

## REVIEWING PATIENT ANTIBODY SCREENING RECORD USING 'Patient Single Antigen Bead Combiner'

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### A Introduction

The Patient\_SAB\_combiner\_0\_9\_december\_2020 program provides a useful tool when reviewing a patient's antibody screening record; it allows analysis of single antigen bead (SAB) results for multiple samples for HLA class I & class II.

Viewing combined results simplifies the process for listing and delisting unacceptable HLA mismatches in highly sensitised patients, reviewing results in an on call scenario and helps base decisions regarding modified acceptance criteria for HLAi transplantation candidates on empirical data.

The Patient\_SAB\_combiner\_0\_9\_december\_2020 was initially called 'anonymiser\_0\_9\_december\_2020' and is therefore referred to by this name in all validation reports.

### B Download of Single Antigen Bead files from Fusion

1. Create folder to hold all SAB files from one patient, using the convention 'SURNAME First name'. Save in an appropriate location:
  - Renal: Z:\Tissue Typing\Renal\Kidney patient antibody assessments and HLAi candidates
  - Cardiothoracic: Z:\Tissue Typing\Thoracic patients (Papworth)\Antibody review
  - Multivisceral: Z:\Tissue Typing\Multivisceral
2. In Fusion 4.4, click 'Reports'
3. Search for patient, e.g. by entering hospital number into 'patient ID' field
4. Enter session date from start of study period of interest or very early date to ensure capture of all patient records, e.g. 01/01/1901
5. Click on '...' next to 'Test Type' field. Within the window that appears, select the 'Single Antigen' box and click 'Ok'
6. Click 'Find'. All SAB files on that patient should appear in the central window.
7. Click 'Speciality' and from the drop-down menu and select "LABScreen – SA MFI"
8. Right click "Include" and "Select All"
9. Click "Export Report"
10. Locate and select the previously created folder (in step 1) where files are to be saved. Click 'Ok'. Click 'Ok' again in response to 'Export completed' message.
11. Repeat steps 9 and 10 again\*. If an 'Export Report' dialogue box appears click 'Yes' to overwrite all existing .csv files.

*\* In the first download, antigen specificities may not be ordered correctly. But the second time the export programme is run, the order is sequential. Beads must be in sequential order because otherwise this would result in incorrect formatting by the Patient\_SAB\_combiner\_0\_9\_december\_2020.*

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### C Combining all SAB files using Patient\_SAB\_combiner\_0\_9\_december\_2020.

- SAB files should be extracted from Fusion into folders for each patient, i.e. all SAB files on that patient in one folder named with patient ID as per section B above.

*Note: a maximum of 200 SAB files can be combined at a time.*

- SAB files should have been checked, ensuring that beads are in correct numerical order and that samples have a 'Sample date'.

*Note: The naming convention of some historical data in Fusion may mean that a 'Sample date' is missing from column E of the programme's 'Results' sheet. These empty cells should be populated by adding dates manually as dates can be inferred from the 'Sample' ID, or a formula can be used to automatically extract data from 'Sample' ID cell if the formatting is consistent.*

*Example formula to use (and modify) if date is in ddmmyy format in first 6 digits of cell:  
=MID(A4,1,2)&"/"&MID(A4,3,2)&"/"&MID(A4,5,2).*

*Alternatively, the missing date can be added in Fusion but the updated file/s will need downloading again from Fusion.*

- Patient\_SAB\_combiner\_0\_9\_december\_2020 (saved to Z:\Tissue Typing\Renal\Kidney patient antibody assessments and HLAi candidates should be saved in the same location as the patient folders (example below).

| <input type="checkbox"/> Name  | Date modified    | Type                  | Size  |
|--|------------------|-----------------------|-------|
|  CAM 123456   | 06/07/2021 16:16 | File folder           |       |
|  CAM 234567   | 06/07/2021 16:16 | File folder           |       |
|  CAM 345678   | 06/07/2021 16:16 | File folder           |       |
| <input checked="" type="checkbox"/>  anonymiser_0_9_december_2020 | 31/12/2020 08:42 | Microsoft Excel 97... | 99 KB |

- Open Patient\_SAB\_combiner\_0\_9\_december\_2020 and click 'Enable content' and then answer 'Yes' to make this file a trusted document.
- Click on the button 'Select folder' and a window will open where the correct patient folder (comprising all patient SAB files) should be selected.
- In column A of the 'Calc' sheet, the SAB files found will be listed, and the total number of files to be analysed will populate the cell next to the 'No of files to process' cell.
- Click on the 'Combine files' button. A loading icon will appear and Excel might flash a few times while the files are being processed, this may take a few seconds/minutes depending on the number of files – do not click anything or try and do anything else on the PC during this time as this may crash the combiner. When the processing is complete, a 'Combining complete' window appears, click 'Ok'.

*Note: If there are any LABscreen mixed files in the folder, a message may appear about no LABScreen files being processed. This is fine and does not affect the combining process.*

- Two extra 'Results' sheets should have appeared at the bottom of the document. One should end with '\_I' and the other '\_II', indicating sheets containing samples tested by SAB I and SAB

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| <p style="text-align: center;"><b>REVIEWING PATIENT ANTIBODY SCREENING RECORD USING<br/>'Patient Single Antigen Bead Combiner'</b></p> |
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It respectively. MFI 'Normal values' will be populated under each bead specificity for each sample tested.

