

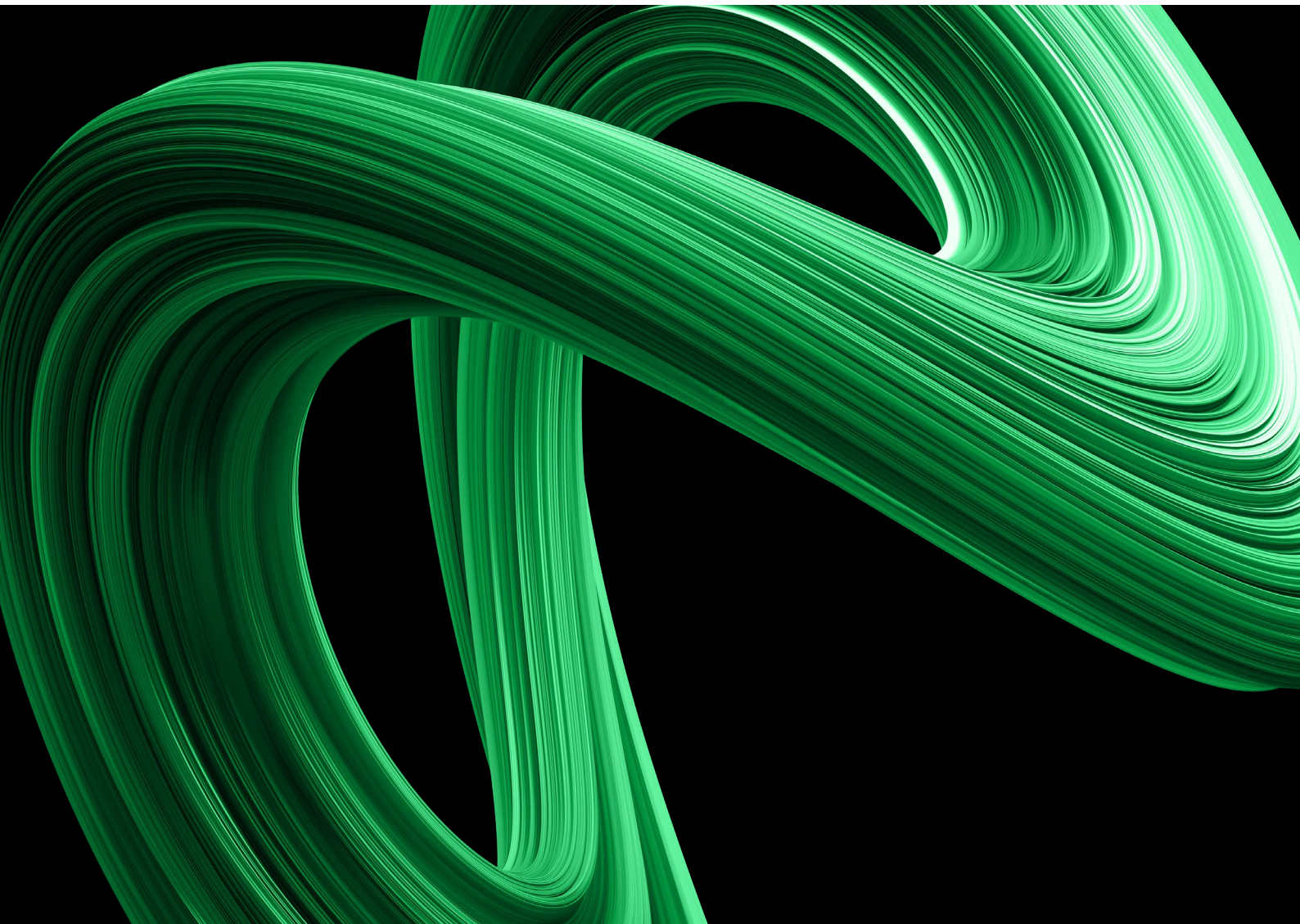


# Myeloid-NDC Assay

*Analysis Guide*

Release: February 2024

For **Research** Use Only. Not for use in diagnostic procedures.





## Myeloid-NDC Assay

This document is intended to guide the users through the analysis of the results generated by the Myeloid-NDC assay and associated bioinformatics analysis application. Although every effort has been made to keep the information correct and up to date, we cannot guarantee that there are no unwilling errors or omissions. We encourage our customers to provide feedback which may lead to modifications and improvements in future versions of this document .

### Warranties

No warranties are provided, explicitly or implied and Univ8 Genomics does not accept any liability for any consequences, direct or indirect arising from the use of the Myeloid-NDC bioinformatics or analytical tools.

### Trademarks

All trademarks are the property of their respective owners.

### Copyright

© 2024 Univ8 Genomics Ltd. All rights reserved.

Univ8® is a Registered Trade Mark of Univ8 Genomics Limited.



