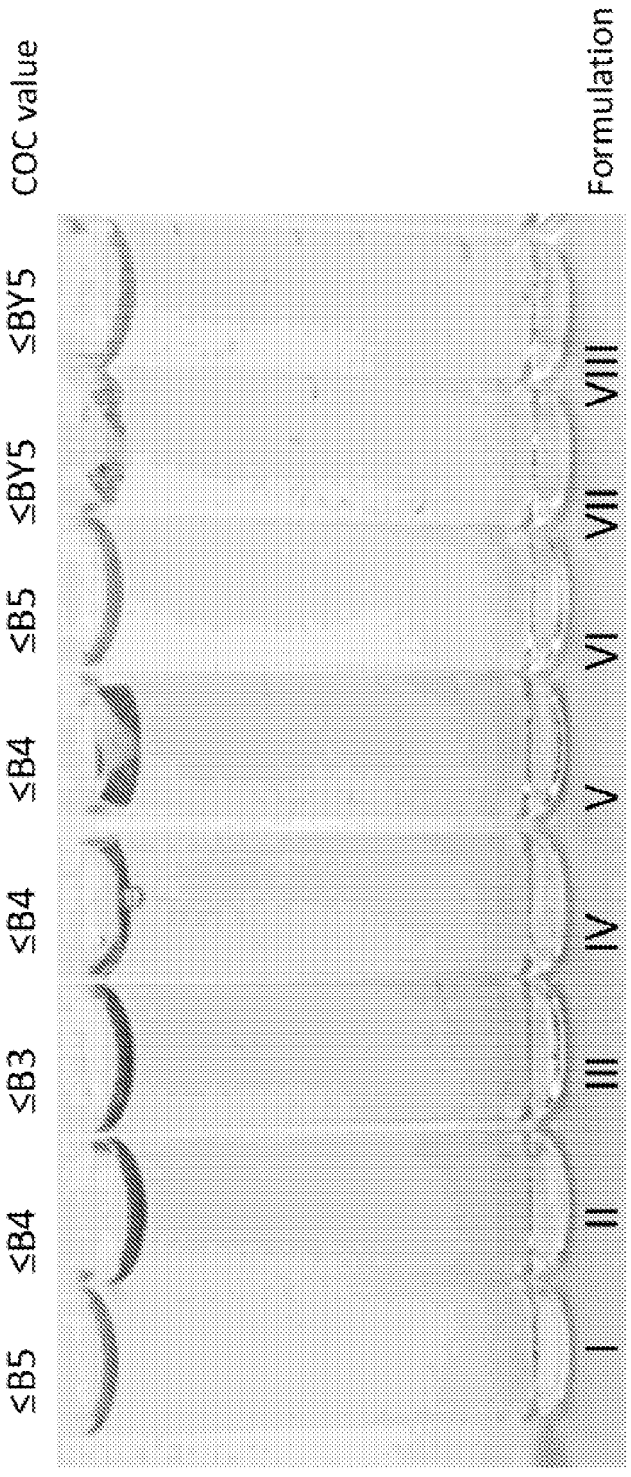

COC value
≤BY5
≤BY5
≤B5
≤B4
≤B4
≤B3
≤B4
≤B5
Formulation
VIII
VII
VI
V
IV
III
II
I