- An MFI value stated as '0' = MFI 0 (nothing detected at this bead)
- An MFI value stated as 'X' = placeholder for absence of this bead (and corresponding specificity) on the lot this sample was tested with. Instead, the alternative bead specificity (if there is one, e.g. DPA1\*02:02, DPB1\*13:01 instead of DPA1\*01:05, DPB1\*13:01) will be at the end of the sheet.

*Note 1: Patient\_SAB\_combiner\_0\_9\_december\_2020 still works when there is just one SAB file to process.*

*Note 2: if a SAB bead lot has two beads with the same specificity (rare – reason unknown why beads are duplicated), the combiner will just select one MFI value from the original data from one of the beads. As the specificity is the same, the MFI values shouldn't vary but this is just something to be aware of.*

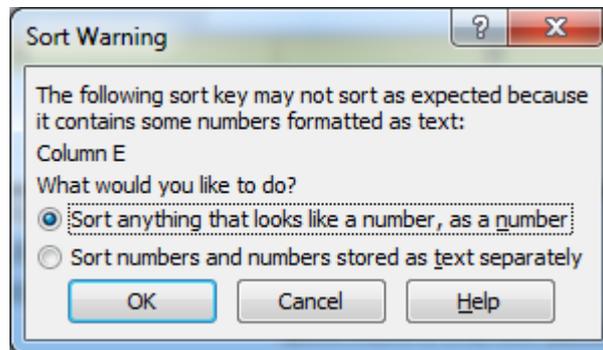
9. Go to 'File' and 'Save as' and save the document to an appropriate location. DO NOT click 'Save' as the patient data would be stored and the Patient\_SAB\_combiner\_0\_9\_december\_2020 would need restoring back to its original blank state.

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### D Organise Data

This needs to be done for each class of antibody.

1. Re-open patient Normalised MFI combiner file & enable contents (should open on the dated results tab).
2. Format cells with MFI data as **Number** and set **Decimal places** to zero. Click OK.
3. For Meditech specimens enter the sample date (identified by TT within the number) - the sample date can be derived directly from the value in the Sample column (B)  
e.g. Sample 040614TT31 was taken on 04/06/2014.
4. Highlight all the data in row 4 to row xx (not rows 1,2,3), on the **Toolbar Data** tab click **Sort** and select:
  - Ensure 'My data has headers' is unchecked.
  - **Sort by** - Column E
  - **Sort on** - Values
  - **Order** = A - Z
  - Click **OK**.
  - Sort Warning appears\*:



- Highlight 'Sort anything that looks like a number, as a number' and click OK
- This sorts the sera by oldest to newest.

\* The warning appears because version **\_0\_9** of the combiner places an apostrophe before the date e.g. **'dd/mm/yyyy**.

5. If any serum dilution results are present cut and paste those rows to the bottom of the list.
6. Highlight all cells containing data using the mouse (you can't use the top left select sheet function) & **Copy**.
  - Open the next empty worksheet within the file, click on cell A1 and paste using **Transpose**.
  - Click **Ok**.
7. Copy and paste patient name into cell A5.
8. Identify and pre & post plasma exchange samples or AO by expanding the columns and checking for PRE, POST or AO after the specimen ID in row 2.
  - If present add to sample date in row 5 e.g. 21/03/2020 PRE or 17/04/20 AO
9. Delete rows 1-4 and adjust column widths as required (width 10 for MFI columns).
10. Highlight all columns and click on the **Toolbar Data** and **Sort**:
  - Tick **my data has headers**:
  - **Sort by** - Select the most recent serum date.
  - **Sort on** - Select Values.
  - **Order** by largest to smallest and Click **OK**.

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11. Highlight the cells containing MFI data & use the Conditional Formatting tool to highlight low, intermediate and high MFI values. From the **Home** tab:
  - Click **Conditional formatting, Highlight cell rules**, in **Greater than** enter **499**, select **Green fill with Dark Green** text and click **OK**.
  - Repeat using value 1999 in **Greater than** and **Yellow Fill with Dark Yellow text**.
  - Repeat using value 4999 in **Greater than** and **Light Red Fill with Dark Red text**.
12. Enter patient MRN to cell A1 and add any additional formatting e.g. screen shots of patient HLA from Epic, previous donor HLA, ETC.
13. Rename tab as '**Analysis**' and save file.
14. When complete move the SAB fusion files into the patient's 'SAB Raw Data' folder (create one if needed).
15. Repeat for second class of antibody if the patient has antibody to HLA class I & II.

NB. If a patient has both HLA class I & II you can copy the analysis sheet from the SAB II combiner & paste into a new sheet in the SAB I combiner spreadsheet for ease of analysis. Just rename the tabs accordingly.

## E Easy way of Identifying Specificities for Listing as Unacceptable Mismatches (UMM)

1. Copy specificities from column B down to agreed cut-off for listing.
2. Paste into new sheet & rename tab as **UMM**.
3. Click on top of column to highlight specificities & then on Data & .
4. Whilst still under the Data screen remove the duplicates, this makes it much easier to enter the UMM & for verification too.

