

SUPPLEMENTAL MATERIAL

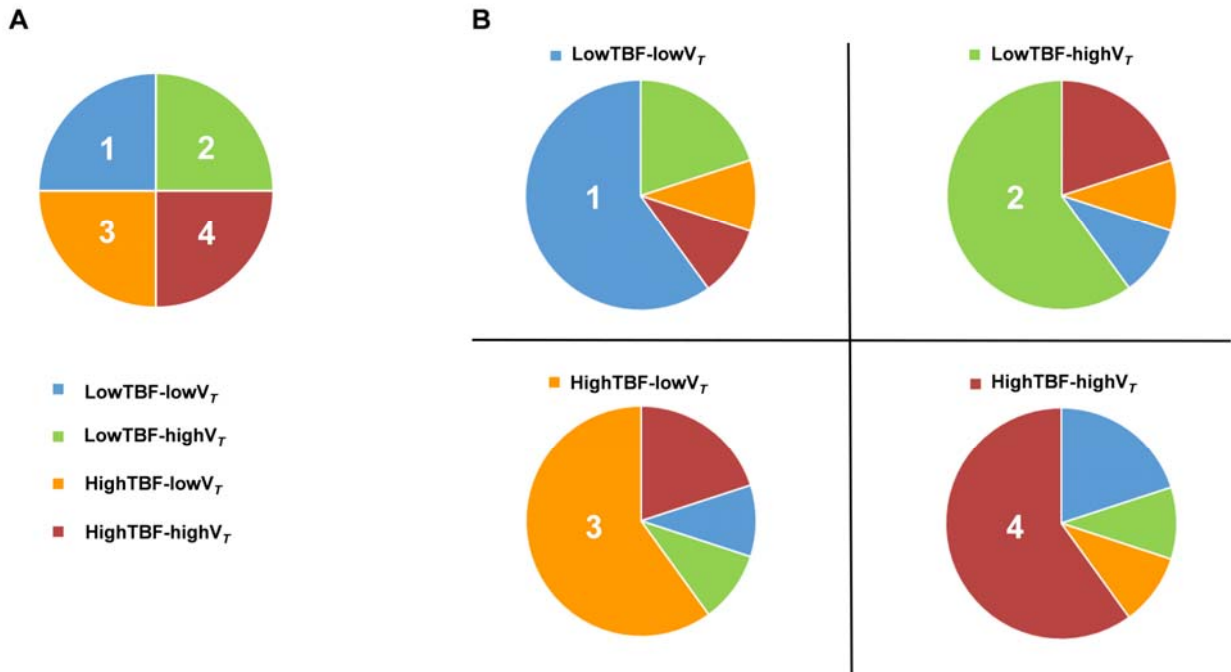
Clustering index

In addition to visual analysis of the level of clustering of voxels into the four categories, a quantitative metric was used to identify the degree of voxel clustering. The clustering index (CI) was obtained by measuring the local entropy of the classified voxels (into 4 categories as explained earlier) (15). This resulted in a CI close to 0.2 when all voxels were uniformly distributed across the lesion, while a CI close to 1.9 referred to random distribution of voxels across the lesion. The latter means that the classified voxels are not well clustered spatially.

