# 2019 BALL-B
# PARTICIPANT SUMMARY

**Evaluation Criteria**

Results for the BALL Survey are not formally evaluated; however, statistics will appear in the Participant Summary for your information.

To provide a timely evaluation of your results, statistics presented in this Participant Summary reflect participant data received by the due date.

Cell markers with less than ten reported results were not included in this Participant Summary.

In the event a result is not graded, a numeric code will appear next to your result. A definition of the code will appear on the first page of your evaluation. Please see "Actions laboratories should take when a PT result is not graded" on page 14.

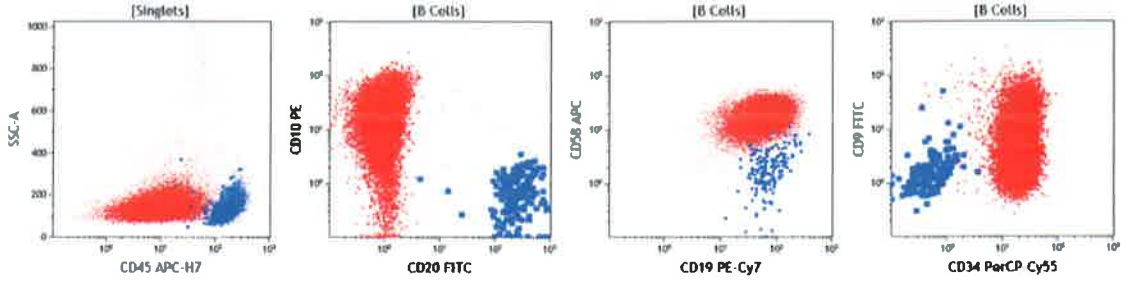
*Note:* To view the discussion images in color, go to cap.org to access the Participant Summary via e-LAB Suite.

## BALL-B Discussion

**Case BALL-04 List Mode Case Positive for MRD at approximately 0.3% of mononuclear cells**
This case contained an abnormal population colored in red. The diagnostic sample contained a population of cells that was CD45 dim, CD10 variable and CD58 bright, CD20 negative, CD38 variable (not shown here) in Tube 1. Tube 2 of the diagnostic sample showed this population to be CD34 positive and CD9 variable and CD13/33 negative. The day 29 sample showed a shift from variable CD9 expression to negative. Antigenic shifts after treatment are not uncommon and have previously been reported. However, this population had an abnormal phenotype, as the cells in Tube 1 were CD10 positive and CD58 bright. The population was also brighter CD19 than the normal lymphocyte population. Tube 2 showed that this CD10+ population was also positive for CD34 and negative for CD13/33, consistent with the original phenotype.

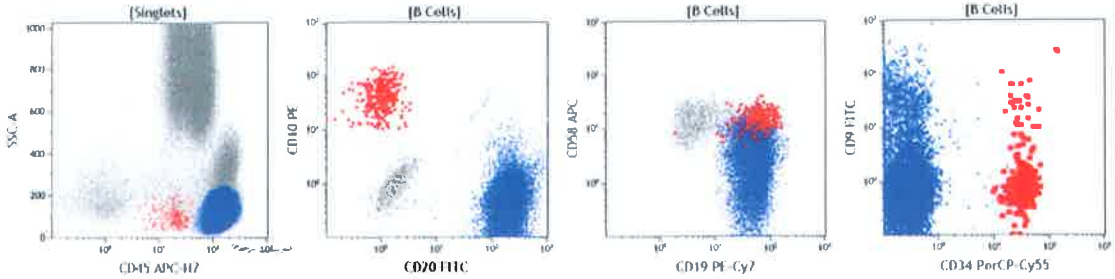
A total of 95.1% (77/81) of participants reported this case as positive, with 4.9% (4/81) calling it negative. Of those calling it positive, the majority reported 0.1 - 0.9%. Participants who reported negative results for this sample should review their analysis procedures and consult recent literature on gating strategy. While this case showed a shift in some of the original markers, the abnormal population continued to express an aberrant phenotype.

**Case 4.** Diagnostic population shown in red



## BALL-B Discussion (cont.)

**Day 29** Blast population shown in red and displayed as rare events



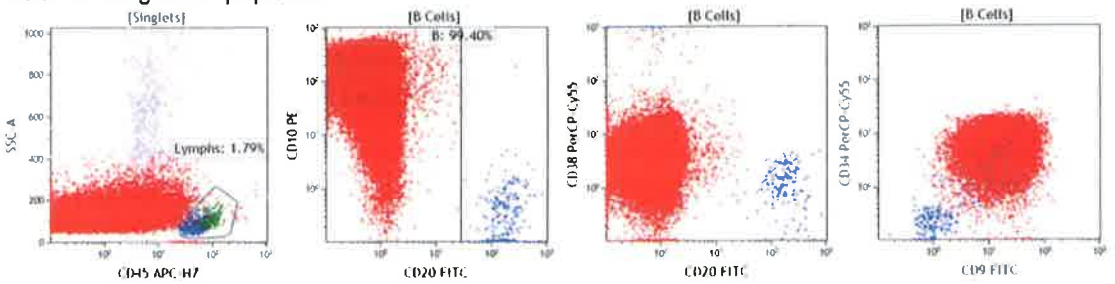

**Case BALL-05 List Mode Case Negative for MRD**

This case did not contain an abnormal CD45 dim blast population or hematogones. However, normal mature B cells and a small population of plasma cells (colored in green) were also clearly visible (CD38 bright, CD10 negative, CD20 negative). The diagnostic population of cells was CD45 dim, CD10 variable and CD58 bright, CD20 negative, CD38 dim/negative in Tube 1. Tube 2 showed a diagnostic population that was CD34 positive, CD13/33 negative and positive for CD9. The day 29 sample was clearly distinct from the prior diagnostic immunophenotype with only normal mature B cells and plasma cells present. (representative dot plots shown below).

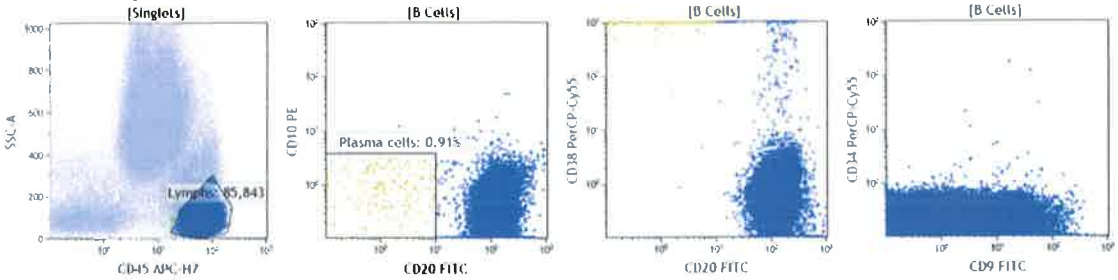
A total of 96.3% (78/81) of participants reported this case as negative, with 3.7% (3/71) calling it positive. Of those calling it positive, the majority reported 0.01 - 0.09%. Participants who reported positive results for this sample should review their analysis procedures.

**Please note that when a sample is negative, the value of abnormal cells is 0 (reported as code 46) and should not be reported as MRD positive. Do not report an immunophenotype if no abnormal population is present.**

**Case 5.** Diagnostic population shown in red




**Case 5.** Day 29 normal population shown in blue



## BALL-B Discussion (cont.)

**Case BALL-06 Positive for MRD at around 5%**

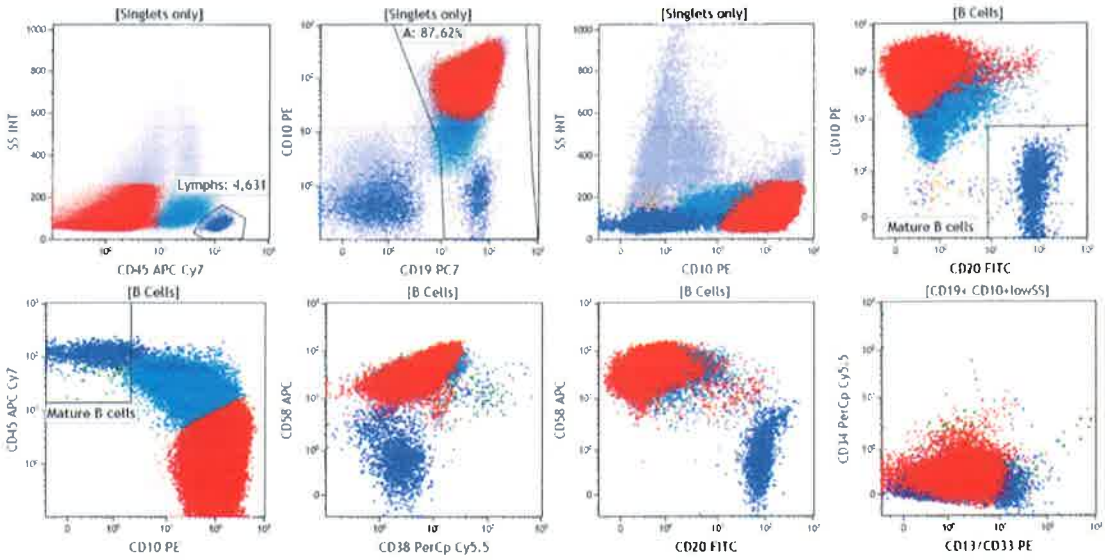
Two sets of plots were provided for this case, one representing the diagnostic immunophenotype at onset of disease, and one representing MRD assessment after the patient had received autologous chimeric antigen receptor T cells expressing anti-CD19 (CAR-T cell therapy). The diagnostic case showed the following abnormal immunophenotype: positive for CD19, CD10 bright, CD9 bright, CD34 and CD38 variable, and CD58 bright; and negative for CD20 and CD45 dim/neg. The MRD tubes did not demonstrate a similar abnormal blast population to the diagnostic sample, but rather showed a population of B-ALL cells with an abnormal immunophenotype: negative for CD19, CD10 bright, CD38 uniform bright, and CD45 dim to negative, with heterogeneous expression of CD20, partial CD34, and bright CD58. A key to identifying these as abnormal cells is to recognize differences in antigen expression intensity between these cells and hematogones which show reduced expression of CD9, CD10, CD34, and CD58 with increased expression of CD20, CD38, and CD45 compared to this CD19 negative population. Another useful feature is that there are no significant populations of CD19-negative normal precursors.

In most cases, it is important when looking at samples from patients on monoclonal or CAR-T therapy to include additional markers to identify the B cell population. CD22 and CD24 are good candidates to add to samples from patients on anti-CD19 therapy. Reference 2 is an excellent example of how to analyze these sample types.

**Of note**: In the kit instructions CD19+ cells are used in the calculation to normalize counts across the 3 tubes. In instances where there are no B cells, or in this case when many of the B cells are CD19 negative, caused confusion for some laboratories as they were unsure what "B cell" number to use in the calculation. The alternative is to use the lymphocyte gate in each of the 3 tubes, which serves the same purpose. Indeed, as a good internal quality assurance check when using either B cells or lymphocytes in the calculation, the MRD percentage should be similar, and if not may indicate a missed abnormal population. Kit instructions will be updated for the 2020 mailing to address this issue.

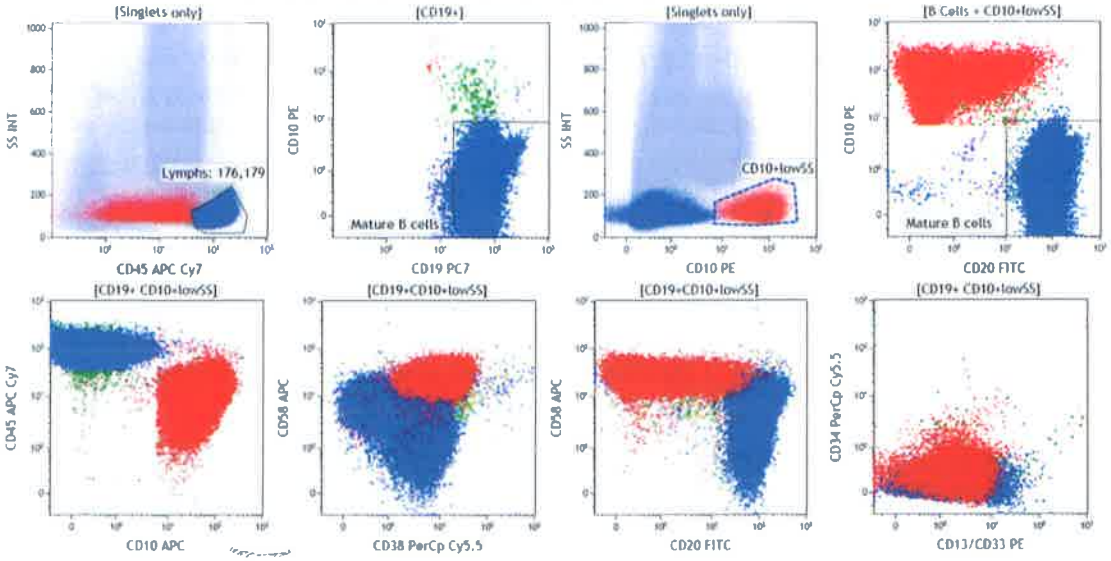
Of the 80 participants who reported a result, 74 (92.5 %) correctly reported this sample as positive, with the remaining 6 (7.5%) participants reporting negative. Of those reporting positive, 4 participants reported in the 0.01 - 0.09% range, 1 in the 0.1 - 0.9% range, 68 participants reported in the 1% - 9.9% range and 1 in the >10% range. The actual value was in the 5% range of mononuclear cells from Tube 3.

**Case 6:** Diagnostic population shown in red



## BALL-B Discussion (cont.)

**Case 6.** Post CAR-T cell immunotherapy; blast population shown in red




### References


1. Borowitz MJ, Pullen DJ, Winick N, Martin PL, Bowman WP, Camitta B. Comparison of diagnostic and relapse flow cytometry phenotypes in childhood acute lymphoblastic leukemia: implications for residual disease detection: a report from the children's oncology group. *Cytometry B Clin Cytom.* 2005 Nov;68(1):18-24.
2. Cherian S, Miller V, McCullouch V, Dougherty K, Fromm JR, Wood BL. A novel flow cytometric assay for detection of residual disease in patients with B-lymphoblastic leukemia/lymphoma post anti-CD19 therapy. *Cytometry B Clin Cytom.* 2018 Jan;94(1):112-120. doi: 10.1002/cyto.b.21482.
3. Keeney M, Wood BL, Hedley BD, et al. A QA program for MRD testing demonstrates that systematic education can reduce discordance among experienced interpreters. *Cytometry B Clin Cytom.* 2018 Mar;94(2):239-249. doi: 10.1002/cyto.b.21528.



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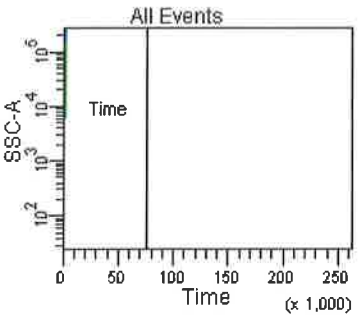
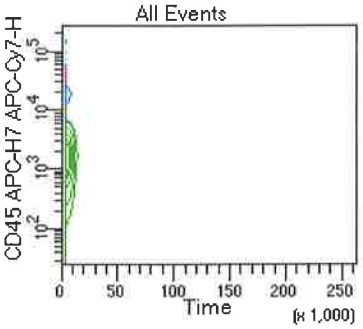
**Diagnostic Immunology and Flow Cytometry Committee**

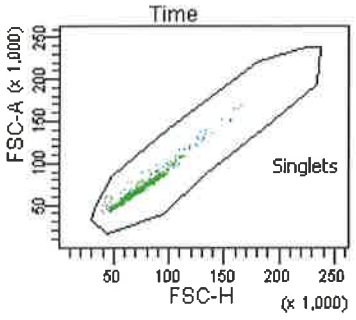
BALL-04 Diagnostic

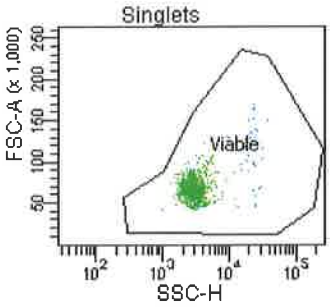


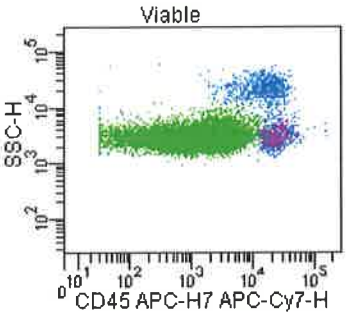



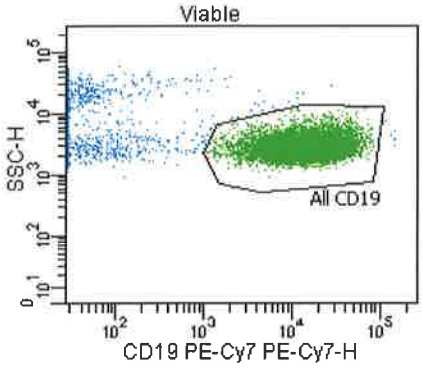

Tube: A-PRE

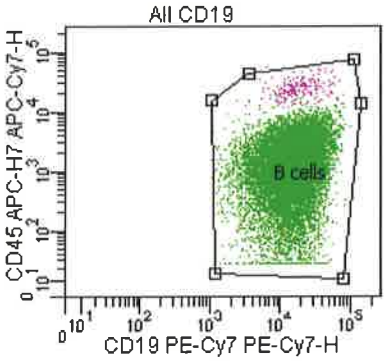
| Population | #Events | %Parent | %Total |
|---|---|---|---|
| All Events | 17,337 | #### | 100.0 |
| Time | 17,337 | 100.0 | 100.0 |
| Singlets | 17,336 | 100.0 | 100.0 |
| Viable | 17,336 | 100.0 | 100.0 |
| All CD19 | 16,011 | 92.4 | 92.4 |
| B cells | 16,005 | 100.0 | 92.3 |
| P1 | 15,808 | 98.8 | 91.2 |

