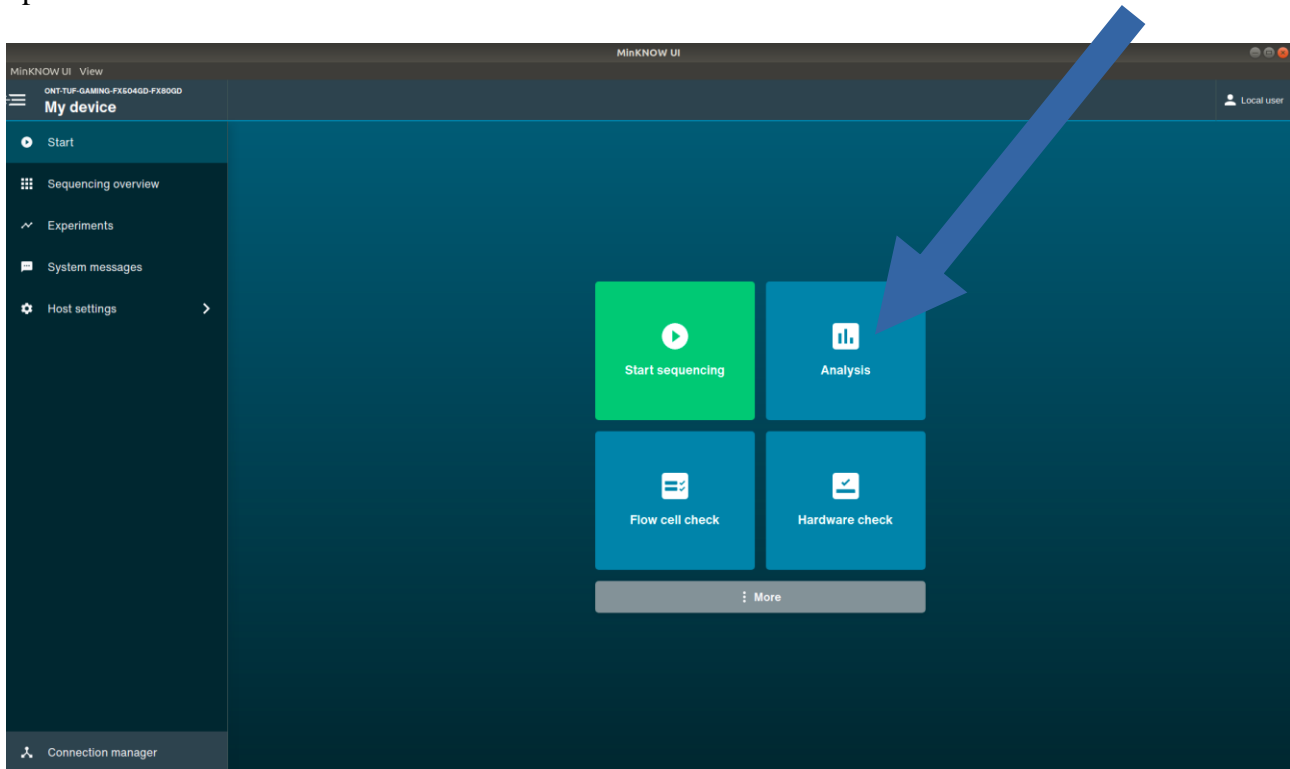


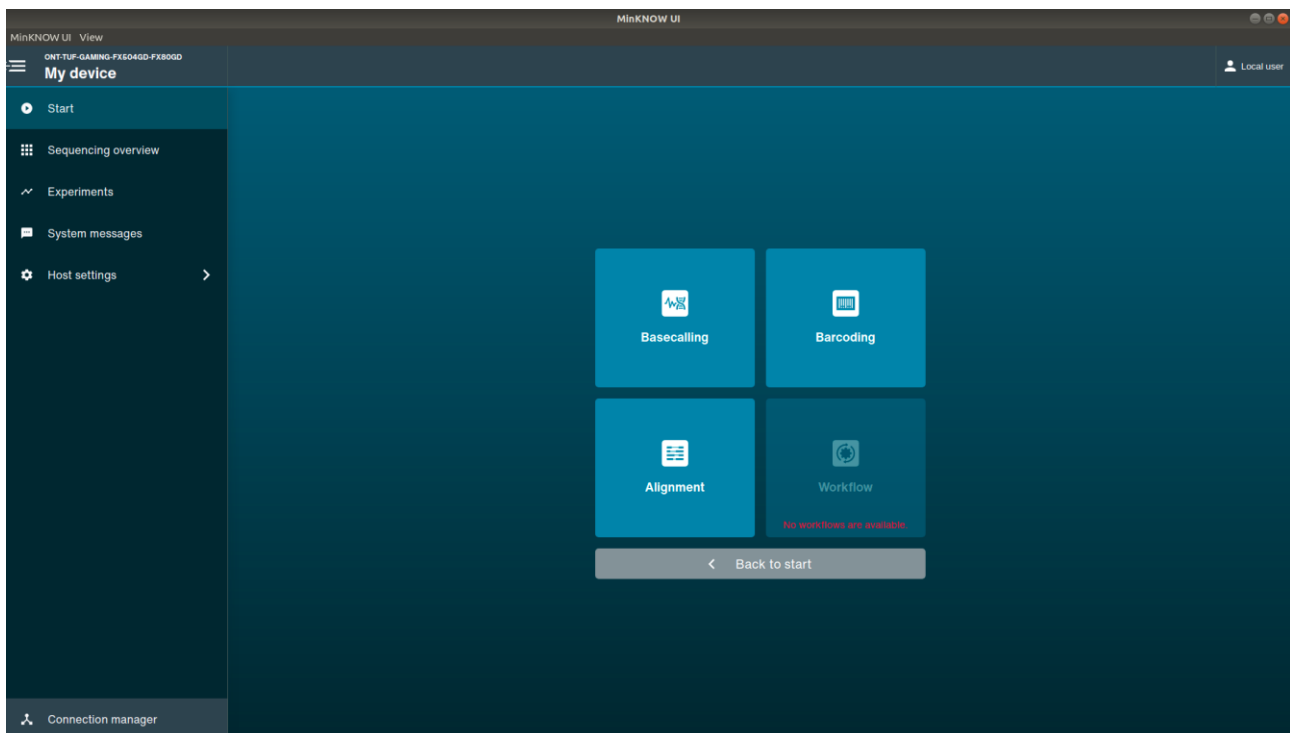
Protocol: How to use Oxford Nanopore MinKNOW API (MinION sequencing)