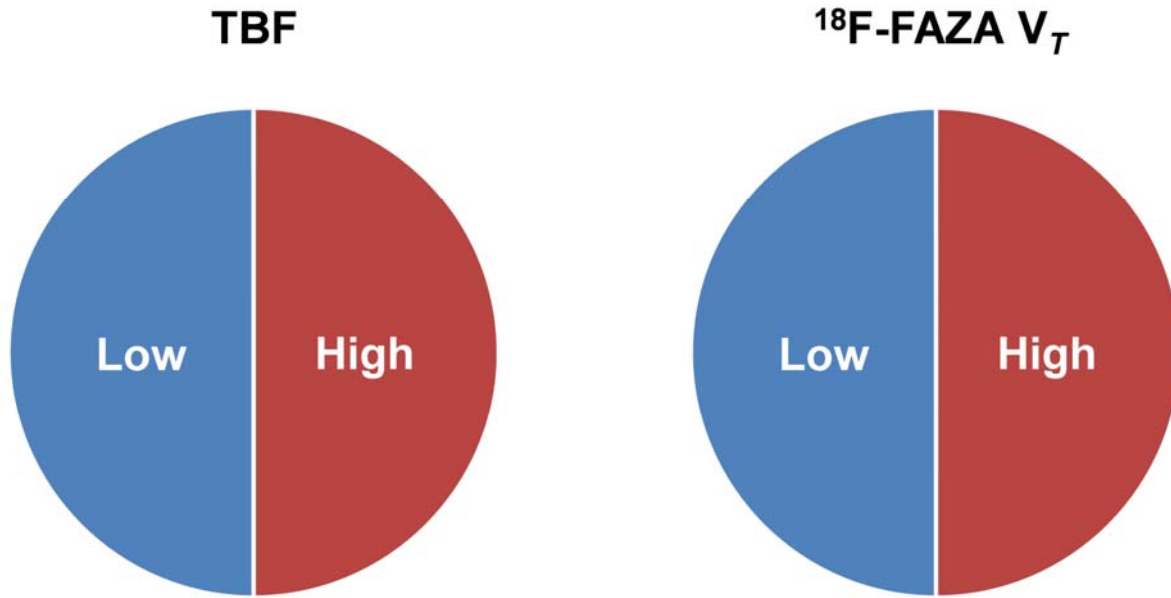
The CI is thus a measure to assess to which extent the classified voxels are clustered within the tumor. Apart from calculating the CI, as described above, the CI was calculated for 3 (theoretical) situations in order to provide the reader with information on the expected range for CI per lesion (Fig. 2). In this way effects of tumor volume and shape can be appreciated as well. The following situations were simulated: (1) a condition where all voxels are classified into one group and maximally clustered within the VOI (Fig. 2A), resulting in a CI of approximately 0; (2) a condition where voxels are classified into four categories and most optimally clustered (Fig. 2B), providing a CI of approximately 0.3; (3) a condition where voxels are classified into four categories, showing random distribution and almost no clustering, resulting in a CI of approximately 2 (Fig. 2C). Once the range of the CI, given by difference in CI for minimal and maximal clustering, has been determined for each lesion, the actual observed clustering can be expressed as a percentage using the following equation:

$$\text{Clustering (\%)} = \left(1 - \left(\frac{\text{CI lesion}}{\text{CI range}} \right) \right) \times 100 \quad (\text{Eq. 1})$$

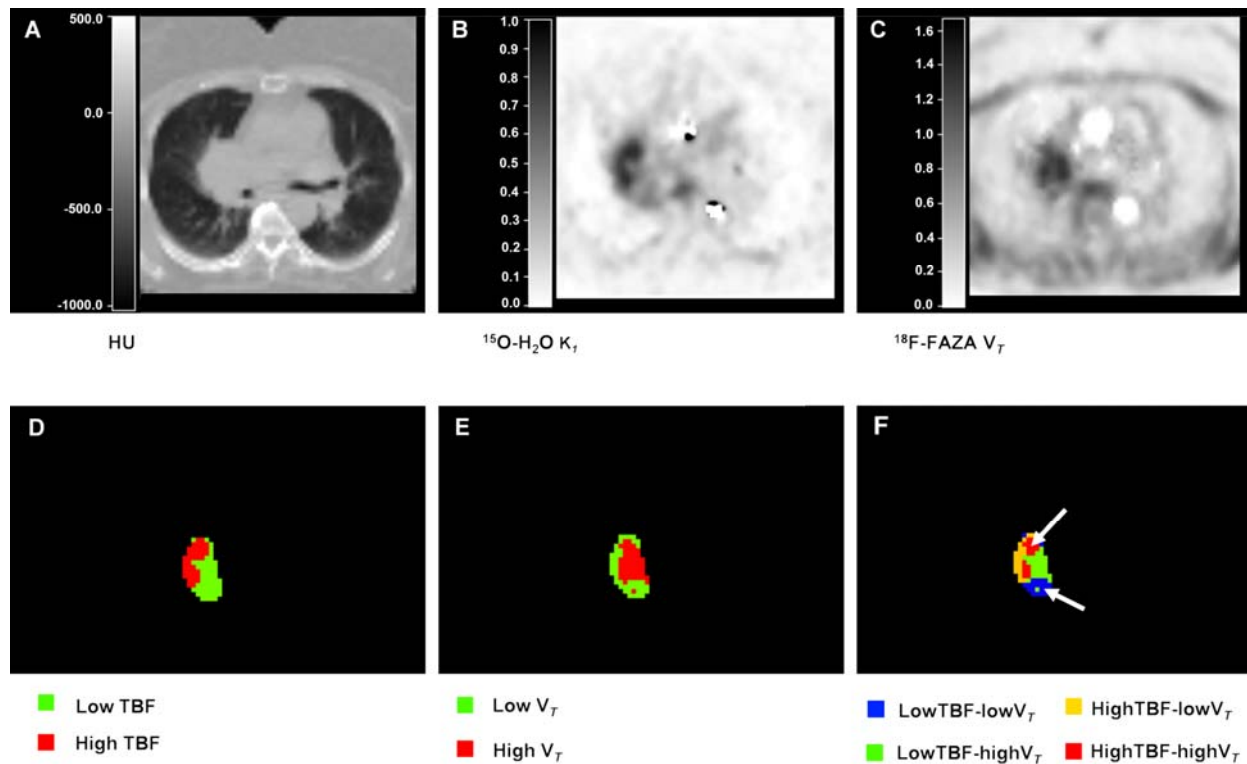
100% clustering corresponds to the condition illustrated in Fig. 2A and 0% clustering represents the condition given in Fig. 2C.