Figure 1

Figure 2

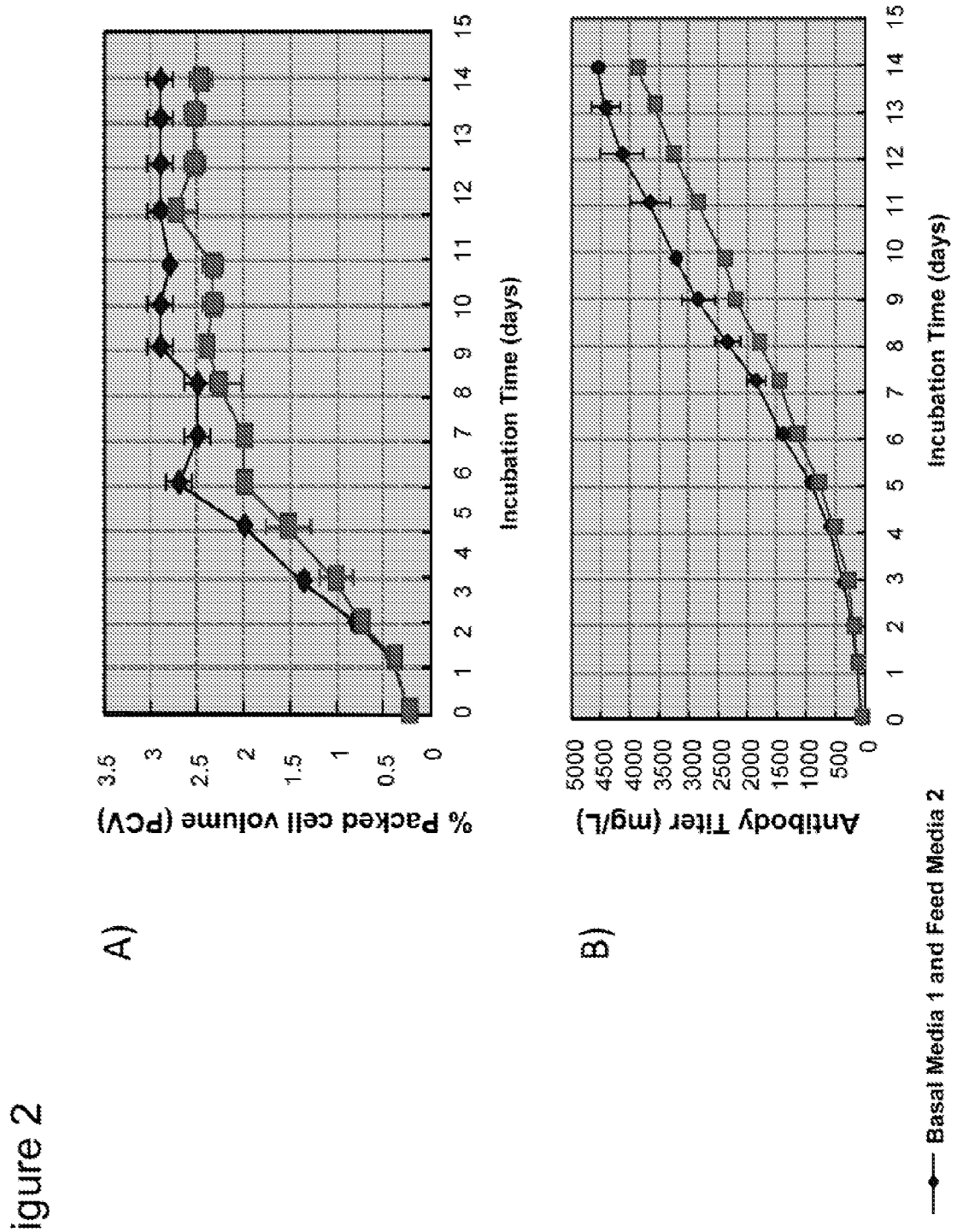

A)
% Packed cell volume (PCV)
3.5
3
2.5
2
1.5
1
0.5
0
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15
Incubation Time (days)
B)
Antibody Titer (mg/L)
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15
Incubation Time (days)
Basal Media 1 and Feed Media 2
Basal Media 3 and Feed Media 4

