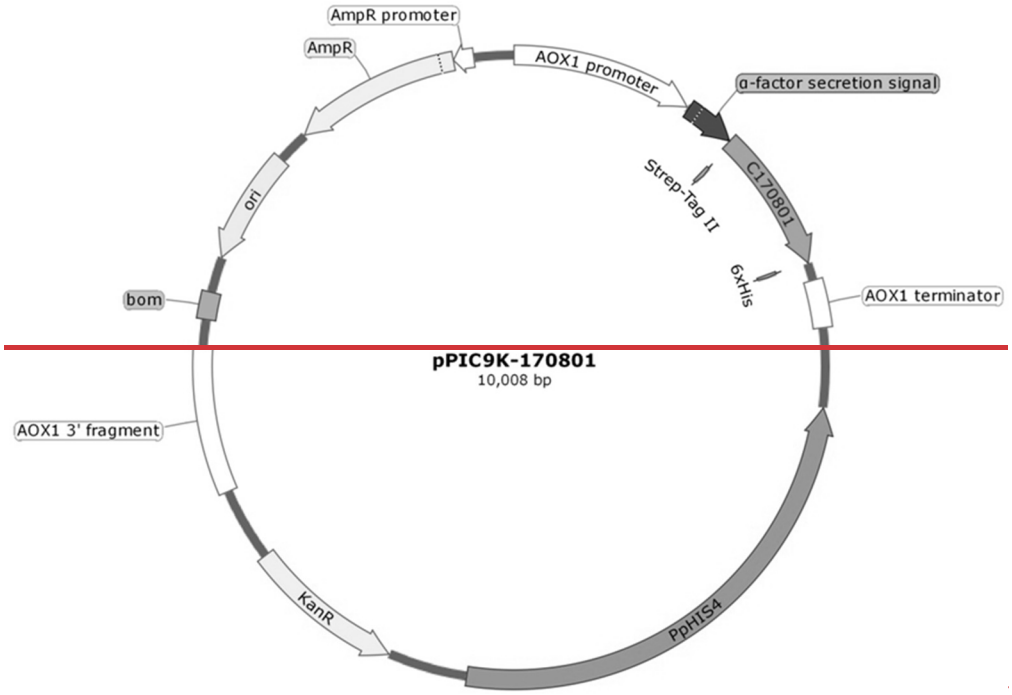

AmpR promoter
AmpR
AOX1 promoter
α-factor secretion signal
Strep-Tag II
C170801
6xHis
AOX1 terminator
ori
bom
pPIC9K-170801
10,008 bp
AOX1 3' fragment
KanR
PpHIS4

FIG. 1

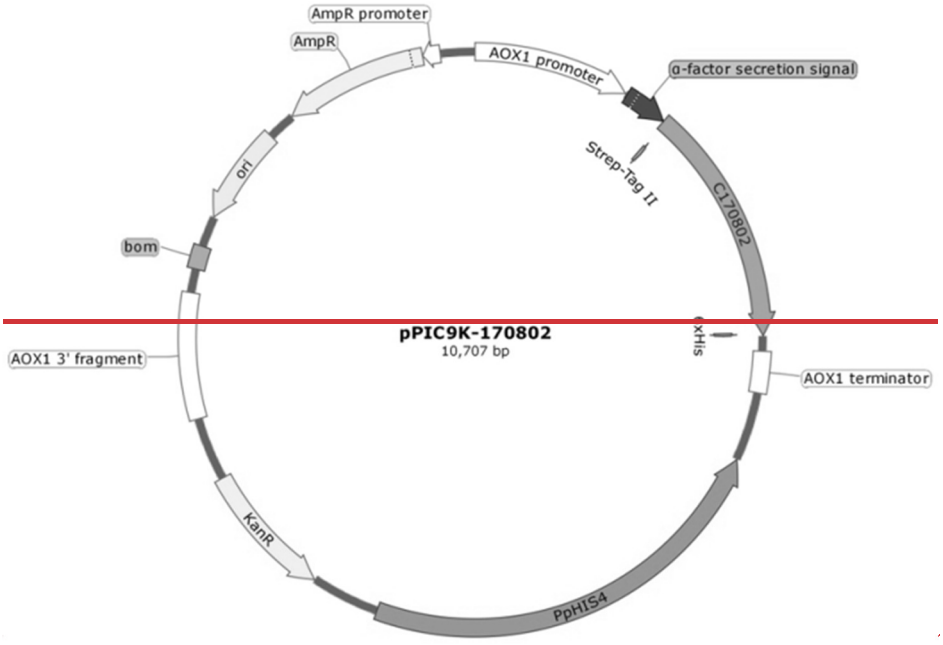

AmpR promoter
AmpR
AOX1 promoter
α-factor secretion signal
Strep-Tag II
C170802
6xHis
AOX1 terminator
ori
bom
pPIC9K-170802
10,707 bp
AOX1 3' fragment
KanR
PpHIS4

FIG. 2

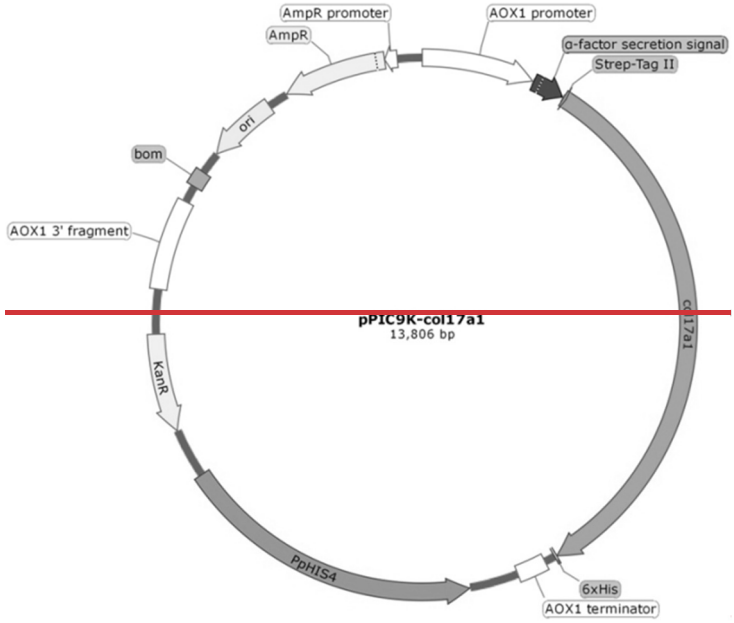

AmpR promoter
AmpR
AOX1 promoter
α-factor secretion signal
Strep-Tag II
ori
bom
AOX1 3' fragment
pPIC9K-col17a1
13,806 bp
col17a1
KanR
PpHIS4
6xHis
AOX1 terminator

FIG. 3

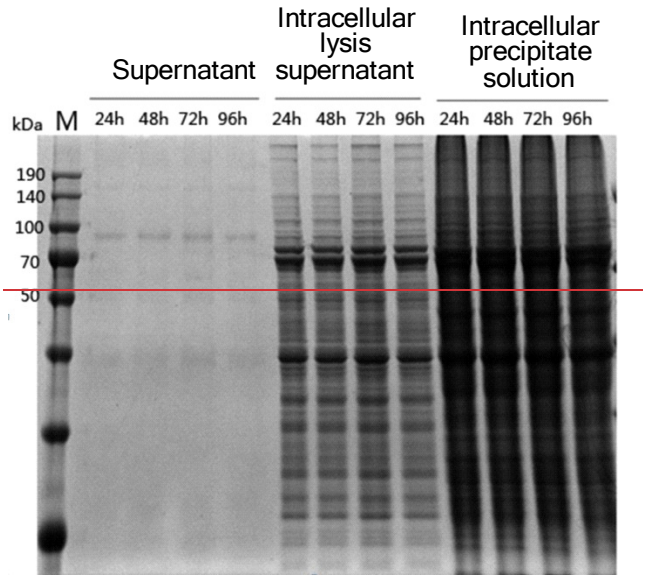

Supernatant
Intracellular lysis supernatant
Intracellular precipitate solution
kDa M
24h 48h 72h 96h
24h 48h 72h 96h
24h 48h 72h 96h
190
140
100
70
50

