

Datasheet

Panexin basic

Defined Serum Replacement

Product	Description	Catalogue-No.	Size
Panexin basic	Defined serum replacement for adherent and non-adherent cells	P04-96090	50 ml
		P04-96900	100 ml
		P04-96950	500 ml

Product description

Panexin basic is a fully defined serum replacement for the cultivation of adherent and non-adherent cells under serum-free culture conditions or to significantly reduce the amount of serum in cell culture. It supports the growth of many cell types in an optimum manner without any extra handling compared to serum.

Storage conditions

Storage: -20°C, in the dark
 Stability: 2 years from date of production
 Size: 50 ml, 100 ml, 500 ml, other sizes on request

Composition

Panexin basic contains purified proteins, lipids, salts, amino acids, trace elements, hormones and a 3-dimensional substrate release system in an optimized formulation. It contains no growth factors, undefined hydrolysates or peptones.

Suitability

Panexin basic is suitable for the cultivation of a variety of adherent and non-adherent cells under serum-free culture conditions or to reduce the necessary FBS amount in cell culture. (see Fig.1)

Effect of Panexin basic in different cell lines

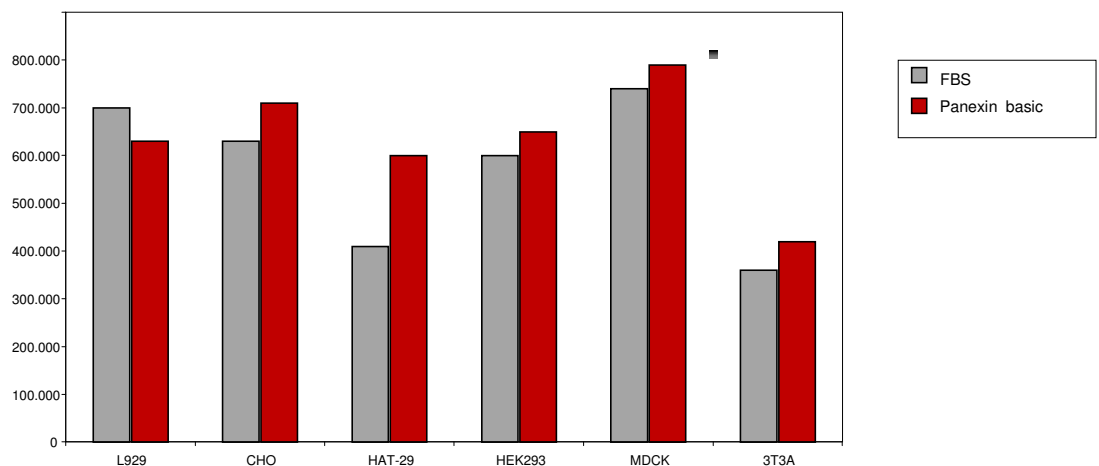


Fig.1 Efficiency and growth stimulation of Panexin basic compared to FBS (10% in DMEM/F12) for different cell types

Special advantages

Panexin basic is designed to replace or to reduce serum in the cell culture in a very simple manner. In most cases there is no need to change the basal medium. As Panexin basic is fully defined and contains no peptones or hydrolysates, lot testing is not necessary anymore. It also allows high reproducibility and simplified downstream process. Panexin basic contains no growth factors and enables defined proliferation and differentiation of stem cells. Characterization studies of growth factors will obtain more reproducible and clearer results. Panexin basic is also useful to develop sensitive cell-based *in vitro* tests and coculture procedures. For cell lines which require specific growth factors these should be added in a concentration as previously used.

Instructions for use

Panexin basic can be stored and used in the same manner as serum.

- Thaw Panexin at maximum 37 °C. Please avoid repeated freeze-thaw cycles!
- To replace serum: Use the same basal medium and the same concentration of Panexin as FBS. The performance can be further improved by optimizing the concentration of Panexin or modifying/changing the basal medium^a.
- To reduce serum concentration: Use the same basal medium and add the same amount of Panexin as the reduced amount of serum, until the minimal necessary concentration of FBS is found (1 to 2.5 % in most cases). The performance can be further improved by optimizing the concentration of Panexin or modifying/changing the basal medium^a (also see adaptation instructions).
- Recommended inoculation cell density $5 \times 10^4 - 10 \times 10^4$ cells /ml for non-adherent cells; $5 \times 10^3 - 20 \times 10^3$ cells/cm² for adherent cells.
- If working with adherent cells: Solve adherent cells as usual from the cell culture vessel (e.g. 0.25% trypsin, Cat.No. P10-033100 or Accutase[®], Cat.No. P10-21100). Once the cells have become round and detach from the surface inactivate trypsin with trypsin inhibitor (Cat.No. P10-034100): Simply resuspend cells in about 1 ml trypsin inhibitor solution for every ml of trypsin solution used for dissociation. Note that Accutase[®] does not need to be inhibited.

Depending on the cell type, some differences in morphology or proliferation rate may be observed with varying standard media. Most applications were performed with RPMI 1640 for non-adherent cells and with DMEM and DMEM/F12 for adherent cells and with these combinations very good growth stimulation was achieved in a range of 5 - 15 % Panexin.

For demanding cells (e.g. primary cells) an adaptation procedure to serum-free condition may be necessary.