# Table of Contents

<b>1. Introduction</b>	<b>4</b>
Intended Use	4
Accessing the Myeloid-NDC analysis application	4-5
<b>2. File Descriptions</b>	<b>6</b>
Myeloid-NDC analysis output files	6
Overview of the Myeloid-NDC 'gathered' files	6-7
<b>3. Analysis</b>	<b>8</b>
Sequence variant (VAR) analysis	8
Structural variant (SV) analysis	8-9
Copy number variant (CNV) analysis	9
Analysis tips	10
Example data files	11-12
<b>4. Supporting Information</b>	<b>13</b>
Abbreviations	13
References	13
Revision History	13



# Introduction

## Intended Use

The Myeloid-NDC assay is a capture-based targeted next generation sequencing (NGS) laboratory assay combined with a customised bioinformatics analysis application for research use only (RUO). It is intended for the integrated and qualitative detection of chromosomal translocations, copy number variants (CNV) and sequence variants from suspected or confirmed cases of myeloid malignancies. The Myeloid-NDC assay has been validated for use with high molecular weight (HMW) genomic DNA (gDNA).

The Myeloid-NDC assay has been designed as an integrated tool covering most genomic alterations recommended for the analysis of acute myeloid leukaemias (AML), myelodysplastic syndromes (MDS) and myeloproliferative neoplasms (MPN)<sup>1-4</sup>. The Myeloid-NDC analysis application provides a simple solution to allow laboratories to produce integrated reports according to the newest guidelines and recommendations.

The Myeloid-NDC assay utilises the same technology and is fully compatible with the EuroClonality-NDC assay, which had undergone a large multi-centre validation across seven European laboratories<sup>5</sup>. This means that laboratories can now process all their suspected haematological malignancy samples at the same time using a universal protocol, reducing labour costs, increasing economy of scales, and speeding turnaround times to produce fully integrated genomic reports.

## Accessing the Myeloid-NDC analysis application

The "Myeloid-NDC analysis" application is available on Illumina's BaseSpace Sequence Hub (BSSH) exclusively for Myeloid-NDC customers. BSSH provides a secure portal for genomic data analysis and storage, with servers across multiple locations to support the requirements of GDPR, CLIA, HIPPA, multiple ISO standards and others. For more information, please refer to Illumina's BSSH help centre on the following link (<https://help.basespace.illumina.com>). For a detailed description of the security and compliance features of Illumina's BSSH, please follow these links ([https://www.illumina.com/content/dam/illumina-marketing/documents/products/flyers/7139\\_AWS\\_Illumina\\_Solution%20Brief\\_SecurityCompliance\\_FINAL\\_20190405.pdf](https://www.illumina.com/content/dam/illumina-marketing/documents/products/flyers/7139_AWS_Illumina_Solution%20Brief_SecurityCompliance_FINAL_20190405.pdf)), (<https://emea.illumina.com/content/dam/>

[illumina/qcs/assembled-assets/marketing-literature/basespace-security-and-privacy-security-brief-m-gl-01959/basespace-security-and-privacy-security-brief-m-gl-01959.pdf](https://www.illumina.com/content/dam/illumina-marketing/documents/products/flyers/7139_AWS_Illumina_Solution%20Brief_SecurityCompliance_FINAL_20190405.pdf)).

To use the Myeloid-NDC analysis application you will first need to contact your Illumina local representative to open a Professional BSSH account for your team. This account will provide iCredits needed to run the Myeloid-NDC analysis application. The computing cost of running the application is 3 iCredits per node/hour; this corresponds approximately to 0.5 iCredits per sample. A BSSH Professional account allows teams to manage access for their members and collaborators, in a secure and controlled fashion. Moreover, access can be customised to different levels for administrators, power users, curators or basic users, helping to implement local security policies as appropriate. The BSSH Professional accounts can be set up in different domains (e.g. EU, US, AUS, etc.), to ensure country-specific data protection compliance (e.g. GDPR), each of them with one "Working Group" (e.g. *A-Team EU*).

Once the BSSH Professional account has been set up, please follow the instructions below to access and run the Myeloid-NDC application:

1. Log onto your regional domain for your team's specific BSSH Professional account with your personal account's username and password (e.g. for EU use <https://eucl.sh.basespace.illumina.com>).
2. On the top right corner, click on your username and select the Working group for your Professional account (e.g. *A-Team EU*).
3. From the top navigation menu, go to "APPS".
4. From the drop-down menu select *All Categories* or *Targeted Sequencing*.
5. On the "Search Apps" field, enter *Univ8* and click the search icon to see all the Univ8 Genomics apps.
6. Click to select the *Myeloid-NDC Analysis App*. **Note:** you can bookmark all the Univ8 Apps for easier access in the future.



7. Launch the *Myeloid-NDC Analysis* App by clicking on the "Launch Application" button.
8. Select the "Samples" option if you want to run samples from a single run (recommended and most common) or "Biosamples" option if you have run the same sample in multiple runs and wish to aggregate the data (not recommended).
9. Provide an *Analysis Name* (or use the generic default with datestamp).
10. Click on 'SELECT SAMPLE(S)' and choose which samples to run by ticking the corresponding boxes and then click "SELECT" to exit. **Note:** you can filter by "Project" to help with sample selection.
11. Click on "SELECT PROJECT" and select an existing project or enter new one.
12. After checking that everything is correct, click on "LAUNCH APPLICATION".
13. The *Myeloid-NDC App* will be launched, and the samples will be run, wait until analysis has completed, approximately 6-10 minutes per sample (an email will be sent to you or the nominated admin account).
14. Download the data to your local server/computer by going to the corresponding "ANALYSIS", clicking on the "File" icon and selecting "DOWNLOAD">"ANALYSIS".
15. Make sure you install the "BaseSpace Sequence Hub Downloader" the first time (or when a new version is available) and then select "All file types including VCF, BAM, & FASTQ" and click "DOWNLOAD".
16. Select the local directory where you want to download the data into and continue.
17. Wait until the download has finished and check for integrity of the data in the corresponding directory.