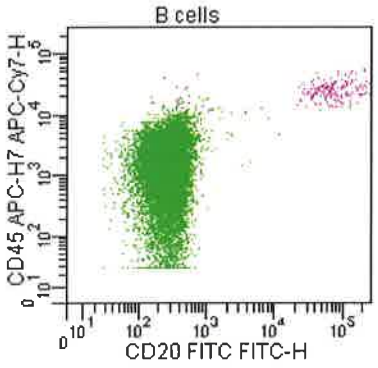


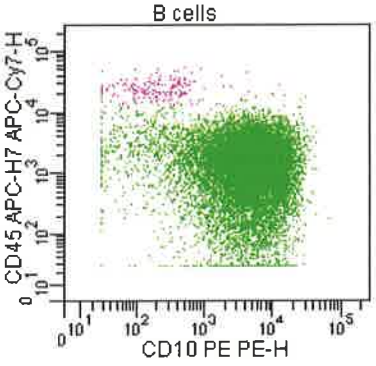



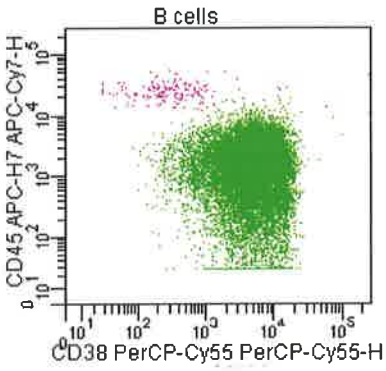



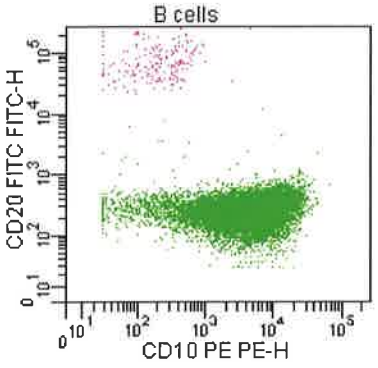



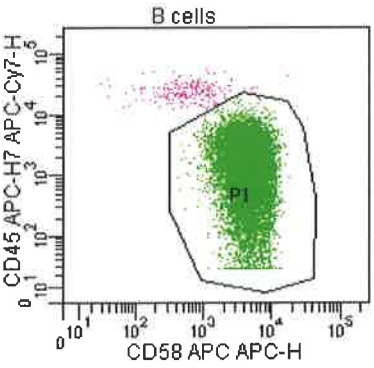



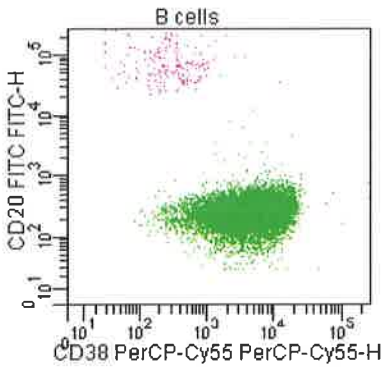



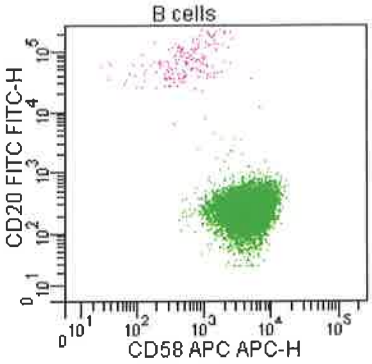



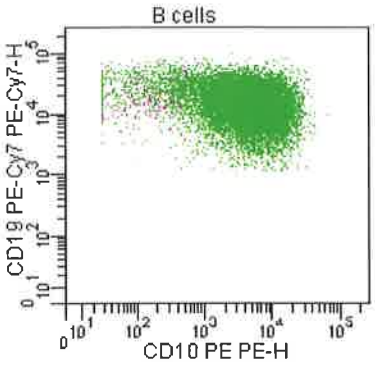



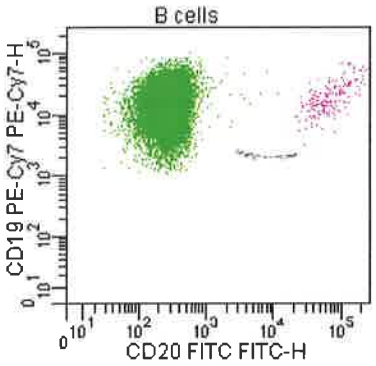



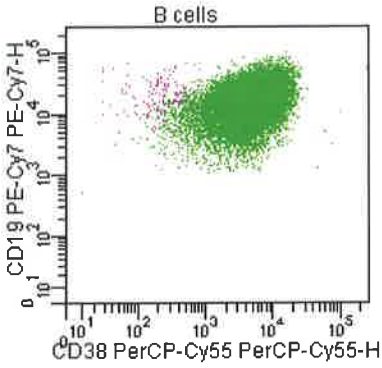



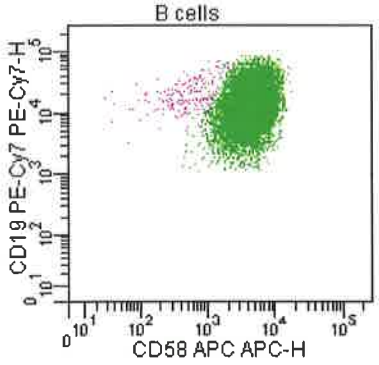


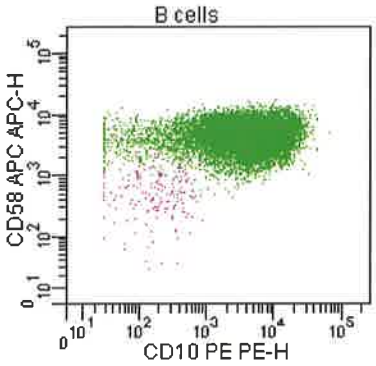



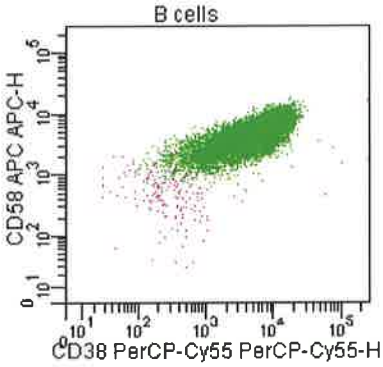



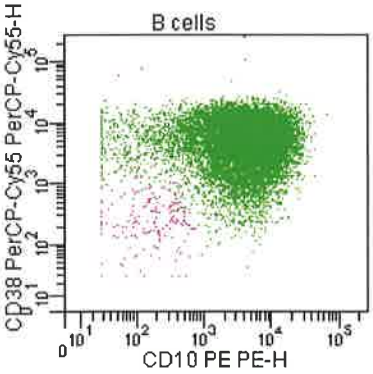

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| Specimen Name: | Case 2 |
| Tube Name: | A-PRE |
| Record Date: | Mar 12, 2016 3:18:58 PM |
| $OP: | HPDevelopmental |
| GUID: | 31d05710-1ce5-4983-a81b-71279261e8c7 |
| Serial Number: | |

| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 17,337 | #### | #### | 100.0 |
| Time | 17,337 | 100.0 | #### | 100.0 |
| Singlets | 17,336 | 100.0 | 100.0 | 100.0 |
| Viable | 17,336 | 100.0 | 100.0 | 100.0 |
| All CD19 | 16,011 | 92.4 | 92.4 | 92.4 |
| B cells | 16,005 | 100.0 | 92.3 | 92.3 |
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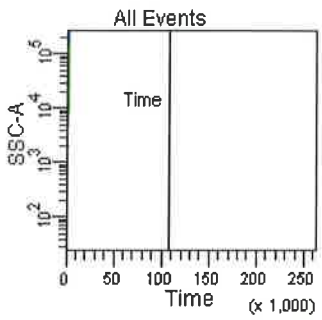
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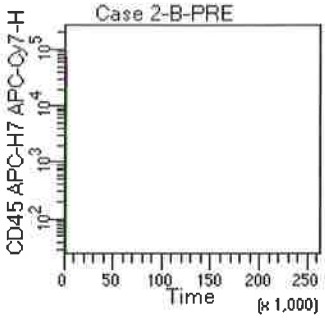
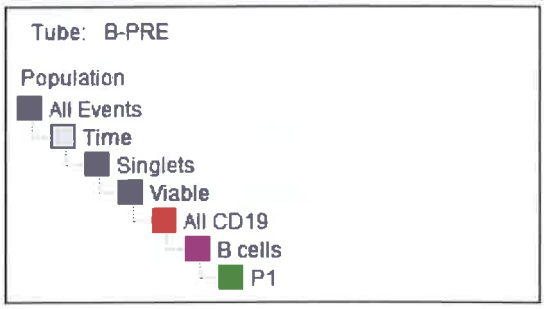
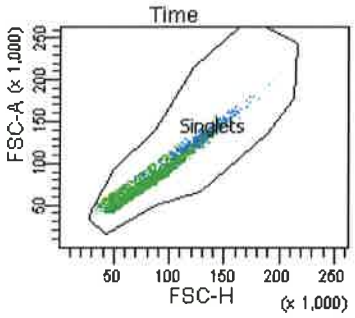
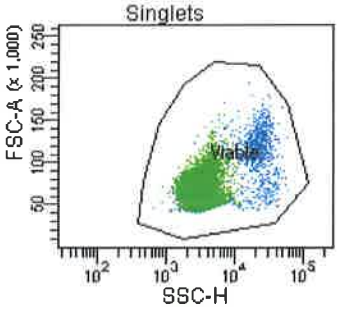
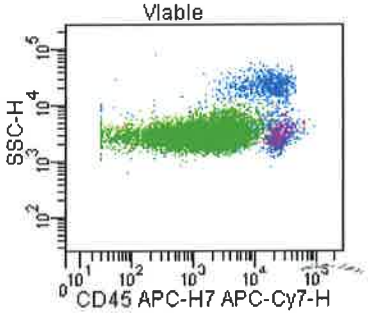
Time
SSC-A
Time
(x 1,000)

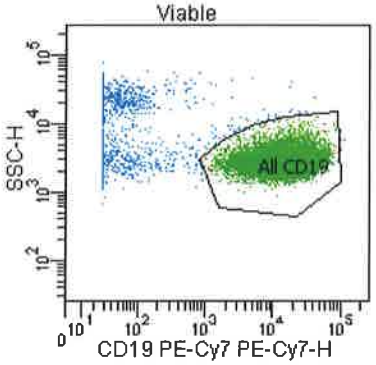

Case 2-B-PRE
CD45 APC-H7 APC-Cy7-H
Time
(x 1,000)


Tube: B-PRE
Population
All Events
Time
Singlets
Viable
All CD19
B cells
P1


Time
Singlets
FSC-A (x 1,000)
FSC-H
(x 1,000)


Singlets
Viable
FSC-A (x 1,000)
SSC-H


Viable
SSC-H
CD45 APC-H7 APC-Cy7-H


Viable
All CD19
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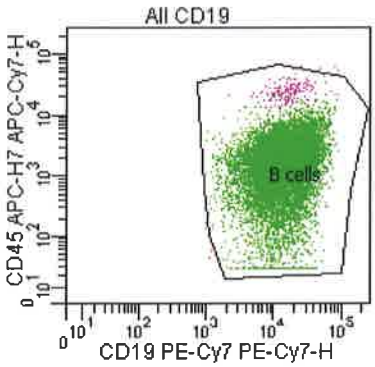
CD19 PE-Cy7 PE-Cy7-H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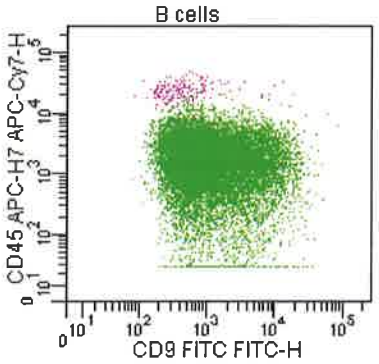
B cells
CD45 APC-H7 APC-Cy7-H
CD19 PE-Cy7 PE-Cy7-H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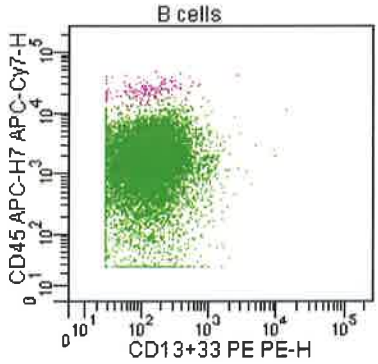
CD9 FITC FITC-H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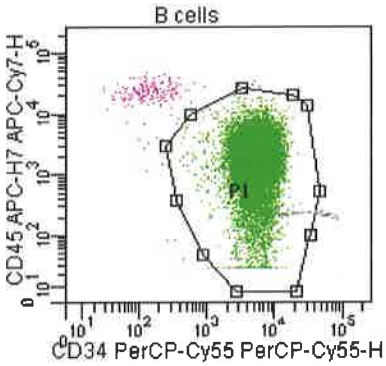
CD13+33 PE PE-H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P1
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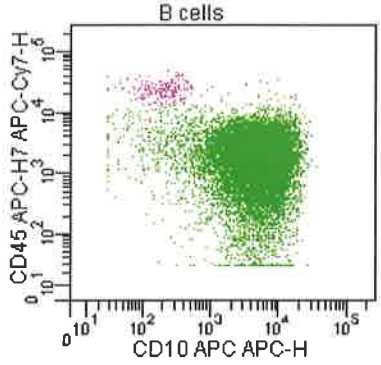
CD34 PerCP-Cy55 PerCP-Cy55-H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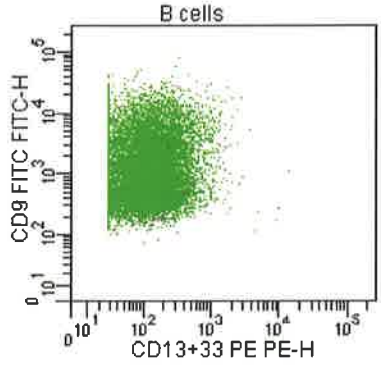
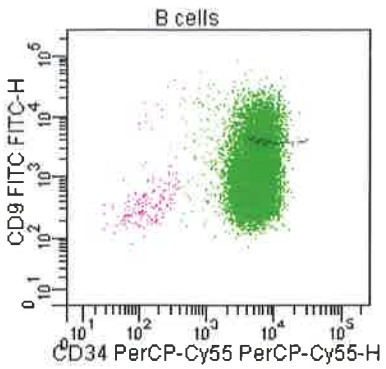
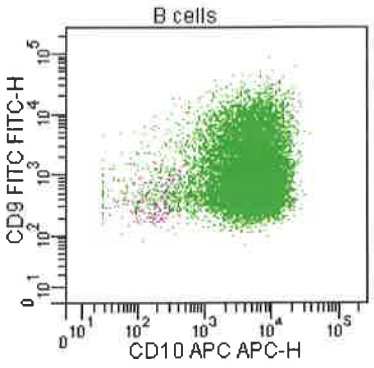
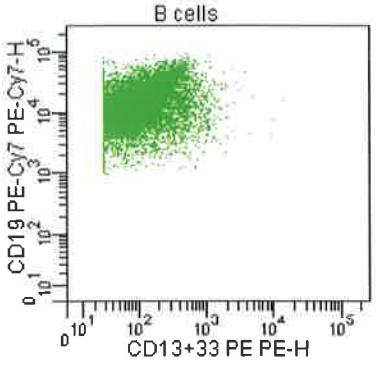
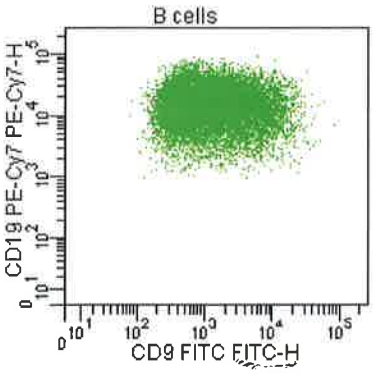
CD10 APC APC-H

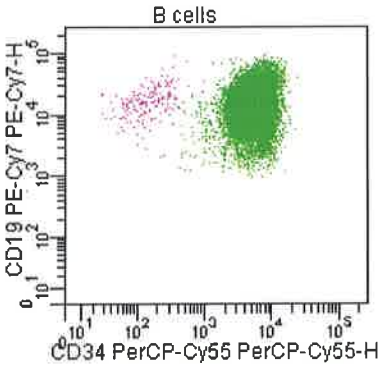

B cells
CD9 FITC FITC-H
CD13+33 PE PE-H

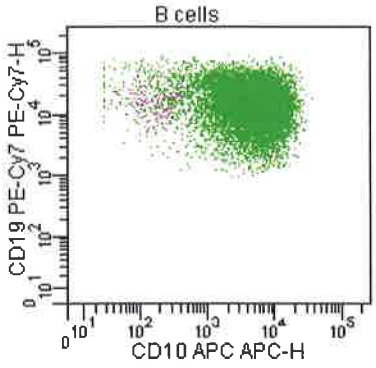


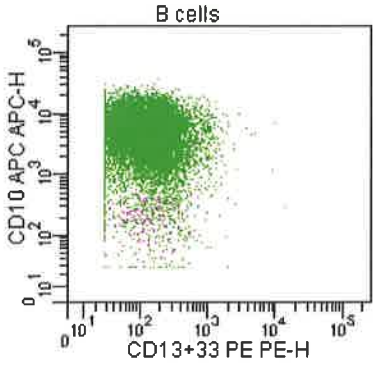



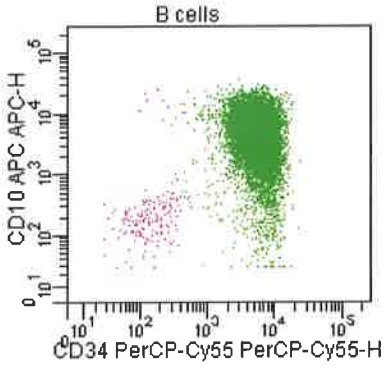



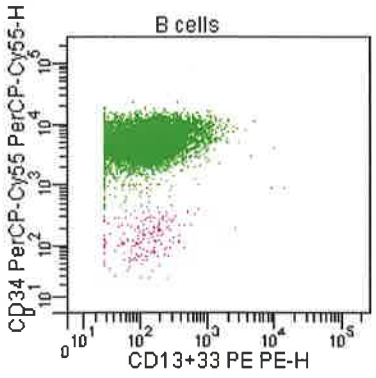



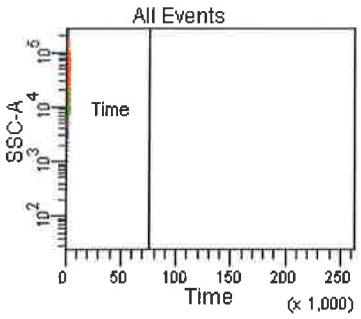



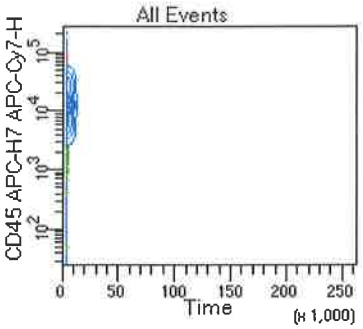




| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 4 |
| Specimen Name: | Case 2 |
| Tube Name: | B-PRE |
| Record Date: | Mar 12, 2016 3:19:22 PM |
| $OP: | HPDevelopmental |
| GUID: | 59d12715-6508-4d0d-801d-2f1a914e1141 |
| Serial Number: | [illegible] |

| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 16,576 | #### | #### | 100.0 |
| Time | 16,576 | 100.0 | #### | 100.0 |
| Singlets | 16,576 | 100.0 | 100.0 | 100.0 |
| Viable | 16,574 | 100.0 | 100.0 | 100.0 |
| All CD19 | 15,227 | 91.9 | 91.9 | 91.9 |
| B cells | 15,217 | 99.9 | 91.8 | 91.8 |
| P1 | 15,032 | 98.8 | 98.7 | 90.7 |

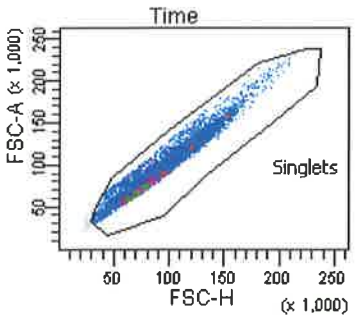
MRD
BALL-04 (Tube 1)

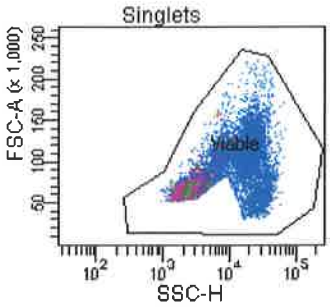


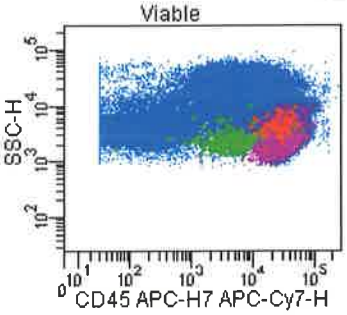



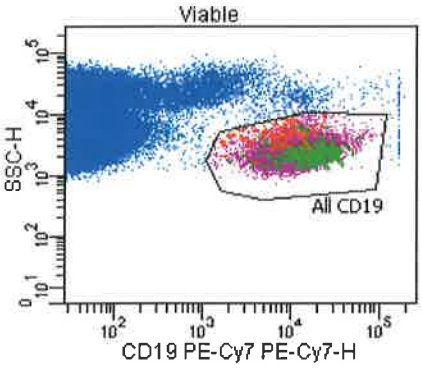

Tube: A-MRD

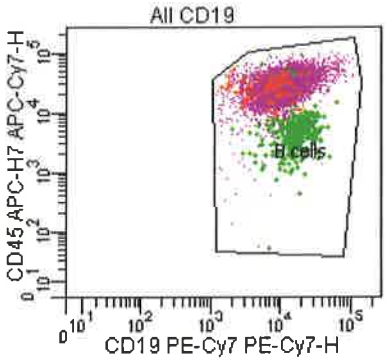
| Population | #Events | %Parent | %Total |
|---|---|---|---|
| All Events | 255,414 | ##### | 100.0 |
| Time | 255,414 | 100.0 | 100.0 |
| Singlets | 253,153 | 99.1 | 99.1 |
| Viable | 253,111 | 100.0 | 99.1 |
| All CD19 | 15,034 | 5.9 | 5.9 |
| B cells | 15,034 | 100.0 | 5.9 |
| P1 | 269 | 1.8 | 0.1 |
| P2 | 72 | 0.5 | 0.0 |