FIG. 4

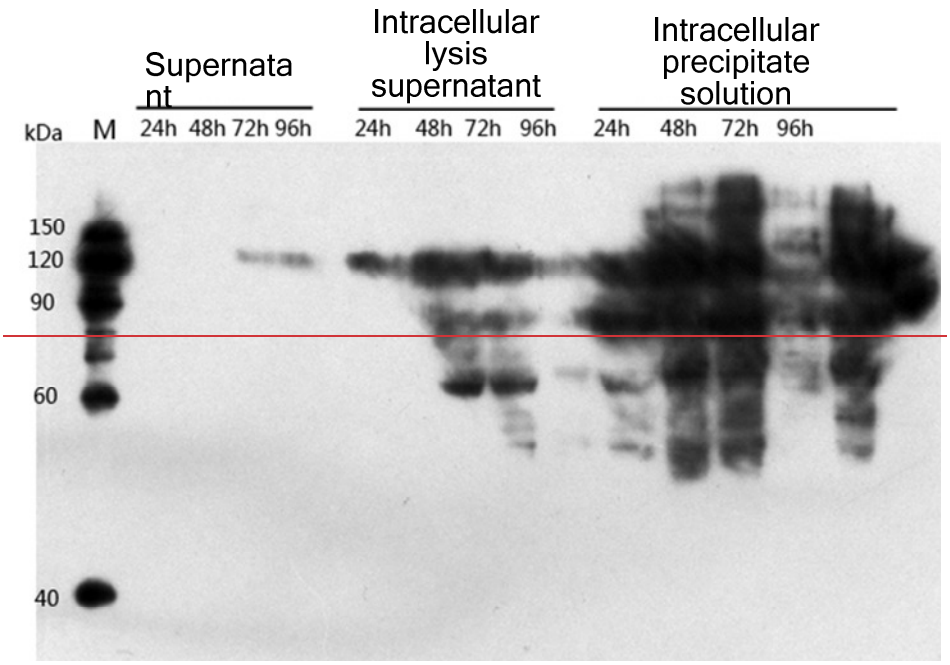

Supernatant
Intracellular lysis supernatant
Intracellular precipitate solution
kDa
M
24h 48h 72h 96h
24h 48h 72h 96h
24h 48h 72h 96h
150
120
90
60
40

FIG. 5

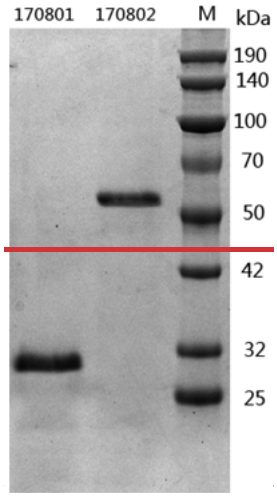

170801
170802
M
kDa
190
140
100
70
50
42
32
25

FIG. 6

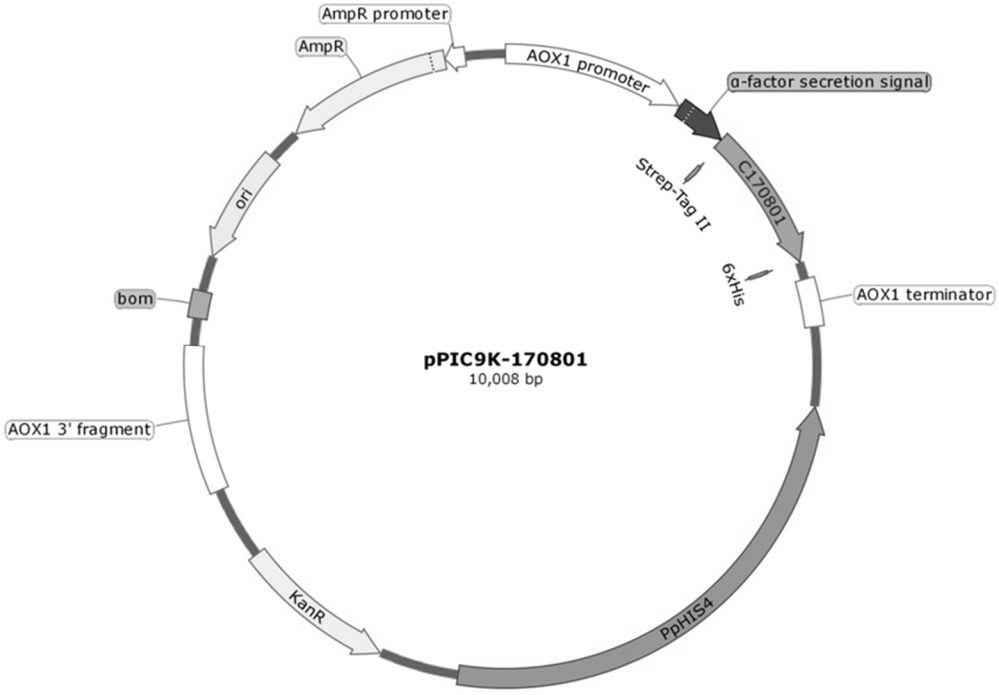

AmpR promoter
AmpR
AOX1 promoter
α-factor secretion signal
Strep-Tag II
C170801
6xHis
AOX1 terminator
ori
bom
pPIC9K-170801
10,008 bp
AOX1 3' fragment
KanR
PpHIS4

FIG. 1

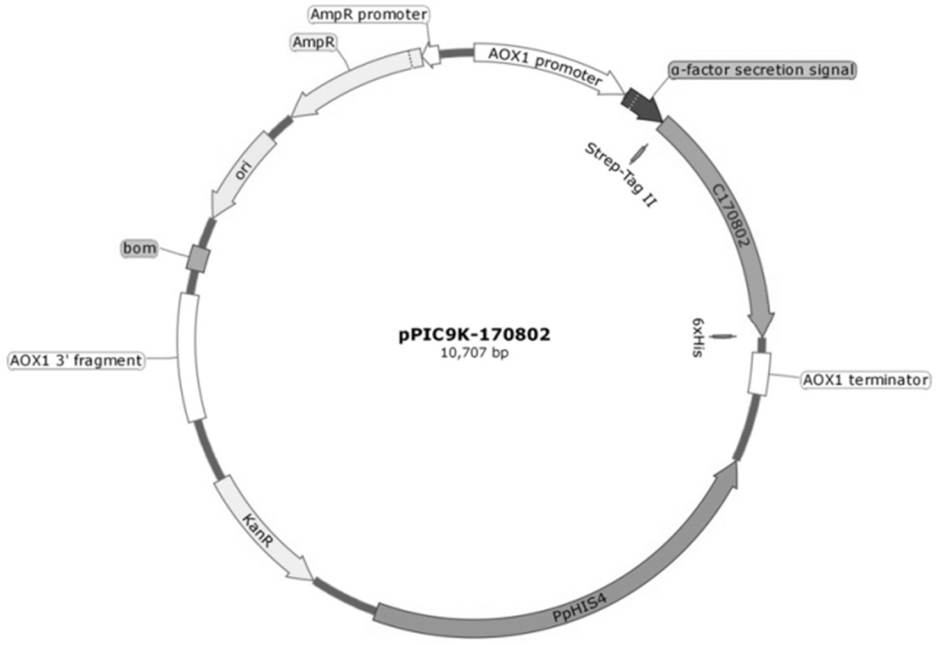

AmpR promoter
AmpR
AOX1 promoter
α-factor secretion signal
Strep-Tag II
C170802
6xHis
AOX1 terminator
ori
bom
pPIC9K-170802
10,707 bp
AOX1 3' fragment
KanR
PpHIS4