Figure 2

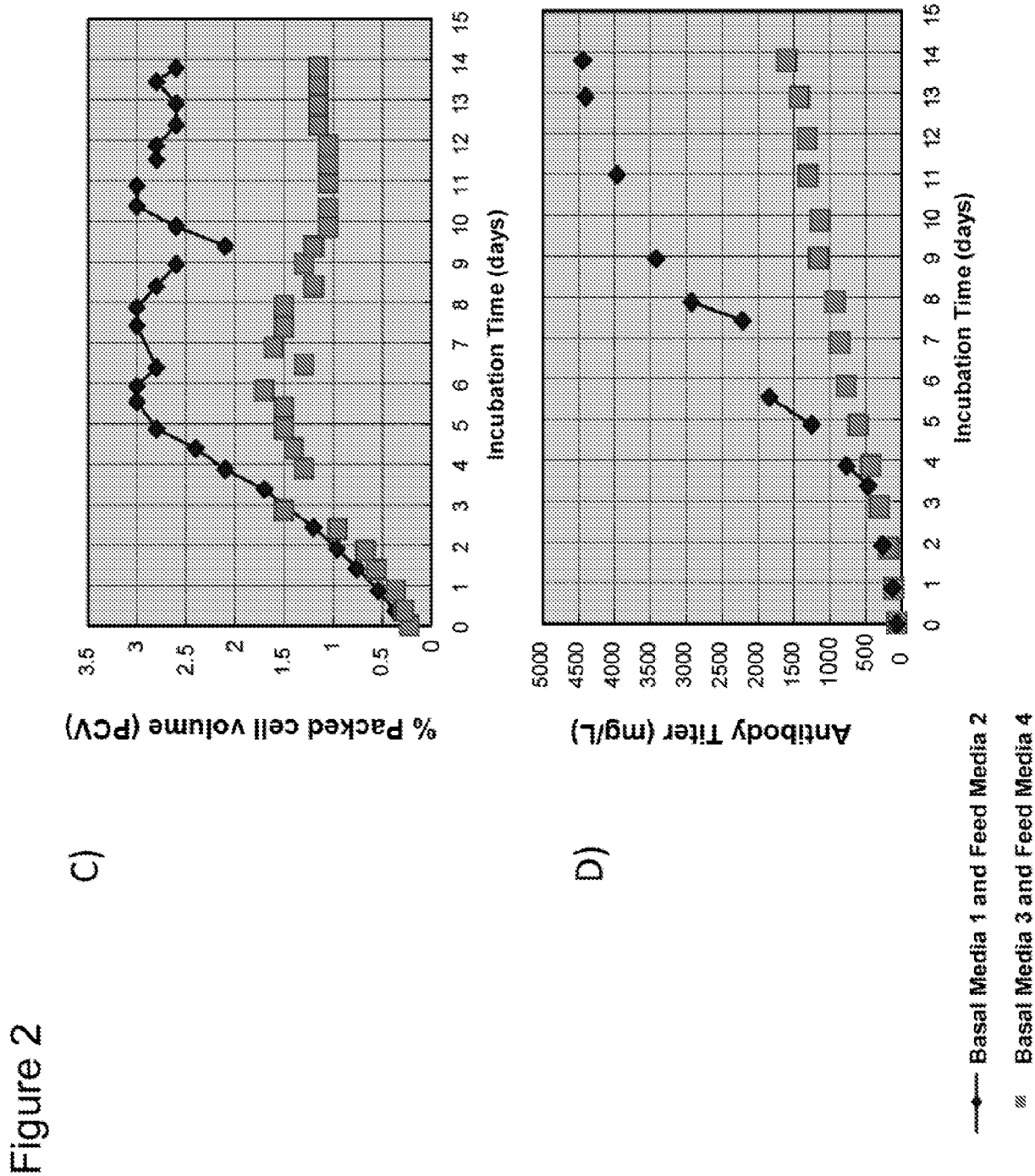
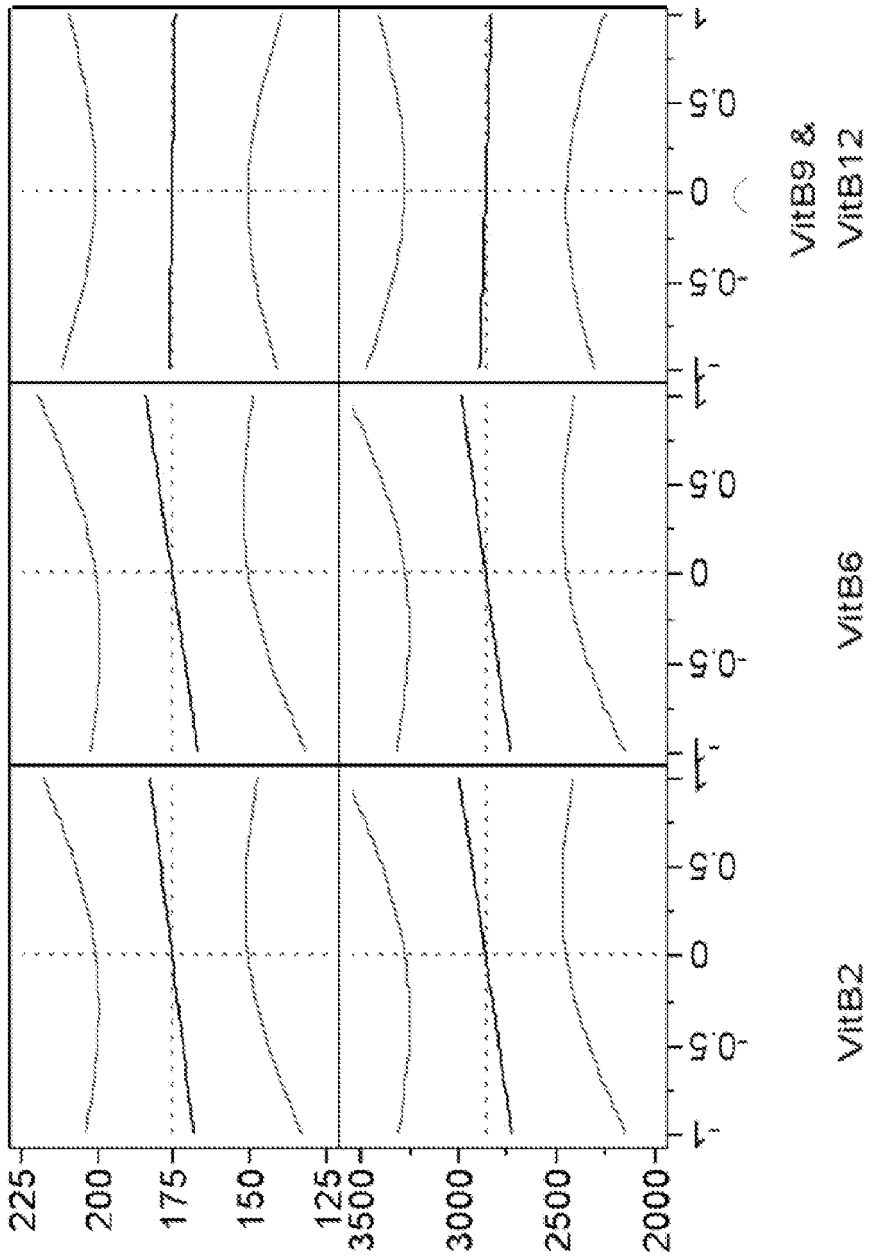

C)
% Packed cell volume (PCV)
Incubation Time (days)
D)
Antibody Titer (mg/L)
Incubation Time (days)
Basal Media 1 and Feed Media 2
Basal Media 3 and Feed Media 4

Figure 3


A)
B)
Final Vol IVPCV
(mL cell-d/L)
Final Titer
(mg/ml)
225
200
175
150
125
3500
3000
2500
2000
-1
-0.5
0
0.5
1
VitB2
VitB6
VitB9 &
VitB12