Users must note that the FASTQ files will not be downloaded as part of the above download instructions and, if the users wish to keep these archived locally, they will need to download from the corresponding area (e.g. from "ANALYSIS" if they have been generated using Illumina's FASTQ generation App).

No data is ever transmitted to or accessible by Univ8 Genomics Ltd. or any third party, and the users are always in full control of their data, which they can choose to store, archive and/or delete from Illumina's BSSH at any time to comply with local security policies. Also note that storing data on Illumina's BSSH has a minimal cost overhead, but this can become significant if large data are stored for long periods of time (please contact Illumina support for detailed and up to date information).

Users must note that Univ8 Genomics Ltd does not warrant in any way, either explicitly or implied, the regulatory security compliance of the data transmitted or stored using the BSSH platform, and Univ8 Genomics Ltd does not accept any liability for any consequences, direct or indirect, arising from the use of the BSSH platform.





## File Descriptions

### Myeloid-NDC analysis output files

The downloaded directory will contain one folder per sample with a series of outputs (file name/extensions in bold):

**gathered.tsv**: this is the main output for analysis of the Myeloid-NDC assay and shows the gathered results for sequence variants (VAR), structural variants (SV) and copy number variants (CNV) in a single integrated file. Files are available in tab-separated-value format (.tsv).

**summaryQC.tsv**: summary metrics per sample, including: total "passed filter" aligned reads, percentage of duplicate reads, mean target unique coverage, percentage of target bases covered at 0x, 100x, 200x, 300x and 500x, percentage of bases on or near target and median insert size.

**bam & bai**: alignment files and their indices required to visualise the data on any genome viewer such as IGV.

**VAR.complete.vcf**: VCF file (v4.3) containing the complete list of sequence variants (VAR) and corresponding annotation. All sequence variants are included here (after removing known artefacts), irrespective of location, functionality, or population frequency.

**VAR.curated.vcf**: VCF file (v4.3) list of sequence variants curated to remove artefacts, most non-functional variants as well as most of the common polymorphic variants and synonymous variants.

**CNV\_plot.html**: these plots include two panels, the top one shows the CNV values for a selection of the main cytogenetic abnormalities; the bottom panel shows the CNV values for all genes/regions included in the Myeloid-NDC assay (these are coloured for ease of use, see below)

### Overview of the Myeloid-NDC 'gathered' files

These files are the main output of the Myeloid-NDC assay and contain information on all events, including sequence, structural and copy number variants. The following data items (see next page) are included in the 'gathered' files:

*Shaded rows indicate the value may have different meaning, depending on the type of variant. \*When analysing sequence variants (VAR) it provides a combination of overall quality (e.g. PASS, f0.02, etc) and whether the variant has been classified as 'curated' or 'complete' (see Analysis section); \*\*Male samples with loss of Y may wrongly be classed as "Female"*



name	VAR example	SV example	CNV example	description
sampleID	sample1_S1	sample2_S2	sample3_S3	sample name as per FASTQ file analysed
sex_calc	Male	Female	Female	expected sex based on the X and Y coverage distribution**
total_reads	11601428	15239996	13785080	total aligned reads passing filter in FASTQ file
pct_duplicates	19.76	21.33	20.55	percentage of duplicated reads
pct_on_target	80.32	80.36	80.48	percentage of bases on or near target
mean_unique_DP	714	942	863	mean unique depth coverage (de-duplicated)
pct_target_bases_0X	0.26	0.78	0.71	percentage of bases covered at 0x
pct_target_bases_100X	99.49	99.08	99.06	percentage of bases covered at 100x
pct_target_bases_200X	98.89	98.88	98.81	percentage of bases covered at 200x
pct_target_bases_300X	97.03	98.27	97.95	percentage of bases covered at 300x
pct_target_bases_500X	78.78	92.87	89.56	percentage of bases covered at 500x
varType	VAR	SV	CNV	type of variant, either sequence (VAR), structural (SV) or copy number (CNV)
quality*	PASS;curatedMyeNDC	HIGH	HIGH	variant quality annotation; for SV and CNV, 'HIGH', 'LOW' and 'FAIL' indicate likelihood of the event being true
chr	chr15	chr22	chr7	chromosome where variant is located
location	90088606	23232388-23232389	50319047-50400635	chromosomal coordinate of the variant
ref	C	G	.	reference genome sequence at location
alt	T	G[chr9:130829912[	.	alternative genomic sequence at location
logR	.	.	0.57	log ratio value (only for CNV)
DPalt	38	307	.	number of alternative/variant reads
DPtotal	797	683	.	total reads at variant position (depth of coverage)
VAF_%	4.76	44.95	99	variant allele frequency (% values for VAR and SV) or quality score for CNV
Gene	IDH2	BCR	IKZF1	gene affected (or closest gene if intergenic)
HGVSc	c.515G>A	.	.	nucleotide sequence nomenclature by HGVS (VAR)
HGVSp	p.Arg172Lys	.	.	protein sequence nomenclature by HGVS (VAR)
variant_class	SNV	BND	.	variant class (e.g. SNV, INV, DEL, INS, DUP, BND, etc)
consequence	missense_variant	BCR & ABL1	CNV_deletion	consequence (VAR and CNV) or genes involved (SV)
impact	MODERATE	.	.	biological impact of VAR (VAR)
transcriptID	ENST00000330062	.	.	ensembl transcript ID (VAR)
MANE_Select	NM_002168.4	.	.	MANE Select transcript ID (VAR)
MANE_PlusClin	-	.	.	MANE Plus Clinical transcript ID (where available) (VAR)
gnomadV3_AF	-	.	.	population frequency in gnomAD v3 (%) (VAR)
clinSig	pathogenic, likely_pathogenic	.	.	clinical significance of the variant according to ClinVar (where available) (VAR)
clinvarID	375987	.	.	variant ID in ClinVar (where available) (VAR)
cosmicID	COSV57468734	.	.	genomic mutation ID in COSMIC (where available) (VAR)
cosmicLID	COSM33733	.	.	variant legacy ID in COSMIC (where available) (VAR)
SIFT	deleterious(0)	.	.	predicted functional consequence by SIFT (VAR)
PolyPhen	damaging(0.998)	.	.	predicted functional consequence by PolyPhen (VAR)
partner_nearest_GENE	.	ABL1	.	partner gene (or nearest gene if intergenic) (SV)
SV_len	.	.	.	length of the SV (for INV, DEL and DUP) (SV)
INS_seq	.	.	.	sequence of the insertion (INS SV only)
INS_Len	.	0	.	length of the insertion (INS SV only)