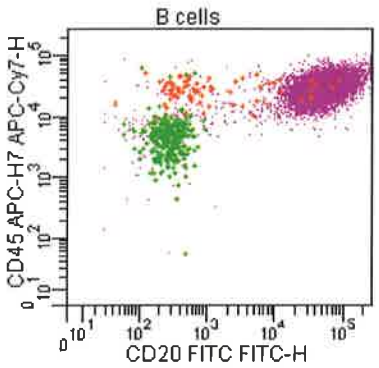


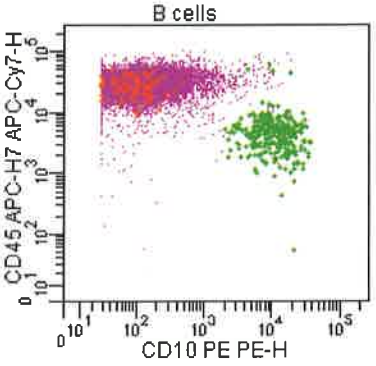



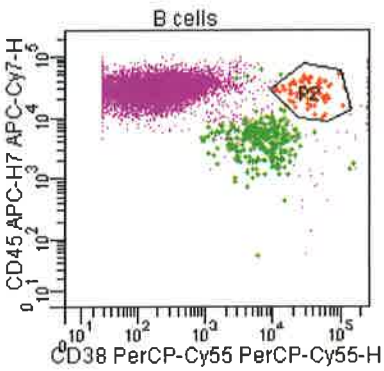



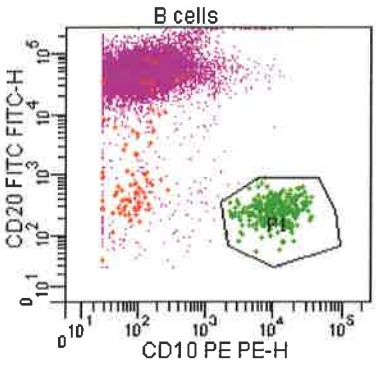



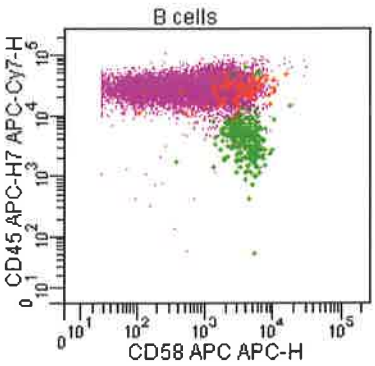



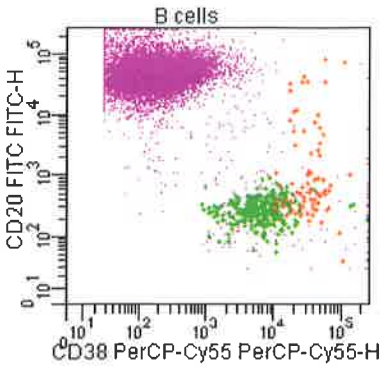



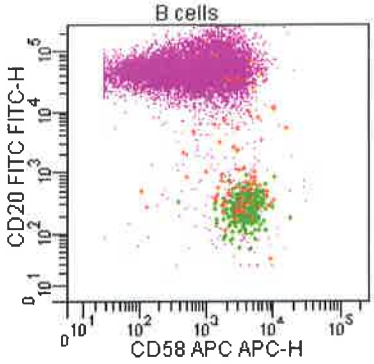



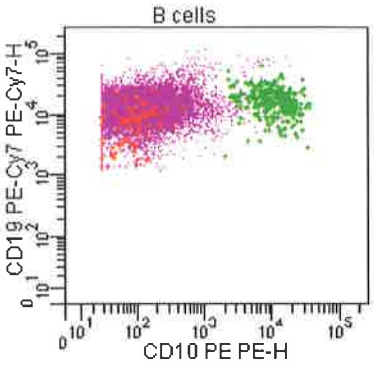



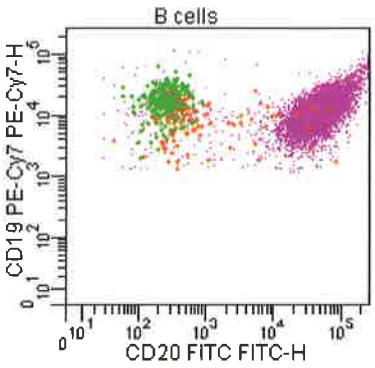



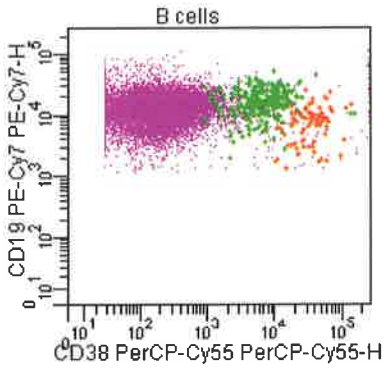



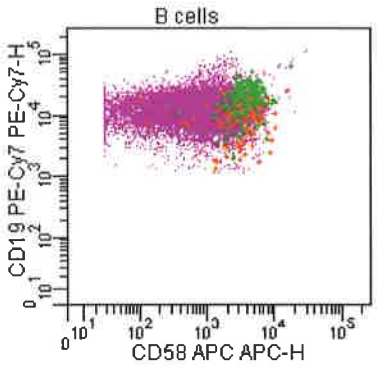


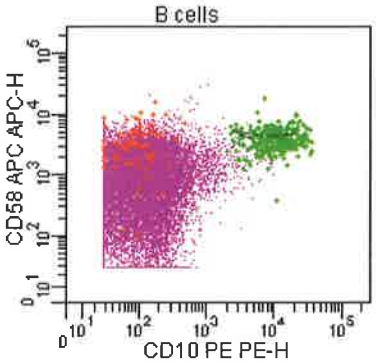



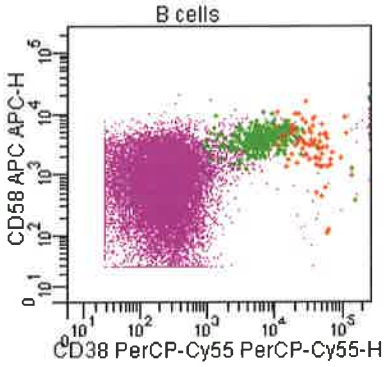



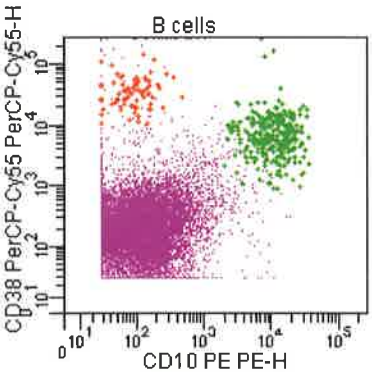



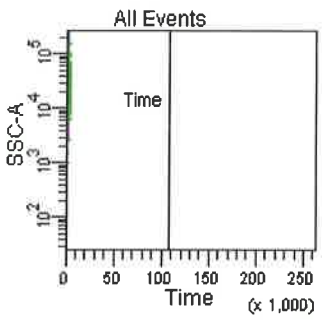



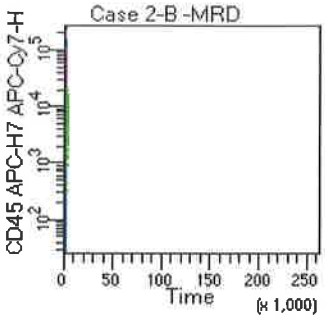



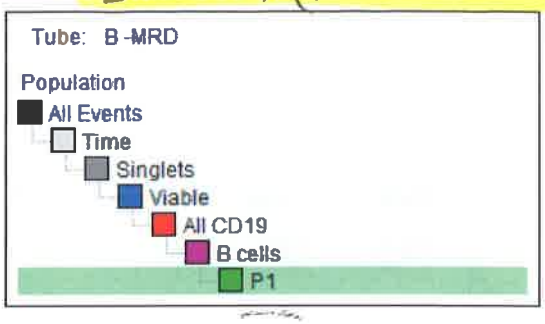



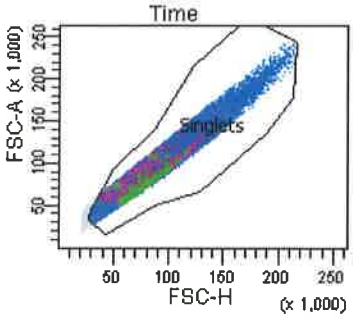

| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 4 |
| Specimen Name: | Case 2 |
| Tube Name: | A-MRD |
| Record Date: | Apr 8, 2016 2:47:38 PM |
| $OP: | HPDevelopmental |
| GUID: | 6aa12585-4761-4e5f-bb53-542143852102 |
| Serial Number: | |

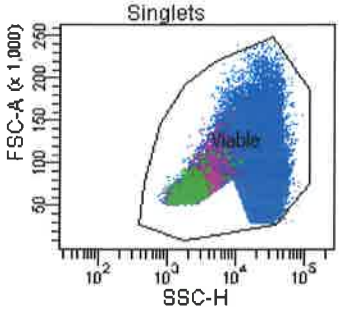
| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 255,414 | #### | #### | 100.0 |
| Time | 255,414 | 100.0 | #### | 100.0 |
| Singlets | 253,153 | 99.1 | 99.1 | 99.1 |
| Viable | 253,111 | 100.0 | 99.1 | 99.1 |
| All CD19 | 15,034 | 5.9 | 5.9 | 5.9 |
| B cells | 15,034 | 100.0 | 5.9 | 5.9 |
| P1 | 269 | 1.8 | 1.8 | 0.1 |
| P2 | 72 | 0.5 | 0.5 | 0.0 |

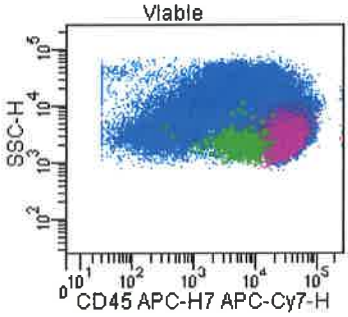

All Events
Time
SSC-A
Time
(x 1,000)

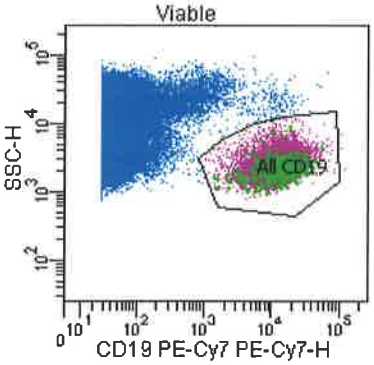

Case 2-B -MRD
CD45 APC-H7 APC-Cy7-H
Time
(x 1,000)

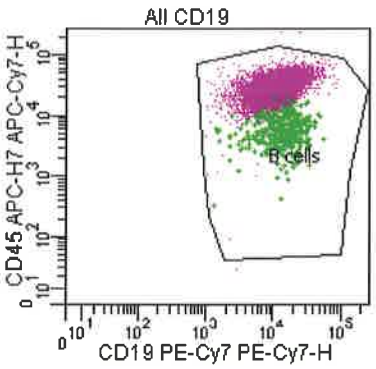

Tube: B -MRD
Population
All Events
Time
Singlets
Viable
All CD19
B cells
P1

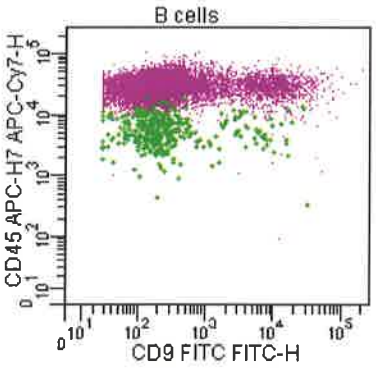

Time
Singlets
FSC-A (x 1,000)
FSC-H
(x 1,000)

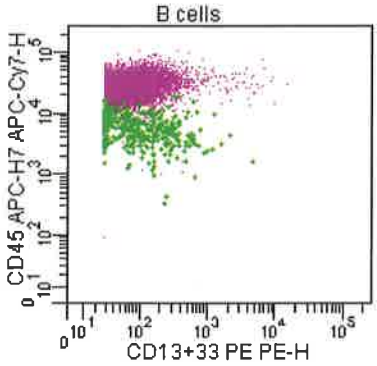

Singlets
Viable
FSC-A (x 1,000)
SSC-H

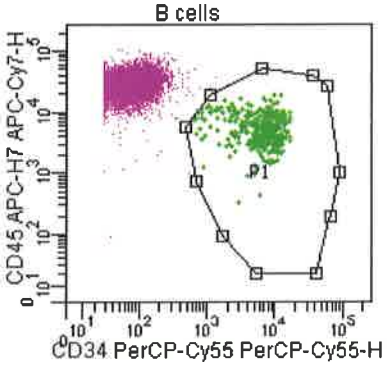

Viable
SSC-H
CD45 APC-H7 APC-Cy7-H

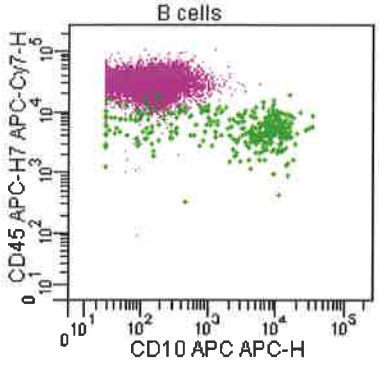

Viable
All CD19
SSC-H
CD19 PE-Cy7 PE-Cy7-H

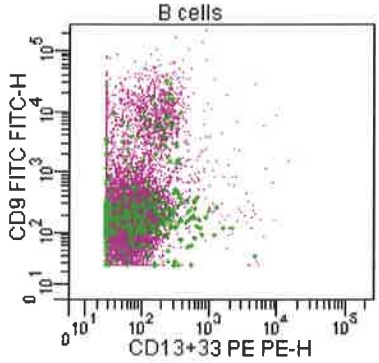

All CD19
B cells
CD45 APC-H7 APC-Cy7-H
CD19 PE-Cy7 PE-Cy7-H

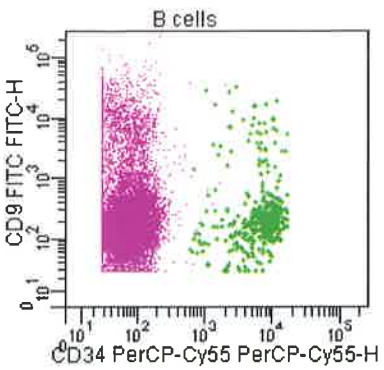

B cells
CD45 APC-H7 APC-Cy7-H
CD9 FITC FITC-H

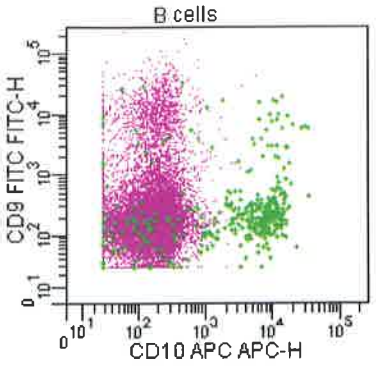

B cells
CD45 APC-H7 APC-Cy7-H
CD13+33 PE PE-H

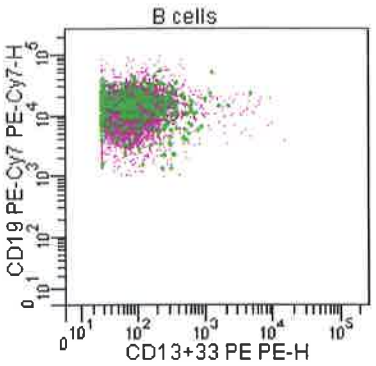

B cells
P1
CD45 APC-H7 APC-Cy7-H
CD34 PerCP-Cy55 PerCP-Cy55-H

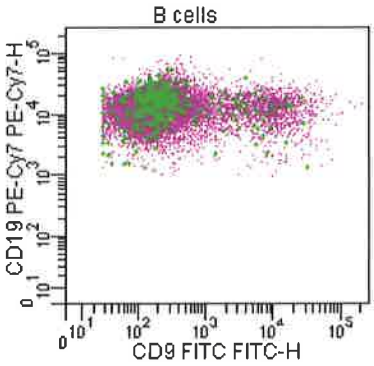

B cells
CD45 APC-H7 APC-Cy7-H
CD10 APC APC-H

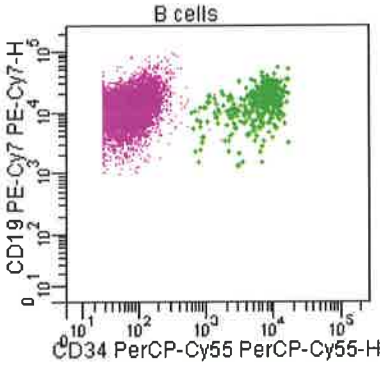

B cells
CD9 FITC FITC-H
CD13+33 PE PE-H

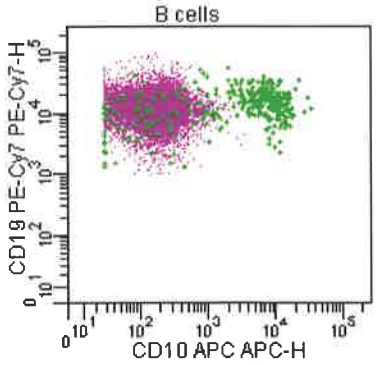


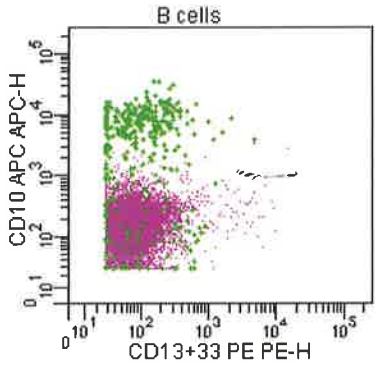



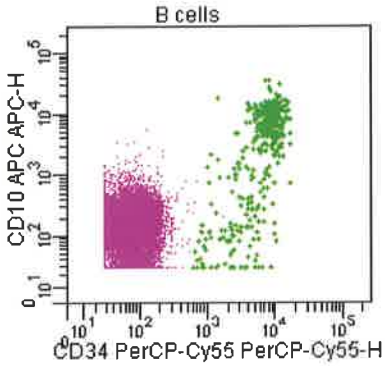



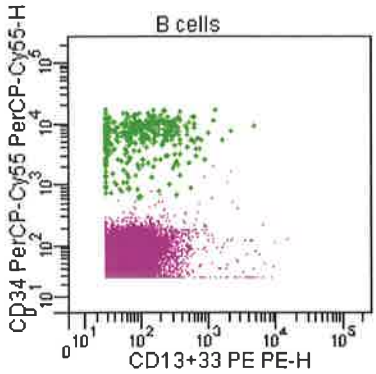



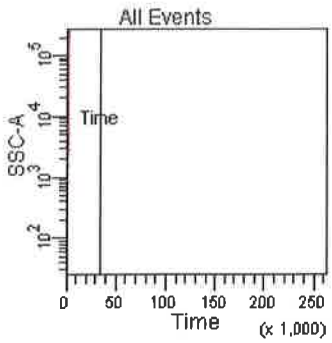



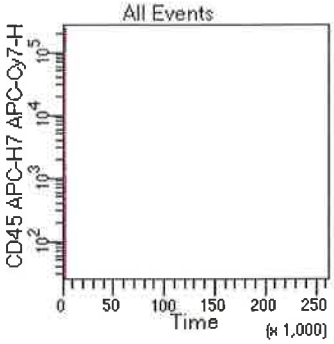



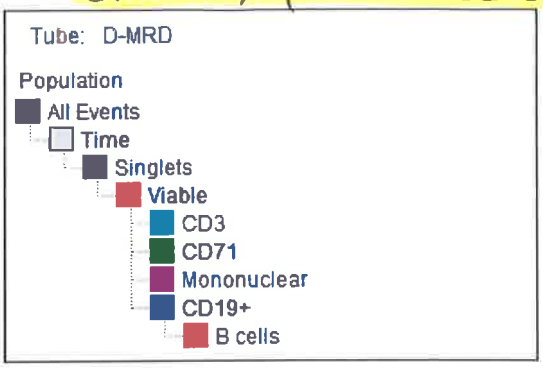



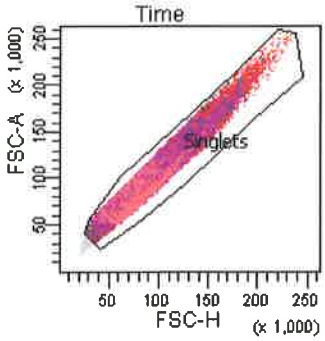

| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 4 |
| Specimen Name: | Case 2 |
| Tube Name: | B -MRD |
| Record Date: | Apr 8, 2016 2:52:04 PM |
| $OP: | HPDevelopmental |
| GUID: | 0a0e5f84-fcab-45cd-bdfe-1814052300ca |
| Serial Number: | |

