Report of Human Cell Line Authentication

（Notice:This authentication report is restricted to the cell sold from Guangzhou Cellcook Biotech Co. ,Ltd, and the date with seal is the date of delivery. ）

?. Sample

Sample Name: labeled as 'SK-MEL-28'

? . Method and Procedure

1. PCR is amplified with STR Multi-amplification Kit (PowerPlex™16HS System);
2. PCR products are assayed with 3100 DNA Analyzer (Applied Biosystems®).
3. Amplification of gene COX1 and electrophoresis are employed to survey the species of the sample.

? . Results

1. The STR profiles of the cell line sample are in the attached table and figure.
2. The search result in ATCC and DSMZ databases.
3. The electrophoresis figure of gene COX1.

SK-MEL-28: ①One loci has tri-alleles (TPOX). Contamination of other human cell line is not found (Figure 1 & Table 1). ②100% matched cell lines are not found in ATCC and DSMZ data banks. The percentage of matching to SK-MEL-28 is 82%. (Figure 2 & Figure 3). ③The sample is a human cell line. Contamination of other species cells are not found in the sample (Figure 4).

Operator: Xiaohua Mo

Auditor: Xuanyi Liang

Guangzhou Cellcook Biotech Co., Ltd

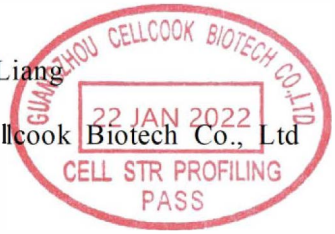

Figure 1. STR profiles of SK-MEL-28 cell line

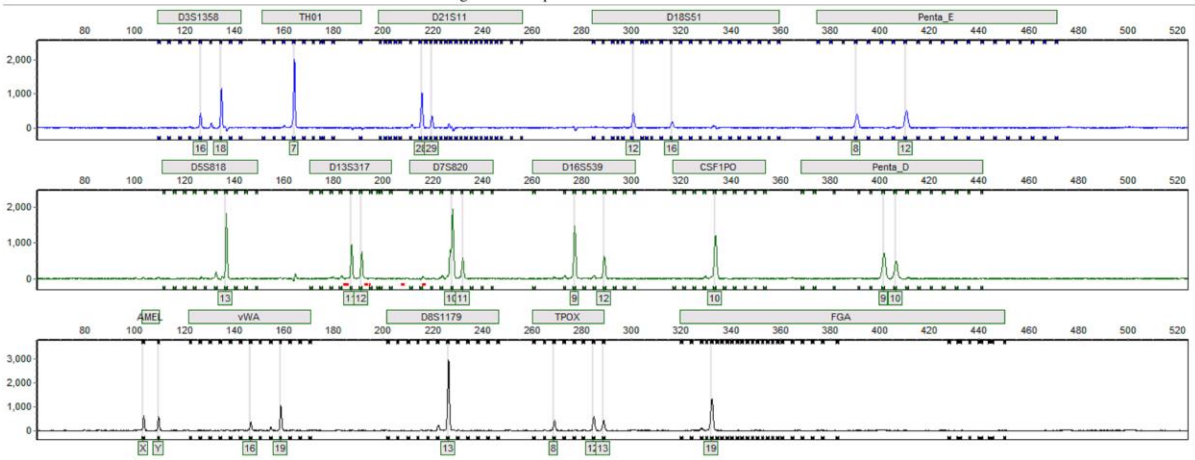

Table 1. STR profiles of SK-MEL-28 cell line

	Allele1	Allele2	Allele3
D3S1358	16	18	
TH01	7		
D21S11	28	29	
D18S51	12	16	
Penta_E	8	12	
D5S818	13		
D13S317	11	12	
D7S820	10	11	
D16S539	9	12	
CSF1PO	10		
Penta_D	9	10	
AMEL	x	y	
vWA	16	19	
D8S1179	13		
TPOX	8	12	13
FGA	19		

Figure 2. Search result in ATCC database

SEARCH THE STR DATABASE

As part of our continuing efforts to characterize and authenticate the cell lines in the Cell Biology collection, ATCC has developed a comprehensive database of short tandem repeat (STR) DNA profiles for all of our human cell lines. View our brief tutorial before starting.