# Analysis

## Sequence variant (VAR) analysis

There are two main ways of analysing sequence variants with the Myeloid-NDC assay, the recommended option is to analyse all variants in an integrated fashion using the 'gathered' files, however you can also analyse sequence variants only using the provided VCF files. There are two VCF v4.3 format files, one labelled 'complete' containing all variants (after removal of potential artefacts), and the other labelled 'curated' containing the more functionally relevant variants (see above under 'output files' section). Both are annotated using VEP on hg38 (ensembl.org) and include the MANE Select transcript (or MANE Plus Clinical) when available.

All variants included in the 'complete' VCF file are included in the 'gathered' files, and there is a flag under the 'quality' column that allows users to filter out 'complete' variants (e.g. if using Microsoft Excel, select the filter tool and filter by 'does not contain' and enter 'complete'), leaving only the 'curated' sequence variants and all the other structural and copy number variants. The variants with the 'curated' flag represent the most likely relevant variants for most analyses, including those that are functional, non-synonymous and present at low allele-frequency in the global population. Conversely, the variants with the 'complete' flag represent non-functional (or at least not clearly functional, such as intronic, regulatory, etc.), and/or synonymous and/or highly polymorphic variants. While the 'curated' flag is recommended for most applications, the 'complete' flag can be used for specific applications or troubleshooting requiring 'identification', such as investigating contaminations, chimerism, or MRD and cfDNA analysis. The 'curated' variants include splice donor/acceptor sites within 2bp of the exon. If you need to analyse wider splice regions, then use the 'complete' list. Note that the 'complete' VCF files contains all variants, while the 'complete' flag in the 'gathered' files contain only the non-'curated' variants.

Variants are reported here down to > 1% variant allele frequency (VAF), although the Myeloid-NDC assay has been validated to report variants at > 4% VAF. Variants present at  $\leq$  4% VAF (or close to 4% VAF) can be the result of background noise and should not be analysed routinely. Nonetheless, the file provides these low-level variants for users to consider when expecting low-level hotspot variants (e.g. *JAK2* p. V617F) or when analysing follow-up samples with known expected variants.

As part of the annotation of the variants, the 'gathered' files include ClinVar and COSMIC entries with the corresponding clinical significance (for ClinVar entries). Note that occasionally, the entry identifier may be provided for a similar variant when a full match is not found (e.g. cases of indels with incorrect HGVS nomenclature or rare variants). Additionally, the population allele frequency is given from gnomAD v3 in percentage values to help identify potential rare polymorphic variants (<0.2% in the global population). It is important to note that a substantial number of hotspot pathogenic variants reported by this assay can have significant frequency in gnomAD due to underlying clonal haematopoiesis or undiagnosed myeloid disorders in the overall population (e.g. *JAK2* p.V617F is present in ~ 0.07% of individuals in gnomAD v3) as well as germline carriers.

## Structural variant (SV) analysis

Only structural variants (SV) that are likely to correspond to real genomic events according to our algorithms are reported. In most cases of myeloid malignancies with substantial neoplastic DNA content, any SV found will be reported with the 'HIGH' quality flag. Occasionally some SV events may be reported with the 'LOW' quality flag, these could represent either genuine clonal SV present at low level in the sample (e.g. low neoplastic DNA content or subclonal SV) or they may be artefactual variants that the user should be able to confirm visually with the .bam files. If a SV containing the 'LOW' flag is present and such variant is not a known or common SV in myeloid malignancies, then it is likely to be an artefact or a 'passenger' event.