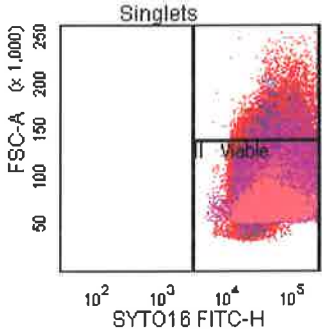
| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 239,128 | #### | #### | 100.0 |
| Time | 239,128 | 100.0 | #### | 100.0 |
| Singlets | 237,766 | 99.4 | 99.4 | 99.4 |
| Viable | 237,729 | 100.0 | 99.4 | 99.4 |
| All CD19 | 14,460 | 6.1 | 6.1 | 6.0 |
| B cells | 14,455 | 100.0 | 6.1 | 6.0 |
| P1 | 360 | 2.5 | 2.5 | 0.2 |

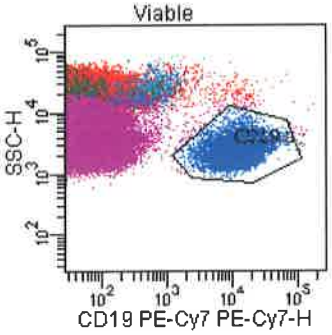


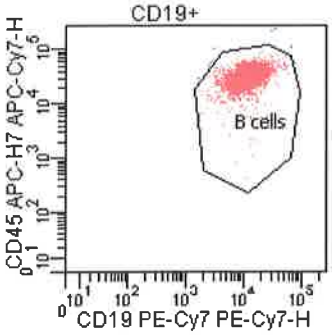



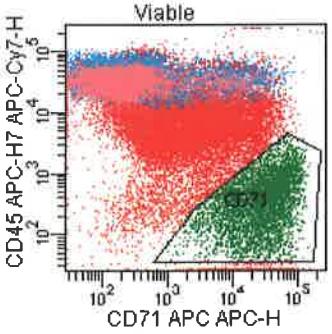



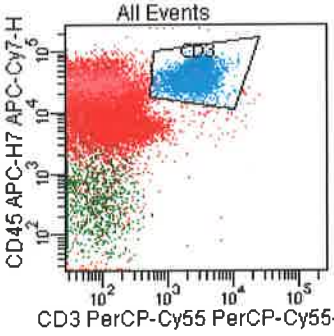



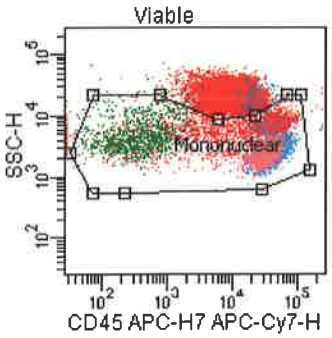



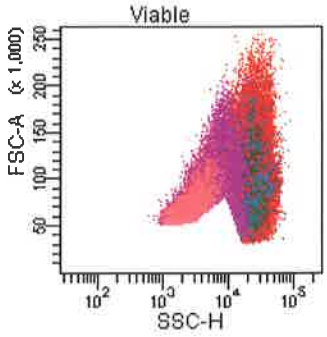



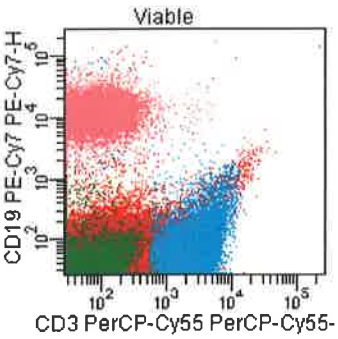



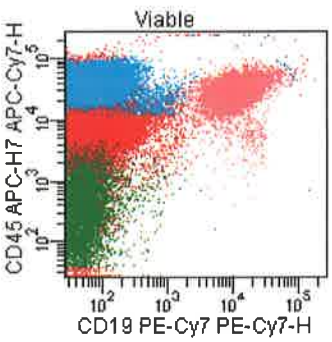



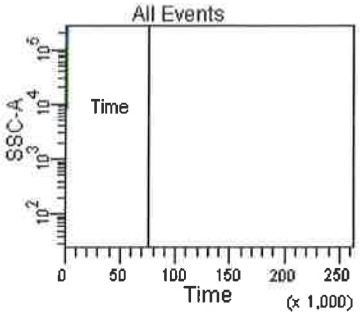



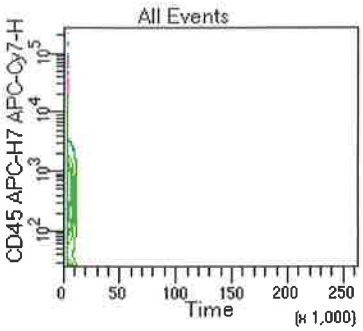

| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 4 |
| Specimen Name: | Case 2 |
| Tube Name: | D-MRD |
| Record Date: | Apr 8, 2016 2:56:32 PM |
| $OP: | HPDevelopmental |
| GUID: | 600004e6-e91b-49c5-95a7-8ea0... |
| Serial Number: | |

| Population | #Events | %Parent | %Grand Par... | %Total |
|---|---|---|---|---|
| All Events | 255,526 | #### | #### | 100.0 |
| Time | 255,526 | 100.0 | #### | 100.0 |
| Singlets | 254,705 | 99.7 | 99.7 | 99.7 |
| Viable | 254,705 | 100.0 | 99.7 | 99.7 |
| CD3 | 38,070 | 14.9 | 14.9 | 14.9 |
| CD71 | 9,668 | 3.8 | 3.8 | 3.8 |
| Mononuclear | 92,494 | 36.3 | 36.3 | 36.2 |
| CD19+ | 16,877 | 6.6 | 6.6 | 6.6 |
| B cells | 16,835 | 99.8 | 6.6 | 6.6 |



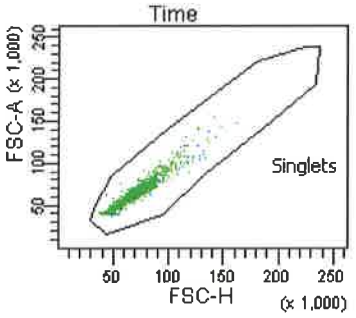



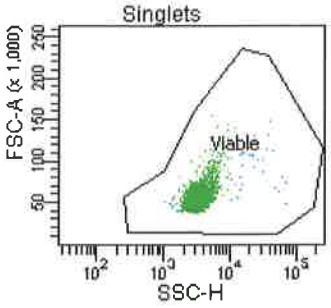


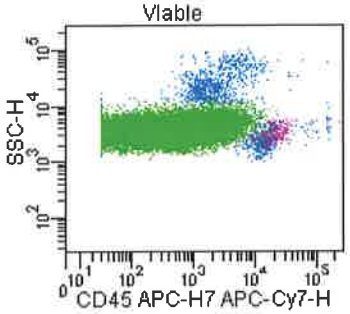



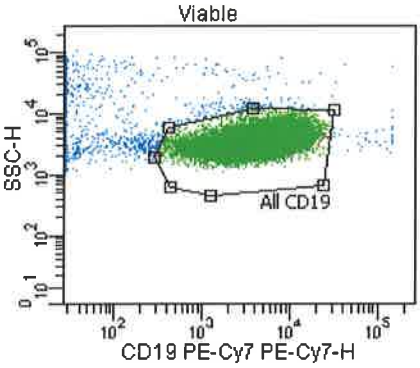

Tube: A-PRE

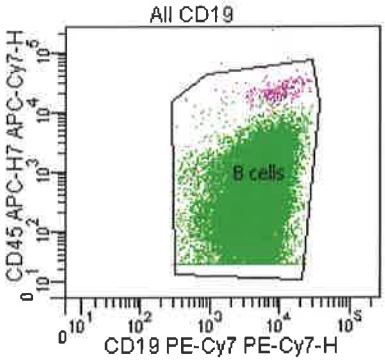
| Population | #Events | %Parent | %Total |
|---|---|---|---|
| All Events | 34,638 | #### | 100.0 |
| Time | 34,638 | 100.0 | 100.0 |
| Singlets | 34,637 | 100.0 | 100.0 |
| Viable | 34,637 | 100.0 | 100.0 |
| All CD19 | 33,002 | 95.3 | 95.3 |
| B cells | 32,992 | 100.0 | 95.2 |
| P1 | 32,778 | 99.4 | 94.6 |

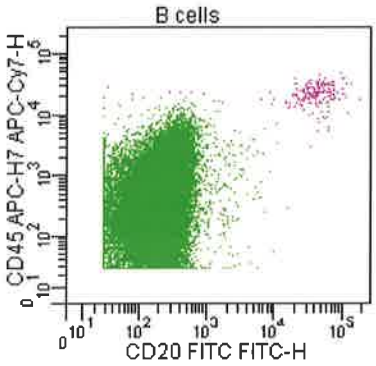


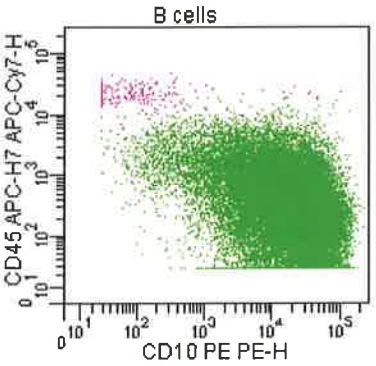



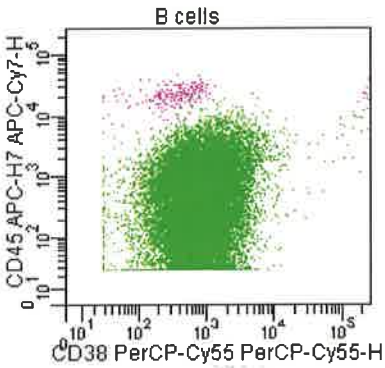



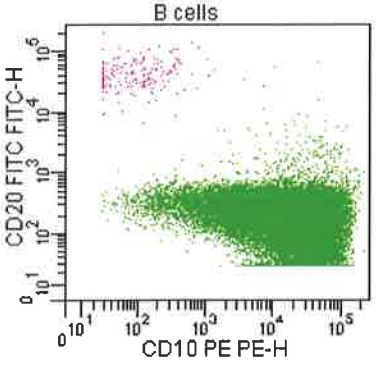



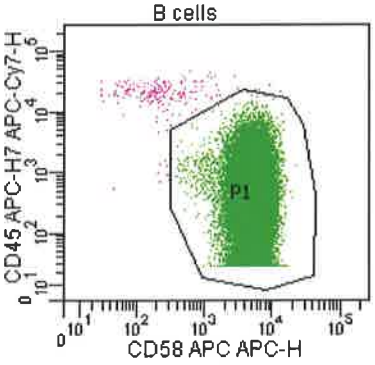



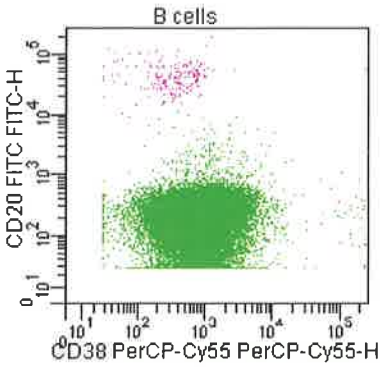



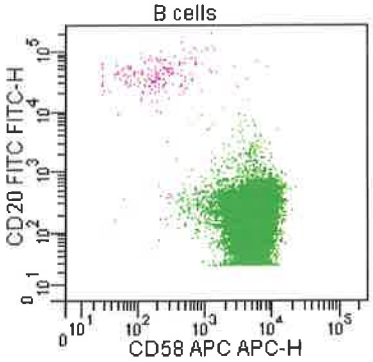



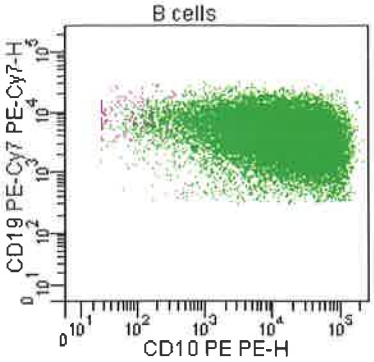



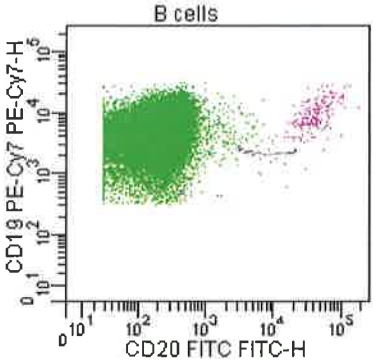



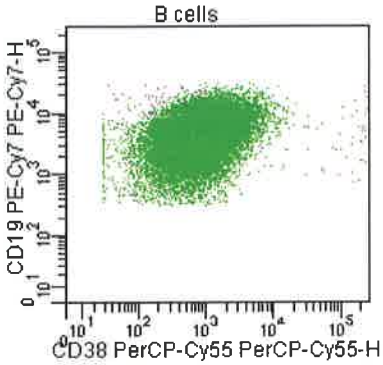



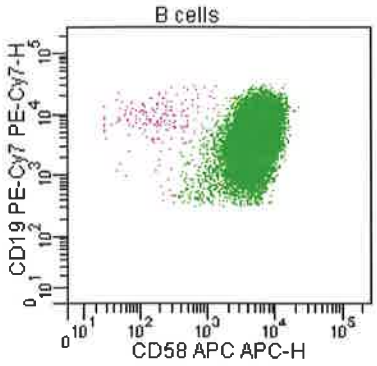


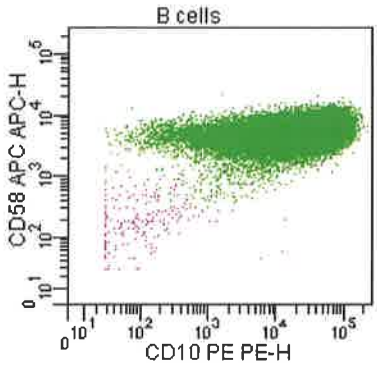



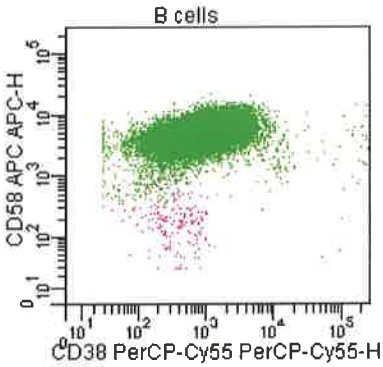



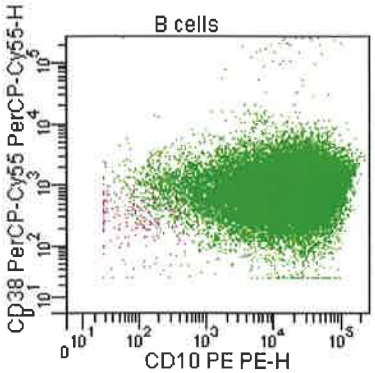



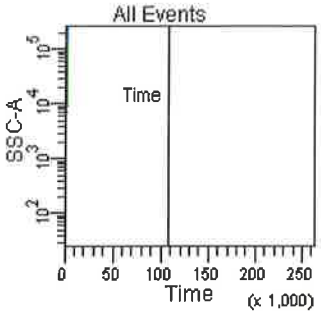



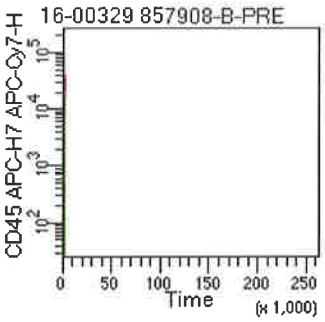



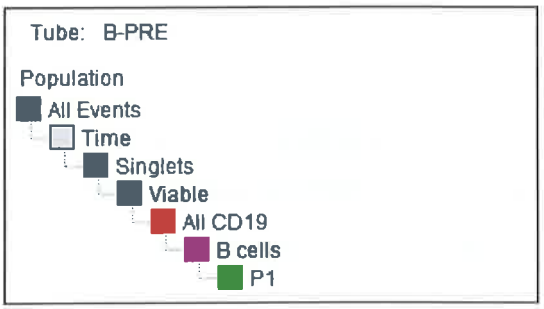



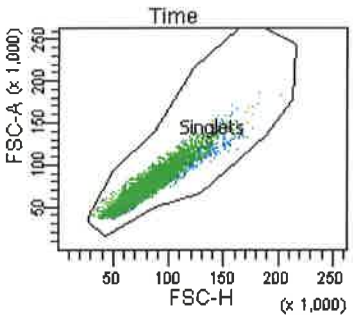

| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 5 |
| Specimen Name: | 16-00329 857908 |
| Tube Name: | A-PRE |
| Record Date: | Jan 7, 2016 5:13:31 PM |
| $OP: | HPDevelopmental |
| GUID: | 050b23cd-64ed-470a-9c49-af13ff97cfaf |
| Serial Number: | |

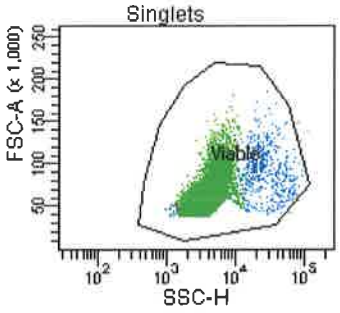
| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 34,638 | #### | #### | 100.0 |
| Time | 34,638 | 100.0 | #### | 100.0 |
| Singlets | 34,637 | 100.0 | 100.0 | 100.0 |
| Viable | 34,637 | 100.0 | 100.0 | 100.0 |
| All CD19 | 33,002 | 95.3 | 95.3 | 95.3 |
| B cells | 32,992 | 100.0 | 95.3 | 95.2 |
| P1 | 32,778 | 99.4 | 99.3 | 94.6 |

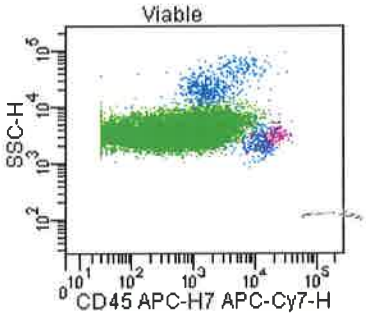

All Events
Time
SSC-A
Time
(x 1,000)