1. STR Profiling Analysis
2. Matching Algorithm
3. Interrogating the Database

Showing 1 - 2 Of 2

PageSize: 100

Add to Cart	%Match	ATCC® Number	Designation	D5S818	D13S317	D7S820	D16S539	vWA	TH01	AMEL	TPOX	CSF1PO
☐	82.0	HTB-72	SK-MEL-28MelanomaHuman	11,13	11,12	9.3,10	9,12	16,19	7	X,Y	8,12	10,12
☐	82.0	CRL-2021	MEG-01MegakaryoblastHuman	13	8	11	9	16	7	X,Y	8,11	10

Add to Cart | Export to Excel

Disclaimer: Reference to this database and the data contained therein may be cited in publications, and ATCC encourages such citation or reference. While every reasonable effort has been made to assure the accuracy of these data, no warranty, express or implied, is made by ATCC as to their accuracy.

While ATCC has largely used the Promega PowerPlex® 1.2 System in the creation of these data and recommends

Figure 3. Search result in DSMZ database

Result of STR matching analysis by your data.

- DSMZ Profile Database -

A graphical presentation is shown at the bottom of this page.

EV	Cell No.	Cell name	Locus names									Figures
			D5S818	D13S317	D7S820	D16S539	VWA	TH01	AM	TPOX	CSF1PO	
	Query (Your Cell)		*13,13*	*11,12*	*10,11*	*9,12*	*16,19*	*7,7*	*x,y*	*8,12,13*	*10,10*	
0.81(30/37)	HTB-72	SK-MEL-28	11,13	11,12	10,10	9,12	16,19	7,7	X,Y	8,12	10,12	-
0.81(30/37)	RCB1930	SK-MEL-28	11,13	11,12	10,10	9,12	16,19	7,7	X,Y	8,12	10,12	-
0.70(26/37)	CRL-2056	CCD-1043Sk	13,13	11,12	10,10	9,11	16,19	7,7	X,Y	8,11	12,12	-
0.65(24/37)	364	MEG-01	13,13	8,8	11,11	9,9	16,16	7,7	X,Y	8,11	10,10	-
0.65(24/37)	CCL-137	HEL 299	11,13	11,12	8,11	10,11	16,16	7,7	X,Y	8,12	7,10	-
0.65(24/37)	CRL-2021	MEG-01	13,13	8,8	11,11	9,9	16,16	7,7	X,Y	8,11	10,10	-
0.65(24/37)	CRL-5867	NCI-H1385 [H1385]	13,13	11,11	8,10	9,11	17,19	7,7	X,X	8,9	10,10	-
0.65(24/37)	IFO50151	MEG-01	13,13	8,8	11,11	9,9	16,16	7,7	X,Y	8,11	10,10	-
0.65(24/37)	SCRC-2002	hES BG01V	10,12	11,12	10,11	9,11	16,17	7,9.3	X,Y	8,8	10,10	-

Figure 4. Authentication of the species of the sample

M: Marker. As the size of 700, 600, 500, 400, 300, 200 and 100bp from up to down.

Nine species are checked, as follow: *Homo sapiens* 391bp, *Cricetulus griseus*315bp, *Macaca mulatta*287bp, *Cercopithecus aethiops*222bp, *Rattus norvegicus*196bp, *Canis familiaris*172bp, *Mus musculus*150bp, *Bos Taurus*102bp, IC 70bp

The sample: The band size is 391bp which matches the size of human.

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细胞 STR 检测报告

一、样品名称/编号：A-375

二、检测项目：STR 基因型检验

三、检测方法：用天根的基因组抽提试剂盒提取 DNA，采用 21-STR 扩增方案扩增，在 Seqstudio 型遗传分析仪上对 STR 位点和性别基因 Amelogenin 进行检测。

四、检验结果：

1.检验基本情况

多等位基因	匹配细胞系	细胞库	EV 值	匹配说明
无	A-375	ExPASy	1.00	完全匹配

多等位基因指三等位及以上基因现象。

本次检测各细胞分型结果良好。

2.样本描述

该株细胞 DNA 分型在细胞系检索中找到完全匹配的细胞系，ExPASy 数据库显示细胞名为 A-375 细胞号对应 CVCL-0132。本次检测在该细胞系中没有发现多等位基因。

3.匹配说明

待测细胞系与收录于 ATCC，DSMZ，JCRB 和 ExPASy 数据库的细胞系 STR 数据进行比对，未收录于以上细胞库的细胞系将无法匹配。

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附表：A-375 细胞的 STR 位点和 Amelogenin 位点的基因分型结果