FIG. 2

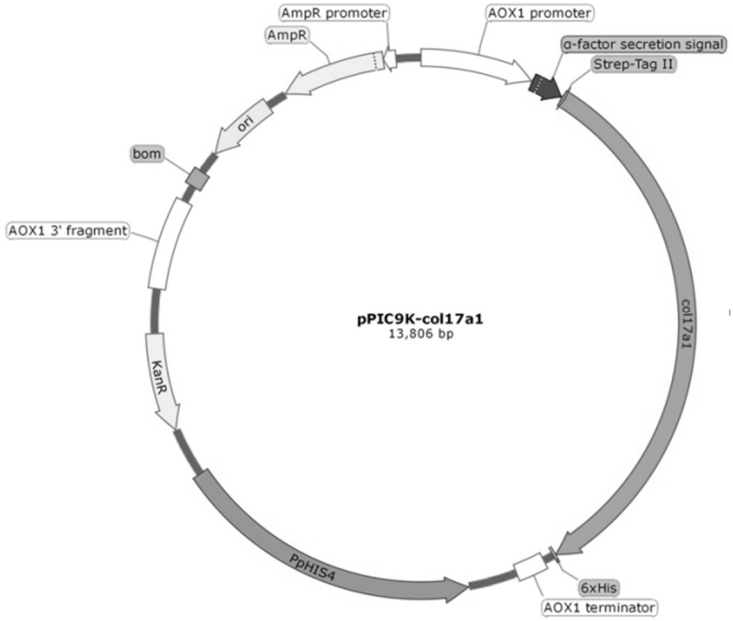

AmpR promoter
AmpR
AOX1 promoter
α-factor secretion signal
Strep-Tag II
ori
bom
AOX1 3' fragment
pPIC9K-col17a1
13,806 bp
col17a1
KanR
PpHIS4
6xHis
AOX1 terminator

FIG. 3

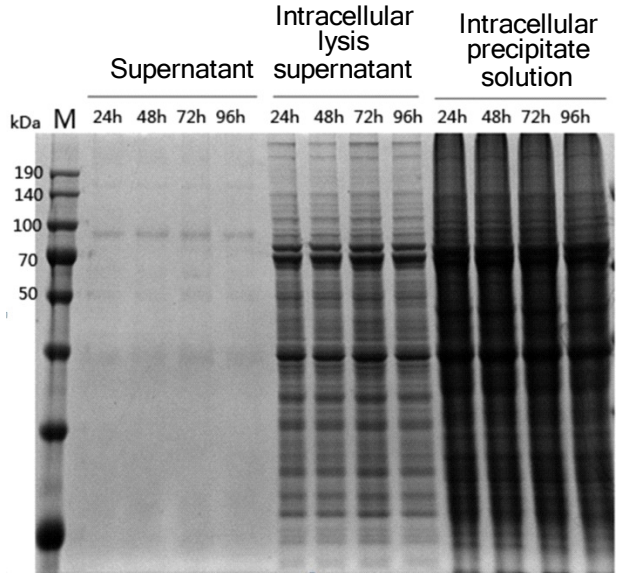

Supernatant
Intracellular lysis supernatant
Intracellular precipitate solution
kDa
M
24h 48h 72h 96h
24h 48h 72h 96h
24h 48h 72h 96h
190
140
100
70
50

FIG. 4

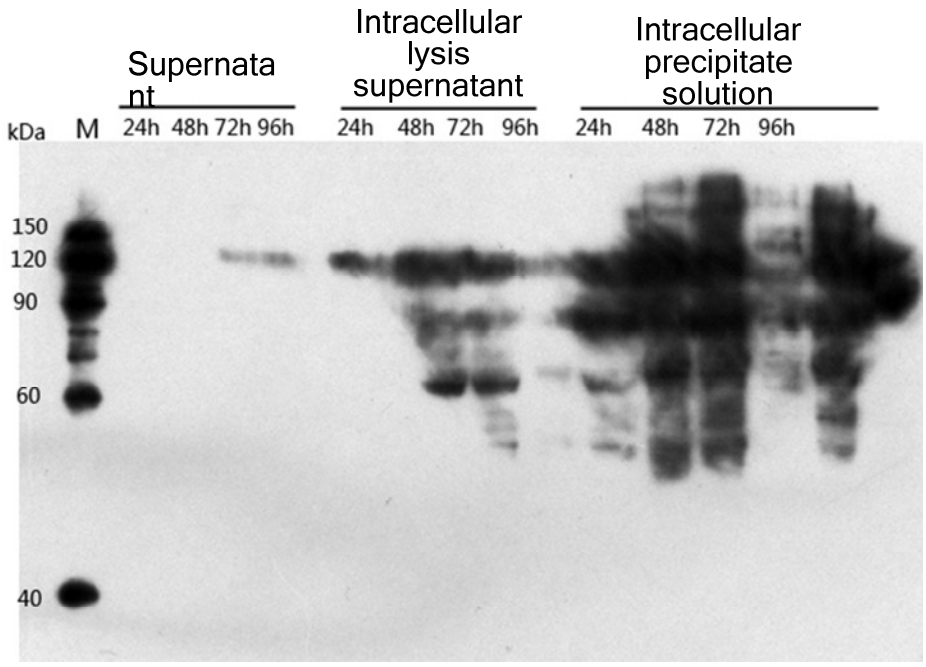

Supernatant
Intracellular lysis supernatant
Intracellular precipitate solution
kDa
M
24h 48h 72h 96h
24h 48h 72h 96h
24h 48h 72h 96h
150
120
90
60
40

FIG. 5

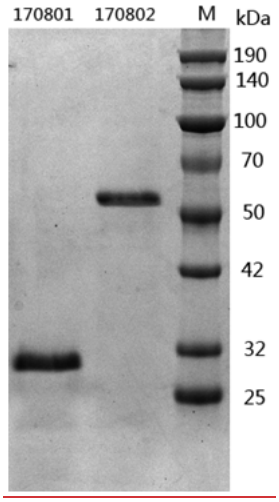

170801
170802
M
kDa
190
140
100
70
50
42
32
25

FIG. 6

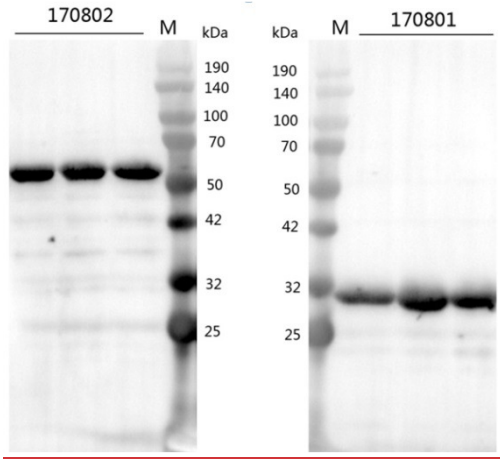

170802
M
kDa
190
140
100
70
50
42
32
25
kDa
M
170801
190
140
100
70
50
42
32
25

FIG. 7

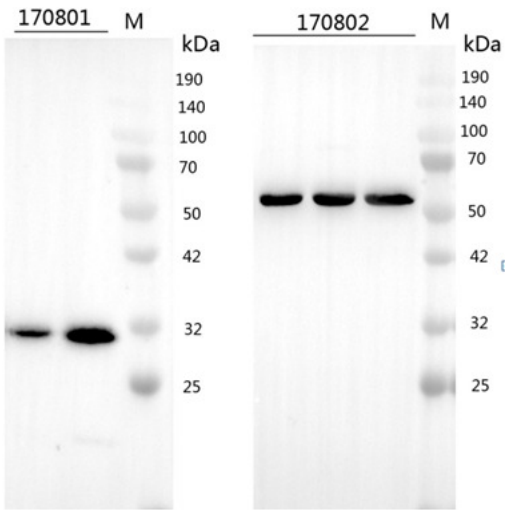

170801
M
kDa
190
140
100
70
50
42
32
25
170802
M
kDa
190
140
100
70
50
42
32
25

FIG. 8

LIJIA_170801S_060320-16							
Hits	Protein Mass	No. of Peptide	Sequence Header	Link	Relative Abundance	Probability	No. of Unique Peptide
1	150725.48	23	>sp|Q9UMD9|COHA1_HUMAN Collagen alpha-1(XVII) chain OS=Homo sapiens GN=COL17A1 PE=1 SV=3	Q9UMD9	100.0%	99.0%	8