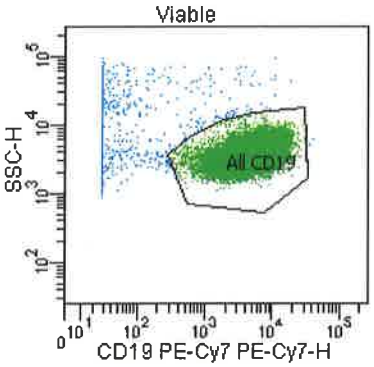

16-00329 857908-B-PRE
CD45 APC-H7 APC-Cy7-H
Time
(x 1,000)

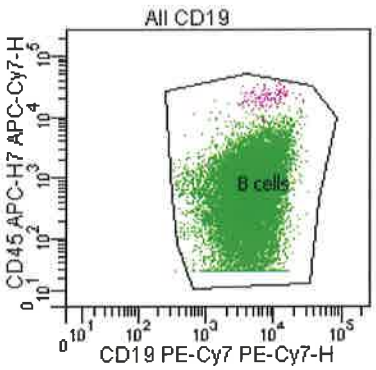

Tube: B-PRE
Population
All Events
Time
Singlets
Viable
All CD19
B cells
P1

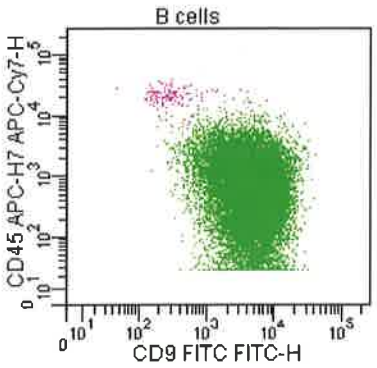

Time
Singlets
FSC-A (x 1,000)
FSC-H
(x 1,000)

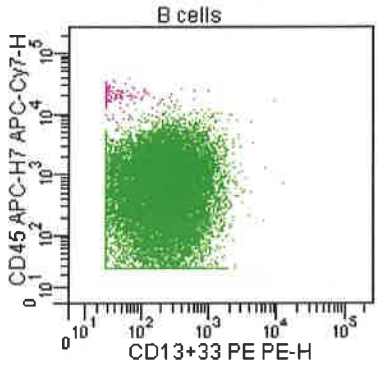

Singlets
Viable
FSC-A (x 1,000)
SSC-H

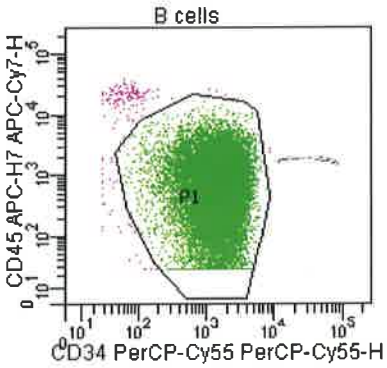

Viable
SSC-H
CD45 APC-H7 APC-Cy7-H

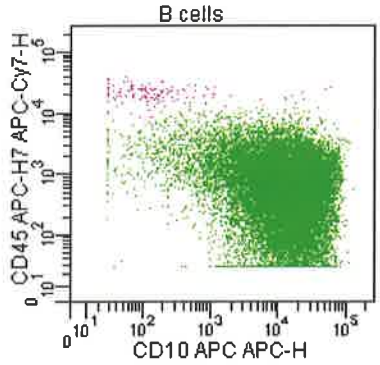

Viable
All CD19
SSC-H
CD19 PE-Cy7 PE-Cy7-H

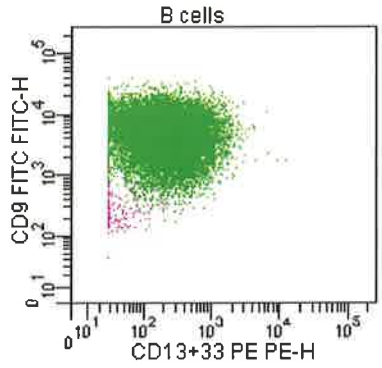

All CD19
B cells
CD45 APC-H7 APC-Cy7-H
CD19 PE-Cy7 PE-Cy7-H

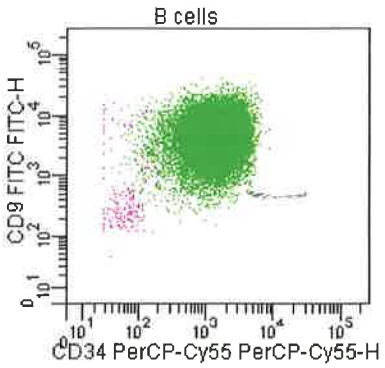

B cells
CD45 APC-H7 APC-Cy7-H
CD9 FITC FITC-H

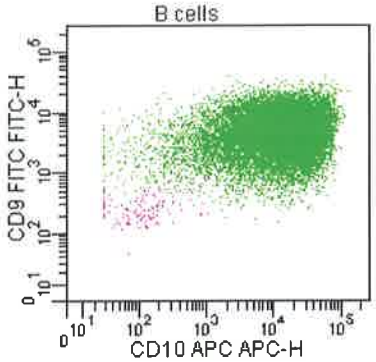

B cells
CD45 APC-H7 APC-Cy7-H
CD13+33 PE PE-H

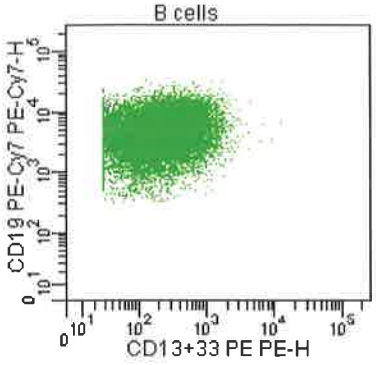

B cells
P1
CD45 APC-H7 APC-Cy7-H
CD34 PerCP-Cy55 PerCP-Cy55-H

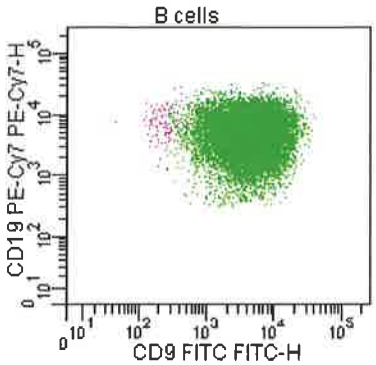

B cells
CD45 APC-H7 APC-Cy7-H
CD10 APC APC-H

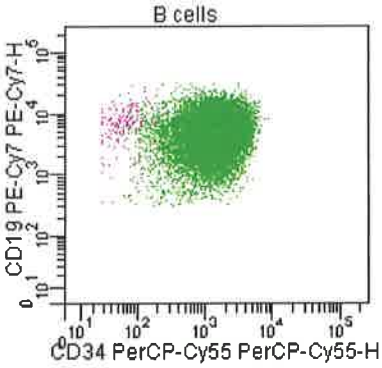

B cells
CD9 FITC FITC-H
CD13+33 PE PE-H

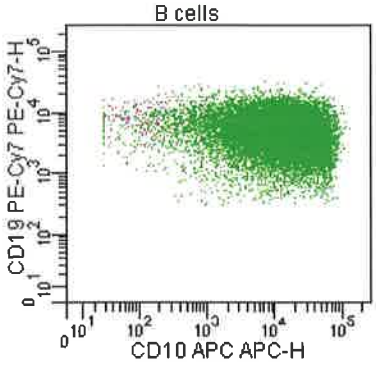


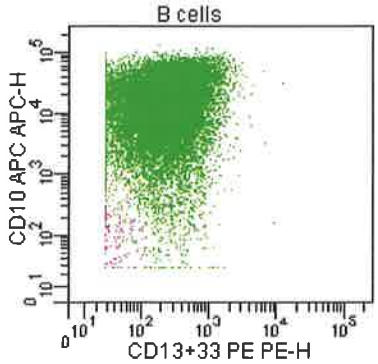



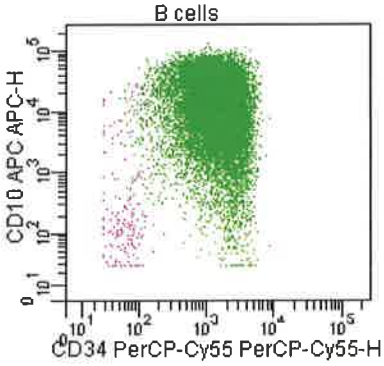



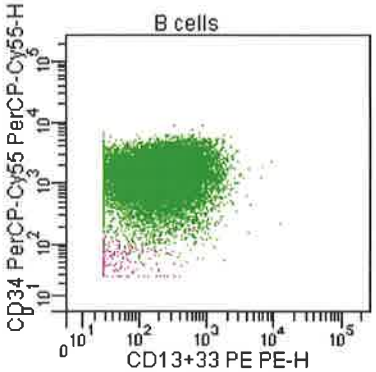



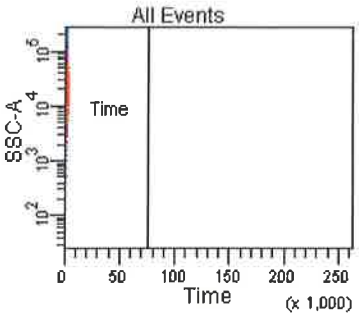



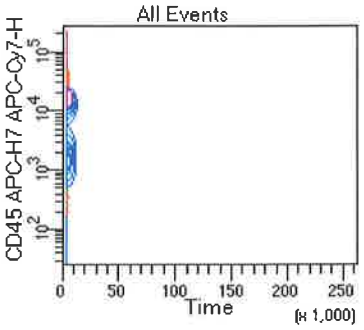




| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 5 |
| Specimen Name: | 16-00329 857908 |
| Tube Name: | B-PRE |
| Record Date: | Jan 7, 2016 5:14:20 PM |
| $OP: | HPDevelopmental |
| GUID: | 22d6c374-6fb5-4b62-a838-028e5d8daa12 |
| Serial Number: | |

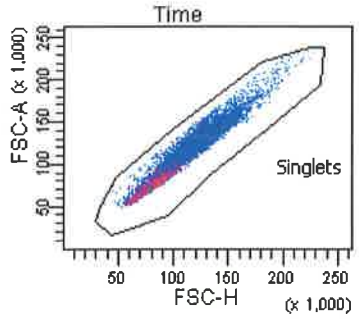
| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 23,560 | #### | #### | 100.0 |
| Time | 23,560 | 100.0 | #### | 100.0 |
| Singlets | 23,559 | 100.0 | 100.0 | 100.0 |
| Viable | 23,550 | 100.0 | 100.0 | 100.0 |
| All CD19 | 22,497 | 95.5 | 95.5 | 95.5 |
| B cells | 22,491 | 100.0 | 95.5 | 95.5 |
| P1 | 22,302 | 99.2 | 99.1 | 94.7 |

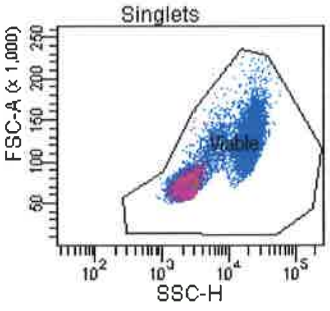


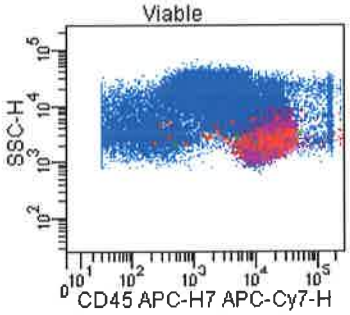



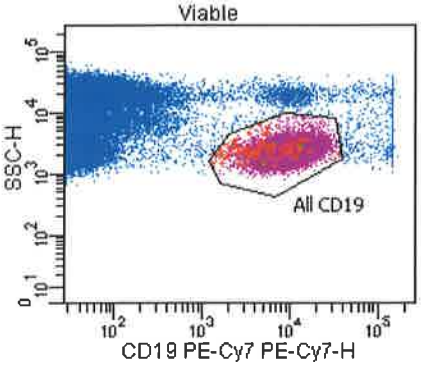

Tube: A-MRD

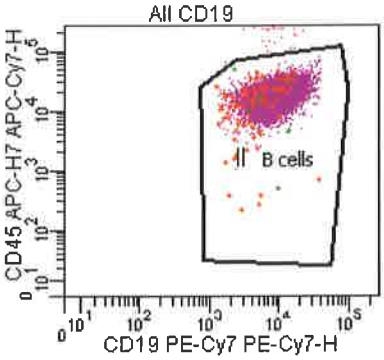
| Population | #Events | %Parent | %Total |
|---|---|---|---|
| All Events | 211,313 | #### | 100.0 |
| Time | 211,313 | 100.0 | 100.0 |
| Singlets | 211,089 | 99.9 | 99.9 |
| Viable | 210,975 | 99.9 | 99.8 |
| All CD19 | 24,121 | 11.4 | 11.4 |
| B cells | 24,094 | 99.9 | 11.4 |
| P1 | 7 | 0.0 | 0.0 |
| P2 | 84 | 0.3 | 0.0 |

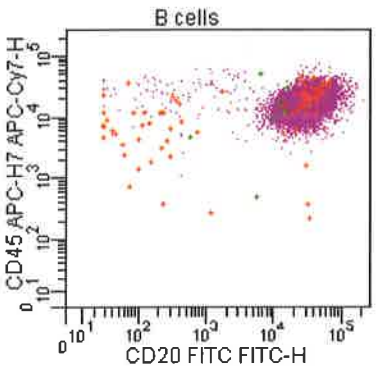


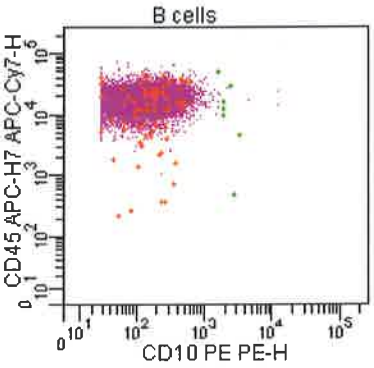



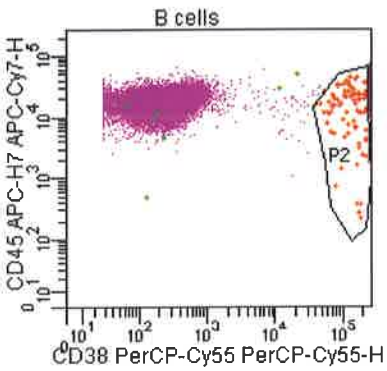



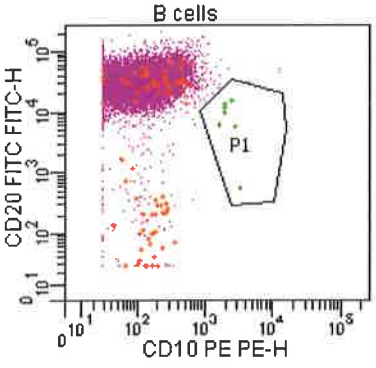



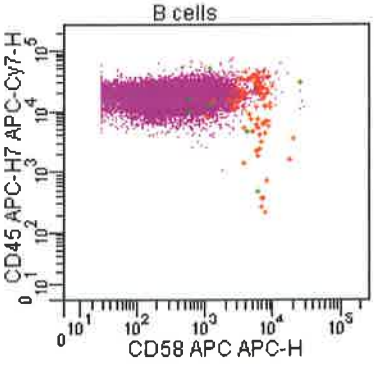



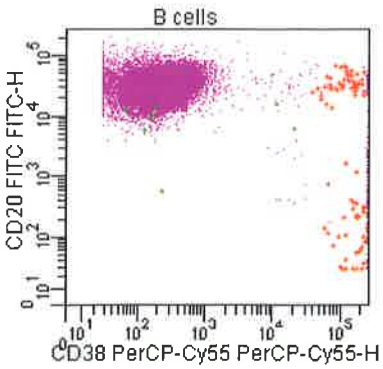



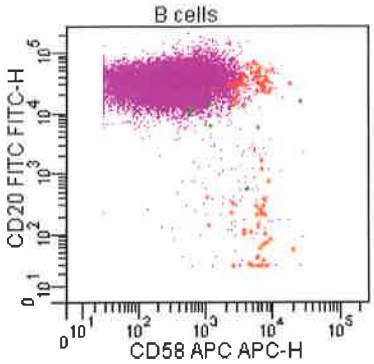



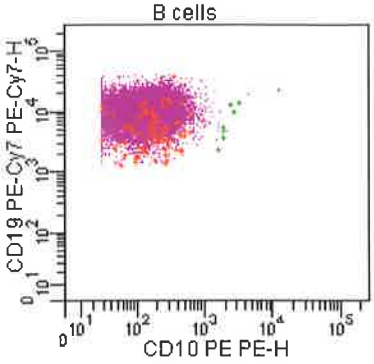



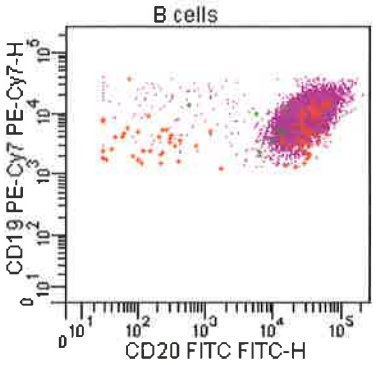



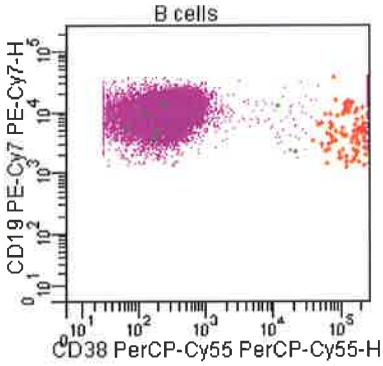



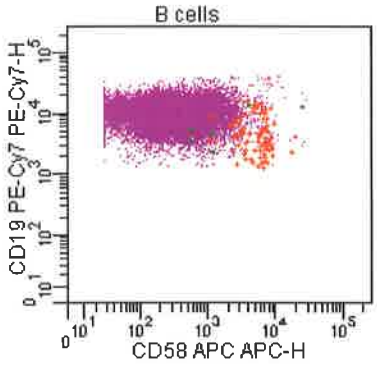


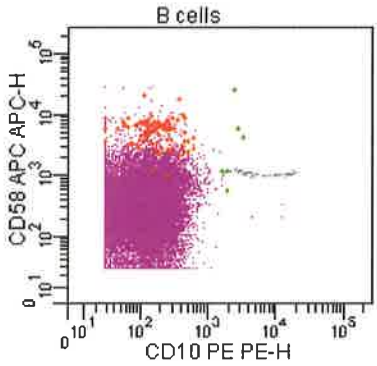



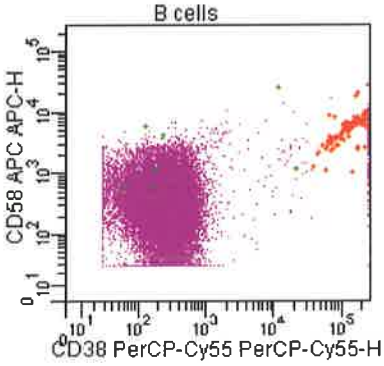



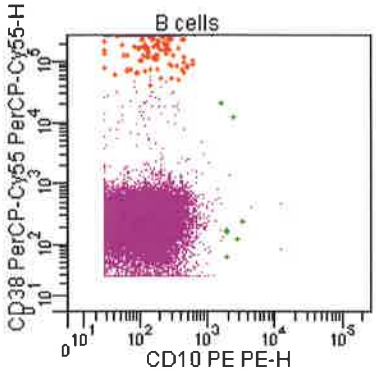



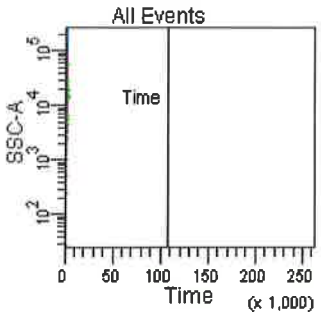



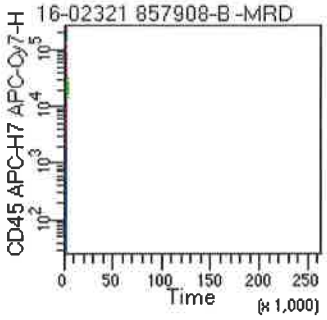



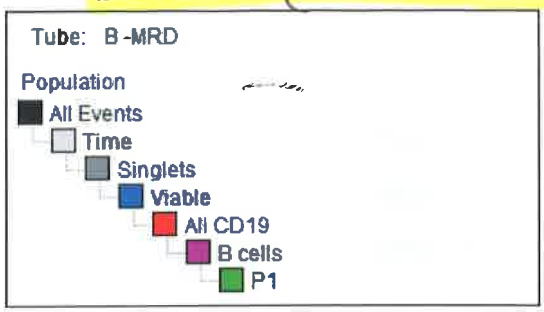



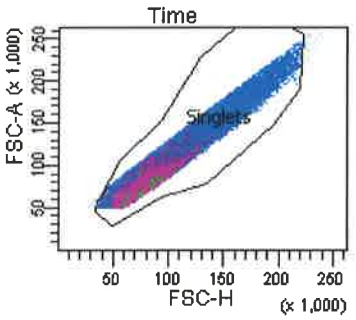

| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 5 |
| Specimen Name: | 16-02321 857908 |
| Tube Name: | A-MRD |
| Record Date: | Feb 3, 2016 2:12:04 PM |
| $OP: | HPDevelopmental |
| GUID: | 801c3f59-0c7e-44e2-b8ff-618864781a80 |
| Serial Number: | |

