

Supplemental material to:

Single-Cell resolved Ploidy and Chromosomal Aberrations in Nonalcoholic Steatohepatitis-(NASH) Induced Hepatocellular Carcinoma and its Precursor Lesions.

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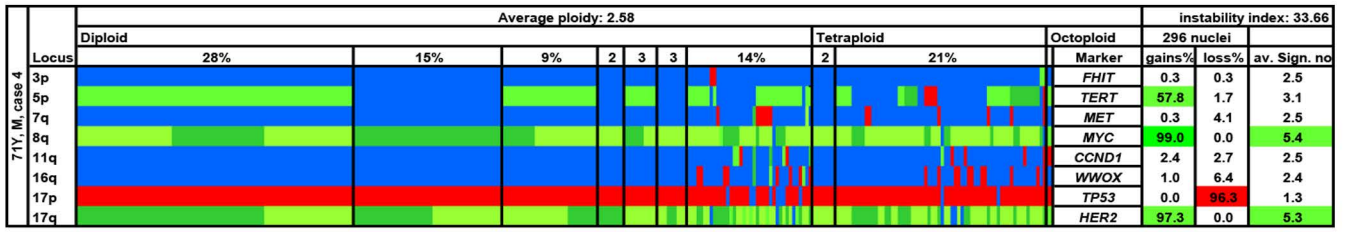
Legends:

Supplementary Figure 1 (A-C) Color display of miFISH analysis with eight gene-specific probes and gallery view showing a case with *HER2* amplifikation. Chart: Green, gains; red, losses; blue, unchanged. **(A)** Detail from left to right: the „locus“ column shows the chromosome arm. Average ploidy (center) and percentages of tumor clones (top left to right) are grouped into diploid, tetraploid and octoploid tumor clones. **(B)** *HER2* signals varied between 4-6 copies/cell, and 8-11 copies in tetraploid cells. Additional *MYC* gains and *TP53* loss. **(C)** Immunohistochemistry staining for *HER2* showed no specific membranous staining.

Supplementary Figure 2 Original FISH tree of case 1. The string of numbers represent the copy numbers of genes as follows: *FHIT-TERT-MET-MYC-CCND1-WWOX-TP53-HER2*. Size of ovals represents respective clone frequency. Root is a normal diploid clone.

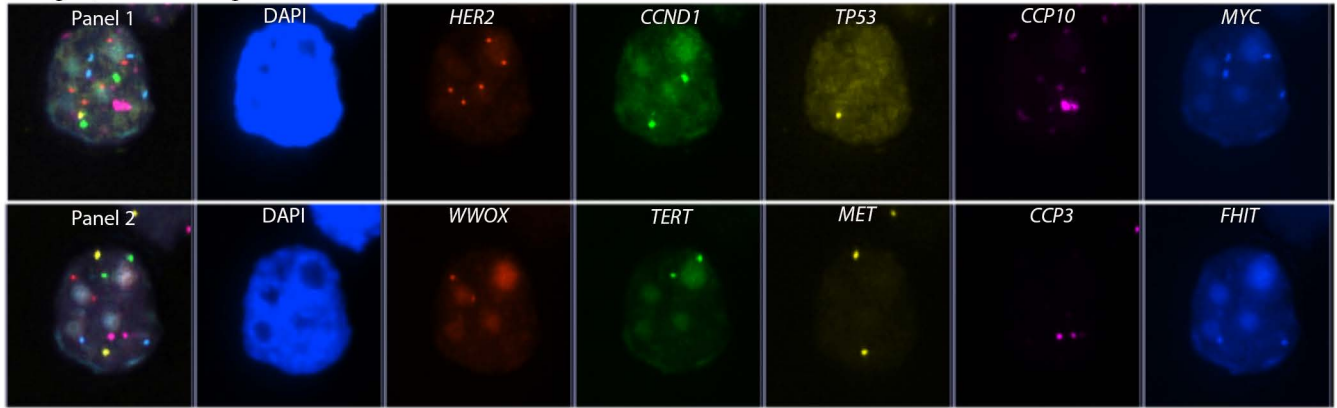
Supplementary Figure 3 Color display of miFISH analysis of all cases that are not shown in Figure 2 and 3. Green, gains; red, losses; blue, unchanged. Detail from left to right: the „locus“ column shows the chromosome arm. Average ploidy (center) and percentages of tumor clones. Instability index: signal patterns/100 nuclei.