SUPPLEMENTAL FIGURE 1. The multiparametric classification was performed using lesional (A) and global (B) thresholds. (A) The lesional threshold provides insight in the intratumor heterogeneity by evaluating the distribution of voxels into four categories in the individual lesion. (B) The global threshold uses absolute perfusion and ^{18}F -FAZA V_T values across all lesions to classify the whole lesion into one of the four categories of the multiparametric classification.



SUPPLEMENTAL FIGURE 2. Classification of voxels into two categories for the parameters TBF and ¹⁸F-FAZA V_T. Voxels are classified as low or high TBF and ¹⁸F-FAZA V_T, respectively.



SUPPLEMENTAL FIGURE 3. An overview of the multiparametric evaluation of TBF and tumor hypoxia. (A) Low dose CT. (B) Parametric image of $^{15}\text{O-H}_2\text{O}$ derived TBF. (C) Parametric image of $^{18}\text{F-FAZA } V_T$. For both (B) and (C): voxels without a signal are located within large arteries and are set to zero. (D) Voxels classified as low or high TBF. (E) Voxels classified as low or high $^{18}\text{F-FAZA } V_T$. (F) Voxels classified into four categories: lowTBF-low V_T , lowTBF-high V_T , highTBF-low V_T and highTBF-high V_T . White arrows indicate distinct areas where TBF and $^{18}\text{F-FAZA } V_T$ were both decreased (blue) or increased (red). Blue areas may reflect regions with flow limited delivery, while red areas indicate regions with high $^{18}\text{F-FAZA}$ uptake, possibly indicating hypoxia, despite having high TBF.

	Lesion	TBF	¹⁸ F-FAZA V _T
Lesional threshold	1	0.33	0.84
	2	0.61	0.94
	3	0.43	1.04
	4	0.34	0.70
	5	0.15	0.63
	6	0.21	0.86
	7	0.41	0.55
	8	0.46	0.99
	9	0.64	0.98
	10	0.34	0.97
	11	0.30	1.09
	12	0.36	0.57
	13	0.21	0.93
Global threshold	1-13	0.37	0.85

SUPPLEMENTAL TABLE 1. Values of the lesional and global thresholds for TBF and ¹⁸F-FAZA V_T.

Lesion	Volume (mL)*	Hypoxic volume fraction (%)**	Highly perfused fraction (%)**	Clustering (%)***	
				<u>A</u>	<u>B</u>
1	19.8	46.1	34.2	37.6	39.4
2	34.6	62.7	86.5	40.6	58.8
3	138.5	75.8	70.9	40.8	54.2
4	35.0	18.5	45.1	39.2	45.3
5	13.7	13.1	0.0	41.4	76.5
6	27.7	51.2	3.7	42.1	62.2
7	12.3	0.0	66.2	22.9	64.8
8	92.9	63.0	64.7	51.0	56.4
9	219.1	61.6	89.8	52.3	72.9
10	178.9	63.3	44.4	56.5	60.3
11	53.5	75.1	32.3	46.1	56.4
12	23.1	0.0	47.9	37.9	67.1
13	30.5	67.0	2.5	36.2	66.3

SUPPLEMENTAL TABLE 2. Quantitative analysis of the multiparametric classification.

* Tumor volume, determined on the whole tumor VOI on the CT scan.

** Hypoxic volume fraction and highly perfused fraction, determined as the fraction of voxels classified above the global threshold, for TBF and ^{18}F -FAZA V_T .

*** Clustering (%) represents to which extent voxels are clustered in tumors, classified according to the lesional (A) and global threshold (B).