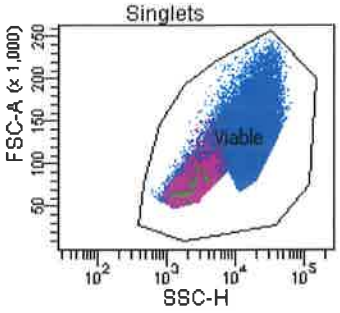
| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 211,313 | #### | #### | 100.0 |
| Time | 211,313 | 100.0 | #### | 100.0 |
| Singlets | 211,089 | 99.9 | 99.9 | 99.9 |
| Viable | 210,975 | 99.9 | 99.8 | 99.8 |
| All CD19 | 24,121 | 11.4 | 11.4 | 11.4 |
| B cells | 24,094 | 99.9 | 11.4 | 11.4 |
| P1 | 7 | 0.0 | 0.0 | 0.0 |
| P2 | 84 | 0.3 | 0.3 | 0.0 |

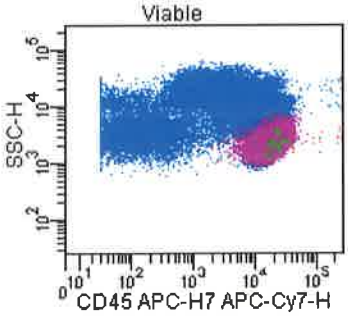

All Events
Time
SSC-A
Time
(x 1,000)

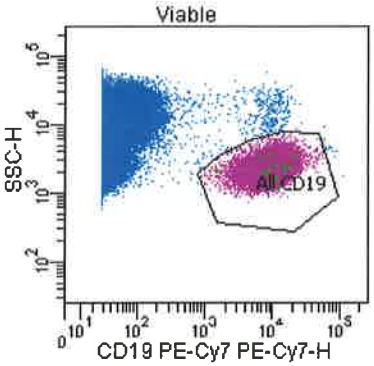

16-02321 857908-B -MRD
CD45 APC-H7 APC-Cy7-H
Time
(x 1,000)

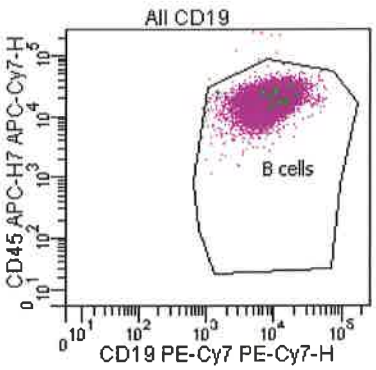

Tube: B -MRD
Population
All Events
Time
Singlets
Viable
All CD19
B cells
P1

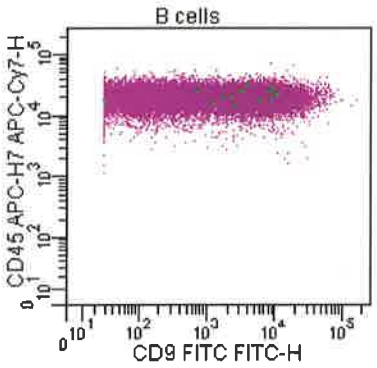

Time
Singlets
FSC-A (x 1,000)
FSC-H
(x 1,000)

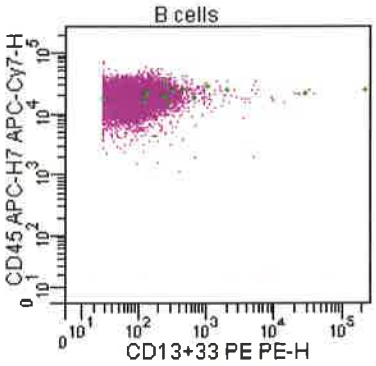

Singlets
Viable
FSC-A (x 1,000)
SSC-H

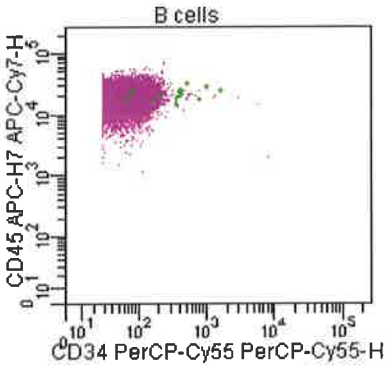

Viable
SSC-H
CD45 APC-H7 APC-Cy7-H

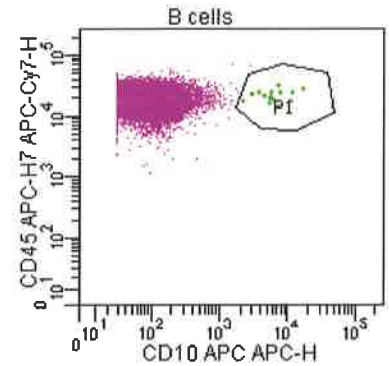

Viable
All CD19
SSC-H
CD19 PE-Cy7 PE-Cy7-H

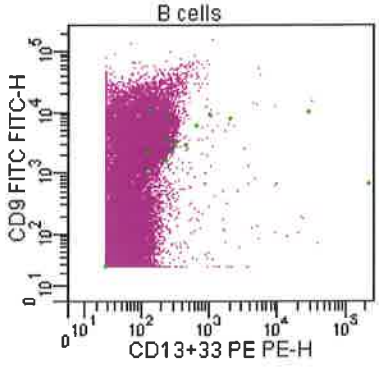

All CD19
B cells
CD45 APC-H7 APC-Cy7-H
CD19 PE-Cy7 PE-Cy7-H

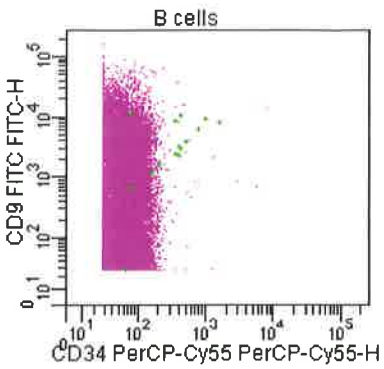

B cells
CD45 APC-H7 APC-Cy7-H
CD9 FITC FITC-H

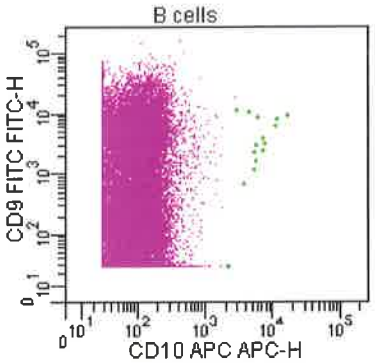

B cells
CD45 APC-H7 APC-Cy7-H
CD13+33 PE PE-H

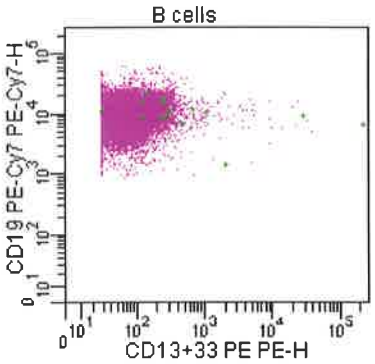

B cells
CD45 APC-H7 APC-Cy7-H
CD34 PerCP-Cy55 PerCP-Cy55-H

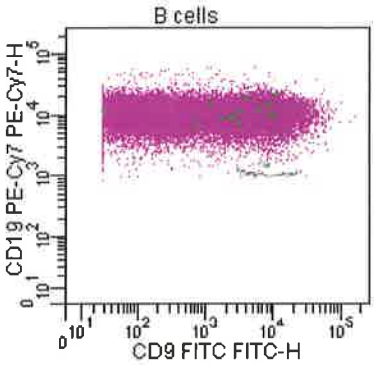

B cells
P1
CD45 APC-H7 APC-Cy7-H
CD10 APC APC-H

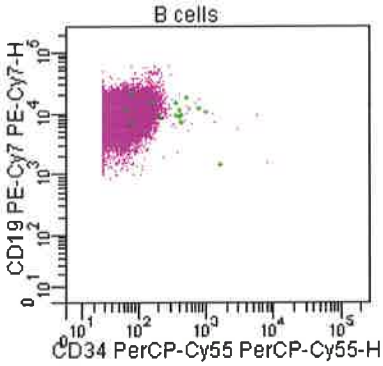

B cells
CD9 FITC FITC-H
CD13+33 PE PE-H

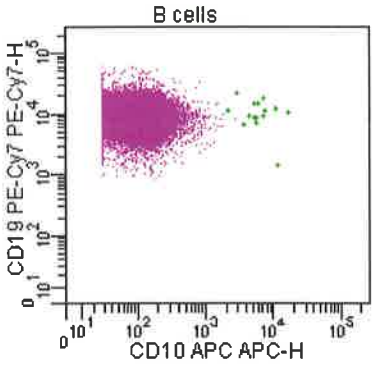


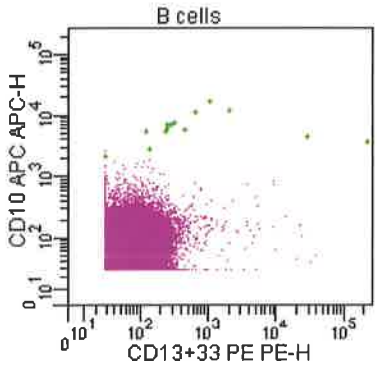



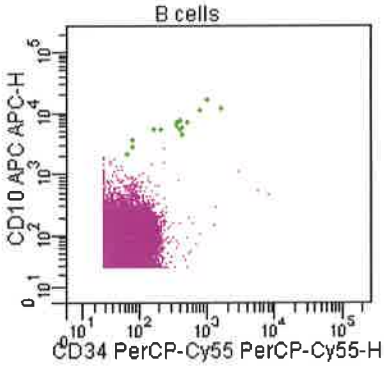



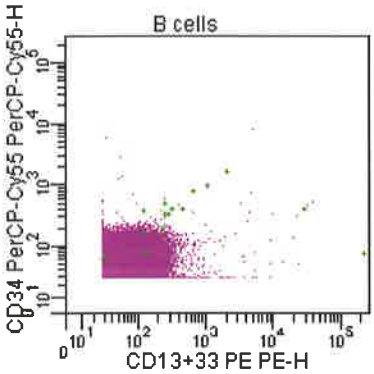



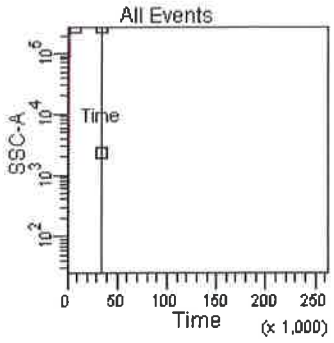



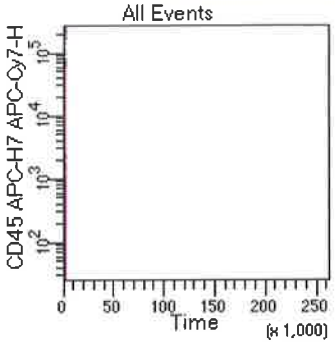



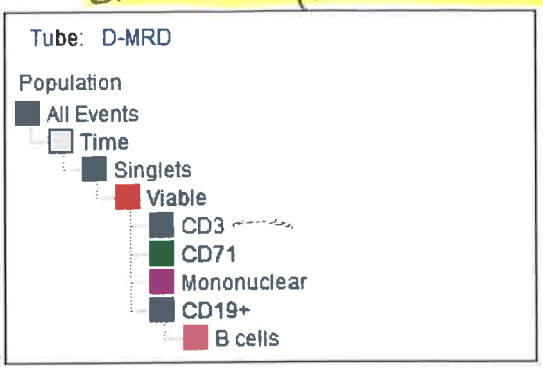



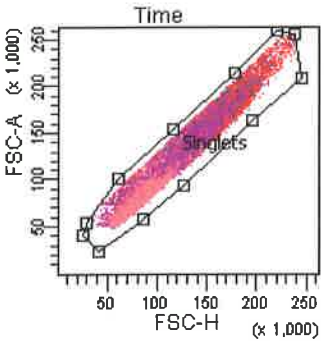

| | |
|---|---|
| **Experiment Name:** | **CAP BALL-B 2019 CASE 5** |
| **Specimen Name:** | **16-02321 857908** |
| **Tube Name:** | **B -MRD** |
| **Record Date:** | **Feb 3, 2016 2:15:00 PM** |
| **$OP:** | **HPDevelopmental** |
| **GUID:** | **c27cc70c-6b7c-4aa5-9b70-a2445c62d4de** |
| Serial Number: | |

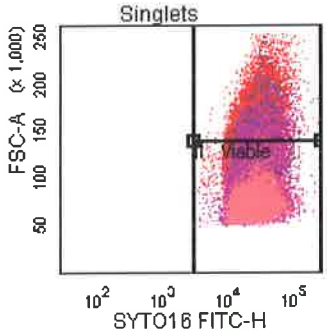
| Population | #Events | %Parent | %Grand Parent | %Total |
|---|---|---|---|---|
| All Events | 202,164 | #### | #### | 100.0 |
| Time | 202,164 | 100.0 | #### | 100.0 |
| Singlets | 201,965 | 99.9 | 99.9 | 99.9 |
| Viable | 201,964 | 100.0 | 99.9 | 99.9 |
| All CD19 | 34,687 | 17.2 | 17.2 | 17.2 |
| B cells | 34,676 | 100.0 | 17.2 | 17.2 |
| P1 | 15 | 0.0 | 0.0 | 0.0 |

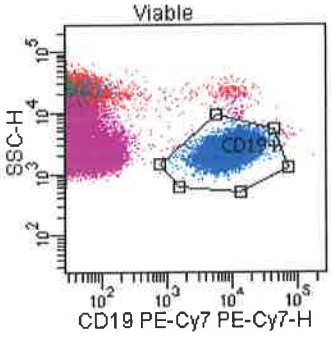


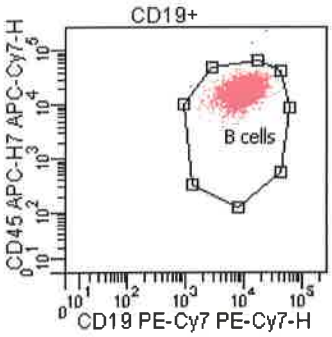



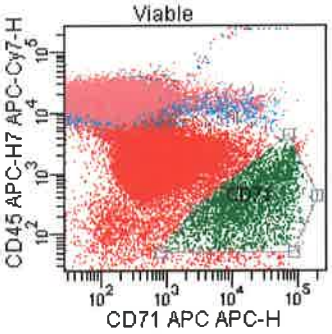



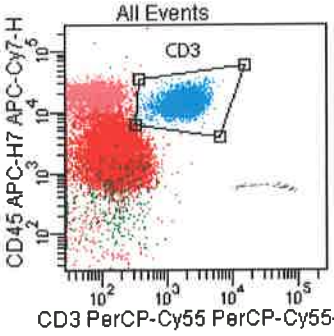



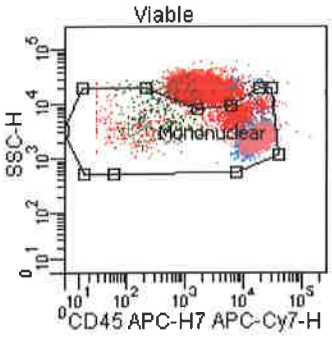



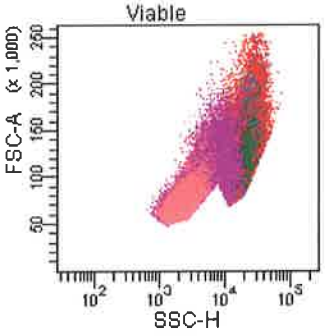



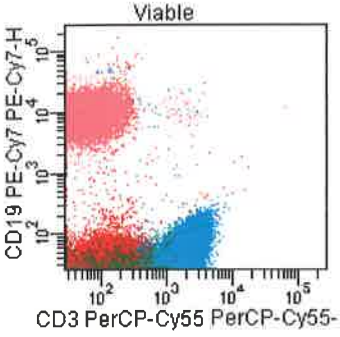



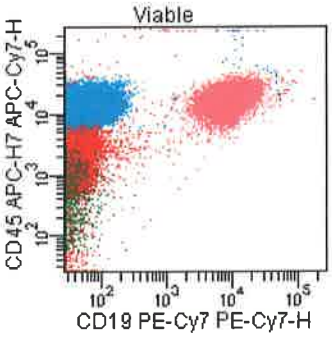




| | |
|---|---|
| Experiment Name: | CAP BALL-B 2019 CASE 5 |
| Specimen Name: | 16-02321 857908 |
| Tube Name: | D-MRD |
| Record Date: | Feb 3, 2016 2:17:28 PM |
| $OP: | HPDevelopmental |
| GUID: | 675e7904-a393-4cf7-8f42-0894c7... |
| Serial Number: | |

| Population | #Events | %Parent | %Grand Par... | %Total |
|---|---|---|---|---|
| All Events | 208,211 | #### | #### | 100.0 |
| Time | 208,211 | 100.0 | #### | 100.0 |
| Singlets | 208,147 | 100.0 | 100.0 | 100.0 |
| Viable | 208,147 | 100.0 | 100.0 | 100.0 |
| CD3 | 48,113 | 23.1 | 23.1 | 23.1 |
| CD71 | 3,215 | 1.5 | 1.5 | 1.5 |
| Mononuclear | 95,931 | 46.1 | 46.1 | 46.1 |
| CD19+ | 32,910 | 15.8 | 15.8 | 15.8 |
| B cells | 32,869 | 99.9 | 15.8 | 15.8 |



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# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

BALL-B 2019 (BALL-06)

