	CaAb	ojects\Default\Method\GC-Dual v.3.met	CaAb	1	1	1	1
5		CAL SMR	2		1	CaBa	ojects\Default\Method\GC-Dual v.3.met	CaBa	1	1	1	1
6		CAL SMR	2		1	CaBb	ojects\Default\Method\GC-Dual v.3.met	CaBb	1	1	1	1
7		CAL SMR	3		1	CaDa	ojects\Default\Method\GC-Dual v.3.met	CaDa	1	1	1	1
8		CAL SMR	3		1	CaDb	ojects\Default\Method\GC-Dual v.3.met	CaDb	1	1	1	1
9		Summary Run	0 n/a		1	R1T1x-NMU	ojects\Default\Method\GC-Dual v.3.met	R1T1x	1	1	1	1
10		Summary Run	0 n/a		1	R1T2x	ojects\Default\Method\GC-Dual v.3.met	R1T2x	1	1	1	1
11		Summary Run	0 n/a		1	R1T3x	ojects\Default\Method\GC-Dual v.3.met	R1T3x	1	1	1	1
12		Summary Run	0 n/a		1	R1T4x	ojects\Default\Method\GC-Dual v.3.met	R1T4x	1	1	1	1
13		Summary Run	0 n/a		1	R2T1x	ojects\Default\Method\GC-Dual v.3.met	R2T1x	1	1	1	1
14		Summary Run	0 n/a		1	R2T2x	ojects\Default\Method\GC-Dual v.3.met	R2T2x	1	1	1	1
15		Summary Run	0 n/a		1	R2T3x	ojects\Default\Method\GC-Dual v.3.met	R2T3x	1	1	1	1
16		Summary Run	0 n/a		1	R2T4x	ojects\Default\Method\GC-Dual v.3.met	R2T4x	1	1	1	1
17		Summary Run	0 n/a		1	R3T1x	ojects\Default\Method\GC-Dual v.3.met	R3T1x	1	1	1	1
18		Summary Run	0 n/a		1	R3T2x	ojects\Default\Method\GC-Dual v.3.met	R3T2x	1	1	1	1
19		Summary Run	0 n/a		1	R3T3x	ojects\Default\Method\GC-Dual v.3.met	R3T3x	1	1	1	1
20		Summary Run	0 n/a		1	R3T4x	ojects\Default\Method\GC-Dual v.3.met	R3T4x	1	1	1	1
21		Summary Run	0 n/a		1	H51x	ojects\Default\Method\GC-Dual v.3.met	H51x	1	1	1	1
22		Summary Run	0 n/a		1	H52x	ojects\Default\Method\GC-Dual v.3.met	H52x	1	1	1	1
23		Summary Run	0 n/a		1	CaBc	ojects\Default\Method\GC-Dual v.3.met	CaBc	1	1	1	1
24		Summary Run	0 n/a		1	CaBd	ojects\Default\Method\GC-Dual v.3.met	CaBd	1	1	1	1
25		Summary Run	0 n/a		1	R4T1x	ojects\Default\Method\GC-Dual v.3.met	R4T1x	1	1	1	1
26		Summary Run	0 n/a		1	R4T2x	ojects\Default\Method\GC-Dual v.3.met	R4T2x	1	1	1	1
27		Summary Run	0 n/a		1	R4T3x	ojects\Default\Method\GC-Dual v.3.met	R4T3x	1	1	1	1
28		Summary Run	0 n/a		1	R4T4x	ojects\Default\Method\GC-Dual v.3.met	R4T4x	1	1	1	1
29		Summary Run	0 n/a		1	R5T1x	ojects\Default\Method\GC-Dual v.3.met	R5T1x	1	1	1	1
30		Summary Run	0 n/a		1	R5T2x	ojects\Default\Method\GC-Dual v.3.met	R5T2x	1	1	1	1
31		Summary Run	0 n/a		1	R5T3x	ojects\Default\Method\GC-Dual v.3.met	R5T3x	1	1	1	1
32		Summary Run	0 n/a		1	R5T4x	ojects\Default\Method\GC-Dual v.3.met	R5T4x	1	1	1	1
33		Summary Run	0 n/a		1	R6T1x	ojects\Default\Method\GC-Dual v.3.met	R6T1x	1	1	1	1
34		Summary Run	0 n/a		1	R6T2x	ojects\Default\Method\GC-Dual v.3.met	R6T2x	1	1	1	1
35		Summary Run	0 n/a		1	R6T3x	ojects\Default\Method\GC-Dual v.3.met	R6T3x	1	1	1	1
36		Summary Run	0 n/a		1	R6T4x	ojects\Default\Method\GC-Dual v.3.met	R6T4x	1	1	1	1
37		Summary Run	0 n/a		1	LS1x	ojects\Default\Method\GC-Dual v.3.met	LS1x	1	1	1	1
38		Summary Run	0 n/a		1	LS2x	ojects\Default\Method\GC-Dual v.3.met	LS2x	1	1	1	1
39		Summary Run	0 n/a		1	CaBe	ojects\Default\Method\GC-Dual v.3.met	CaBe	1	1	1	1
40		Summary Run	0 n/a		1	CaBf	ojects\Default\Method\GC-Dual v.3.met	CaBf	1	1	1	1
41		Summary Run	0 n/a		1	NitrA	ojects\Default\Method\GC-Dual v.3.met	NitrA	1	1	1	1
42		Summary Run	0 n/a		1	NitrB	ojects\Default\Method\GC-Dual v.3.met	NitrB	1	1	1	1
43		Summary End	0 n/a		1	NitrB	ojects\Default\Method\GC-Dual v.3.met	NitrB	1	1	1	1

20. Now go to the correct Data folder you designated earlier for the old sequence. Copy all of the data files that are in your new sequence (the sample names and file names in the sequence edit window are the same) from this Data folder. Paste these data files into your new Data folder for the new sequence. Using the previous example, go to the SVS data folder (at whatever correct date you are on) and copy all of the files from Air1 to NitrB (can use “last modified” organization to make this easier) which should correspond to SVC’s vials. Then paste the copied files into the SVC Data folder.

21. The files are now moved to the correct folder, so go back to EZStart (which should have your new sequence opened) and go to *Sequence: Properties*. Designate the path: and data: to the same folder you just moved those files to. Click Apply and then Okay. Go to *File: Sequence: Save* (always a good idea to save frequently).

22. **Always** make sure in the sequence edit window that your first run under “Run Type” says “Summary Begin”. If it does not say Summary Begin, then double click on the box for the first run under “Run Type”. Check the

Summary Begin option. On the right hand side of this window, you then need to select the correct template. The template I used is located at *C: EZStart: Template: Summary-Test22.tpl*, but I saved this template over to *C: EZStart: Projects: Great Plains Methane: Help: GPMreport v.2.tpl* if you prefer that location. Select one of these templates hit Open and then Okay on the next two windows. Also make sure that the last run in the sequence says "Summary End" in the "Run Type" within the sequence edit window.

23. Go to *Sequence: Process*, check the review option, and then hit Start to process the new sequence. Note any errors or problems like before and make sure to go through the whole list.
24. Then you can open the Summary Report just like before in Excel (it will of course be in the folder you just designated with the path: in Properties). Make sure to save the excel file like before. Now you have separated a sequence file into two different plots and generated an excel file for both of these plots.