The presence of genuine SV can be confirmed visually using IGV or any other genome visualisation tool and we recommend performing this step in all instances. It can also help ascertain the exact makeup of the breakpoint and whether it is a balanced translocation or not. The main limitation of SV analysis is the capture of at least one of the regions involved in the translocation. In particular, the Myeloid-NDC assay can only detect SV in the regions described in the Quick Reference Guide. Additionally, there are areas that are not fully covered or where the breakpoints may fall outside of the regions covered; for example up to 10-15% of *KMT2A* breakpoints or up to 5-10% of *BCR::ABL1* translocations (particularly rare breakpoints) could be potentially missed. On the other hand, since this assay does





not require growing metaphases and has a very low limit of detection, it can detect up to 10-15% more translocations than conventional cytogenetics and FISH.

A key strength of the integrated analysis provided by the Myeloid-NDC assay is that analysis of SV can be complementary to the analysis of sequence variants and copy number variants in some instances. For example, large insertions/duplications, such as *FLT3*-ITD will be easily detected as a SV (reported as 'FLT3 & FLT3'), even if it has not been reported as a sequence variant (VAR), and similarly for large deletions. In relation to copy number, some common variants such as *KMT2A*-PTD can be reported as a SV (if at least one breakpoint of the duplication falls within the captured regions of *KMT2A*) in addition to the corresponding CNV. In fact, due to the lower limit of detection for SV, the sensitivity for such variants is significantly improved by the SV reporting.

### Copy number variant (CNV) analysis

For each sample, there is a CNV plot in HTML format, which should be always used to ascertain the quality of the CNV calls and confirm the CNV data. The plots highlight consecutive chromosomes in different colours and, where applicable, the short (p) and long (q) arms of each chromosome are highlighted in different shades of the same colour, to help identify the location of each alteration and whether consecutive gain/losses represent a whole arm event (e.g. deletion of the whole 5q arm or just *EGRI* and/or *PDGFRB*).

Regions with no evidence of copy number alterations are dotted in grey, where altered regions are shown as dots in different colours, according to the likelihood of CNV being present: green are regions that may be deleted ( $\log R: 0.7-0.8$ ), blue regions are likely deleted ( $\log R < 0.7$ ), orange represent potential gains ( $\log R: 1.3-2$ ) and red are areas likely to be amplified ( $\log R > 2$ ). To make analysis easier, by hovering over the grey/colour dots for a particular region/gene, the user will see a pop-up box (coloured according to the CNV call) with magnified information containing the chromosomal region (and gene for bottom panel) and the  $\log R$  value.

Similar to SV analysis, CNV events reported in the gathered file can have different quality flags, including 'HIGH', 'LOW' and 'FAIL'. These flags give an indication of the total number of CNV events in both panels of the CNV plot. If the number of events is relatively small, the assumption is that those CNV calls will be of 'HIGH' quality. Conversely, if the number of CNV events is large, they will be assumed to be of 'LOW' quality or even 'FAIL'. Note that the user should always review the CNV plots in the html file to review the quality of the CNV analysis and calls. For example, samples with complex karyotypes may lead to many CNV calls of genuine high quality when reviewing the plot, yet they will appear as 'LOW' or even 'FAIL' in the 'gathered' file due to the unusually

high number of events. On the other hand, a normal CNV profile can be "noisy", resulting in a few artefactual copy number calls made by the algorithm, which they will be flagged as 'HIGH' in the 'gathered' file due to the low number of CNV events.

The performance of the CNV analysis using the Myeloid-NDC assay is higher on samples with neoplastic DNA content higher than 40%. The main limitations of the CNV analysis are: i) they have only been validated in HMW-DNA; ii) they can be highly affected by DNA quality and purity (ddH<sub>2</sub>O elution is strongly recommended); iii) CNV analysis is significantly affected by hyperdiploidy and CNV data should not be analysed with this assay in samples with >20 regions altered; iv) under- or over-clustering, as well as read length different to 2 x 75bp, can have a negative effect in CNV analysis (e.g. if using significantly more or less samples than recommended or different sequencing instruments and strategies).



## Analysis tips

**VCF compatibility:** the VCF files contain only sequence variant information in v4.3 format.

**Integrated analysis:** the 'gathered' files contain all the results for sequence, structural and copy number variants, making it unique in its ability to support users performing an integrated analysis of the genomic data. Users can filter and sort by any given header, to simplify and speed up analysis, according to their needs. It is extremely helpful to see all the types of variants together, to have a holistic view of the entire genomic profile of a sample. Due to the large number of individual events, this is best achieved by following two simple filtering steps:

- Filter the 'quality' column to omit any variant that contains the keyword "complete"; this will show only 'curated' VAR, without filtering out SV/CNV events. Note: if using Microsoft Excel, use the filter tool and filter by 'does not contain' and enter the word "complete".
- Filter the 'VAF\_%' column to include values 'Greater than' and enter the number "3"; this will show VAR and SV with VAF > 3% and 'HIGH' and 'LOW' CNV events. It is important to remember that the recommended limit of detection of the assay is 4%, and therefore a significant number of artefacts may appear below that level. Note: if you select instead 'Greater or equal than' "3", then the 'FAIL' CNV events will also be reported, as they are allocated a value of "3" by the application to support users that have optimised the CNV outputs.