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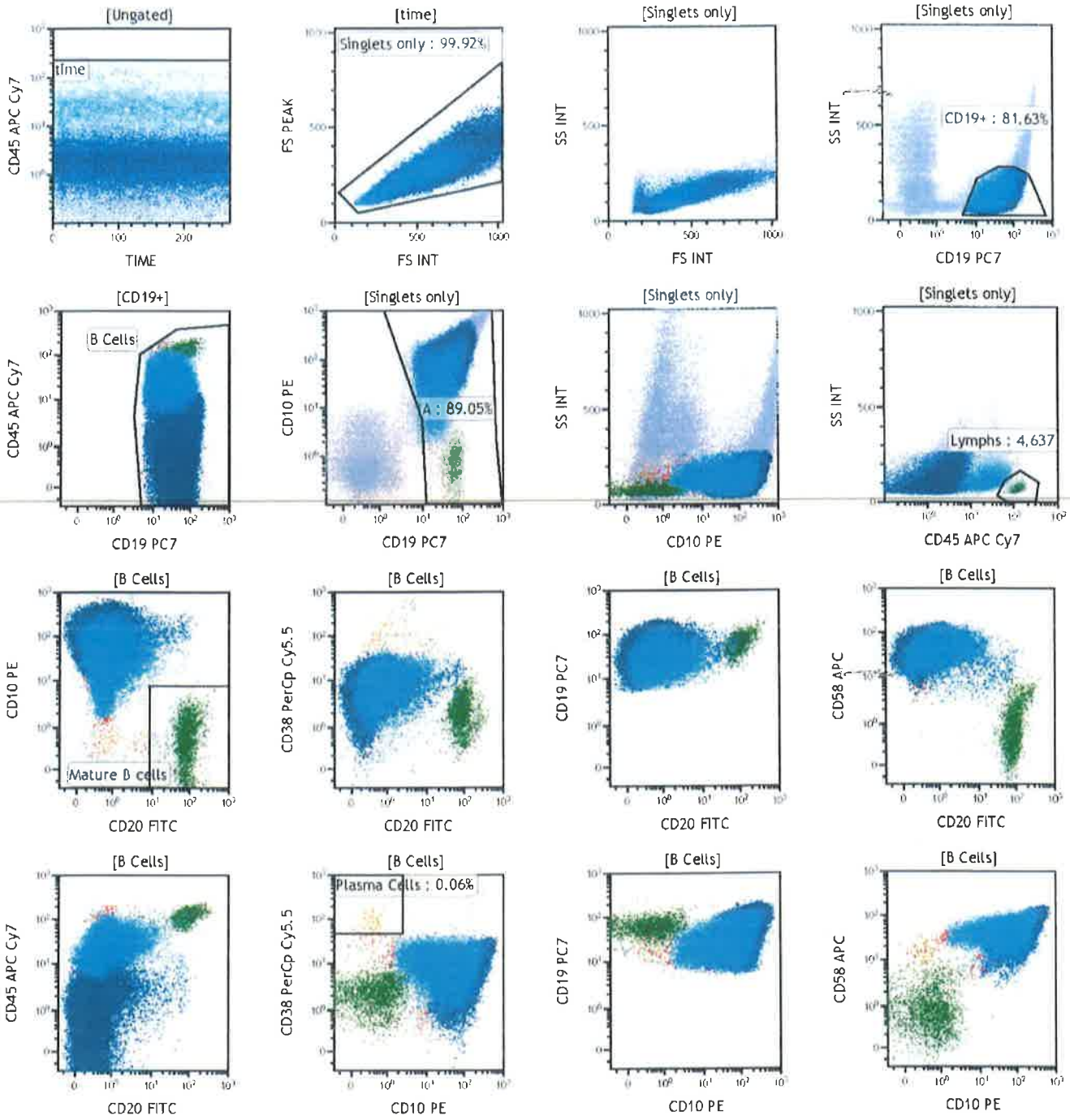
BALL-06

## Diagnostic Phenotype at Onset of Disease

### Tube 1



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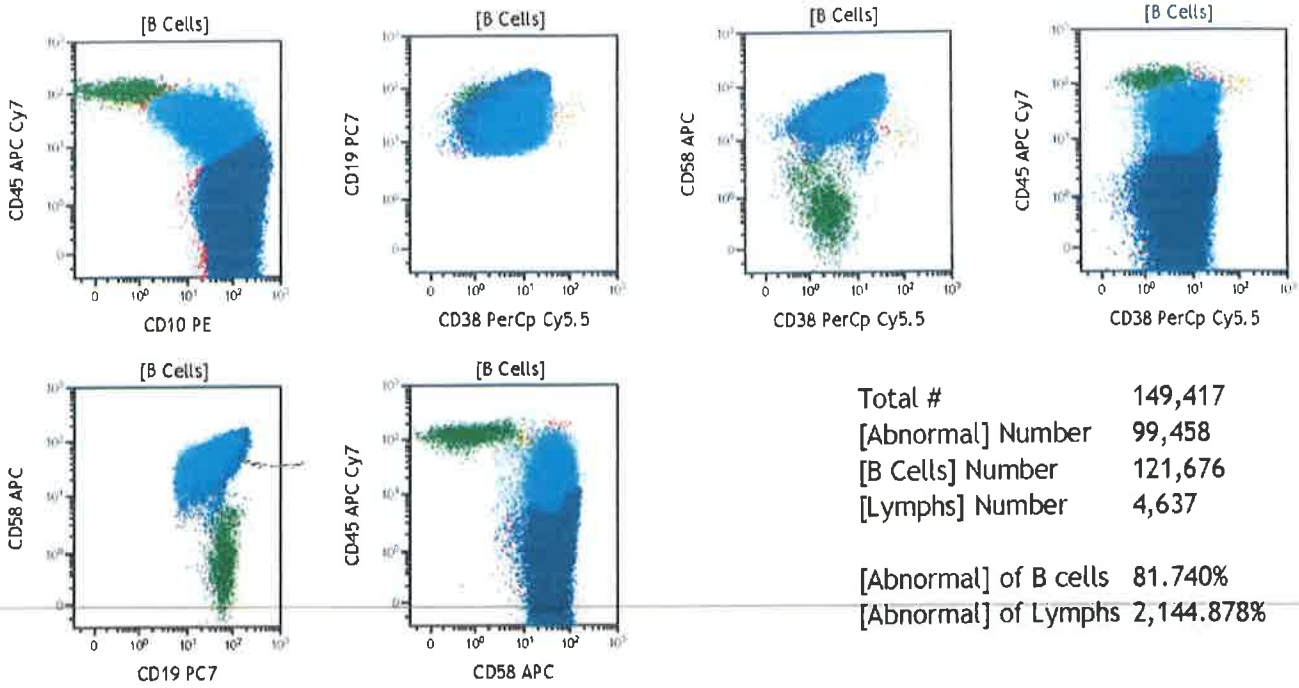
# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

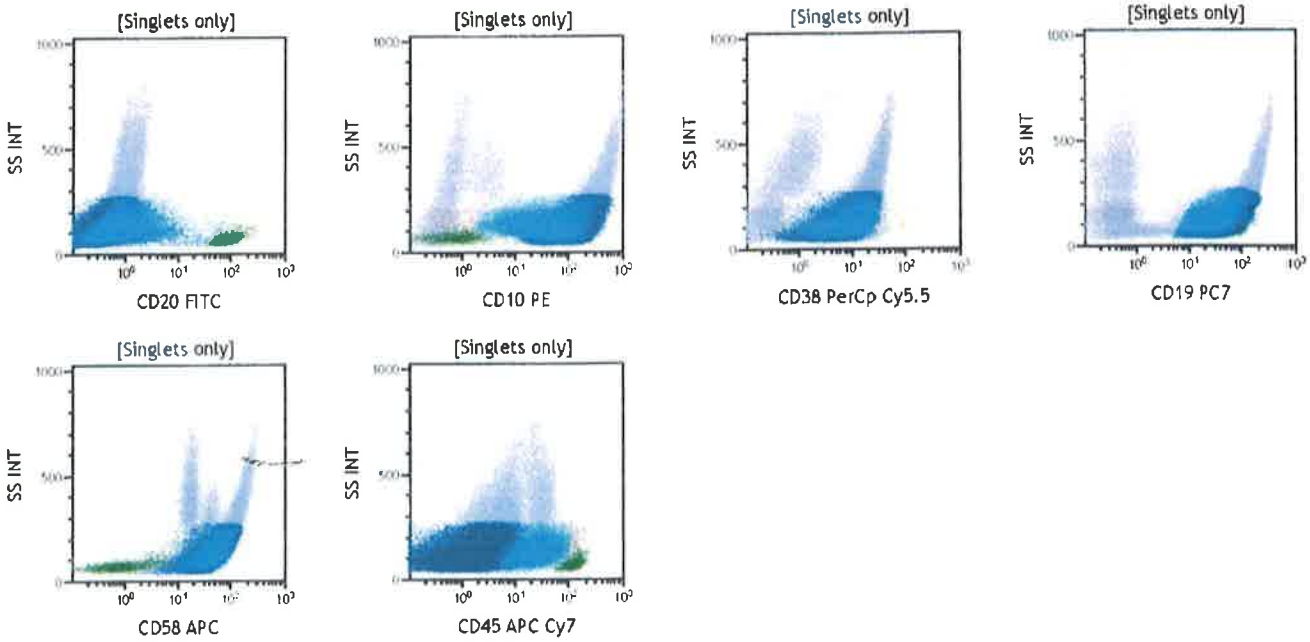
BALL-B 2019 (BALL-06)



## Diagnostic Phenotype at Onset of Disease
## Tube 1, cont'd




| | |
|---|---|
| Total # | 149,417 |
| [Abnormal] Number | 99,458 |
| [B Cells] Number | 121,676 |
| [Lymphs] Number | 4,637 |
| [Abnormal] of B cells | 81.740% |
| [Abnormal] of Lymphs | 2,144.878% |



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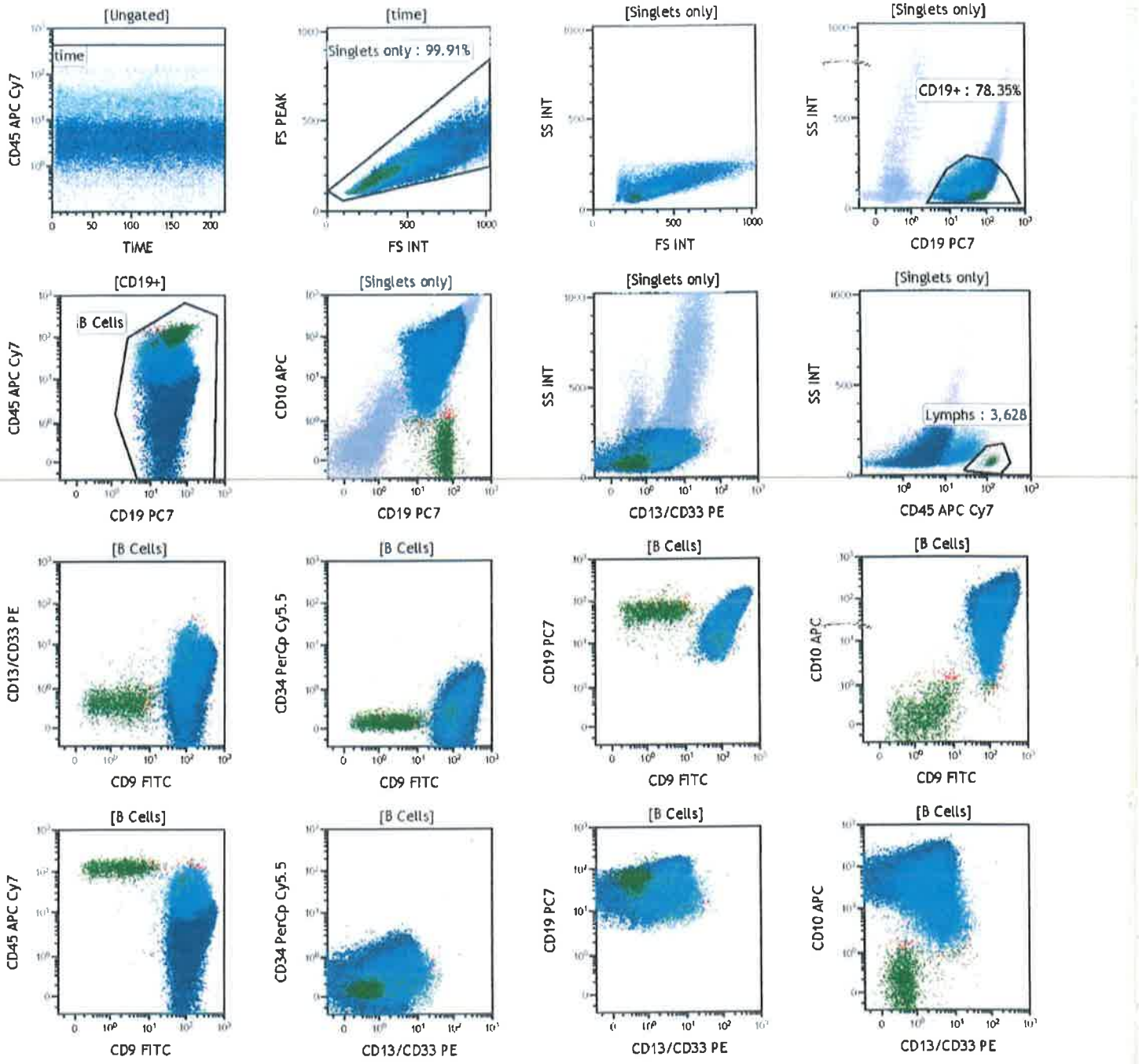
# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

BALL-B 2019 (BALL-06)



## Diagnostic Phenotype at Onset of Disease
## Tube 2



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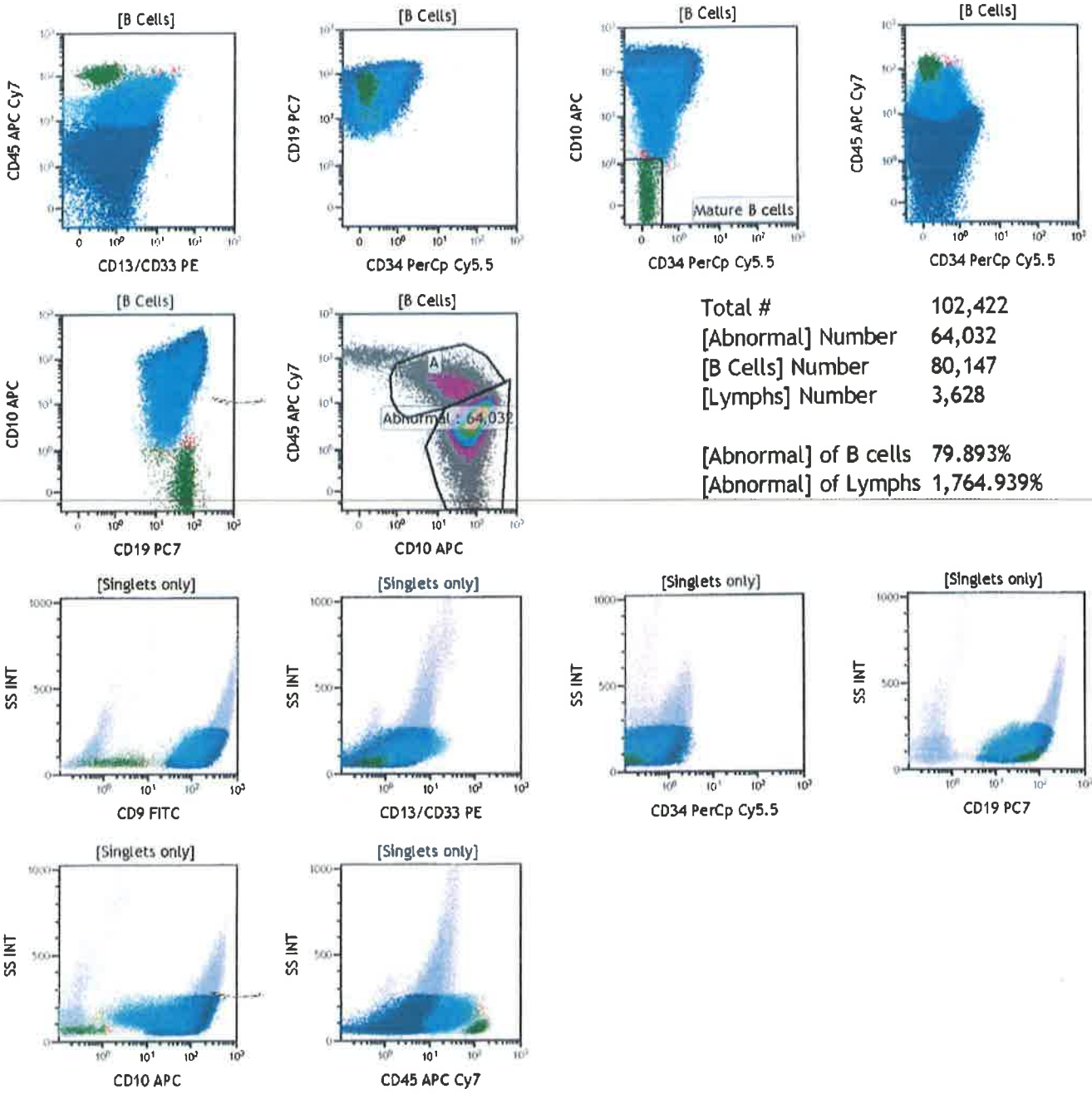
# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

BALL-B 2019 (BALL-06)



## Diagnostic Phenotype at Onset of Disease
## Tube 2, cont'd




| | |
|---|---|
| Total # | 102,422 |
| [Abnormal] Number | 64,032 |
| [B Cells] Number | 80,147 |
| [Lymphs] Number | 3,628 |
| [Abnormal] of B cells | 79.893% |
| [Abnormal] of Lymphs | 1,764.939% |

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# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

BALL-B 2019 (BALL-06)

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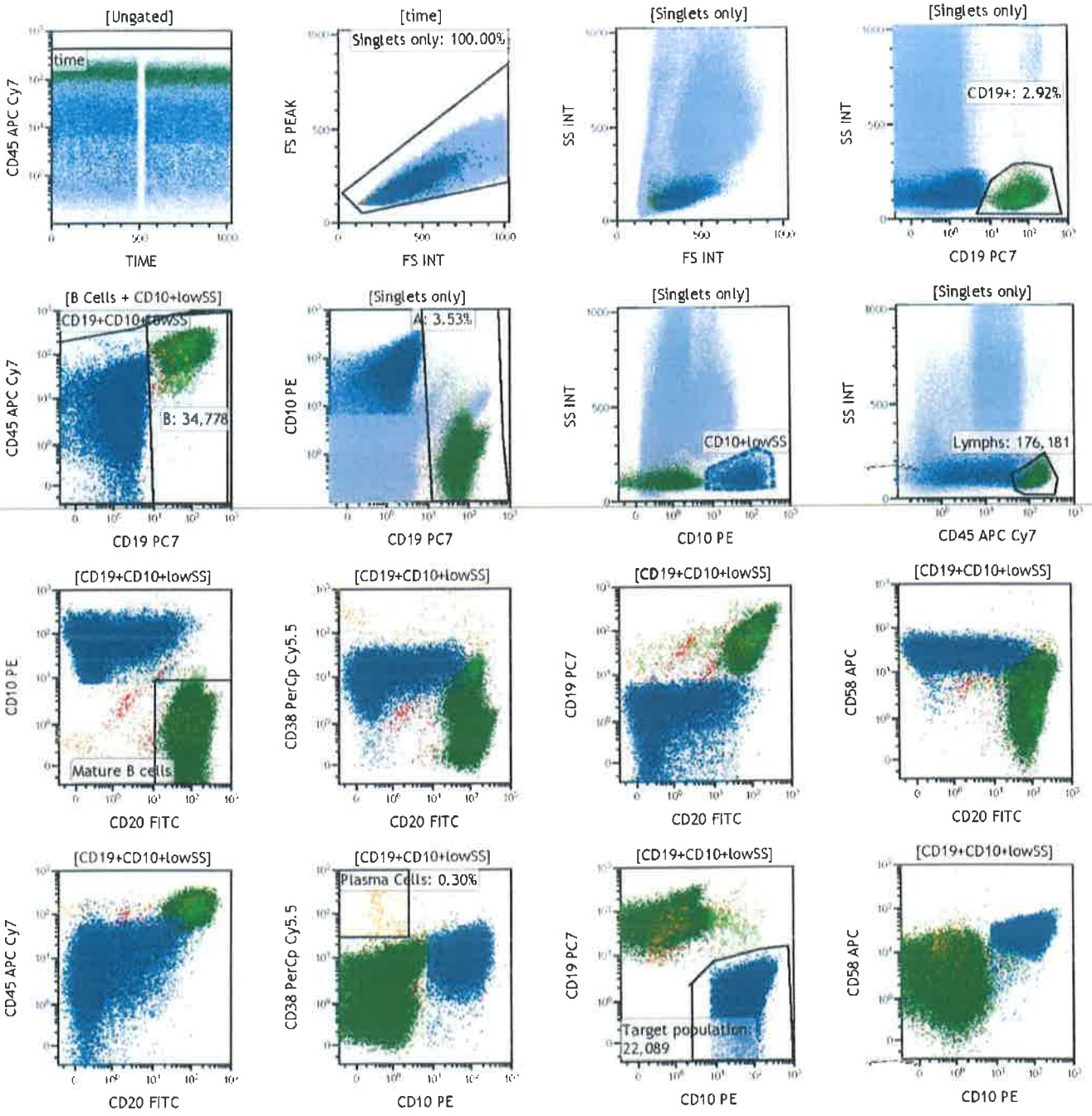
## Day 29 - Tube 1

*Note:* Patient has received autologous chimeric antigen receptor T cells expressing anti-CD19 (CAR-T cell therapy).