1) Start Step

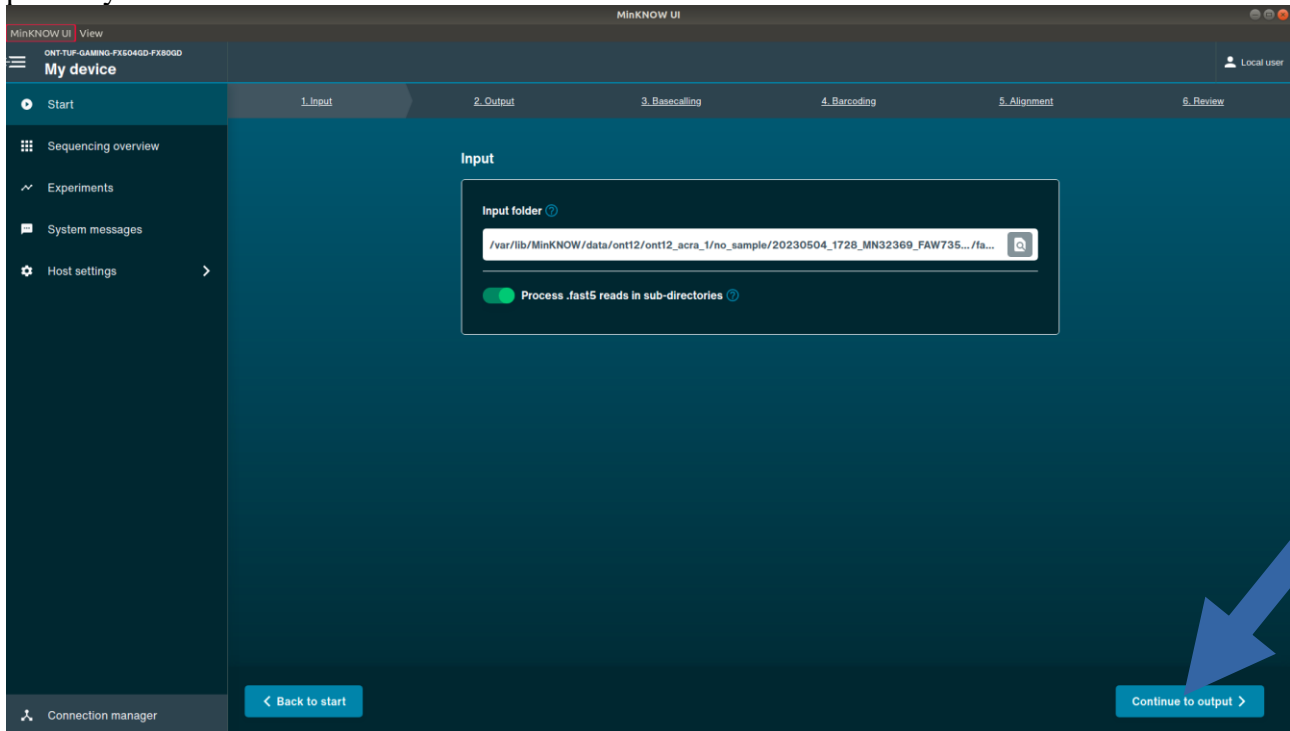
When you are on the “Start” menu of the MinKNOW application, you can select the “Analysis” option.



