

Table 1. The voxel based morphologic parameters based on 3D reconstruction

ID #	Long name	Short name	Symbol	Unit	Definition
1	Cell grid perimeter	CLGP	GP_c	μm	$GP_c = N_{s,cyto} a_{vxl} \quad (1)$
2	Cell surface area	CLSF	$S_c \quad (4)$	μm^2	$S_c = N_{s,cyto} S_{vxl} \quad (2)$
3	Cell volume	CLVM	$V_c \quad (3)$	μm^3	$V_c = (N_{v,cyto} + N_{v,nucl} + N_{v,mito}) v_{vxl} \quad (3)$
4	Cell surface to volume ratio	CLSV	SVr_c	μm^{-1}	$SVr_c = S_c / V_c$
5	Cell surface irregularity index	CLSI	SI_i_c	$\mu\text{m}^{-1/2}$	$ISI_c = GP_c / \sqrt{V_c}$
6	Cell equivalent spherical radius	CLER	ER_c	μm	$ER_c = \left(\frac{3V_c}{4\pi}\right)^{1/3}$
7	Cell volume sphericity index	CLVS	VSi_c	-	$VSi_c = 4\pi ER_c^2 / S_c = (36\pi V_c^2)^{1/3} / S_c$
8	Average distance of cell membrane voxels to centroid	CLDM	$\langle R_c \rangle$	μm	$\langle R_c \rangle = \sum_{i=1}^{N_{s,cyto}} R_c(i) / N_{s,cyto} \quad (4)$
9	Standard deviation of R_c	CLDS	ΔR_c	μm	$\Delta R_c = \left\{ \frac{1}{N_{s,cyto}} \sum_{i=1}^{N_{s,cyto}} (R_c(i) - \langle R_c \rangle)^2 \right\}^{1/2}$
10	Cell protrusion volume	CLPV	V_p	μm^3	$V_{cp} = V_c - V_{c0} \quad (5)$
11	Cell protrusion number	CLPN	N_p	-	
12	Nuclear grid perimeter	NRGP	GP_n	μm	$GP_n = N_{s,nucl} a_{vxl}$
13	Nuclear surface area	NRSF	S_n	μm^2	$S_n = N_{s,nucl} S_{vxl}$
14	Nuclear volume	NRVM	V_n	μm^3	$V_n = N_{v,nucl} v_{vxl}$
15	Nuclear surface to volume ratio	NRSV	SVr_n	μm^{-1}	$SVr_n = S_n / V_n$
16	Nuclear surface irregularity index	NRSI	SI_i_n	$\mu\text{m}^{-1/2}$	$ISI_n = GP_n / \sqrt{V_n}$
17	Nuclear equivalent spherical radius	NRER	ER_n	μm	$ER_n = \left(\frac{3V_n}{4\pi}\right)^{1/3}$
18	Nuclear volume sphericity index	NRVS	VSi_n	-	$VSi_n = 4\pi ER_n^2 / S_n = (36\pi V_n^2)^{1/3} / S_n$
19	Average distance of nuclear membrane voxels to centroid	NRDM	$\langle R_n \rangle$	μm	$\langle R_n \rangle = \sum_{i=1}^{N_{s,nucl}} R_n(i) / N_{s,nucl}$
20	Standard deviation of R_n	NRDS	ΔR_n	μm	$\Delta R_n = \left\{ \frac{1}{N_{s,nucl}} \sum_{i=1}^{N_{s,nucl}} (R_n(i) - \langle R_n \rangle)^2 \right\}^{1/2}$
21	Mitochondrial grid perimeter	MNGP	GP_m	μm	$GP_m = N_{s,mito} a_{vxl}$
22	Mitochondrial surface area	MNSF	S_m	μm^2	$S_m = N_{s,mito} S_{vxl}$
23	Mitochondrial volume	MNVM	V_m	μm^3	$V_m = N_{v,mito} v_{vxl}$
24	Mitochondrial surface to volume ratio	MNSV	SVr_m	μm^{-1}	$SVr_m = S_m / V_m$
25	Mitochondrial surface irregularity index	MNSI	SI_i_m	$\mu\text{m}^{-1/2}$	$ISI_m = GP_m / \sqrt{V_m}$
26	Mitochondrial equivalent spherical radius	MNER	ER_m	μm	$ER_m = 3 / SVr_m$
27	Nucleus-to-cell centroid distance	NCCD	CD_{nc}	μm	$D_{nc} = \mathbf{r}_{nc} - \mathbf{r}_{cc} \quad (6)$
28	Nucleus-to-cell volume ratio	NCVR	Vr_{nc}	-	$Vr_{nc} = V_n / V_c$
29	Mitochondrion-to-cell volume ratio	MCVR	Vr_{mc}	-	$Vr_{mc} = V_m / V_c$

(1) $a_{vxl} = \text{voxel side length} = d_x = d_y \approx d_z$, where d_x and d_y is the pixel size along x- and y-axis, respectively, while d_z is the distance between two neighboring interpolated slices. The side length d_z is obtained by requiring $d_z \cdot (N_{int} + 1) \approx z_{\text{step}} \cdot f_{\text{scale}}$ with N_{int} as the integer number of interpolated slices between two raw slices and $z_{\text{step}} = 0.5 \mu\text{m}$ as the translation step size along z-axis, $f_{\text{scale}} = 0.87$ is the correction factor for light refraction.

(2) $N_{s,xxxx} = \text{number of surface or membrane voxels for the organelle } xxxx$, $S_{vxl} = 1.414 \cdot a_{vxl} d_z = \text{area of the diagonal plane of one voxel}$ (see figure below) as the average surface area of the membrane voxels. A surface voxel of a specific organelle is defined as the one

which has at least one of the six neighboring voxels belong to another organelle or the host medium outside of the cell.