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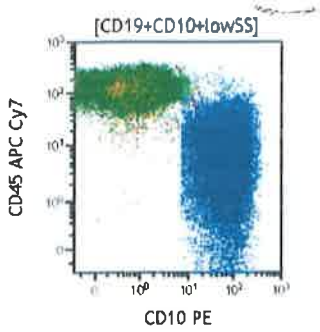
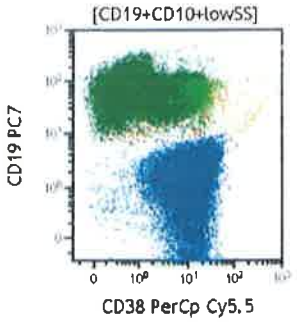
# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

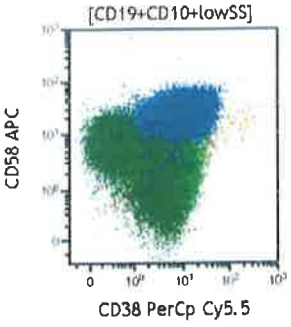
BALL-B 2019 (BALL-06)

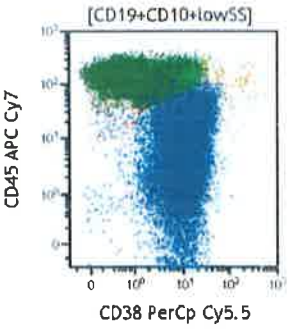


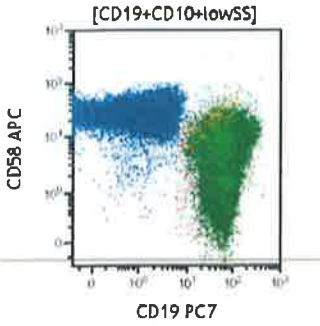
## Day 29 - Tube 1, cont'd

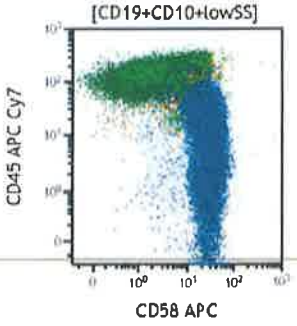


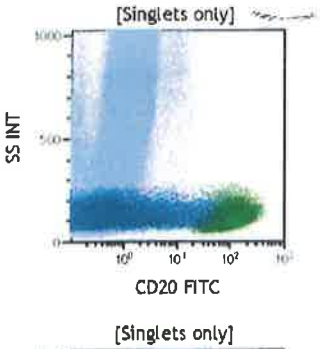



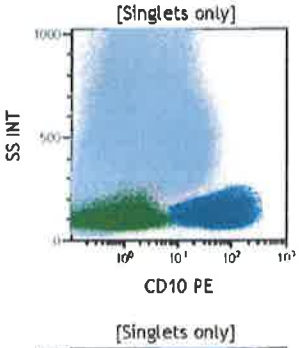



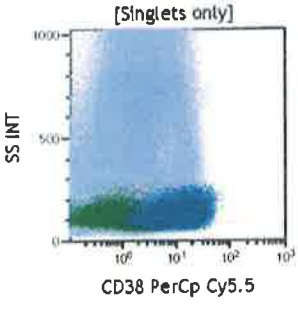



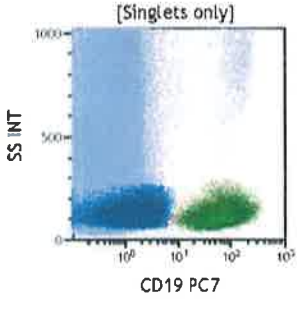



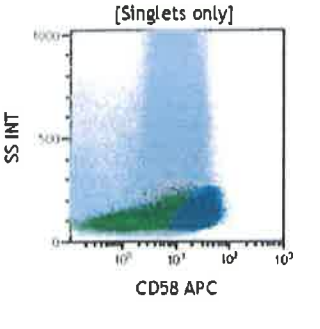

| | |
|---|---|
| Total # | 1,191,934 |
| Target population | 22,401 |
| [CD19+CD10+lowSS] Number | 56,862 |
| [Lymphs] Number | 176,181 |
| Target popualtion"of B cells" | 39.40% |
| Target population "of Lymphs" | 12.71% |

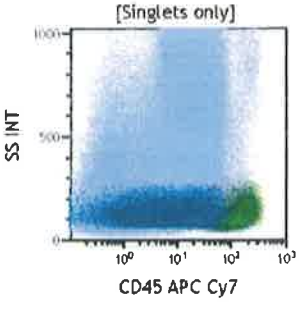


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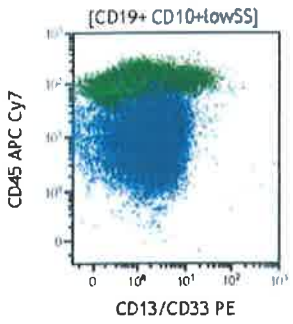
# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

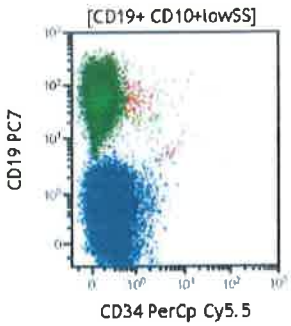
BALL-B 2019 (BALL-06)

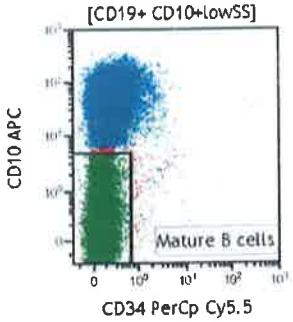


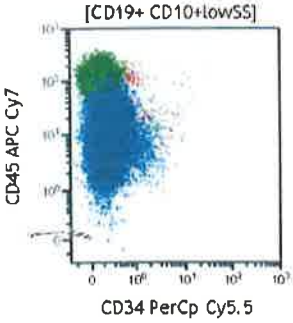
BALL-06

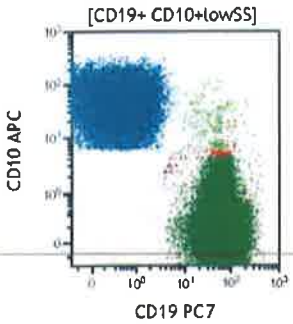
## Day 29 - Tube 2, cont'd

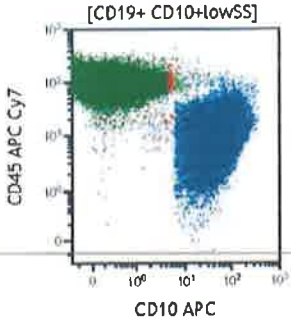


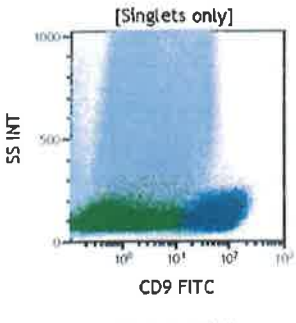



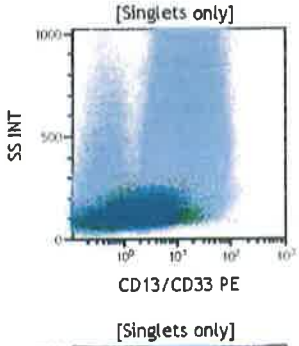



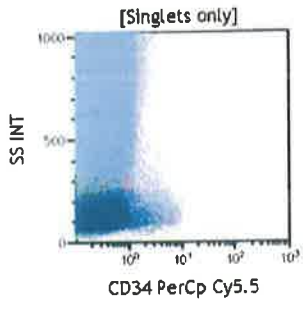



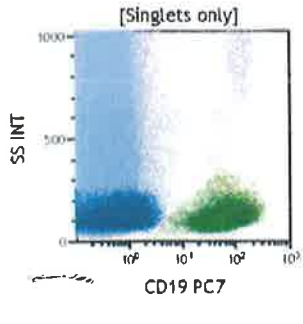



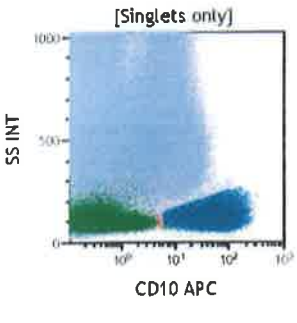

| | |
|---|---|
| Total # | 1,208,861 |
| [Target population] N... | 22,054 |
| [CD19+ CD10+lowSS]... | 60,127 |
| [Lymphs] Number | 184,350 |
| [Target population] o... | 36.68% |
| [Target population] o... | 11.96% |

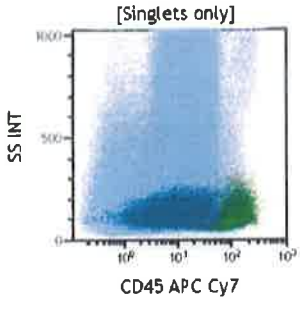


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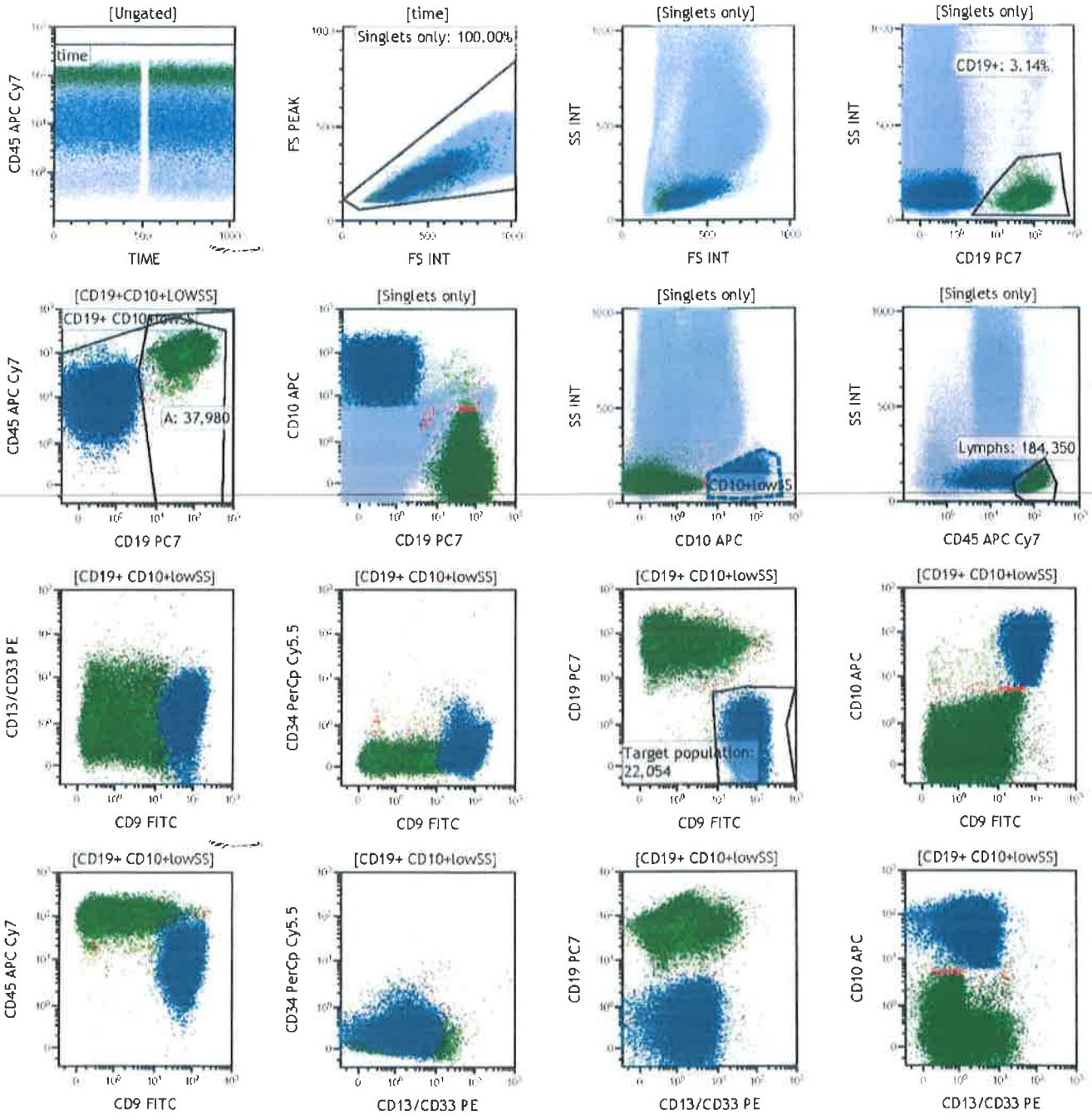
# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

BALL-B 2019 (BALL-06)

CAP 2019

## Day 29 - Tube 2



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# FLOW CYTOMETRY B-ALL MINIMAL RESIDUAL DISEASE

BALL-B 2019 (BALL-06)

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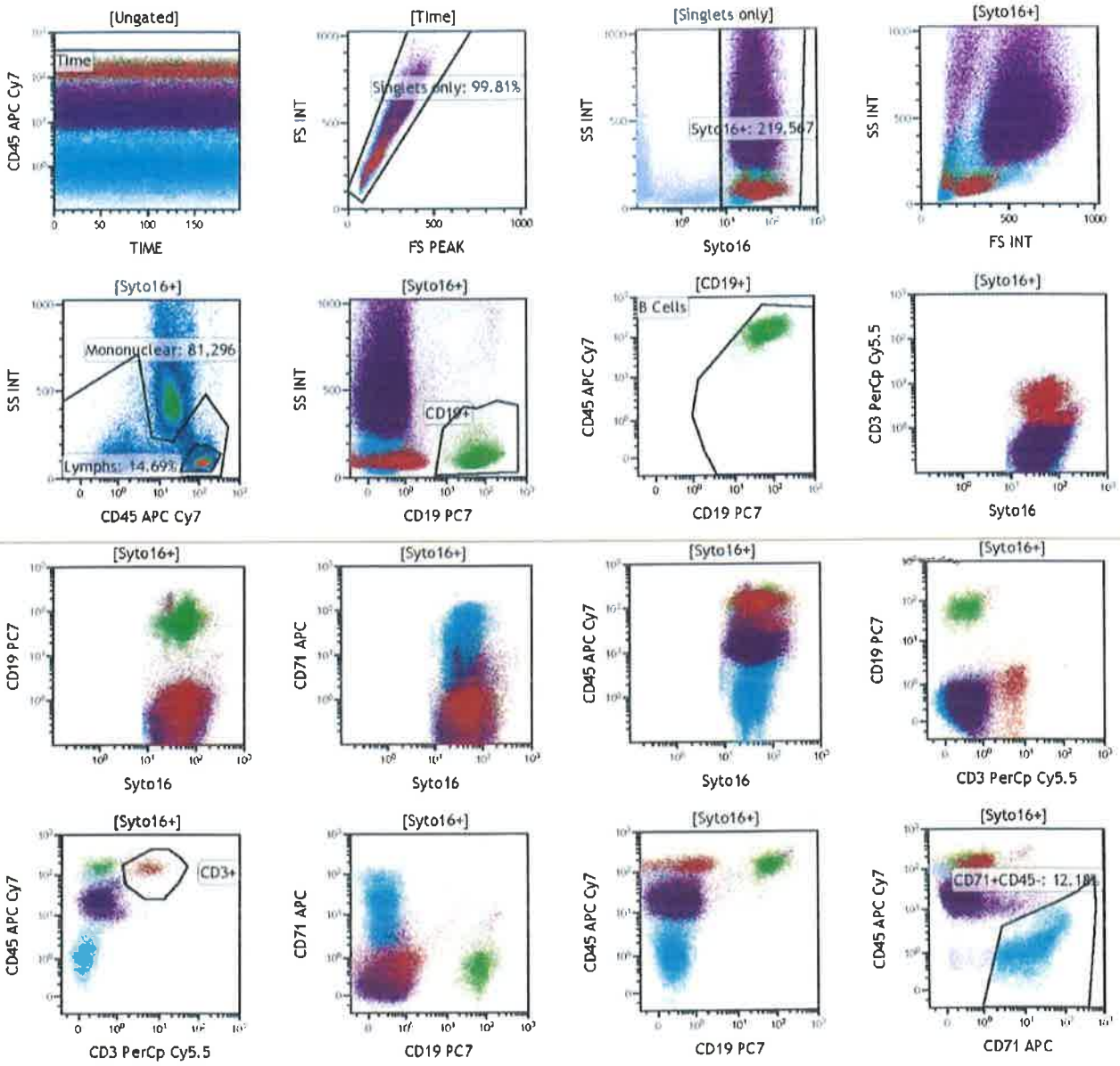
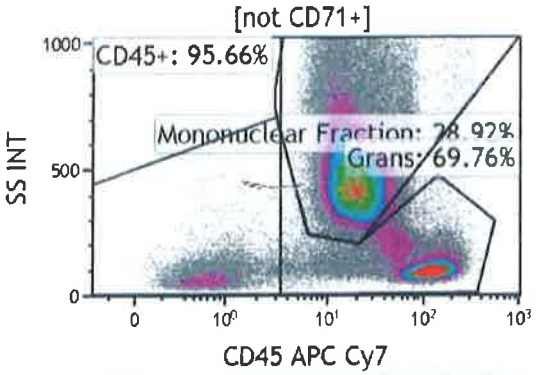
**Day 29 - Tube 3**

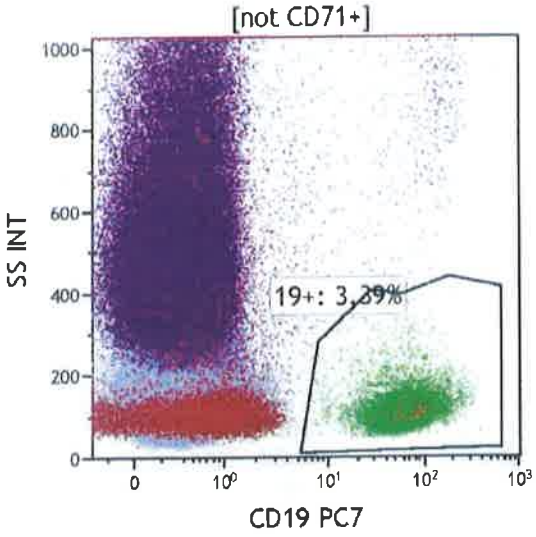



| | |
|---|---|
| Total # | 265,012 |
| [B Cells] Number | 6,491 |
| [Lymphs] Number | 32,259 |
| [Mononuclear] Number | 81,296 |
| [CD3+] Number | 11,320 |

## Day 29 - Tube 3, cont'd




| Gate | Number |
|---|---|
| All | 192,814 |
| CD45+ | 184,440 |
| Grans | 134,512 |
| Mononuclear Fraction | 55,771 |




Differential for reporting

| | |
|---|---|
| Total Lymphocytes | 9.26 % of Total Leukocytes |
| Total Grans | 69.76 % of Total Leukocytes |
| Total of rest | 20.98 % of Total Leukocytes |