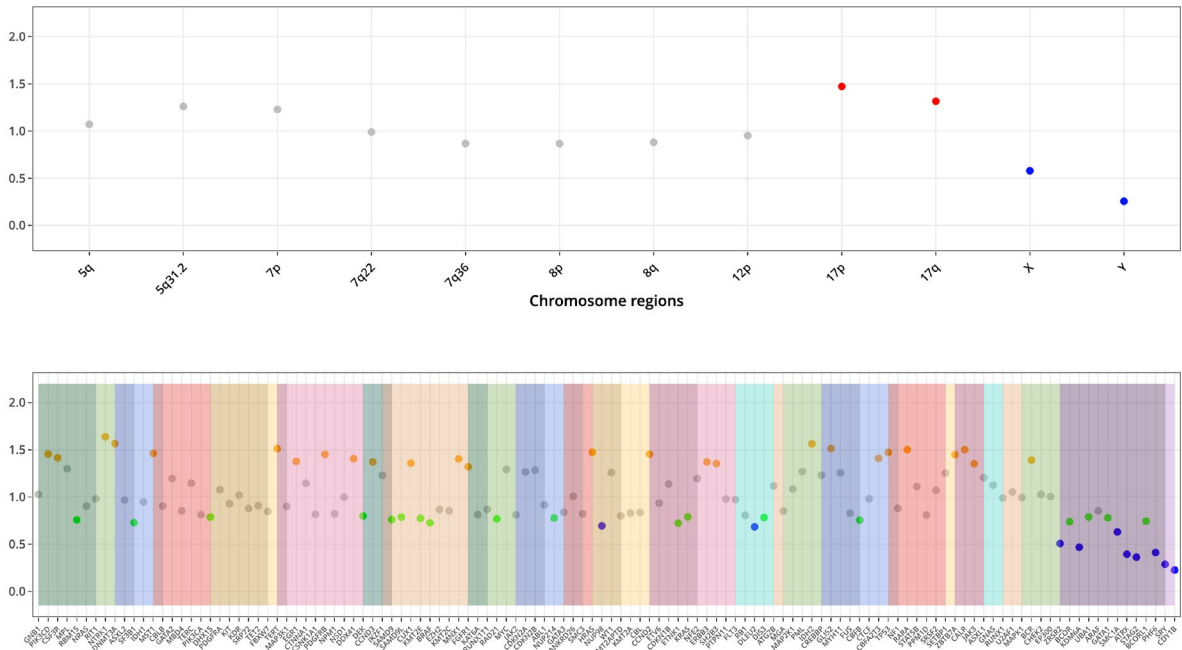
**False positive and false negative VAR and SV results:** to achieve the best balance between sensitivity and specificity it is not possible to eliminate entirely all false positive/negative results. For VAR and SV analysis, when detected at low VAF values, we recommend focussing only on those 'known' or 'hotspot' variants to reduce false positive findings. In fact, for known hotspot variants (e.g. *JAK2* p.V617F, *KIT* p.D816V, *FLT3*-ITD, etc.), the 'VAF\_%' filter may be removed to show variants > 1% VAF.

**Analysis of CNV:** CNV analysis is the part of the Myeloid-NDC assay that is most susceptible to performance issues related to DNA quality, elution in ultra-pure H<sub>2</sub>O, library and/or hybridisation quality, number of samples hybridised/sequenced, etc. Therefore, we strongly recommend always reviewing the CNV profiles (.html files) to ensure the overall quality for each sample is good enough to report the findings showed on the 'gathered' files. If the CNV plots produce consistently poor results (e.g. noisy profiles and/or with some calls just above the thresholds, see CNV figure 1) we recommend users review their protocols, specifically DNA elution in ultra-pure H<sub>2</sub>O, library preparation and hybridisation/sequencing multi-plexing, or they contact us for further support. If the protocol is working well, the CNV profiles should look similar to those below (see CNV figures 2 and 3).

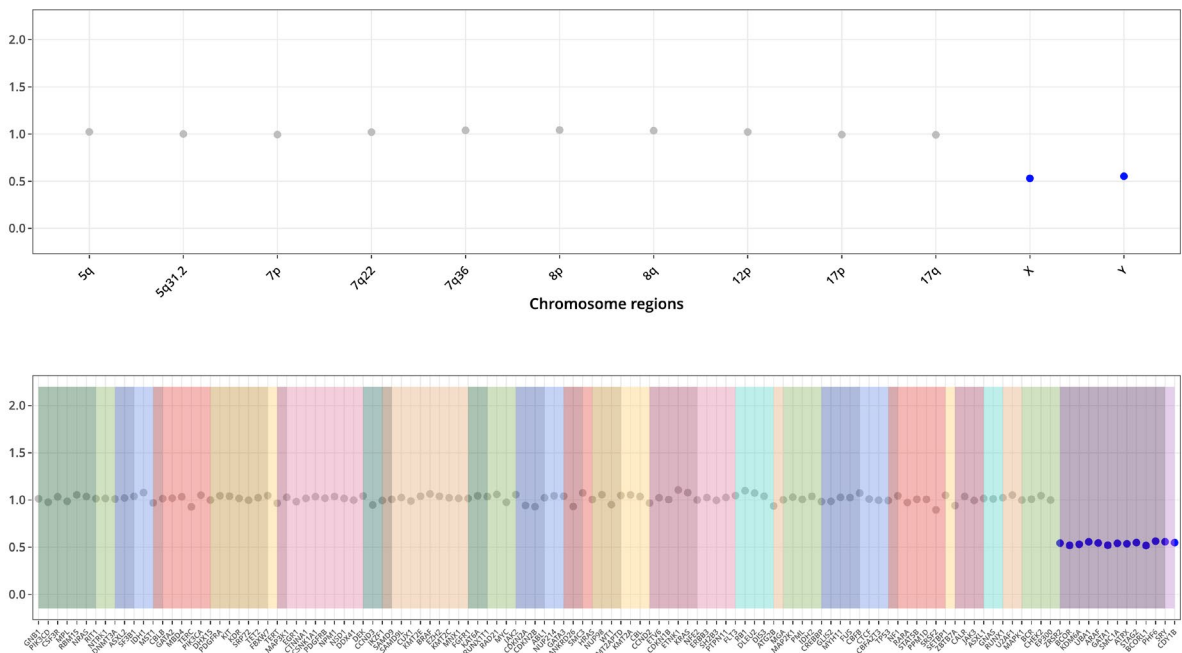
We recommend particular caution when calling single CNV events that have a logR value just above/below the thresholds for gain/loss calling (i.e. 1.3 or 0.8), as individual regions/genes can easily be artefacts. However, if all the genes for a whole chromosome, or large arm with several genes per region, have values just above 1.3 or below 0.8, then it is more likely that it represents a genuine gross chromosomal alteration (see CNV figure 4).