Adaptation instructions for Panexin basic

Precondition for a successful transition are vital cells (trypan blue exclusion staining), which should be harvested in the logarithmic growth phase. If 10% FBS was used in the original protocol,

Step 1: 7.5 % FBS + 2.5 % Panexin

- Seed cells at $5 \times 10^4 - 10 \times 10^4$ cells/ml (non-adherent cells), or at $5 \times 10^3 - 20 \times 10^3$ cells/cm² (adherent cells).
- Observe cells under a microscope at about 90 % confluence passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 2: 5 % FBS + 5 % Panexin

- Seed cells at $5 \times 10^4 - 10 \times 10^4$ cells/ml (non-adherent cells), or at $5 \times 10^3 - 20 \times 10^3$ cells/cm² (adherent cells).
- Observe cells under a microscope, at about 90 % confluence passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 3: 2.5 % FBS + 7.5 % Panexin

- Seed cells at $5 \times 10^4 - 10 \times 10^4$ cells/ml (non-adherent cells), or at $5 \times 10^3 - 20 \times 10^3$ cells/cm² (adherent cells).
- Observe cells under a microscope, at about 90 % confluence, passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 4: 1 % FBS + 9 % Panexin

- Seed cells at $5 \times 10^4 - 10 \times 10^4$ cells/ml (non-adherent cells), or at $5 \times 10^3 - 20 \times 10^3$ cells/cm² (adherent cells).
- Observe cells under a microscope, at about 90 % confluence, passage the cells for another 2-3 passages.

If normal growth is obtained transfer cells into:

Step 5: 10 % Panexin

- Seed cells at $5 \times 10^4 - 10 \times 10^4$ cells/ml (non-adherent cells), or at $5 \times 10^3 - 20 \times 10^3$ cells/cm² (adherent cells).
- Observe cells under a microscope.

For some cells an adaptation to serum-free conditions is difficult to reach or even impossible.

The following measures may help to facilitate a successful adaptation:

- Reseeding with a higher cell amount (about 2x to 4x of the usual cell density).
- Addition of growth factors (if known, which factors have a positive effect on the relevant cells).
- Coating the culture dishes or flasks with attachment factors (e.g. fibronectin, laminin, collagen, gelatine, etc).
- Change the basal medium. Note: A change of the basal medium to a richer or more complex formulation may be all that is needed to achieve growth in serum free condition.

Table 1: Comparison of Cell Growth in 10% Panexin basic in different Basal Media versus Cell Growth in 10% FBS in different Basal Media

Cell Line	Origin	Basal Medium	Percentage of Growth 10% Panexin basic	Percentage of Growth 10% FBS
HEK 293 T	Renal cells, human embryonic	DMEM/F12	105%	100%
		alpha-MEM	76%	
		DMEM	62%	
MDCK	Renal cells, canine	DMEM/F12	102%	100%
		McCoy's 5A	91%	
		alpha-MEM	106%	

MDBK	Renal cells, bovine	RPMI 1640	122%	100%
		McCoy's 5A	135%	
		DMEM	131%	
L 929	Fibroblasts, mouse	DMEM	97%	100%
		RPMI 1640	78%	
		Ham's F-12	128%	
HT-29	Colon Carcinoma, human	IMDM	108%	100%
		DMEM/F12	98%	
		alpha-MEM	96%	
HeLa S3	Cervix carcinoma epithel, human	Glasgow MEM	106%	100%
		IMDM	72%	
		EMEM	100%	
CHOO	Ovarial cells epithel, Chinese hamster	DMEM/F12	106%	100%
		IMDM	97%	
		alpha-MEM	82%	
CHO-Luc	Ovarial cells epithel, Chinese hamster, transfected	IMDM	86%	100%
		DMEM	97%	
		alpha-MEM	84%	
3T3A	Fibroblasts, mouse	RPMI 1640	98%	100%
		McCoy's 5A	72%	
		DMEM/F12	97%	
MCF-7	Mammary carcinoma, human	Ham's F-12	292%	100%
		DMEM/F12	176%	
		RPMI 1640	214%	
RAW 264.7	Macrophages, mouse	McCoy's 5A	40%	100%
		DMEM/F12	67%	
		alpha-MEM	38%	
U-937	Lymphoma, human	alpha-MEM	107%	100%
		DMEM/F12	15%	
		DMEM	20%	
MM6	Monocytes, human	RPMI 1640	120%	100%
		McCoy's 5A	143%	
		DMEM/F12	118%	
HL-60	Promyelocytic leukemia cells, human	RPMI 1640	92%	100%
		DMEM/F12	14%	
		DMEM	11%	
X63-Ag8	Myeloma	DMEM	94%	100%
		RPMI 1640	97%	
		DMEM/F12	29%	

References

For cell line specific references please see our homepage (www.pan-biotech.com)

Technical support

For technical support, questions or remarks please contact your local PAN-Biotech partner or the technical department of PAN-Biotech via email (info@pan-biotech.com) or phone +49-8543-601630.

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^a As a basal medium, standard media such as RPMI 1640, DMEM (high or low glucose), DMEM/F12, etc. can be used. Make sure that L-glutamine is present in sufficient quantity (supplement L-glutamine as needed)