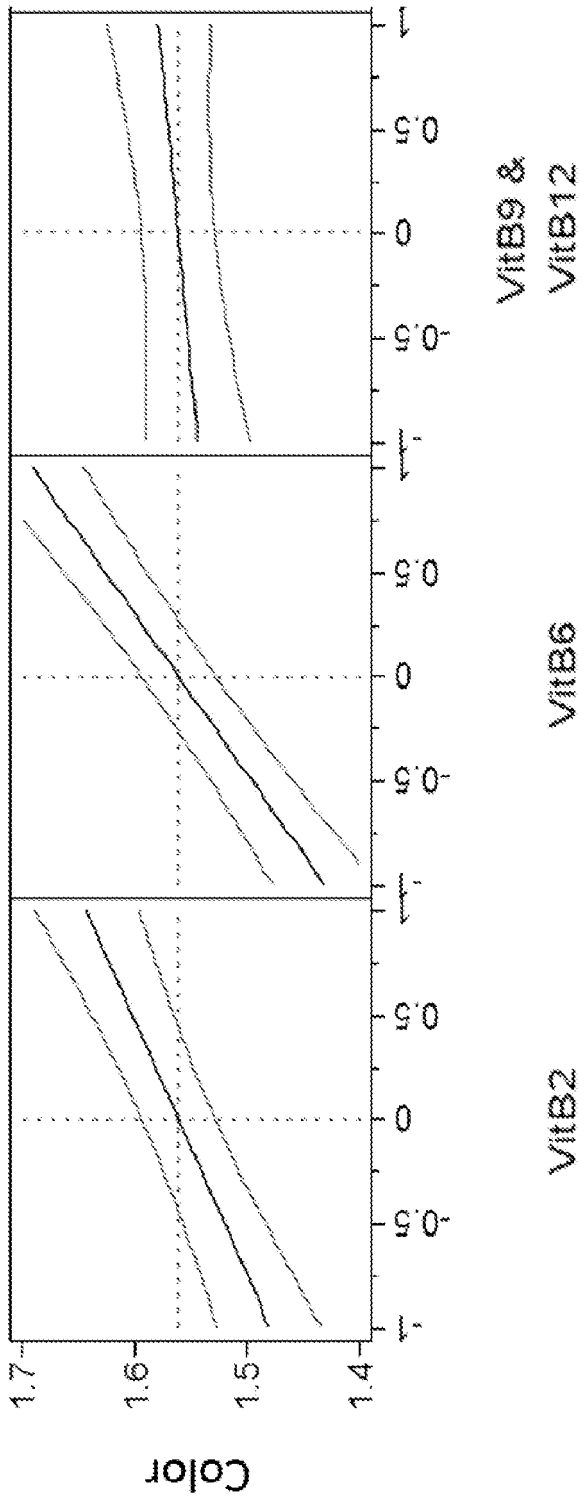


Figure 4

Figure 5

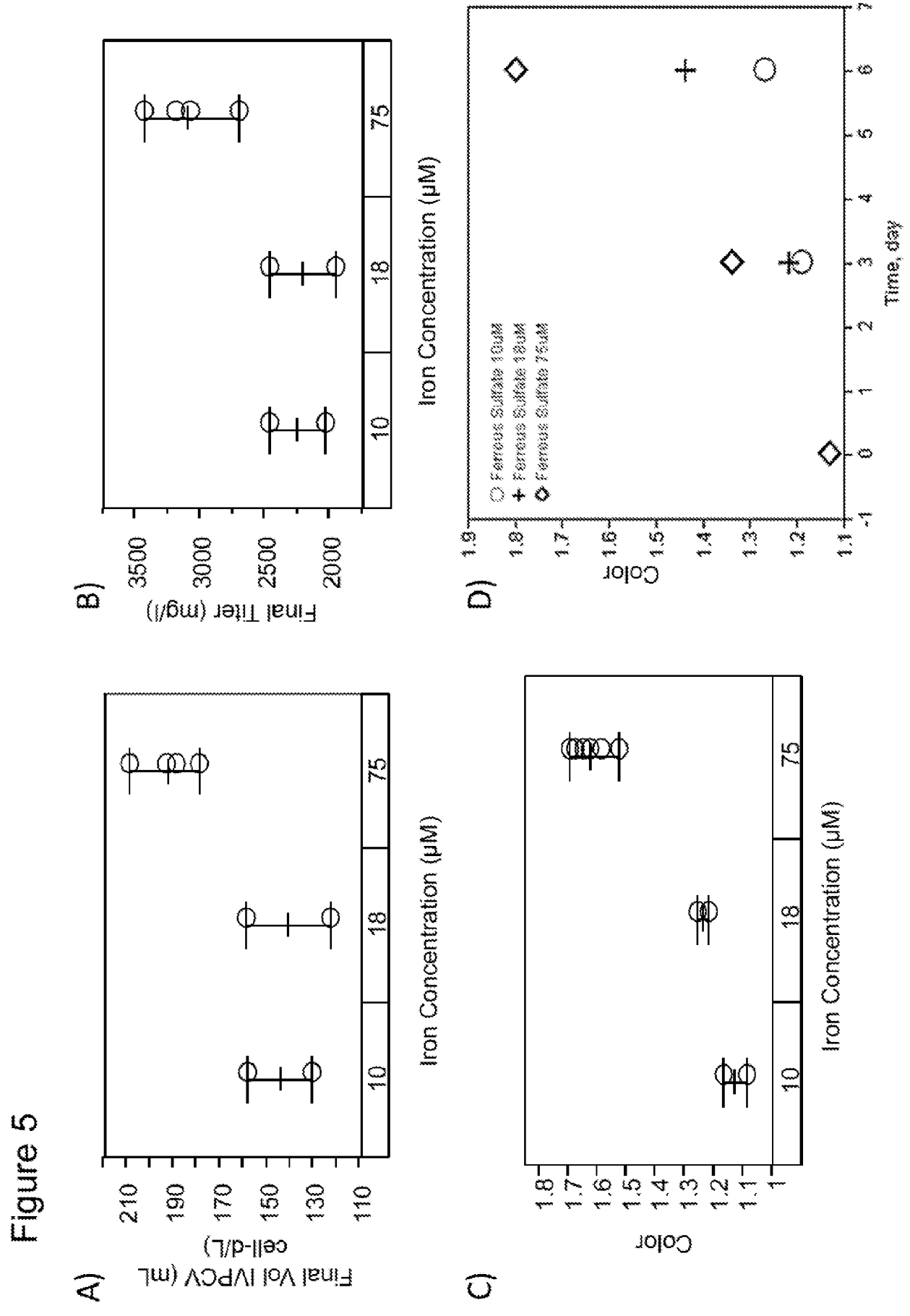

A)
Final Vol IVPCV (mL cell-d/L)
210
190
170
150
130
110
10
18
75
Iron Concentration (µM)
B)
Final Titer (mg/l)
3500
3000
2500
2000
10
18
75
Iron Concentration (µM)
C)
Color
1.8
1.7
1.6
1.5
1.4
1.3
1.2
1.1
1
10
18
75
Iron Concentration (µM)
D)
Color
1.9
1.8
1.7
1.6
1.5
1.4
1.3
1.2
1.1
Ferrous Sulfate 10uM
Ferrous Sulfate 18uM
Ferrous Sulfate 75uM
-1
0
1
2
3
4
5
6
7
Time, day