**CNV figure 1:** example of a **poor quality CNV profile** from a sample with normal male karyotype. Notice how in this extreme case many regions and genes are called as CNV (e.g. orange, green, red or blue), even though many times poor quality CNV profiles may only show a few 'artefactual' calls

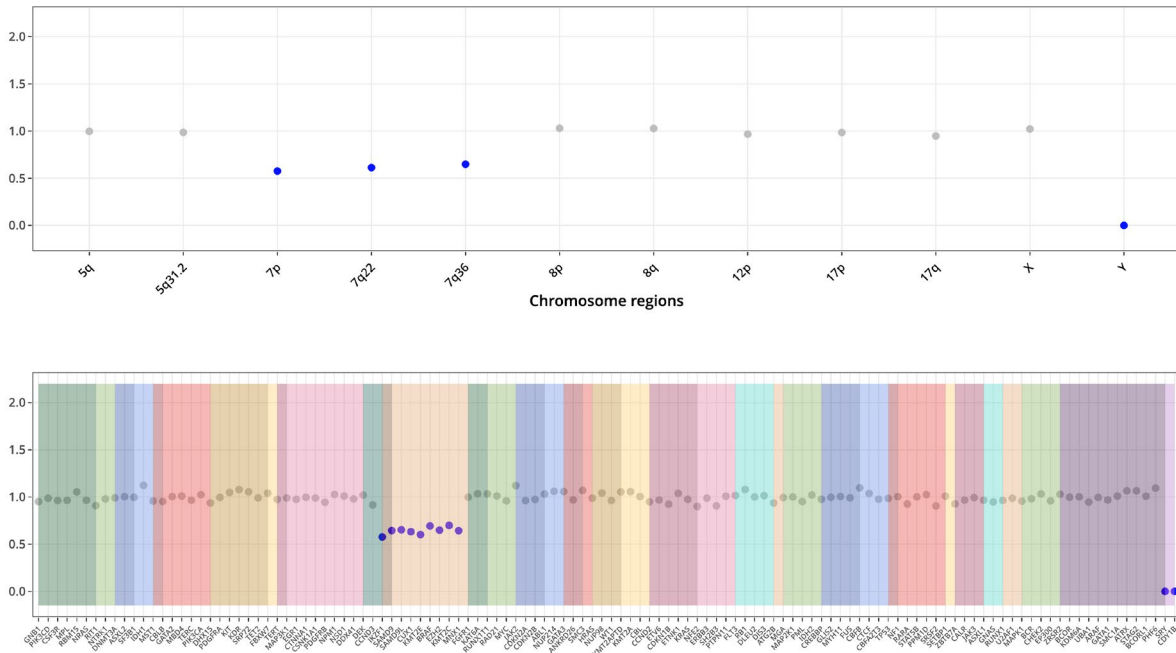


**CNV figure 2:** example of a **good quality CNV profile** from a sample with normal male karyotype.

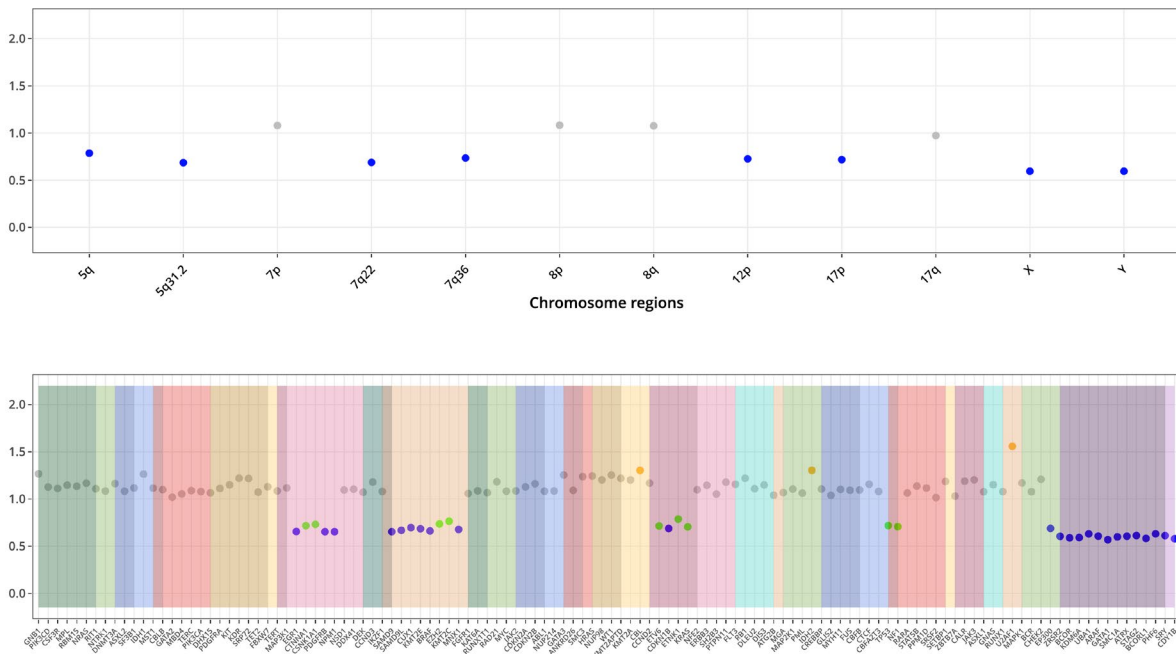




**CNV figure 3:** example of a **good quality CNV profile** from a sample with abnormal female karyotype. The profile is quite clean, with most regions in grey hovering around the diploid value of '1.0' and only chromosome 7p and 7q regions showing a CNV\_deletion (i.e. loss of chromosome 7) at an approximate logR of 0.6 (below the 0.7 value considered for 'deletion').



**CNV figure 4:** example of a **good quality CNV profile** from a sample with abnormal male karyotype. The profile is still good but more noisy than figures 2 and 3, likely due to a large number of deletions (including 5q, 7q, 12p, 17p and 22q (EP300) deletions), which artificially push the logR values for the rest of the genomic regions in the assay, producing artefactual 'gain' CNV calls (orange dots) just over the 1.3 threshold. Note one region with logR value over 1.5 likely representing a genuine 'gain' in this example. Note also how for the genes involved in the deletions, logR values in the bottom panel fluctuate between 'green' (<0.8) and 'blue' (<0.7), as the deletions sit around logR values of 0.75 (top panel)



**Example data files**

Please visit the Resources section of [www.univ8genomics.com](http://www.univ8genomics.com) to download example files from several cell lines analysed with the Myeloid-NDC assay. These are cell lines from lymphoid rather than myeloid disorders, and they have been analysed also with the EuroClonality-NDC assay, allowing users to employ the same samples for internal verification and quality control processes.