A

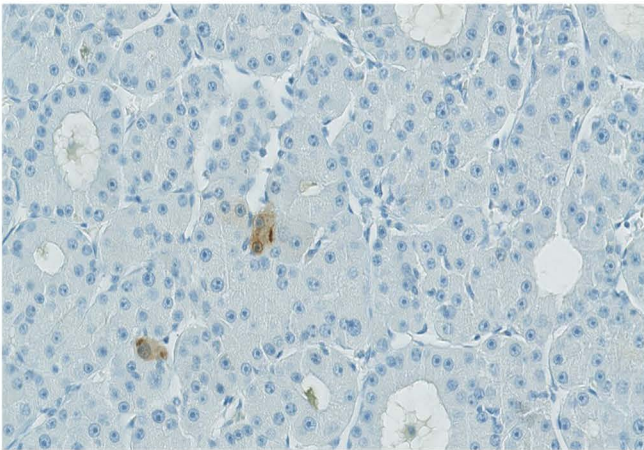


HER2 gain, TP53 loss MYC gain

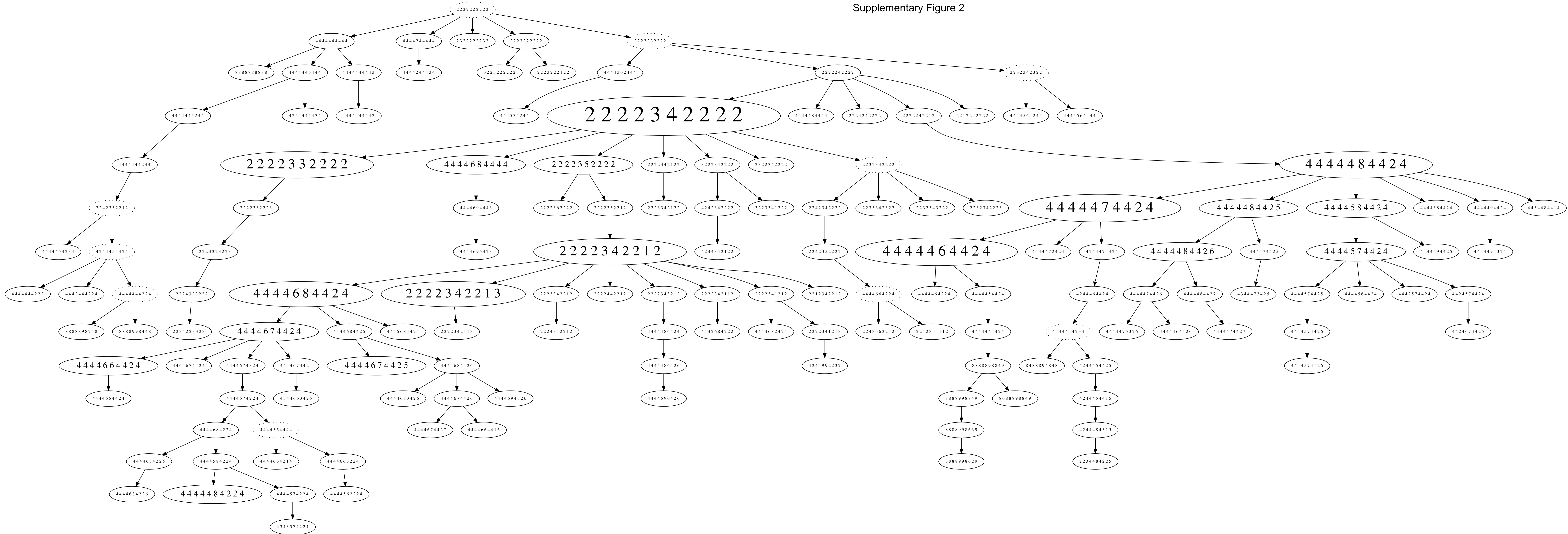
B



C



Supplementary Figure 2



		Average ploidy: 4.05										Instability index: 26.17			
Locus	Dip Tetraploid										gene	298 nuclei		av.Sig.no	
	4%	32%			33%	5%	5%	4%	32%			gains%	losses%		
69Y, M, case 2	3p											FHIT	10	2	4.1
	5p											TERT	0	14	3.8
	7q											MET	6	10	3.9
	8q											MYC	3	5	3.9
	11q											CCND1	1	3	3.9
	16q											WWOX	0	93	2.1
	17p											TP53	0	99	2.0
	17q											HER2	0	74	3.1

		Average ploidy: 2.38										Instability index: 36.4			
Locus	Diploid										gene	250 nuclei		av.Sig.no	
	14%	12%	8%	7%	5%	4	3	32%				14%	gains%		losses%
74, M, case 3	3p											FHIT	3	8	2.5
	5p											TERT	16	4	2.9
	7q											MET	3	6	2.5
	8q											MYC	7	11	2.7
	11q											CCND1	13	3	2.7
	16q											WWOX	44	3	2.3
	17p											TP53	2	26	2.3
	17q											HER2	26	24	2.8

		Average ploidy: 2.38										Instability index: 36.4			
Locus	Diploid										Gene	250 nuclei		av.Sig.no	
	14%	12%	8%	7%	5%	4	3	32%				14%	gains%		losses%
74, M, case 3	3p											FHIT	3.2	7.6	2.5
	5p											TERT	16	3.6	2.9
	7q											MET	2.8	6	2.5
	8q											MYC	7.2	11	2.7
	11q											CCND1	13.2	3.2	2.7
	16q											WWOX	44	3.2	2.3
	17p											TP53	2	26	2.3
	17q											HER2	26	24	2.8

		Average ploidy: 2.07										instability index: 19.11			
Locus	Diploid										gene	293 nuclei		av. Sig.no	