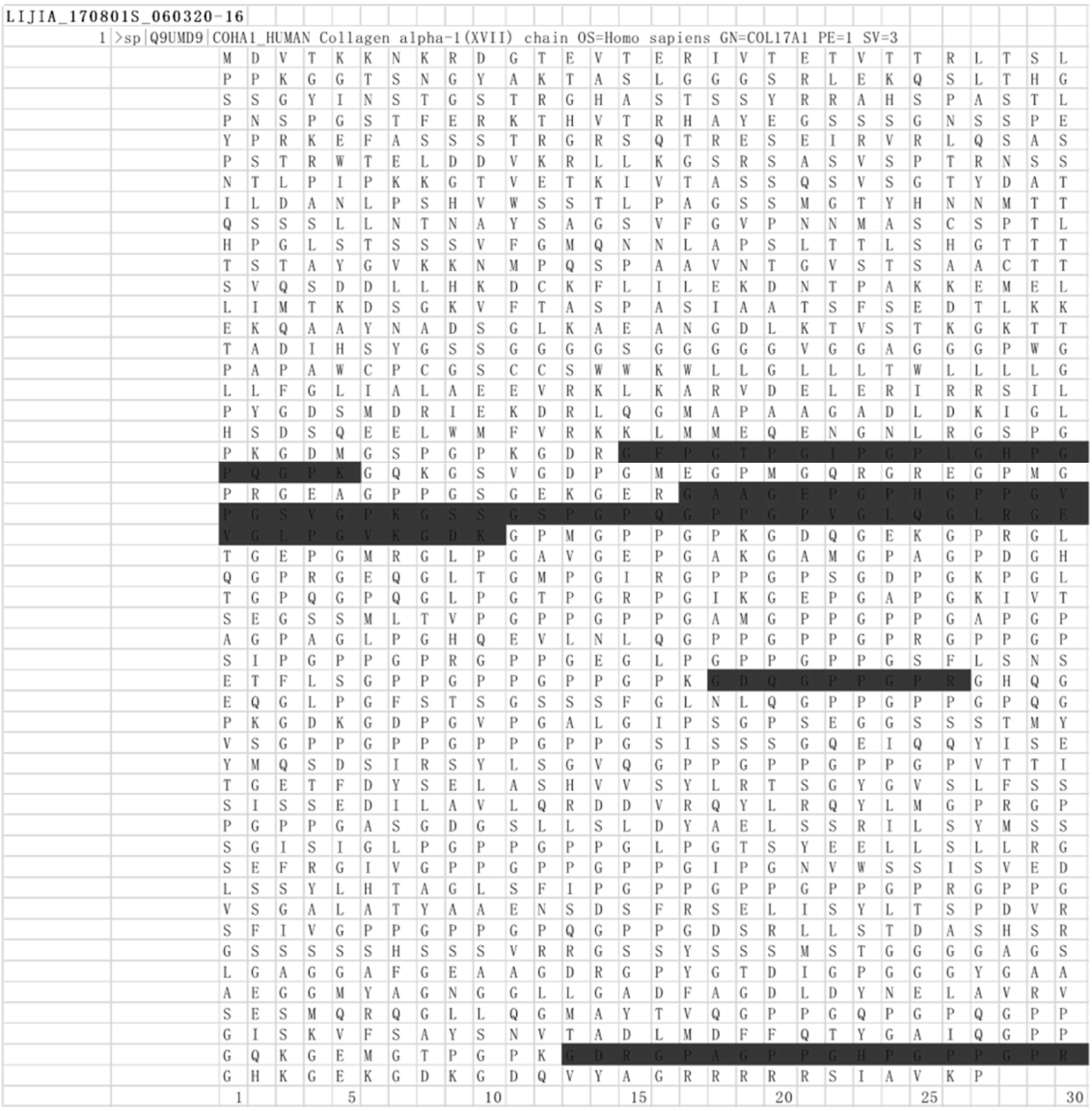


FIG. 9

LIJIA_170802S_062620						
Hits	Protein Mass	No. of Peptide	Sequence Header	Link	Relative Abundance	Probability	No. of Unique Peptide
1	150725.48	17	>sp|Q9UMD9|COHA1_HUMAN Collagen alpha-1(XVII) chain OS=Homo sapiens GN=COL17A1 PE=1 SV=3	Q9UMD9	100.0%	99.0%	5

LIJIA 170802S 062620

1 >sp|Q9UMD9|COHA1_HUMAN Collagen alpha-1(XVII) chain OS=Homo sapiens GN=COL17A1 PE=1 SV=3