2) Then you can select either “**Basecalling**” or “**Barcoding**” option from the following frame:

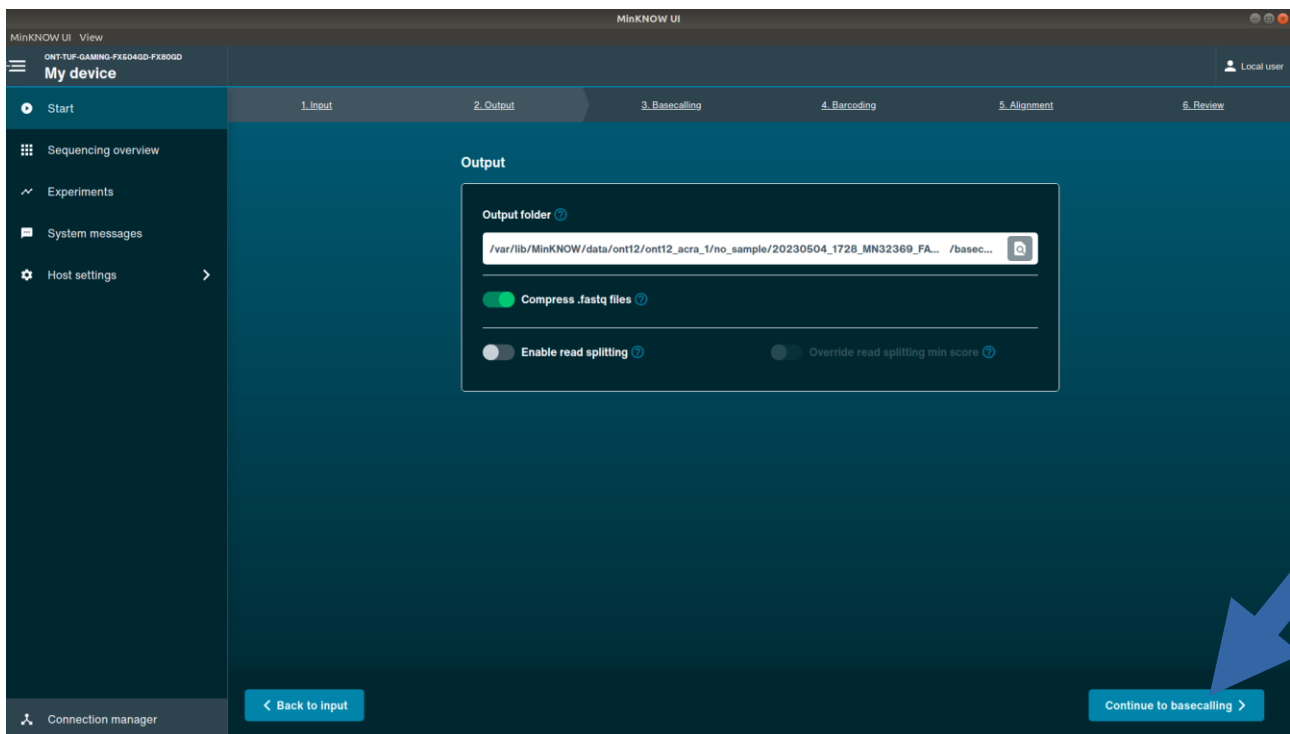


3) When you select the "**Basecalling**" option, you'll be taken to this page, where you can enter the path to your **fast5** folder as follows:



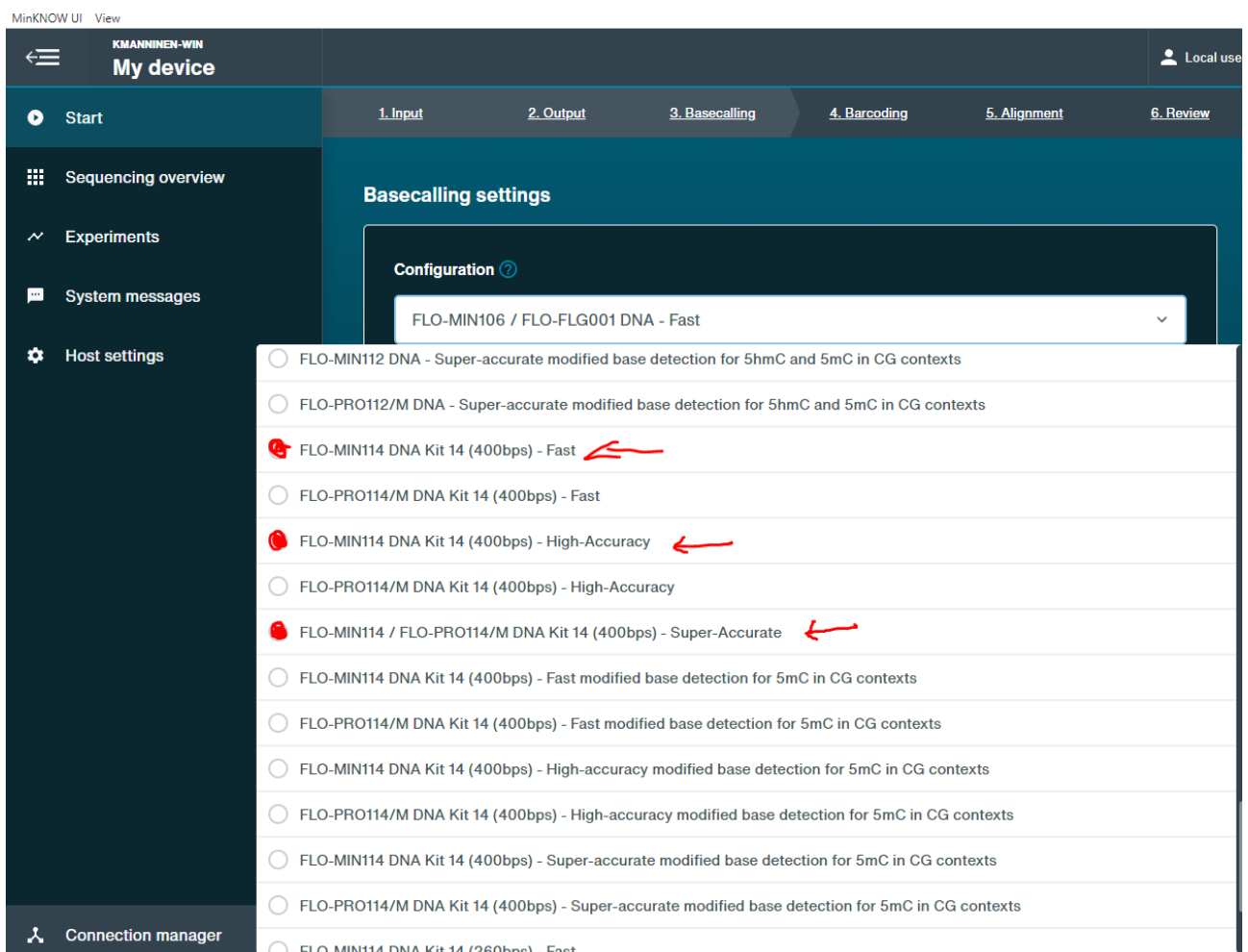
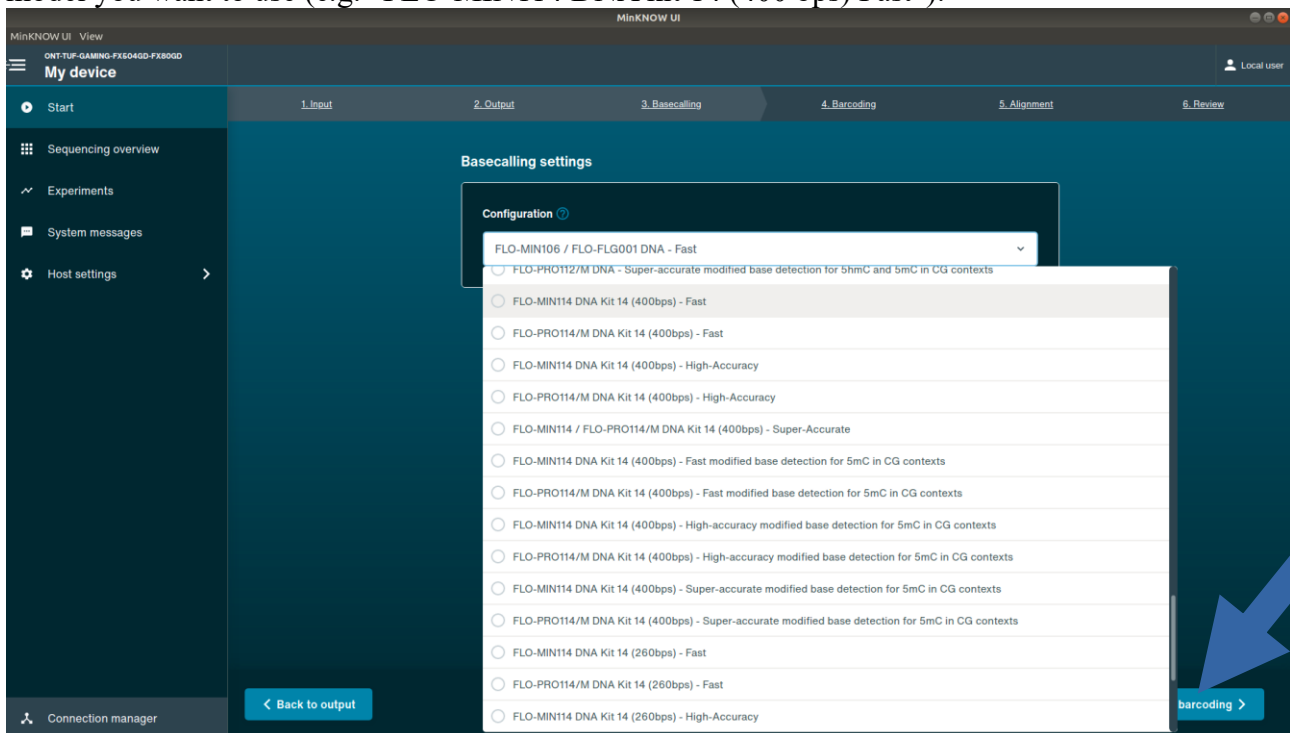
*Please note that the option “Process .fast5 reads in sub-directories” is enabled. Then you should click on “**Continue to output**”.