A-375 细胞			
Marker	Allele 1	Allele 2	Allele 3
D19S433	13	14.2	
D5S818	12	12	
D21S11	29	30	
D18S51	12	17	
D6S1043	11	14	
AMEL	X	X	
D3S1358	15	17	
D13S317	11	14	
D7S820	9	9	
D16S539	9	9	
CSF1PO	11	12	
Penta D	9	15	
D2S441	11	11	
vWA	16	17	
D8S1179	11	14	
TPOX	8	10	
Penta E	10	12	
TH01	8	8	
D12S391	18	21	
D2S1338	16	24	
FGA	23	23	

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附图：ExPASy 数据库比对结果

Accession	Name	N° Markers	Score	Amel	CSF1PO	D2S1338	D3S1358	D5S818	D7S820	D8S1179	D13S317	D16S539	D18S51	D19S433	D21S11	FGA	Penta D	Penta E	TH01	TPOX	vWA
NA	Query	NA	NA	x	11,12			12	9		11,14	9							8	8,10	16,17
CVCL_0132	A-375	8	100.00%	X	11,12	16,24	15,17	12	9	11,14	11,14	9	12,17	13,14.2	29,30	23	9,15	10,12	8	8,10	16,17
CVCL_UD29	A-375 KRAS p.G13D	8	100.00%	X	11,12			12	9		11,14	9							8	8,10	16,17
CVCL_UR34	A-375 KRAS p.G13D-Luc 2	8	100.00%	X	11,12			12	9		11,14	9							8	8,10	16,17

分型方案及位点分布

	方案 1	方案 2	方案 3	方案 4
1	D19S433	AMEL	D2S441	TH01
2	D5S818	D3S1358	vWA	D12S391
3	D21S11	D13S317	D8S1179	D2S1338
4	D18S51	D7S820	TPOX	FGA
5	D6S1043	D16S539	Penta E	
6		CSF1PO		
7		Penta D		