M	D	V	T	K	K	N	K	R	D	G	T	E	V	T	E	R	I	V	T	E	T	V	T	T	R	L	T	S	L
P	P	K	G	G	T	S	N	G	Y	A	K	T	A	S	L	G	G	G	S	R	L	E	K	Q	S	L	T	H	G
S	S	G	Y	I	N	S	T	G	S	T	R	G	H	A	S	T	S	S	Y	R	R	A	H	S	P	A	S	T	L
P	N	S	P	G	S	T	F	E	R	K	T	H	V	T	R	H	A	Y	E	G	S	S	S	G	N	S	S	P	E
Y	P	R	K	E	F	A	S	S	S	T	R	G	R	S	Q	T	R	E	S	E	I	R	V	R	L	Q	S	A	S
P	S	T	R	W	T	E	L	D	D	V	K	R	L	L	K	G	S	R	S	A	S	V	S	P	T	R	N	S	S
N	T	L	P	I	P	K	K	G	T	V	E	T	K	I	V	T	A	S	S	Q	S	V	S	G	T	Y	D	A	T
I	L	D	A	N	L	P	S	H	V	W	S	S	T	L	P	A	G	S	S	M	G	T	Y	H	N	N	M	T	T
Q	S	S	S	L	L	N	T	N	A	Y	S	A	G	S	V	F	G	V	P	N	N	M	A	S	C	S	P	T	L
H	P	G	L	S	T	S	S	S	V	F	G	M	Q	N	N	L	A	P	S	L	T	T	L	S	H	G	T	T	T
T	S	T	A	Y	G	V	K	K	N	M	P	Q	S	P	A	A	V	N	T	G	V	S	T	S	A	A	C	T	T
S	V	Q	S	D	D	L	L	H	K	D	C	K	F	L	I	L	E	K	D	N	T	P	A	K	K	E	M	E	L
L	I	M	T	K	D	S	G	K	V	F	T	A	S	P	A	S	I	A	A	T	S	F	S	E	D	T	L	K	K
E	K	Q	A	A	Y	N	A	D	S	G	L	K	A	E	A	N	G	D	L	K	T	V	S	T	K	G	K	T	T
T	A	D	I	H	S	Y	G	S	S	G	G	G	G	S	G	G	G	G	G	V	G	G	A	G	G	G	P	W	G
P	A	P	A	W	C	P	C	G	S	C	C	S	W	W	K	W	L	L	G	L	L	L	T	W	L	L	L	L	G
L	L	F	G	L	I	A	L	A	E	E	V	R	K	L	K	A	R	V	D	E	L	E	R	I	R	R	S	I	L
P	Y	G	D	S	M	D	R	I	E	K	D	R	L	Q	G	M	A	P	A	A	G	A	D	L	D	K	I	G	L
H	S	D	S	Q	E	E	L	W	M	F	V	R	K	K	L	M	M	E	Q	E	N	G	N	L	R	G	S	P	G
P	K	G	D	M	G	S	P	G	P	K	G	D	R	G	F	P	G	T	P	G	T	P	G	P	L	G	H	P	G
P	Q	G	P	K	G	Q	K	G	S	V	G	D	P	G	M	E	G	P	M	G	Q	R	G	R	E	G	P	M	G
P	R	G	E	A	G	P	P	G	S	G	E	K	G	E	R	G	A	A	G	E	P	G	P	H	G	P	P	G	V
P	G	S	V	G	P	K	G	S	S	G	S	P	G	P	Q	G	P	P	G	P	V	G	L	Q	G	L	R	G	E
V	G	L	P	G	V	K	G	D	K	G	P	M	G	P	P	G	P	K	G	D	Q	G	E	K	G	P	R	G	L
T	G	E	P	G	M	R	G	L	P	G	A	V	G	E	P	G	A	K	G	A	M	G	P	A	G	P	D	G	H
Q	G	P	R	G	E	Q	G	L	T	G	M	P	G	I	R	G	P	P	G	P	S	G	D	P	G	K	P	G	L
T	G	P	Q	G	P	Q	G	L	P	G	T	P	G	R	P	G	I	K	G	E	P	G	A	P	G	K	I	V	T
S	E	G	S	S	M	L	T	V	P	G	P	P	G	P	P	G	A	M	G	P	P	G	P	P	G	A	P	G	P
A	G	P	A	G	L	P	G	H	Q	E	V	L	N	L	Q	G	P	P	G	P	P	G	P	R	G	P	P	G	P
S	I	P	G	P	P	G	P	R	G	P	P	G	E	G	L	P	G	P	P	G	P	P	G	S	F	L	S	N	S
E	T	F	L	S	G	P	P	G	P	P	G	P	P	G	P	K	G	D	Q	G	P	P	G	P	R	G	H	Q	G
E	Q	G	L	P	G	F	S	T	S	G	S	S	S	F	G	L	N	L	Q	G	P	P	G	P	P	G	P	Q	G
P	K	G	D	K	G	D	P	G	V	P	G	A	L	G	I	P	S	G	P	S	E	G	G	S	S	S	T	M	Y
V	S	G	P	P	G	P	P	G	P	P	G	P	P	G	S	I	S	S	S	G	Q	E	I	Q	Q	Y	I	S	E
Y	M	Q	S	D	S	I	R	S	Y	L	S	G	V	Q	G	P	P	G	P	P	G	P	P	G	P	V	T	T	I
T	G	E	T	F	D	Y	S	E	L	A	S	H	V	V	S	Y	L	R	T	S	G	Y	G	V	S	L	F	S	S
S	I	S	S	E	D	I	L	A	V	L	Q	R	D	D	V	R	Q	Y	L	R	Q	Y	L	M	G	P	R	G	P
P	G	P	P	G	A	S	G	D	G	S	L	L	S	L	D	Y	A	E	L	S	S	R	I	L	S	Y	M	S	S
S	G	I	S	I	G	L	P	G	P	P	G	P	P	G	L	P	G	T	S	Y	E	E	L	L	S	L	L	R	G
S	E	F	R	G	I	V	G	P	P	G	P	P	G	P	P	G	I	P	G	N	V	W	S	S	I	S	V	E	D
L	S	S	Y	L	H	T	A	G	L	S	F	I	P	G	P	P	G	P	P	G	P	P	G	P	R	G	P	P	G
V	S	G	A	L	A	T	Y	A	A	E	N	S	D	S	F	R	S	E	L	I	S	Y	L	T	S	P	D	V	R
S	F	I	V	G	P	P	G	P	P	G	P	Q	G	P	P	G	D	S	R	L	L	S	T	D	A	S	H	S	R
G	S	S	S	S	H	S	S	S	V	R	R	G	S	S	Y	S	S	S	M	S	T	G	G	G	G	A	G	S	
L	G	A	G	G	A	F	G	E	A	A	G	D	R	G	P	Y	G	T	D	I	G	P	G	G	G	Y	G	A	A
A	E	G	G	M	Y	A	G	N	G	G	L	L	G	A	D	F	A	G	D	L	D	Y	N	E	L	A	V	R	V
S	E	S	M	Q	R	Q	G	L	L	Q	G	M	A	Y	T	V	Q	G	P	P	G	Q	P	G	P	Q	G	P	P
G	I	S	K	V	F	S	A	Y	S	N	V	T	A	D	L	M	D	F	F	Q	T	Y	G	A	I	Q	G	P	P
G	Q	K	G	E	M	G	T	P	G	P	K	G	D	R	G	P	A	G	P	P	G	H	P	G	P	P	G	P	R
G	H	K	G	E	K	G	D	K	G	D	Q	V	Y	A	G	R	R	R	R	R	S	I	A	V	K	P			
1				5					10					15					20					25					30	

FIG. 10

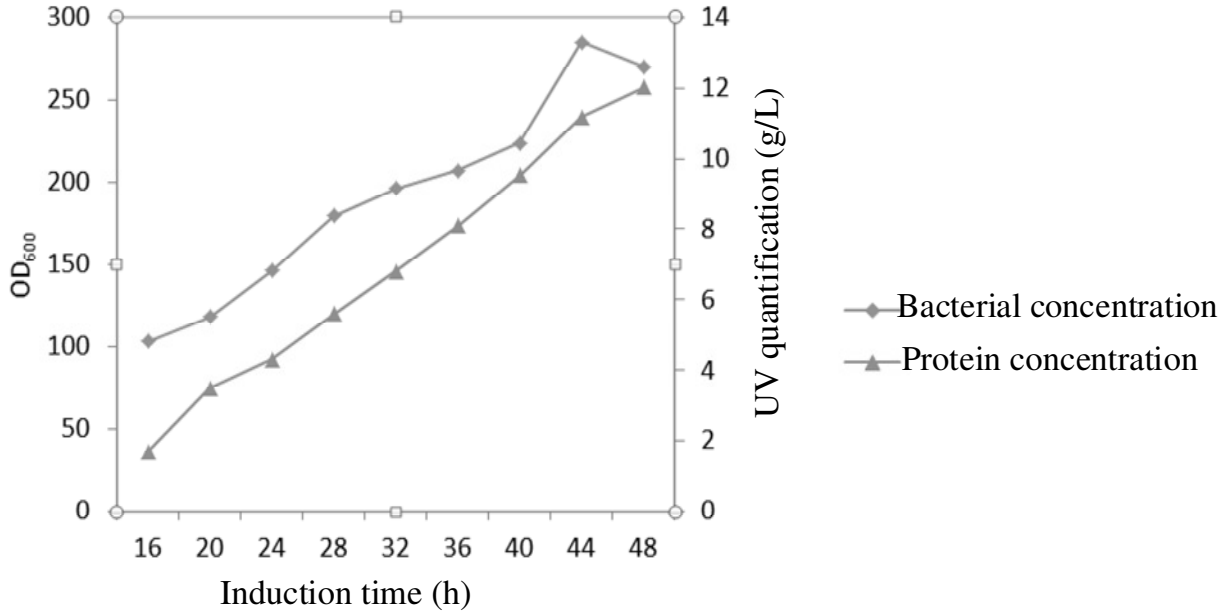

300
250
200
150
100
50
0
OD600
14
12
10
8
6
4
2
0
UV quantification (g/L)
16 20 24 28 32 36 40 44 48
Induction time (h)
Bacterial concentration
Protein concentration

FIG. 11

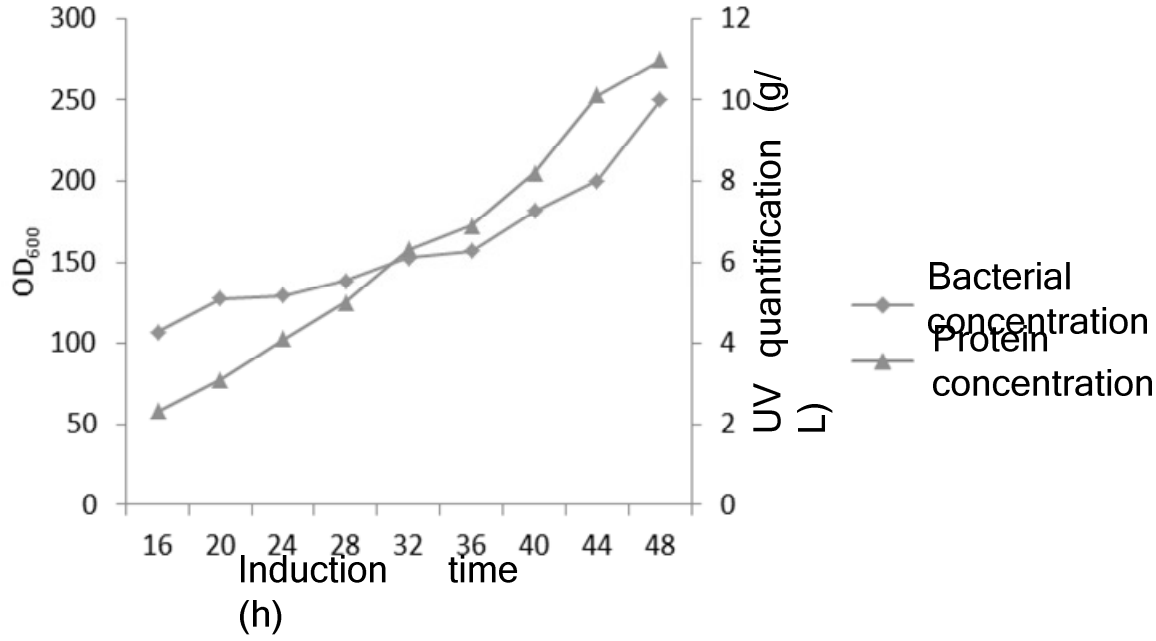

300
250
200
150
100
50
0
OD600
12
10
8
6
4
2
0
UV quantification (g/L)
16 20 24 28 32 36 40 44 48
Induction time (h)
Bacterial concentration
Protein concentration