(3) $N_{v,xxxx}$ = number of volume voxels for the organelle xxxx which includes the surface voxels and interior voxels, v_{voxel} =volume of voxel= $d_x d_y d_z$.

(4) $R_c = |\mathbf{r}_{cs}(i) - \mathbf{r}_{cc}|$ with $\mathbf{r}_{cs}(i) = (x_{cs}(i), y_{cs}(i), z_{cs}(i))d_{av}$ as the position vector of the i th voxel on the cell surface or membrane and $\mathbf{r}_{cc} = (x_{cc}, y_{cc}, z_{cc})d_{av}$ as the position vector of the cell centroid, $d_{av} = (d_x + d_y + d_z)/3$. The component coordinates of \mathbf{r}_{cc} are defined as $x_{cc} = \sum_{i=1}^{N_{v,cell}} x(i) / N_{v,cell}$ with $N_{v,cell} = N_{v,cyto} + N_{v,nucl} + N_{v,mito}$, etc..

(5) V_{c0} = cell volume after removing the protrusions through the opening morphology operation (an erosion followed by a dilation using the same structuring element for both operations) on the confocal image slices.

(6) $\mathbf{r}_{nc} = (x_{nc}, y_{nc}, z_{nc})d_{av}$ is the position vector of the nuclear centroid with its components defined as $x_{nc} = \sum_{i=1}^{N_{v,nucl}} x(i) / N_{v,nucl}$, etc..

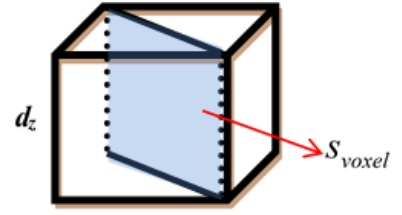


Table 2. The GLCM and pixel intensity parameters extracted from an input image

ID #	Long name	Short name	Definition ⁽¹⁾
1	Angular second moment (or energy or homogeneity)	ASM	$\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} \{p(i, j)\}^2$
2	Contrast (or inertia)	CON	$\sum_{k=0}^{G-1} k^2 p_{x-y}(k)$
3	Correlation	COR	$\frac{\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (i - \mu_x)(j - \mu_y) p(i, j)}{\sigma_x \sigma_y} = \frac{\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (ij) p(i, j) - \mu_x \mu_y}{\sigma_x \sigma_y}$
4	Variance	VAR	$\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (i - \mu_x)^2 p(i, j) = \sum_{i=0}^{G-1} (i - \mu_x)^2 p_x(i)$
5	Inverse difference moment (or local homogeneity)	IDM	$\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} \frac{1}{1 + (i - j)^2} p(i, j)$
6	Sum average (or mean value of 1 st and 2 nd gray-levels)	SAV	$\sum_{k=0}^{2G-2} k p_{x+y}(k) = \mu_x + \mu_y = 2\mu_x$
7	Sum entropy	SEN	$-\sum_{k=0}^{2G-2} p_{x+y}(k) \cdot \log(p_{x+y}(k))$
8	Sum variance	SVA	$\sum_{k=0}^{2G-2} (k - SAV)^2 p_{x+y}(k)$
9	Entropy	ENT	$-\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} p(i, j) \cdot \log(p(i, j))$
10	Difference entropy	DEN	$-\sum_{k=0}^{G-1} p_{x-y}(k) \cdot \log(p_{x-y}(k))$
11	Difference variance	DVA	$\sum_{k=0}^{G-1} k^2 p_{x-y}(k) - (\sum_{k=0}^{G-1} k p_{x-y}(k))^2$
12	Dissimilarity	DIS	$\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} i - j p(i, j)$
13	Cluster shade	CLS	$\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (i + j - \mu_x - \mu_y)^3 p(i, j) = \sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (i + j - 2\mu_x)^3 p(i, j)$
14	Cluster prominence	CLP	$\sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (i + j - \mu_x - \mu_y)^4 p(i, j) = \sum_{i=0}^{G-1} \sum_{j=0}^{G-1} (i + j - 2\mu_x)^4 p(i, j)$
15	Maximum probability	MAP	$\max(p(i, j))$
16	Relative minimum pixel intensity	IMIN	$\min(J(x, y)) / \text{mean}((J(x, y)))$
17	Relative maximum pixel intensity	IMAX	$\max(J(x, y)) / \text{mean}((J(x, y)))$
18	Mean value of pixel intensity	IMEA	$\text{mean}((J(x, y)))$

⁽¹⁾ $J(x, y)$ is a 12-bit image before normalization, $I(z, y)$ is the associated normalized 8-bit image as the input image for calculation of GLCM $p(i, j)$. In addition, the following functions are used in the above definitions with i or $j = 0, 1, 2, \dots, G-1$ and G as the number of gray levels. $G=255$ for 8-bit gray level images $I(x, y)$:

$$p_x(i) = \sum_{j=0}^{G-1} p(i, j), p_y(j) = \sum_{i=0}^{G-1} p(i, j), p_{x+y}(k) = \sum_{\substack{i=0 \\ i+j=k}}^{G-1} \sum_{j=0}^{G-1} p(i, j), p_{x-y}(k) = \sum_{\substack{i=0 \\ |i-j|=k}}^{G-1} \sum_{j=0}^{G-1} p(i, j),$$

$$\mu_x = \sum_{i=0}^{G-1} i p_x(i), \sigma_x^2 = \sum_{i=0}^{G-1} (p_x(i) - \mu_x)^2, \mu_y = \sum_{j=0}^{G-1} j p_y(j), \sigma_y^2 = \sum_{j=0}^{G-1} (p_y(j) - \mu_y)^2.$$