4) You can then select the output folder (usually named “**basecalling**”), and run the basecalling using default options (with option “**compress .fastq files**” enabled).



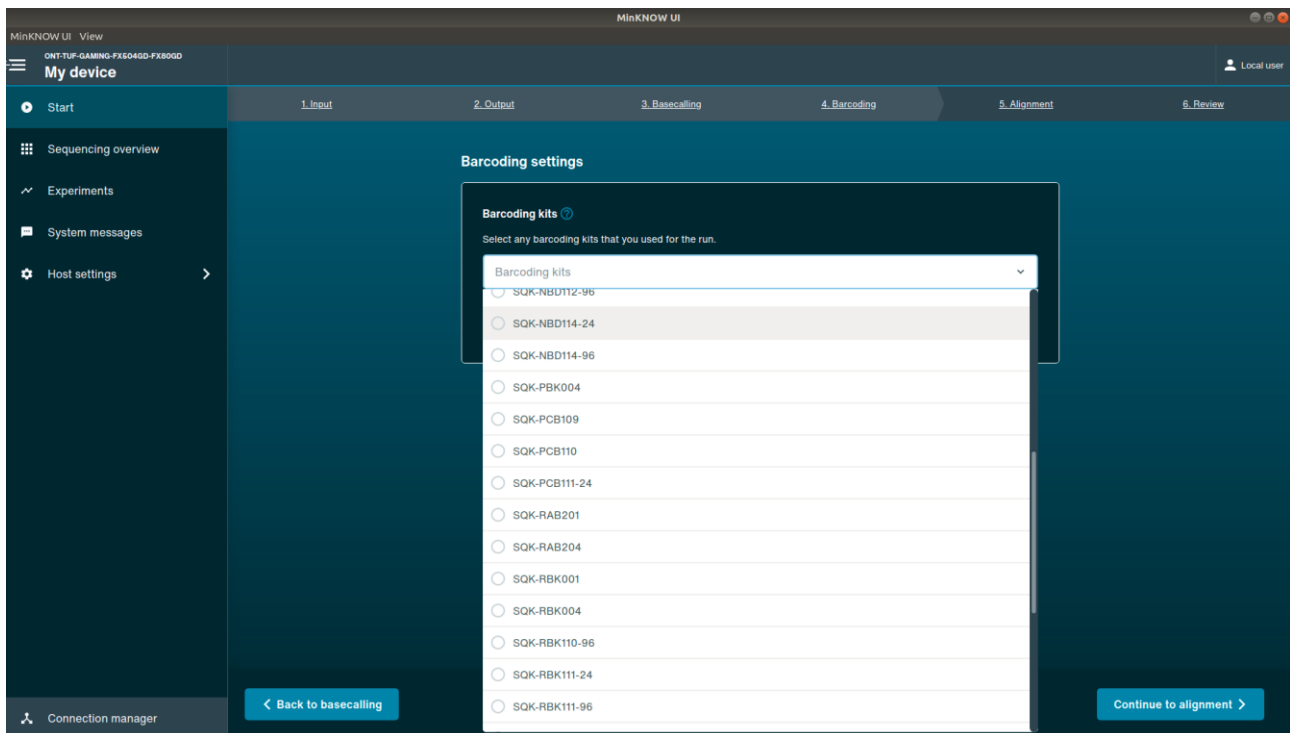
Then you should click on “**Continue to basecalling**”.

5) The next step is to select “**basecalling settings**”. Please select the correct configuration - FLO-MIN114 DNA kit 14 (400 bps) Fast/High-Accuracy/Super-Accurate, and choose the basecalling model you want to use (e.g. “FLO-MIN114 DNA kit 14 (400 bps) Fast”).

