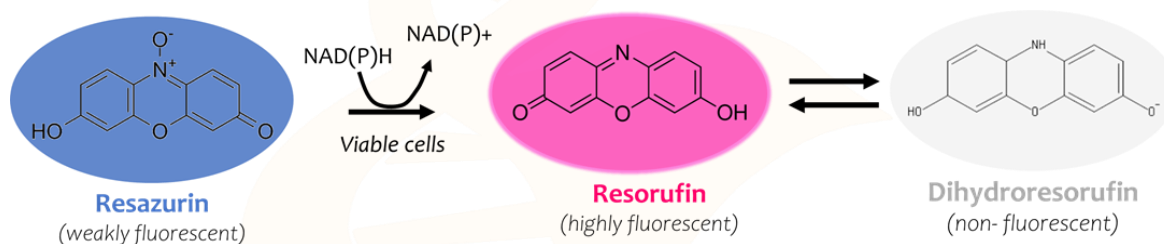


Frequently Asked Questions

Resazurin Viability Assay Kit

1. How Resazurin Viability Assay Kit works?

Resazurin Viability Assay Kit is based in the use of cell permeable compound resazurin. Weakly fluorescent resazurin is irreversibly reduced to the highly fluorescent resorufin at neutral pH by viable cells. Blue resazurin is reduced in a first step to the highly fluorescent pink resorufin and the reaction could be detected by colorimetry (absorbance at 540-570 nm) or by fluorometry (excitation 570 nm/ emission 585 nm).



2. Is resazurin a cell toxic compound?

Resazurin is generally regarded as nontoxic compound at low concentrations and brief incubation periods. In some cell lines, there is a reduction in glucose uptake in resazurin assay incubated in long times, in order of days¹. If the assay is incubated too long, resorufin will further reduce to di-hydro-resorufin, a non-fluorescent product of the second reduced state.

¹ Cytotechnology, 2015, 67(1): 13-7. doi: 10.1007/s10616-013-9664-1.

3. Is phenol red in the culture medium compatible with the Resazurin Viability Assay Kit?

Yes. Phenol red does not interfere with the resazurin reaction nor does it affect detection.

4. Which advantages and disadvantages offer Resazurin Viability Assay kit?

Resazurin Viability Assay Kit

Advantages	Disadvantages
- Applicable to bacteria, yeast, fungi, protozoa, plant cells. - Applicable to a number of cell lines. - Applicable to cell suspension and adherent cells. - Readout colorimetric or fluorometric. - Highly sensitive. - Rapid, easy and safe assay.	- Fluorescence interference by test compounds - Not applicable to thawed cells. - Since the signal gradually increases over time, a decrease in cell viability during incubation time cannot be detected.

5. How many cells must be seeded to use Resazurin Viability Assay kit?

The next table* includes some useful information to seed cell in 96-well plates

Cell line	Description	Time Post-Seeding	Fluorescence		Absorbance	
			Cells/ well	Z'	Cells/ well	Z'
A-549	Human Non-small Cell Lung Adeno-carcinoma	72 h	780-1.560	0.88	780-1.560	0.73
DU-145	Human Prostate Carcinoma	24 h	6.300-12.500	0.88	6.300-12.500	0.80
		48 h	3.100-6.300	0.90	6.300-12.500	0.79
		72 h	1.560-3.100	0.89	2.300-3.125	0.75
HCC-44	Human Non-small Cell Lung Carcinoma	48 h	780-1.560	0.85	1.560-3.100	0.75
		72 h	780-1.560	0.81	780-1.560	0.55
HeLa	Human Cervical Adenocarcinoma	48 h	3.400-6.800	0.78	14.000-27.000	0.75
		72 h	1.300-2.600	0.84	2.600-5.300	0.63
PC-3	Human Prostate Adenocarcinoma	24 h	13.000-25.000	0.82	13.000-25.000	0.89
		48 h	6.300-12.500	0.69	6.300-12.500	0.69
		72 h	3.100-6.300	0.79	3.100-6.300	0.59
SKOV-3	Human Ovarian Adenocarcinoma	72 h	1.300-2.600	0.76	2.600-5.300	0.70
St-T1b	Human Endometrial Stroma, immortalized	72 h	3.300-6.600	0.81	6.600-13.000	0.69
T98-G	Human Glioblastoma	48 h	6.300-12.500	0.62	6.300-12.500	0.68
		72 h	3.100-6.300	0.64	6.300-12.500	0.55
		96 h	1.600-3.100	0.64	3.100-6.300	0.50
U-251 MG	Human Glioblastoma	48 h	6.300-12.500	0.78	13.000-25.000	0.73
		72 h	3.100-6.300	0.58	3.100-6.300	0.53
		96 h	1.600-3.100	0.63	1.600-3.100	0.75

* taken from Biosensors 2022, 12(4), 196.