FIG. 12

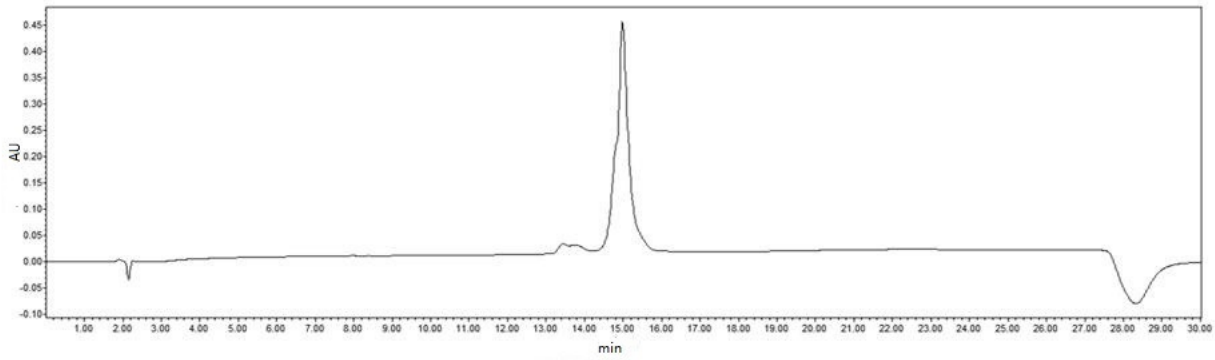

AU
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
-0.05
-0.10
1.00 2.00 3.00 4.00 5.00 6.00 7.00 8.00 9.00 10.00 11.00 12.00 13.00 14.00 15.00 16.00 17.00 18.00 19.00 20.00 21.00 22.00 23.00 24.00 25.00 26.00 27.00 28.00 29.00 30.00
min