**Performance specifications**

Refer to the Quick Reference Guide for the Myeloid-NDC assay for details on expected metrics and analytical performance of the assay.



## Abbreviations

<b>CNV</b>	Copy number variation
<b>HMW</b>	High-molecular weight
<b>VAR</b>	Sequence variant (e.g. single nucleotide variant or small indels)
<b>SV</b>	Structural variant

## References

1. Arber DA, Orazi A, Hasserjian RP, Borowitz MJ, Calvo KR, Kvasnicka HM, et al. International Consensus Classification of Myeloid Neoplasms and Acute Leukemias: integrating morphologic, clinical, and genomic data. *Blood*. 2022 Sep 15;140(11):1200-1228. doi: 10.1182/blood.2022015850. PMID: 35767897; PMCID: PMC9479031.
2. Döhner H, Wei AH, Appelbaum FR, Craddock C, DiNardo CD, Dombret H, et al. Diagnosis and management of AML in adults: 2022 recommendations from an international expert panel on behalf of the ELN. *Blood*. 2022 Sep 22;140(12):1345-1377. doi: 10.1182/blood.2022016867. PMID: 35797463.
3. Bernard E, Tuechler H, Greenberg PL, Hasserjian RP, Arango Ossa JE, Nannya Y, et al. Molecular International Prognostic Scoring System for myelodysplastic syndromes. *NEJM Evid*. 2022 doi: 10.1056/EVIDoa2200008.
4. Gerds AT, Gotlib J, Ali H, Bose P, Dunbar A, Elshoury A, et al. Myeloproliferative Neoplasms, Version 3.2022, NCCN Clinical Practice Guidelines in Oncology. *J Natl Compr Canc Netw*. 2022 Sep;20(9):1033-1062. doi: 10.6004/jnccn.2022.0046. PMID: 36075392.
5. Stewart JP, Gazdova J, Darzentas N, Wren D, Proszek P, Fazio G, et al; EuroClonality-NGS Working Group. Validation of the EuroClonality-NGS DNA capture panel as an integrated genomic tool for lymphoproliferative disorders. *Blood Adv*. 2021 Aug 24;5(16):3188-3198. doi: 10.1182/bloodadvances.2020004056. PMID: 34424321.

## Revision History

Release	Summary of change	Date
2400014 r01	First released	February 2024

## Technical Assistance

Univ8 Genomics Ltd  
63 University Road  
Belfast  
Northern Ireland

**Email:** [contact@univ8genomics.com](mailto:contact@univ8genomics.com)

**Web:** [univ8genomics.com](http://univ8genomics.com)