检测人：张葡萄

审核人：殷世腾

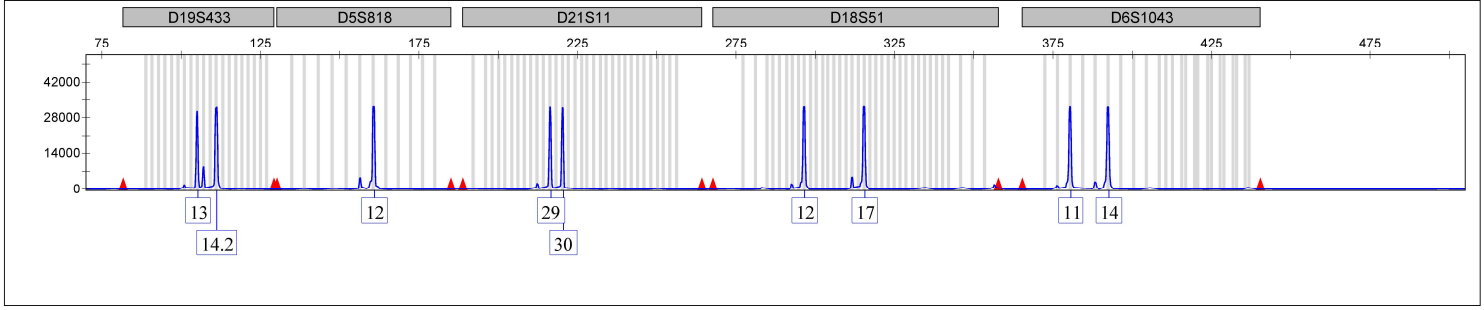
签发日期：2023.06.12

河南省工业微生物菌种工程技术研究中心

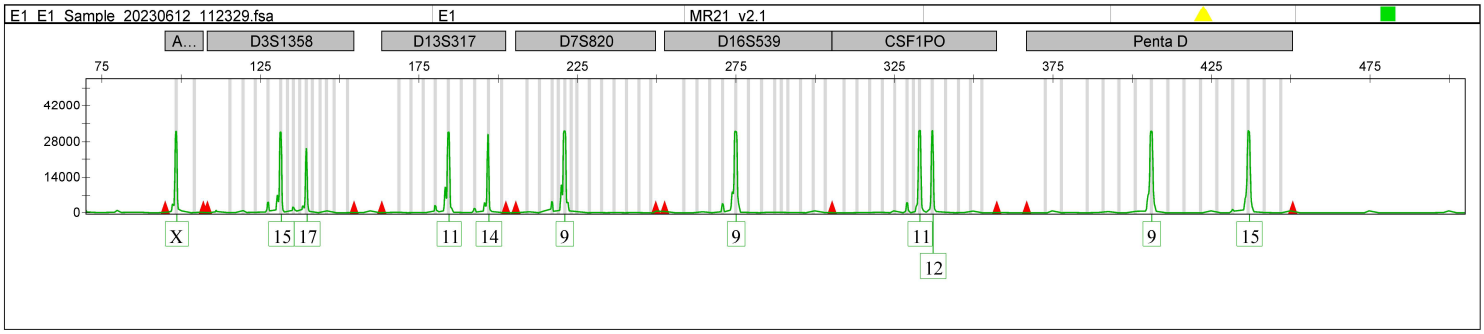
Henan Engineering Research Center of Industrial Microbiology

网址：www.bncc.com 电话：400-6699-833

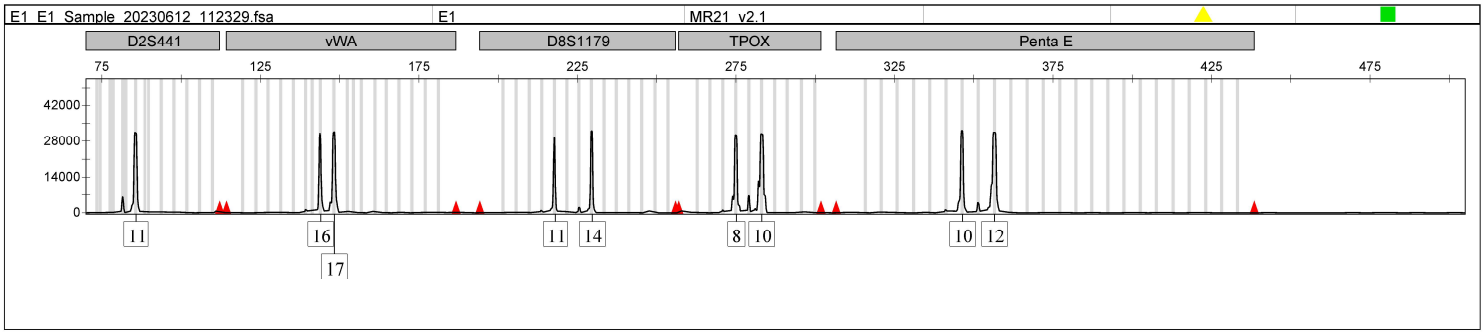
Sample File	Sample Name	Panel	SQI	OS	SQ
E1_E1_Sample_20230612_112329.fsa	E1	MR21_v2.1			



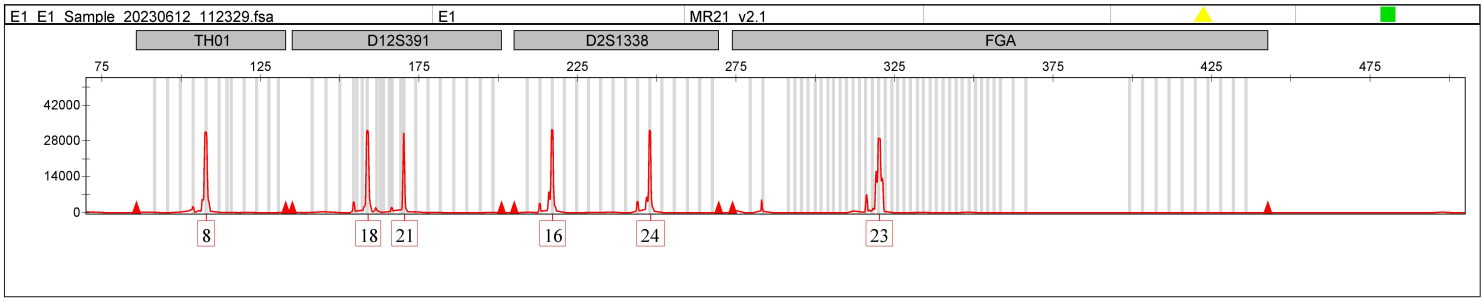
E1_E1_Sample_20230612_112329.fsa | E1 | MR21_v2.1



E1_E1_Sample_20230612_112329.fsa | E1 | MR21_v2.1



E1_E1_Sample_20230612_112329.fsa | E1 | MR21_v2.1



The above is a Chinese A375 cell STR testing report from Henan Engineering Research Center of Industrial Microbiology of China. To better understand, we have translated this report into English and presented below.