FIG. 13

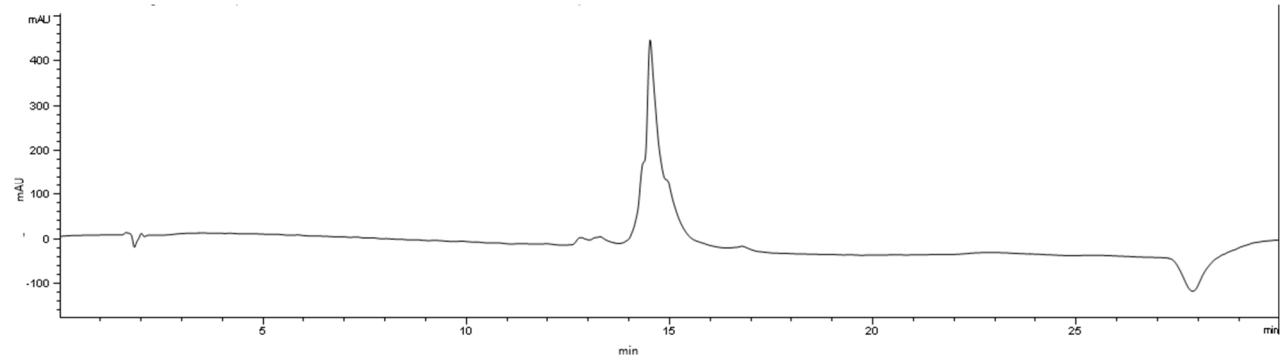


FIG. 14

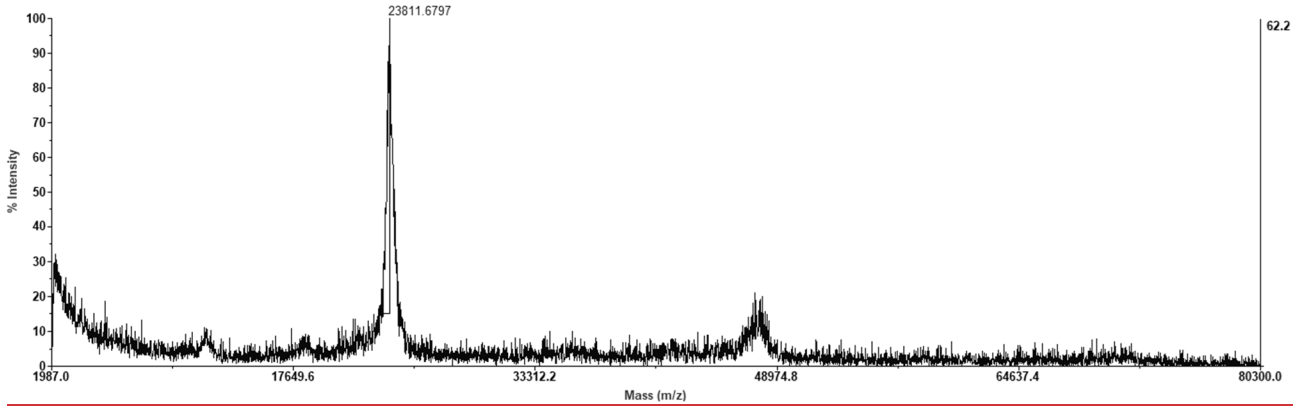


FIG. 15

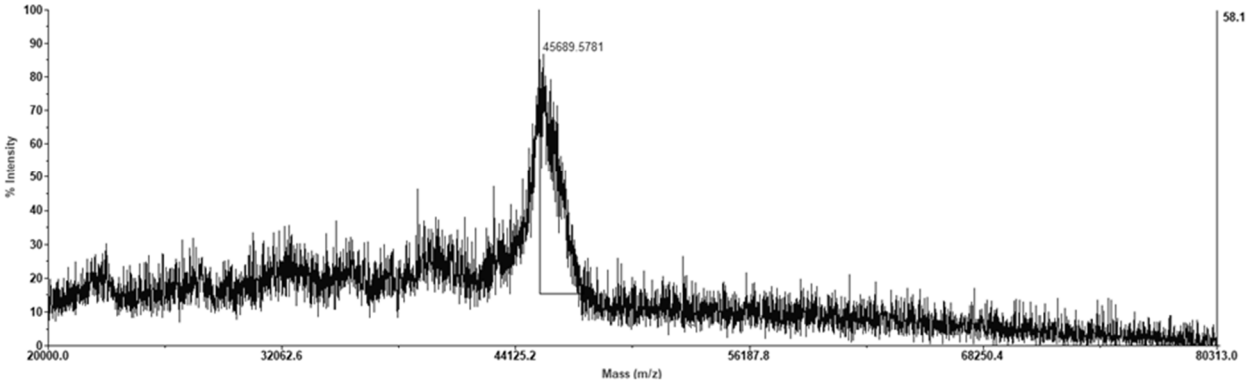


FIG. 16

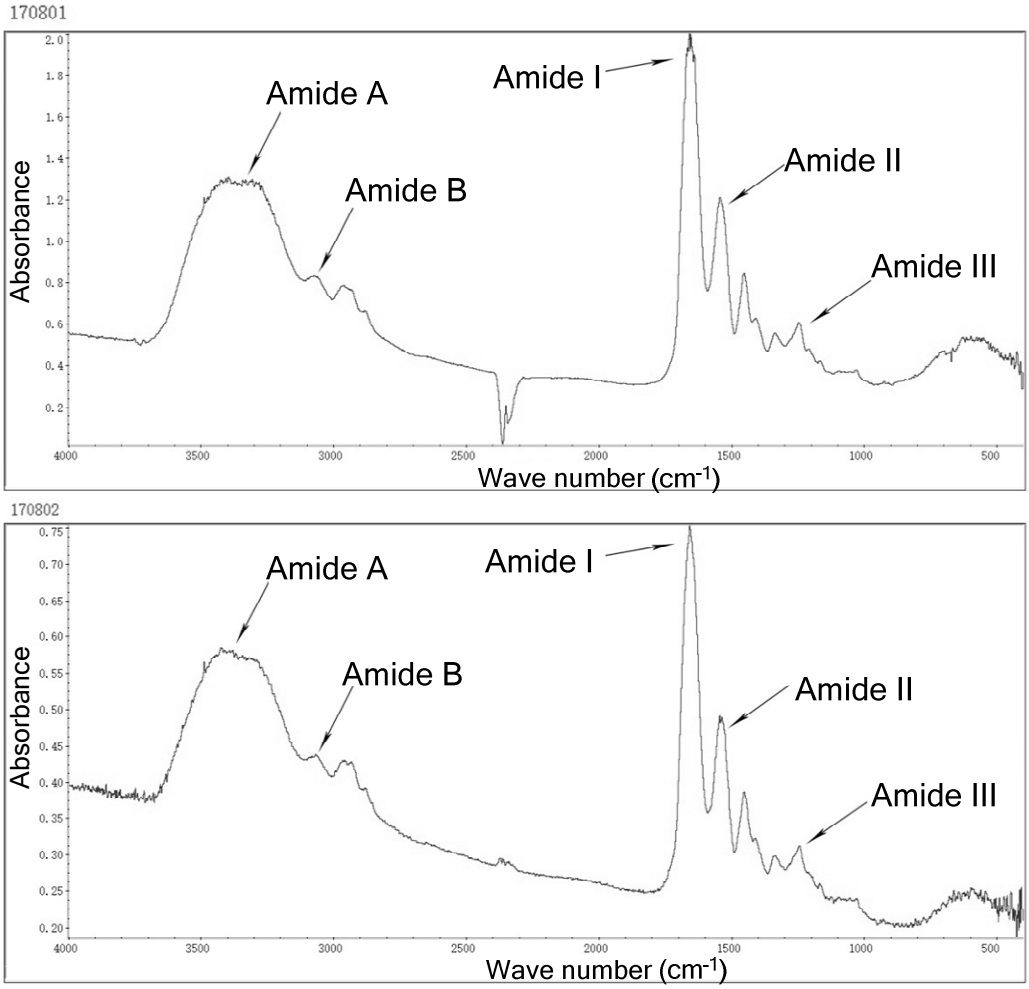

170801
Amide A
Amide I
Amide B
Amide II
Amide III
Absorbance
Wave number (cm-1)
170802
Amide A
Amide I
Amide B
Amide II
Amide III
Absorbance
Wave number (cm-1)

FIG. 17

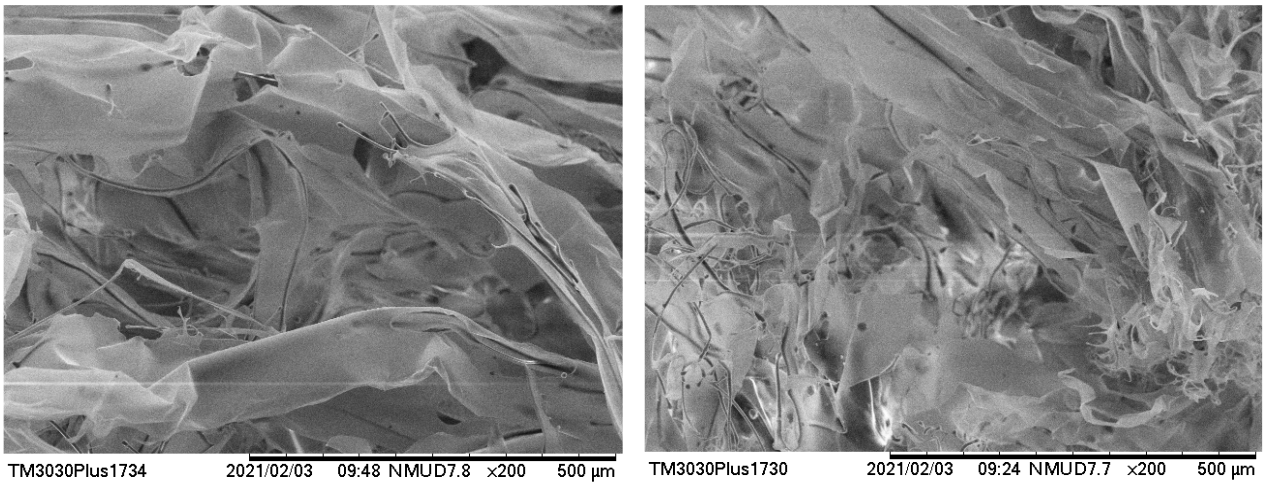

TM3030Plus1734 2021/02/03 09:48 NMUD7.8 ×200 500 µm
TM3030Plus1730 2021/02/03 09:24 NMUD7.7 ×200 500 µm

FIG. 18

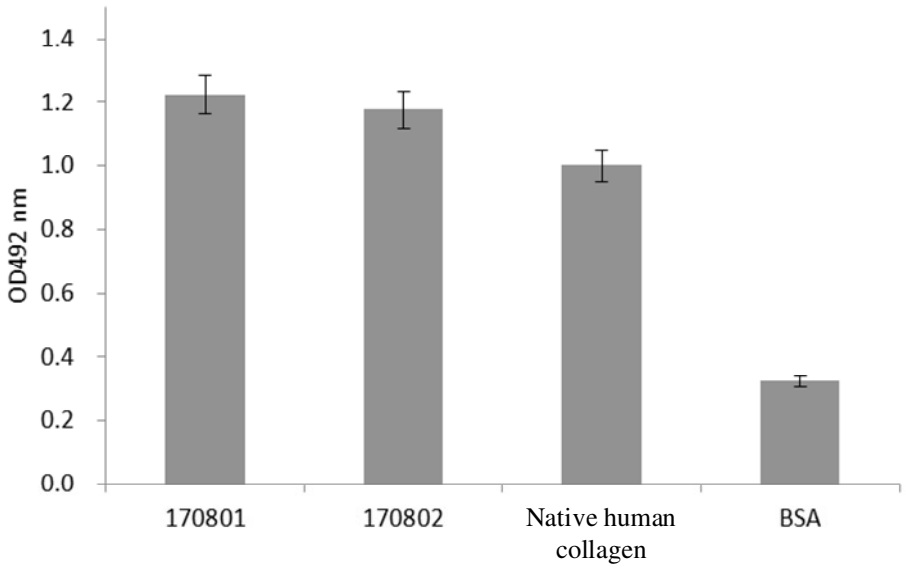

1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
OD492 nm
170801
170802
Native human collagen
BSA