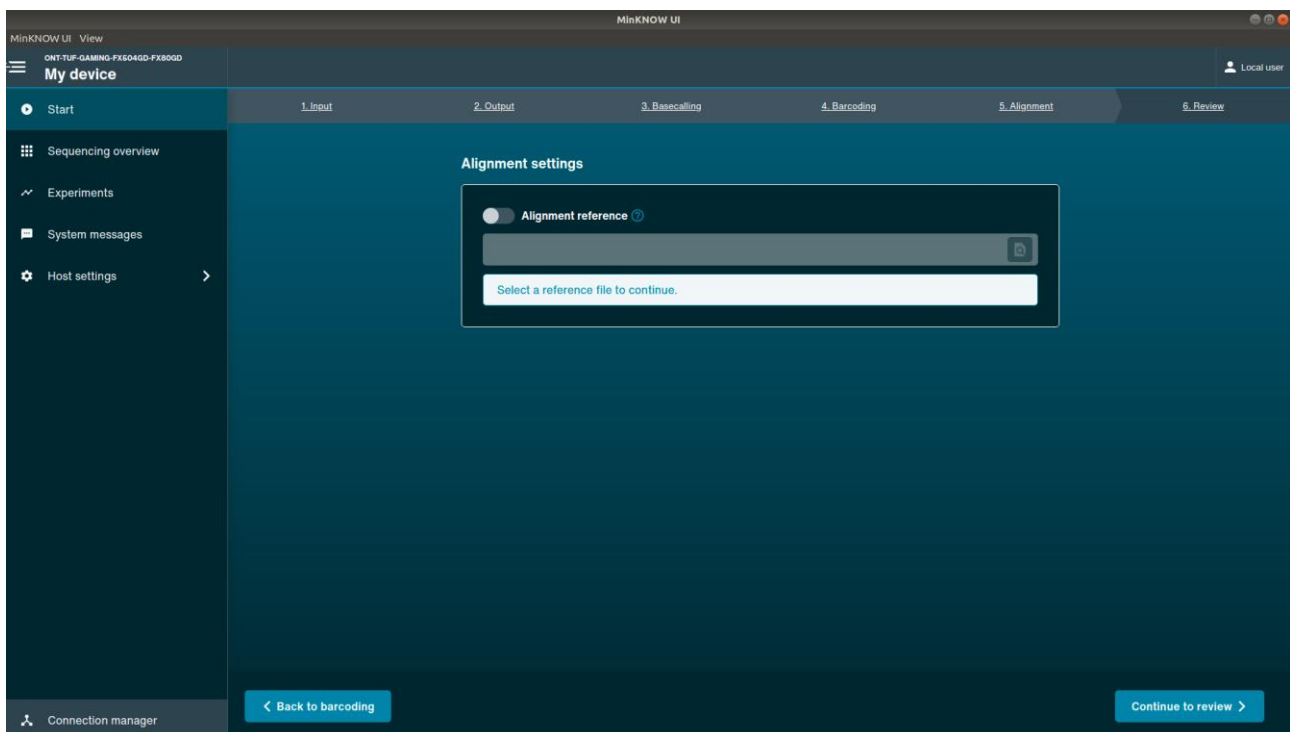


Other possible options (only one option can be selected): Then you should click on “**continue to barcoding**”.

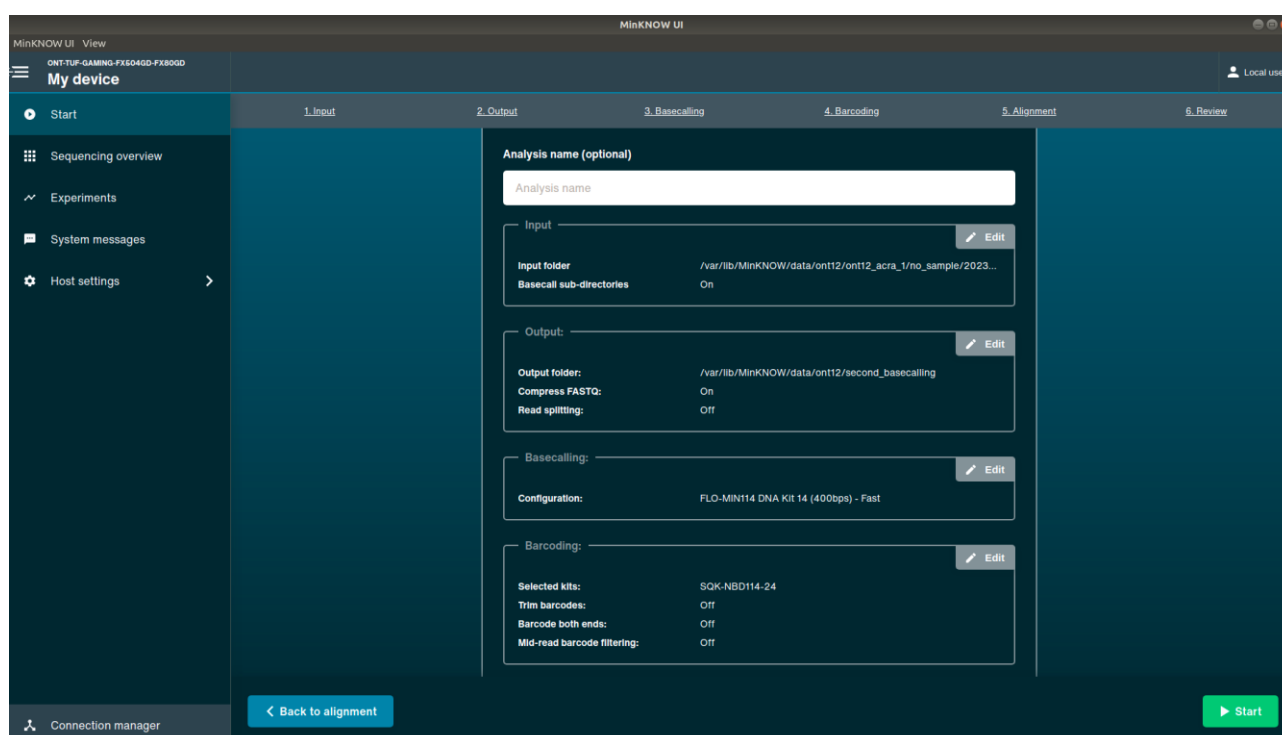
6) The next step is to select “**barcoding settings**” using the “**SQK-NBD114-24**” barcoding kit. Then you can click on “**Continue to alignment**”.