Figure 6

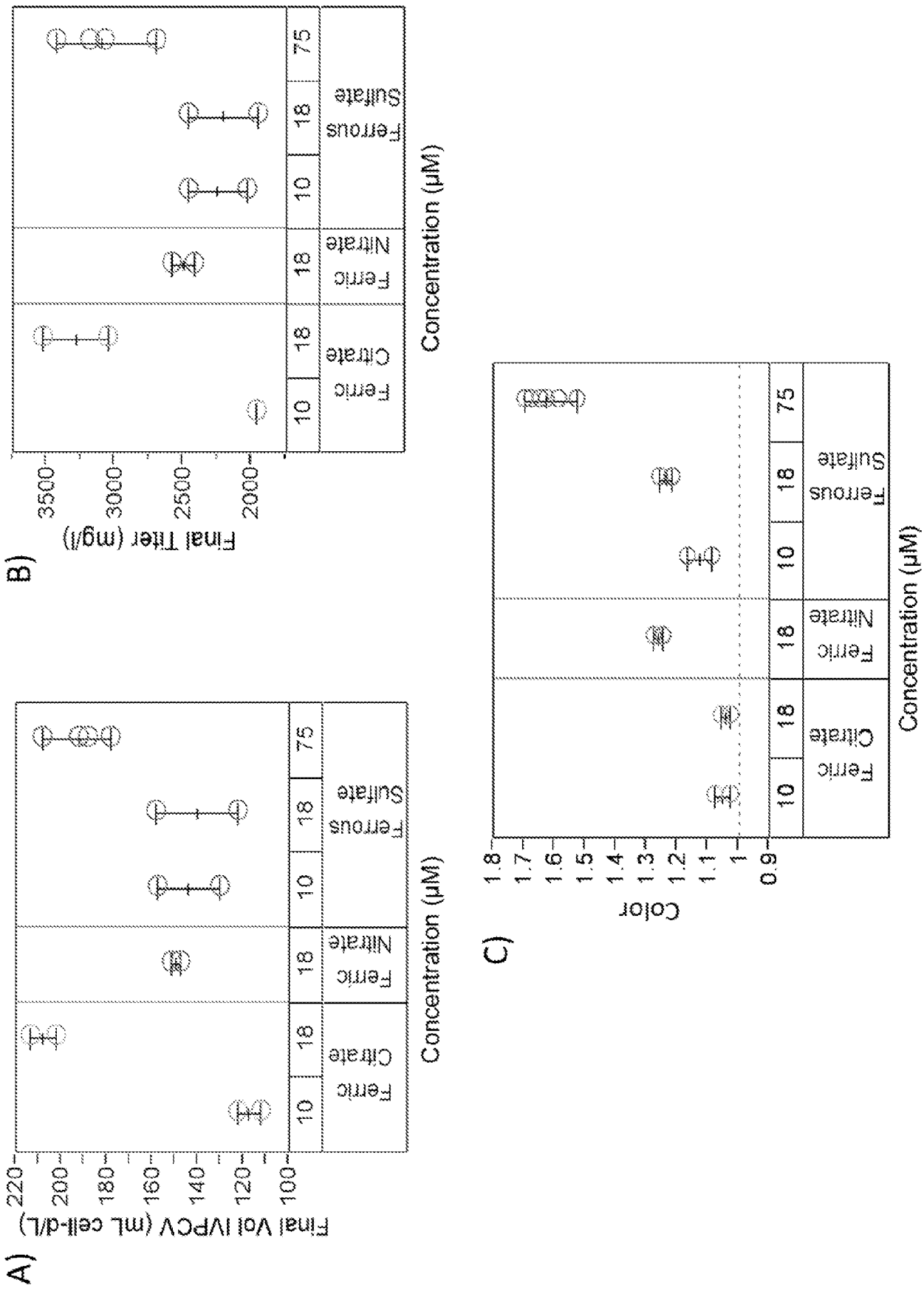
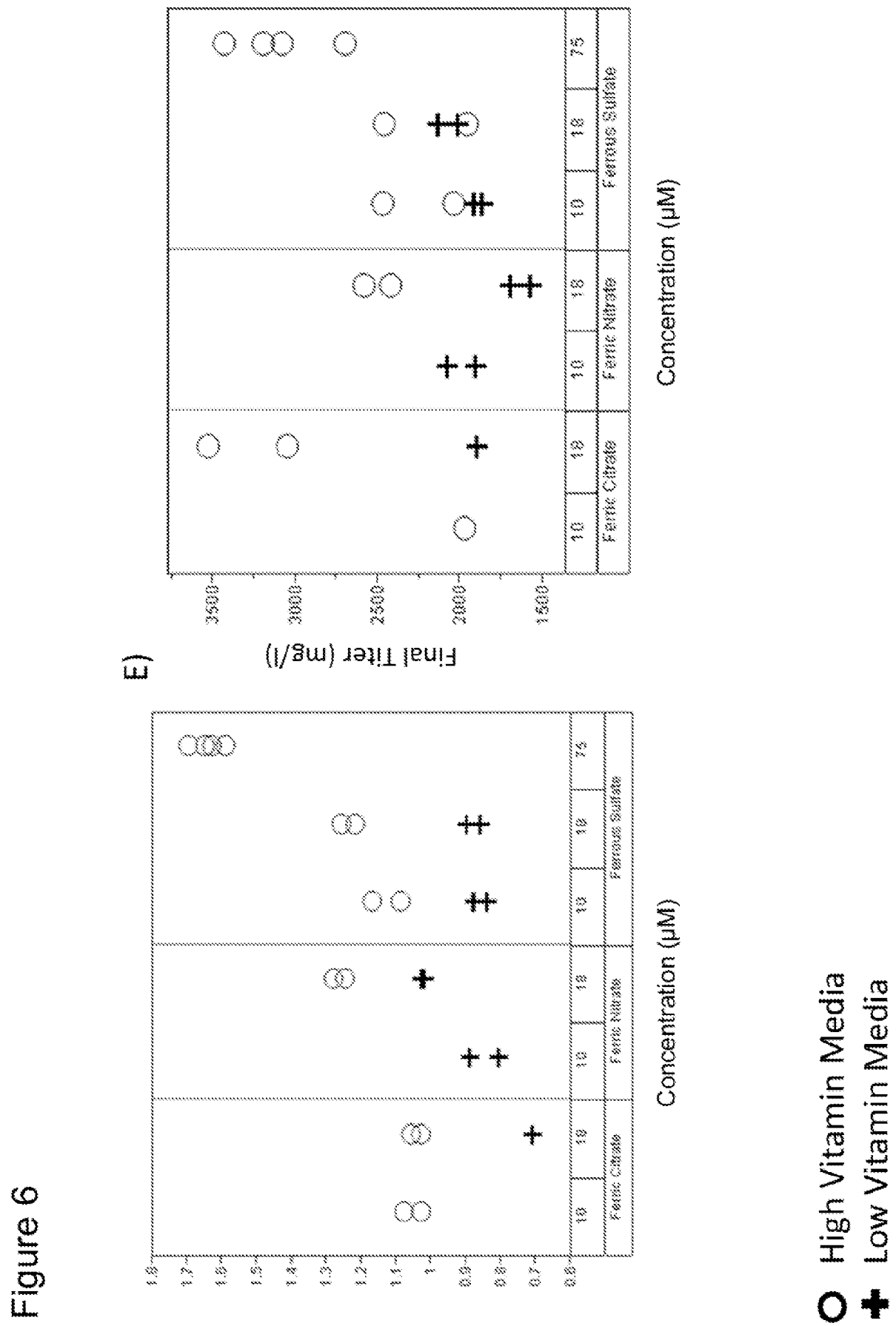