	33%	17%			5%	4%	3	3	3	2		2	2		23%
60Y, M, case 7	3p											FHIT	0	0	2.1
	5p											TERT	91	0	3.8
	7q											MET	78	1	3.0
	8q											MYC	1	1	2.0
	11q											CCND1	17	1	2.2
	16q											WWOX	0	35	1.8
	17p											TP53	0	84	1.5
	17q											HER2	13	2	2.2

		Average ploidy: 3.17										Instability index: 24.08					
Locus	Diploid										gene	191 nuclei		av. Sign.			
	25%	10%	6%	4%	4	4	3	3	3	2		2	11%		7%	4	13%
64Y, F, case 9	3p											FHIT	0	1	2.9		
	5p											TERT	23	1	3.2		
	7q											MET	3	6	3.1		
	8q											MYC	0	26	2.8		
	11q											CCND1	12	2	3.1		
	16q											WWOX	1	6	3.0		
	17p											TP53	0	71	2.4		
	17q											HER2	0	1	2.8		

		Average ploidy: 2.23										Instability index: 6.55			
Locus	Diploid										gene	290 nuclei		av. Sig. no	
	79%											3	3		7%
62Y, M, case 10	3p											FHIT	0	1	2.1
	5p											TERT	1	3	2.1
	7q											MET	0	4	2.1
	8q											MYC	1	2	2.1
	11q											CCND1	1	1	2.1
	16q											WWOX	0	3	2.1
	17p											TP53	0	99	1.1
	17q											HER2	0	0	2.1

		Average Ploidy: 2.08										Instability Index: 14.2			
Locus	Diploid										gene	300 nuclei		AvgSig	
	70.0%											13.3%	14.0%		gains%
54Y, M, case 11	3p											FHIT	1%	2%	2.1
	5p											TERT	2%	3%	2.1
	7q											MET	82%	0%	2.9
	8q											MYC	1%	2%	2.1
	11q											CCND1	99%	0%	9.4
	16q											WWOX	0%	1%	2.1
	17p											TP53	0%	96%	1.1
	17q											HER2	2%	1%	2.1