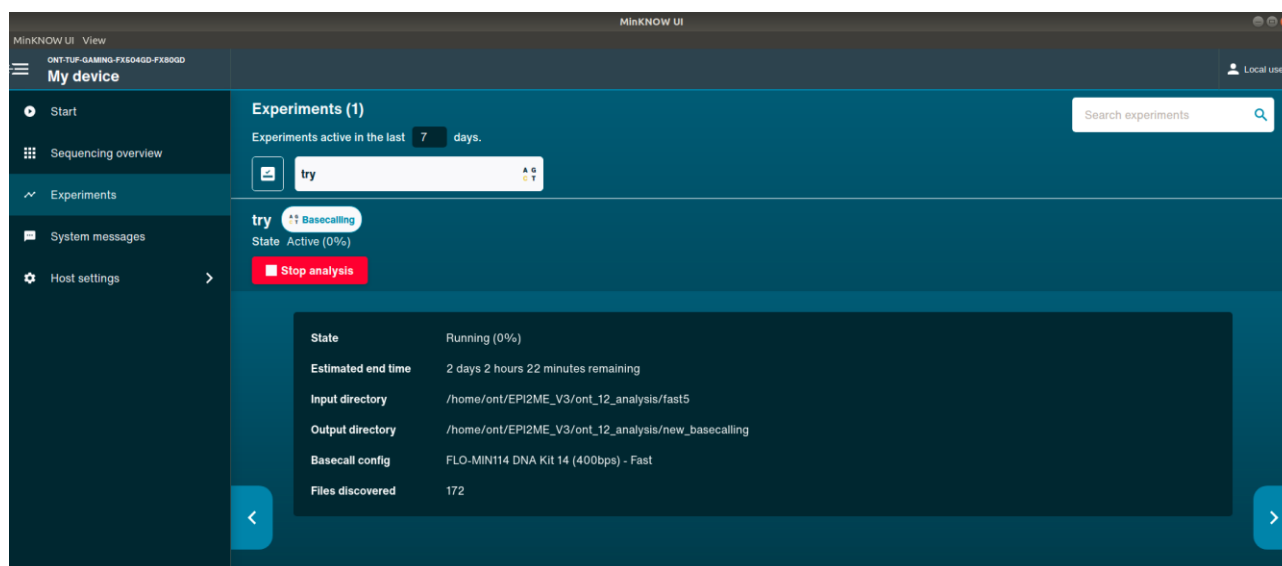
7) You can avoid the next step and click on “**Continue to review**”.



8) After this step, a “**Review**” page will be displayed, and you will be able to “**Start**” the analysis:



9) Finally, you will be prompted to the running page:



Please note that it is also possible to run the “**barcoding**” directly from .fastq files generated by Guppy (or another software). You will just have to start by selecting “**barcoding**” option in step 2. Please note that it is also possible to run the “**barcoding**” directly from .fastq files generated by Guppy (or another software). You will just have to start by selecting “**barcoding**” option in step 2. Note that if you encounter some issues for running the analysis, you can ask a bioinformatician to change permissions for the folder corresponding to the analysis (e.g. “sudo chmod 777 ont12/”). For deeper analyses, several bioinformatics tools could be used (you can also contact the Oxford Nanopore Technologies support team. You can visit the following links for further details:

- <https://github.com/nanoporetech>
- <https://a-slide.github.io/pycoQC/>