Cell STR Testing Report

1. Sample Name/ID: A-375
2. Testing Item: STR Genotyping Test
3. Testing Method: DNA extraction was performed using TianGen's genomic extraction kit. The 21-STR amplification protocol was applied, and the STR loci as well as the gender gene Amelogenin were analyzed using the Seqstudio genetic analyzer.

4. Testing Result：

(1) General information

Multi-allelic genes	Matched cell line	Cell bank	EV value	Matching explanation
None	A-375	ExPASy	1.00	Complete match

Multi-allelic gene refers to the phenomenon of having three or more alleles.
The cell typing results for this test are satisfactory.

(2) Sample Description

The DNA typing of this cell strain found a cell line that is a perfect match in the cell line retrieval, and the ExPASy database showed that the cell is named A-375 with the corresponding cell number CVCL-0132. No multi-allelic genes were found in this cell line during this test.

(3) Matching Explanation

The cell line under test was compared with the STR data of cell lines recorded in the ATCC, DSMZ, JCRB, and ExPASy databases. Cell lines that are not included in the aforementioned cell line repositories will not be able to find a match.

Appendix: Genotyping results of STR loci and Amelogenin locus for A-375 cells.

A-375 cells			
Marker	Allele 1	Allele 2	Allele 3
D19S433	13	14.2	
D5S818	12	12	
D21S11	29	30	
D18S51	12	17	
D6S1043	11	14	
AMEL	X	X	
D3S1358	15	17	
D13S317	11	14	
D7S820	9	9	
D16S539	9	9	
CSF1PO	11	12	
Penta D	9	15	
D2S441	11	11	
vWA	16	17	
D8S1179	11	14	
TPOX	8	10	
Penta E	10	12	
TH01	8	8	
D12S391	18	21	
D2S1338	16	24	
FGA	23	23	

Appendix: Comparison results with the ExPASy database.

Genotyping scheme and locus distribution

	Plan 1	Plan2	Plan3	Plan4
1	D19S433	AMEL	D2S441	TH01
2	D5S818	D3S1358	vWA	D12S391
3	D21S11	D13S317	D8S1179	D2S1338
4	D18S51	D7S820	TPOX	FGA
5	D6S1043	D16S539	Penta E	
6		CSF1PO		
7		Penta D		

Tested by: Putao Zhang

Reviewed by: Shiteng Yin

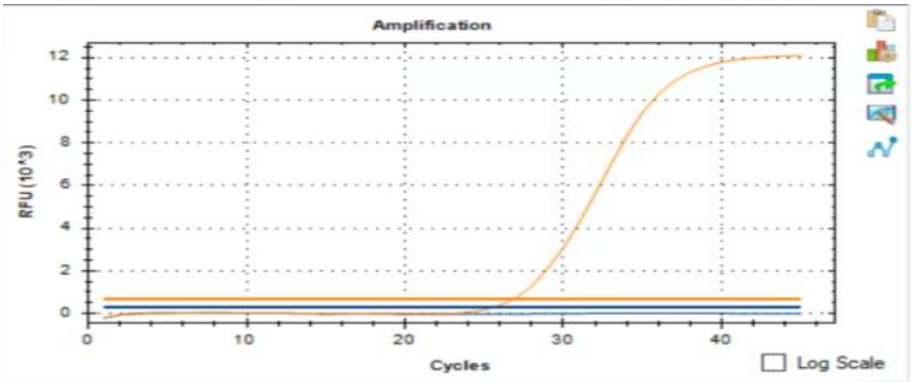
Date of Issuance: 2023.06.12

Henan Engineering Research Center of Industrial Microbiology

Website：www.bncc.com Telephone：400-6699-833

细胞支原体检测报告

检测单位	河南省工业微生物菌种工程技术研究中心		
检测方法	实时荧光定量 PCR 法		
检测项目	A375 细胞支原体检测		
质检员	代文灵	**审核员**	何立胜
检验日期	2023.09.14	**报告日期**	2023.09.14

检测结果	**CT 值**	**通道**	**CT 值**
		FAM	N/A
		ROX	26.86
	曲线图		

检验及判定依据	FAM Channel	ROX Channel	Interpretation
	Positive control	Ct＜35	Reagents works
	Positive control	Ct＞35	Reagents not work
	Sample no Ct	Ct＜35	Negative
	Sample Ct＜40	Ct＜35	Positive
	Sample no Ct	Ct＞35	PCR inhibition

检测结论	**A375 细胞未受到支原体污染**

The above is a Chinese report from the Industrial Microbial Strain Engineering Technology Research Center regarding the detection of mycoplasma in A375 cells. To better understand, we have translated this report into English and presented below.

