**Supplementary material and methods:**

***IDH1* R132 and *IDH2* R172 genotyping using the Fast IDH method**

DNA was isolated using a Roche Cobas® kit and a Cobas® DNA sample preparation kit for both MPS approaches. qPCR amplification was performed using 1 μl or 5 μl total DNA (depending on whether the DNA concentration was > 30 ng/μl or > 10 ng/μl) using Thermo-Start Taq DNA Polymerase (ThermoFisher Scientific, 1U), 10X PCR Thermo-Start Buffer (ThermoFisher Scientific) MgCl2 (ThermoFisher Scientific), dNTP (Bioline), andEvaGreen 20x (Biotinium). The PCR primers were manufactured by Generi-Biotech and are not shown due to the ongoing licencing process of technology to company Biovendor Group (Brno, Czech Republic). Kit is expected to be commercially available in Q3/2021. For one PCR multiplex reaction (*IDH1* and *IDH2* reactions) the conditions were as follows: 95 ° C for 2 min, then 40 cycles, profile: 15 seconds at 95 °C, 30 seconds at 62 °C and 30 seconds at 72 °C (FAM channel fluorescence scanning), finally incubating for 5 minutes at 72 °C and melting from 60 °C to 95 °C with scanning fluorescence in FAM channel at
0.5 °C. After amplification, the QIAquick PCR Purification Kit (Hilden, GER, QIAGEN) was used to purify amplicons. The product concentration was determined by Qubit 2.0 HS DNA kit (Invitrogen). The sequencing library was diluted and denatured with 0.1 M NaOH (DNA concentration 10 μM). The library contained approximately the equivalent quantities of each sample and up to nine samples can be pooled (labelled by 8-base index) according to enclosed number of indexes within our kit. The indexed sequencing allows pooling of many libraries from different samples where unique index sequence is distinguished by Illumina sequencing platform processes for downstream analysis which was used in both methods. The sequencing was carried out on the Illumina MiSeq using the MiSeq V2 Nano 300 bp or V3 150 bp sequencing kit using 2 x 75 bp sequencing reads with custom sequencing primers, that are the part of upper mentioned technology licencing. The analysis was performed using the MiSeq Reporter software using the Somatic Variant Caller function. Output vcf files were processed using Excel (Microsoft). In order to identify mutations in *IDH1* and *IDH2* genes in exones 4 for both variants, the criteria were set to narrow the area of mutations present in codons 132 and 172 with frequency of > 5 % and with coverage >1000.

***IDH1* R132 and *IDH2* R172 genotyping using Nextera XT Library Prep Kit**

To enable *IDH1* R132 and *IDH2* R172sequencing, PCR was used to amplify the corresponding DNA sequence. The PCR master mix consisted of 0.5x EvaGreen® Dye (Biotium Inc., Fremont, CA, USA), 1U ThermoStart polymerase with 1x corresponding buffer (ThermoScientific, Waltham, MA), 200 μM dNTPs (Bioline, London, UK), 2 mM MgCl2 (ThermoScientific, Waltham, MA), and 0.25 μM of each primer pair listed in Table S1 (Generi Biotech, The Czech Republic). Approximately 20 ng DNA and DEPC-water (ThermoScientific, Waltham, MA) were added for the final volume of 20 μl. Because of the primers’ differing Tm values, touchdown PCR was performed using a LightCycler®480 (Roche Diagnostics, Mannheim, Germany). The amplification conditions were as follows: initial denaturation at 95 °C for 15 min, ten ‑1°C/cycle touchdown cycles of 15s each (starting at 95 °C), then 35 cycles of 95 °C for 10 s, 60 °C for 15 s, and 72 °C for 15 s. LightCycler®480 (Roche Diagnostics, Mannheim, Germany) was used for data evaluation, and product quality control was performed using QIAxcel® (QIAGEN, Hilden, Germany).

PCR products were purified using the QIAquick PCR Purification Kit (QIAGEN, Hilden, Germany) and amplicon concentrations were measured with an IMPLEN NanoPhotometer Pearl® (Implen, München, Germany). Library preparation was performed using the Nextera™ XT Library Prep Kit (Illumina Inc., San Diego, CA, USA) and Nextera™ XT Index Kit (Illumina Inc., San Diego, CA, USA) according to the manufacturer’s protocol which allows to pool up to 384 samples labelled by 2x8-bases index. PCR products with indexes were purified using the QIAquick PCR Purification Kit (QIAGEN, Hilden, Germany) and their final concentration was measured with Qubit® 2.0 (Life Technologies, MA, USA). Sequencing and analysis were performed using the MiSeq and MiSeq Reporter software using the Somatic Variant Caller function, respectively, with the same parameters as were used with the fastIDH method.

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| Gene  | Primer | DNA sequence | amplicon lenght |
| *IDH1 R132* | forward | 5´-CGGTCTTCAGAGAAGCCATTAT-3´ | 100 bp |
|  | reverse | 5´-TCACATTATTGCCAACATGACTTAC-3´ |
| *IDH2 R172* | forward | 5´-AAACATCCCACGCCTAGTCC-3´ | 200 bp |
|  | reverse | 5´-AGGTCAGTGGATCCCCTCTC-3´ |

Table S1. Primers used for *IDH1* R132 and *IDH2* R172 amplification by the Nextera XT method.