A)
Final Vol IVPCV (mL cell-d/L)
220
200
180
160
140
120
100
Ferric Citrate
Ferric Nitrate
Ferrous Sulfate
10
18
10
18
18
75
Concentration (μM)
B)
Final Titer (mg/l)
3500
3000
2500
2000
Ferric Citrate
Ferric Nitrate
Ferrous Sulfate
10
18
10
18
18
75
Concentration (μM)
C)
Color
1.8
1.7
1.6
1.5
1.4
1.3
1.2
1.1
1
0.9
Ferric Citrate
Ferric Nitrate
Ferrous Sulfate
10
18
10
18
18
75
Concentration (μM)

Figure 6


D)
Color
1.8
1.7
1.6
1.5
1.4
1.3
1.2
1.1
1
0.9
0.8
0.7
0.6
10
10
Ferric Citrate
10
10
Ferric Nitrate
10
10
Ferrous Sulfate
Concentration (μM)
E)
Final Titer (mg/l)
3500
3000
2500
2000
1500
10
10
Ferric Citrate
10
10
Ferric Nitrate
10
10
Ferrous Sulfate
Concentration (μM)
High Vitamin Media
Low Vitamin Media

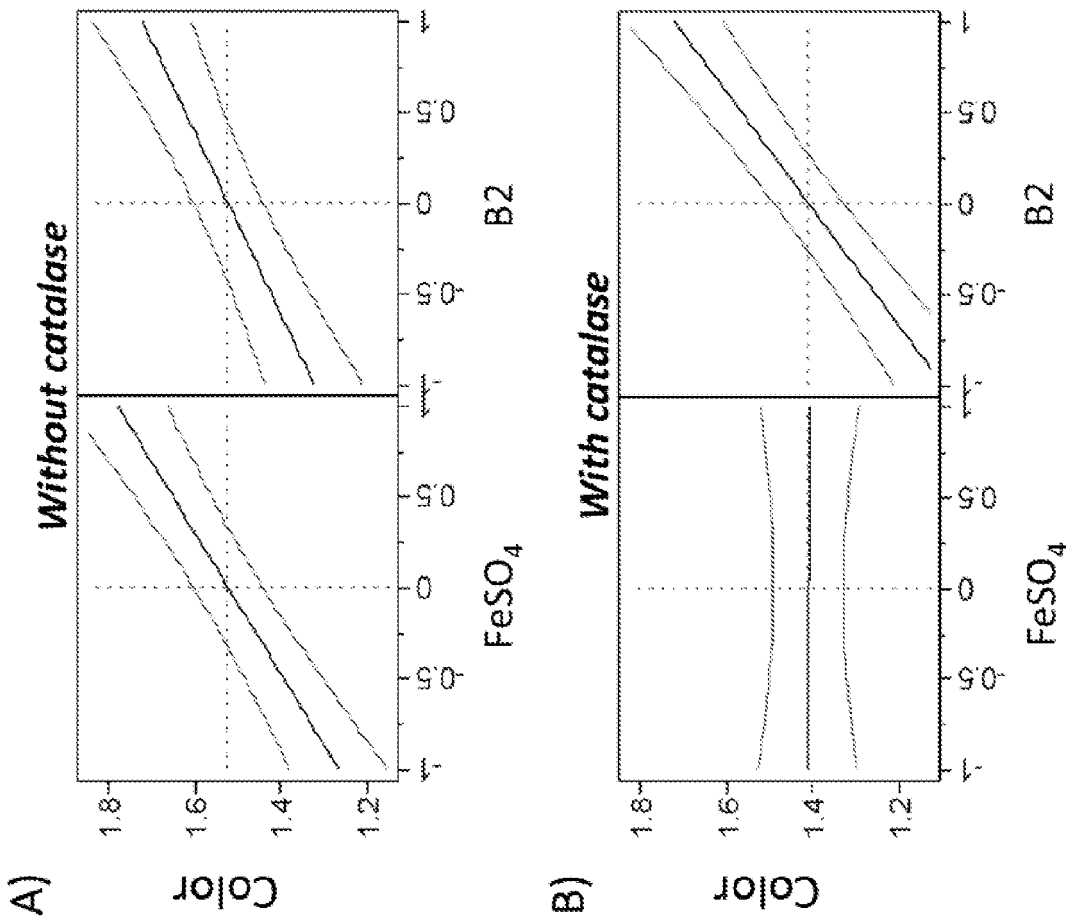

A)
Without catalase
Color
1.8
1.6
1.4
1.2
-1
-0.5
0
0.5
1
FeSO4
B2
B)
With catalase
Color
1.8
1.6
1.4
1.2
-1
-0.5
0
0.5
1
FeSO4
B2

Figure 7

Figure 8

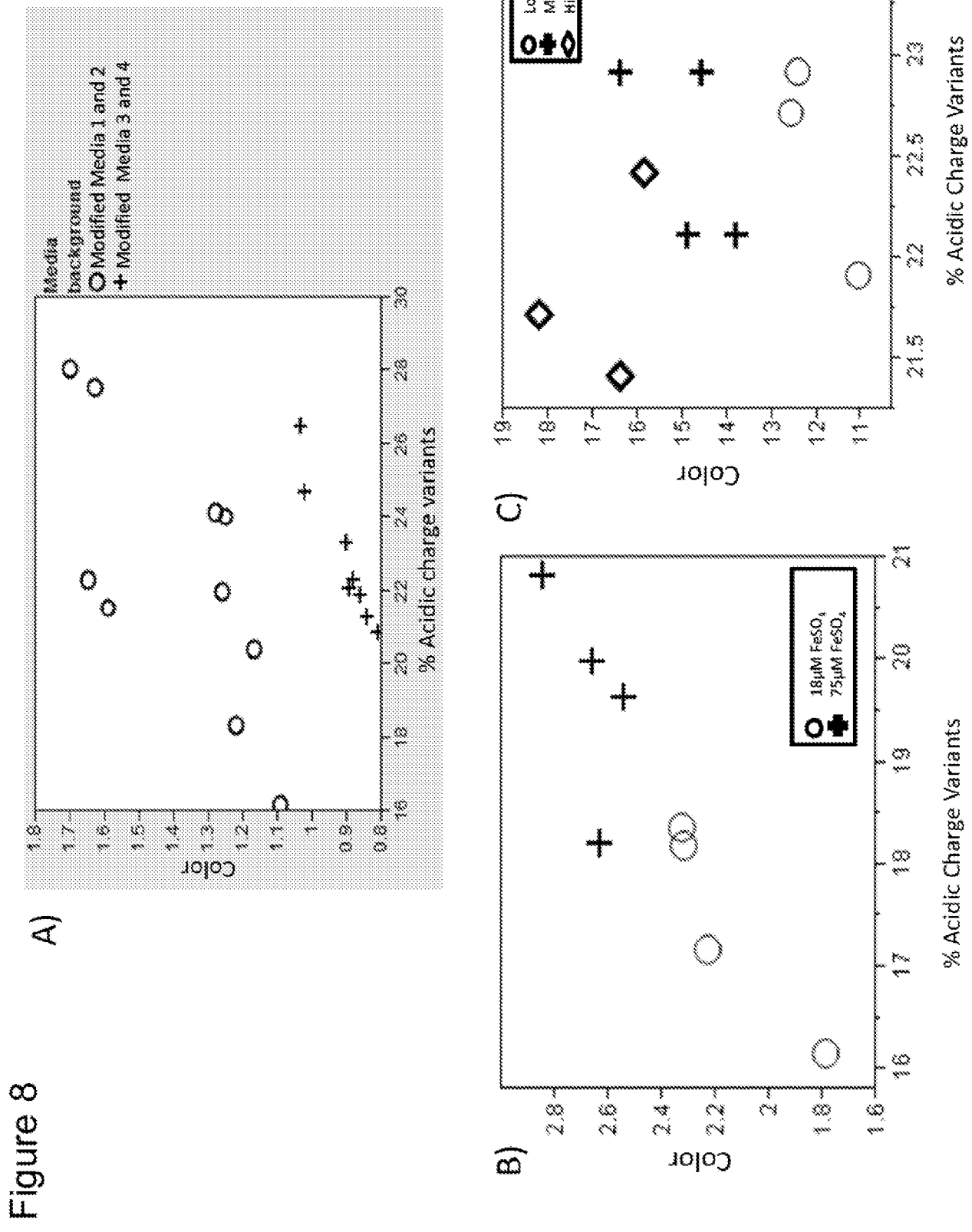
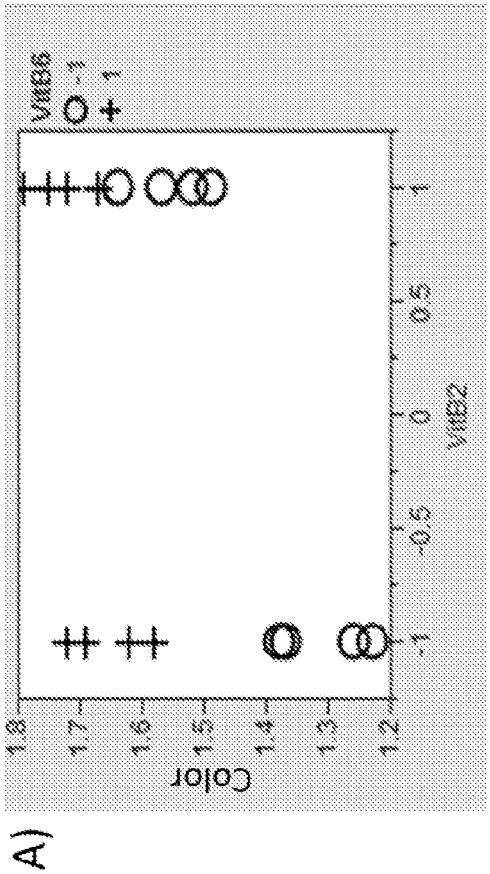