Cell Mycoplasma Detection Reports

<table>
<tr><td colspan="2">Department</td><td colspan="4">Industrial Microbial Strain Engineering Technology Research Center of Henan Province, China</td></tr>
<tr><td colspan="2">Method</td><td colspan="4">Real-time fluorescent quantitative PCR method</td></tr>
<tr><td colspan="2">Test Item</td><td colspan="4">Mycoplasma detection of A375 cells</td></tr>
<tr><td colspan="2">Quality inspector</td><td>Wenling Dai</td><td>Auditor</td><td colspan="2">Lisheng He</td></tr>
<tr><td colspan="2">Inspection date</td><td>2023.09.14</td><td>Report time</td><td colspan="2">2023.09.14</td></tr>
<tr><td rowspan="4">Result</td><td rowspan="3">CT Value</td><td colspan="2">Channel</td><td colspan="2">CT Value</td></tr>
<tr><td colspan="2">FAM</td><td colspan="2">N/A</td></tr>
<tr><td colspan="2">ROX</td><td colspan="2">26.86</td></tr>
<tr><td>Curve figure</td><td colspan="4"></td></tr>
<tr><td colspan="2" rowspan="6">Inspection and judgment basis</td><td>FAM Channel</td><td>ROX Channel</td><td colspan="2">Interpretation</td></tr>
<tr><td>Positive control</td><td>Ct＜35</td><td colspan="2">Reagents works</td></tr>
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<tr><td>Sample no Ct</td><td>Ct＞35</td><td colspan="2">PCR inhibition</td></tr>
<tr><td colspan="2">Conclusion</td><td colspan="4">A375 cells were not contaminated by mycoplasma</td></tr>
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细胞支原体检测报告

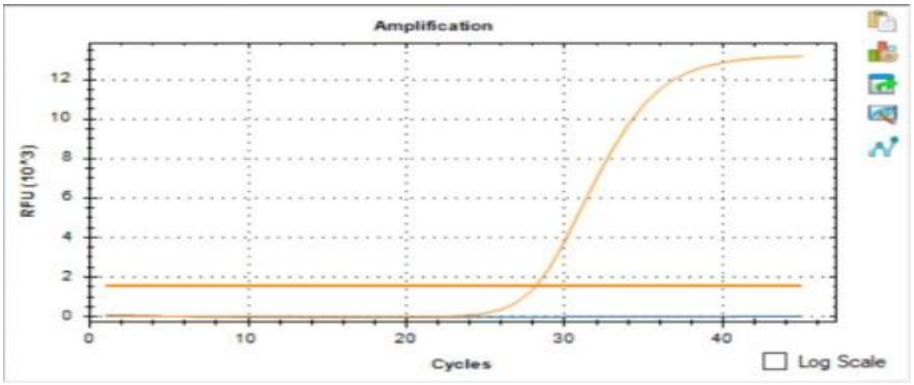
检测单位	河南省工业微生物菌种工程技术研究中心		
检测方法	实时荧光定量 PCR 法		
检测项目	SK-MEL-28 细胞支原体检测		
质检员	代文灵	**审核员**	何立胜
检验日期	2023.09.14	**报告日期**	2023.09.14

检测结果

CT 值

通道	**CT 值**
FAM	N/A
ROX	28.06

曲线图



检验及判定依据

FAM Channel	ROX Channel	Interpretation
Positive control	Ct＜35	Reagents works
Positive control	Ct＞35	Reagents not work
Sample no Ct	Ct＜35	Negative
Sample Ct＜40	Ct＜35	Positive
Sample no Ct	Ct＞35	PCR inhibition

检测结论	SK-MEL-28 细胞未受到支原体污染

The above is a Chinese report from the Industrial Microbial Strain Engineering Technology Research Center regarding the detection of mycoplasma in SK-MEL-28 cells.
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<tr><td colspan="3">FAM</td><td colspan="2">N/A</td></tr>
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