FIG. 19

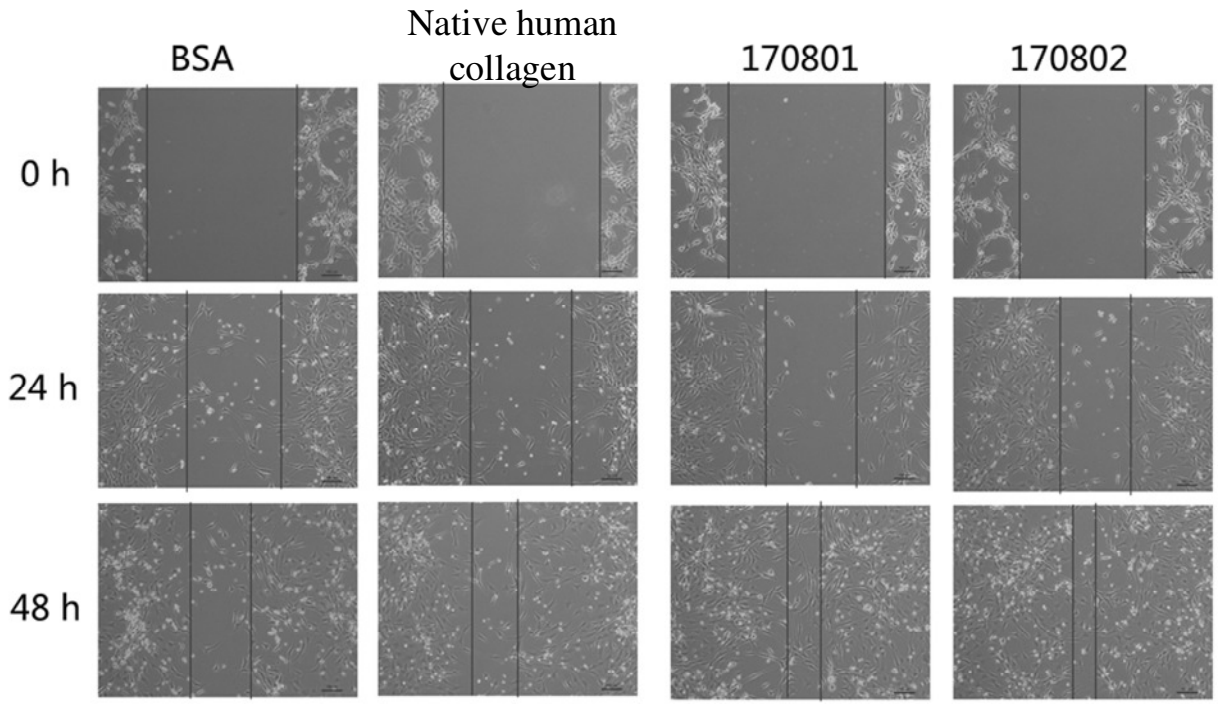

BSA
Native human collagen
170801
170802
0 h
24 h
48 h

FIG. 20

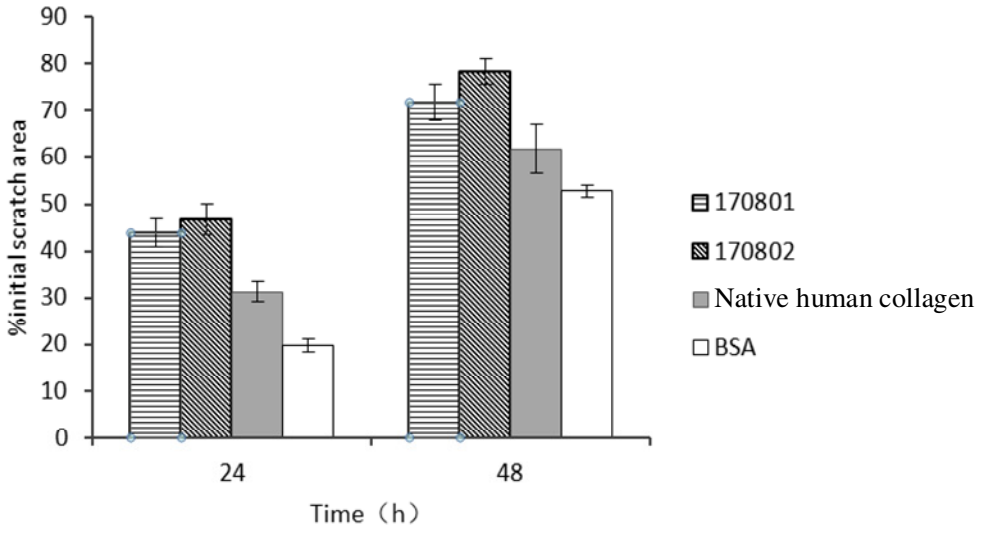

90
80
70
60
50
40
30
20
10
0
%initial scratch area
24
48
Time（h）
170801
170802
Native human collagen
BSA

FIG. 21

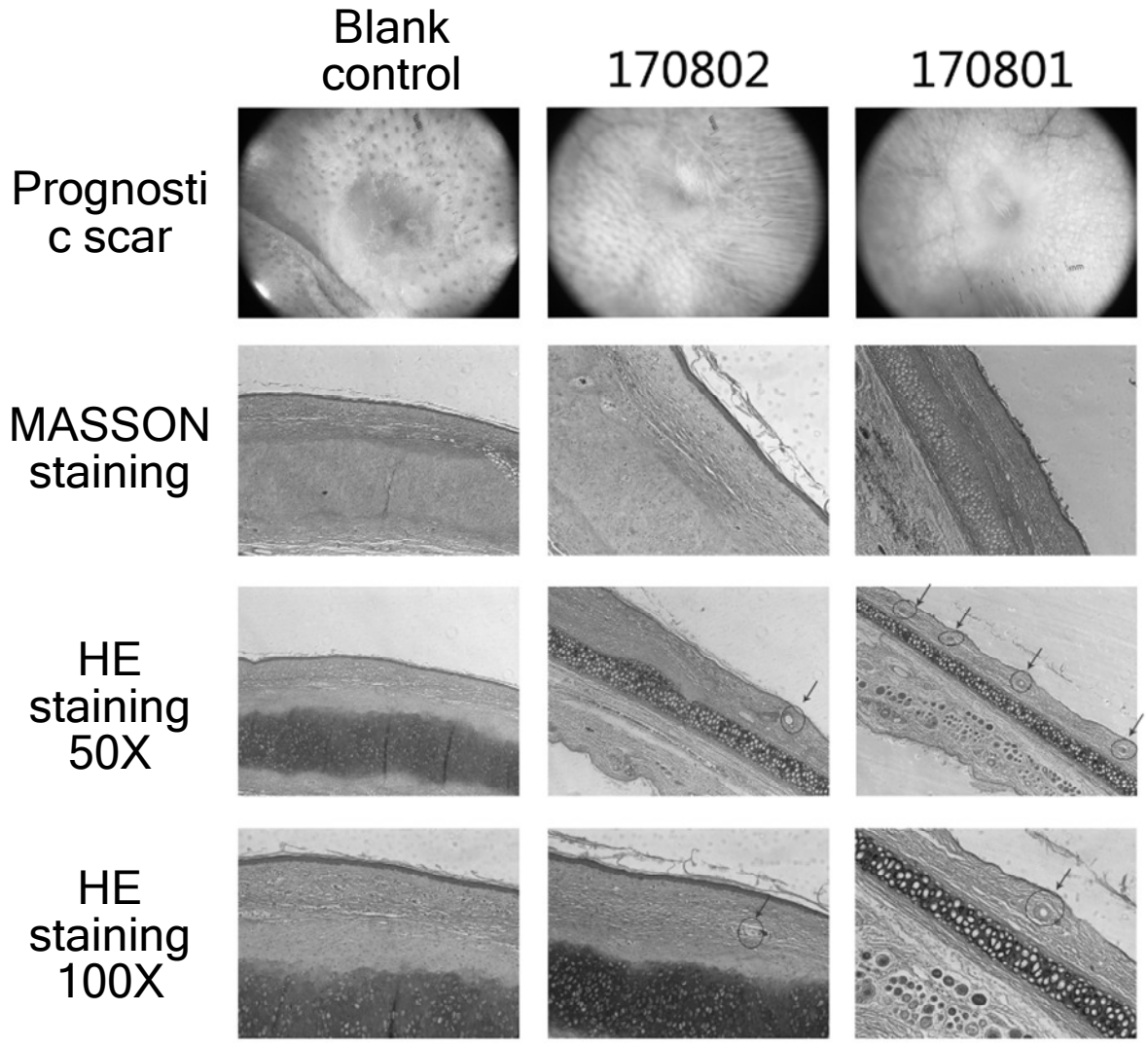

Blank control
170802
170801
Prognosti c scar
MASSON staining
HE staining 50X
HE staining 100X

FIG. 22