A)
Media background
Modified Media 1 and 2
Modified Media 3 and 4
Color
% Acidic charge variants
B)
18μM FeSO4
75μM FeSO4
Color
% Acidic Charge Variants
C)
Low Vitamin
Medium Vitamin
High Vitamin
Color
% Acidic Charge Variants

Figure 9


A)
VitB6
-1
1
Color
1.8
1.7
1.6
1.5
1.4
1.3
1.2
VitB2
1
0.5
0
-0.5
-1

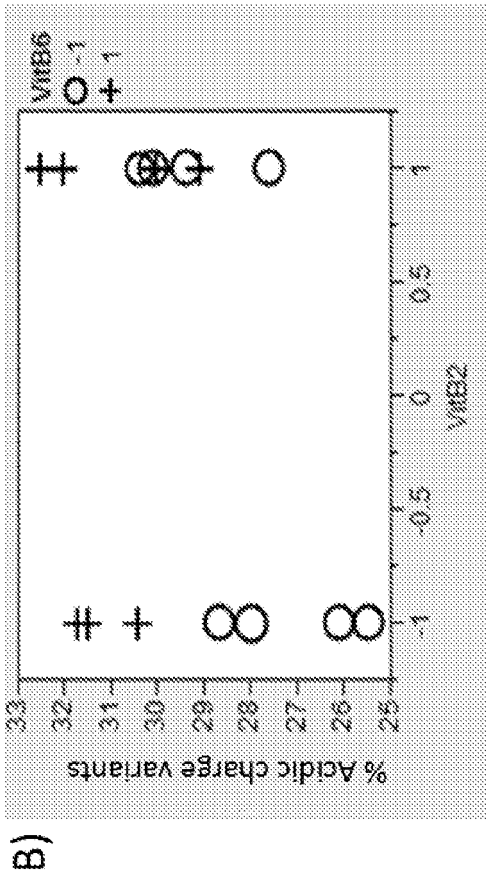
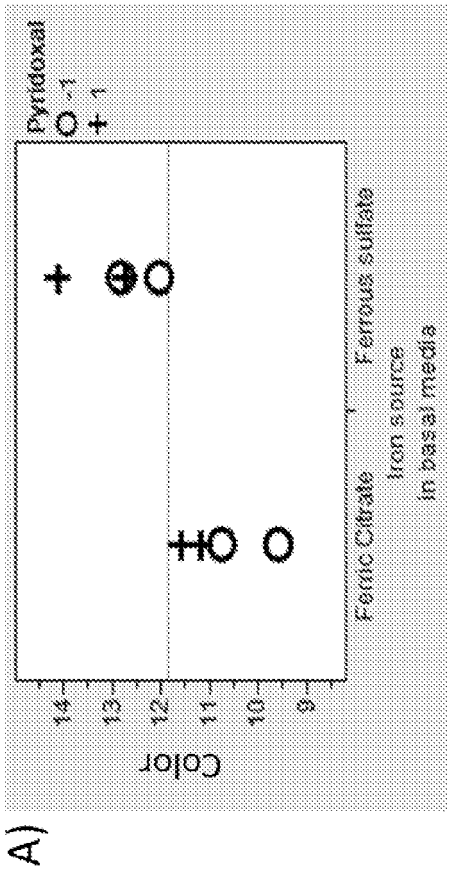

B)
VitB6
-1
1
% Acidic charge variants
33
32
31
30
29
28
27
26
25
VitB2
1
0.5
0
-0.5
-1

Figure 10


A)
Color
14
13
12
11
10
9
Ferric Citrate
Ferrous sulfate
Iron source
in basal media
Pyridoxal
-1
1

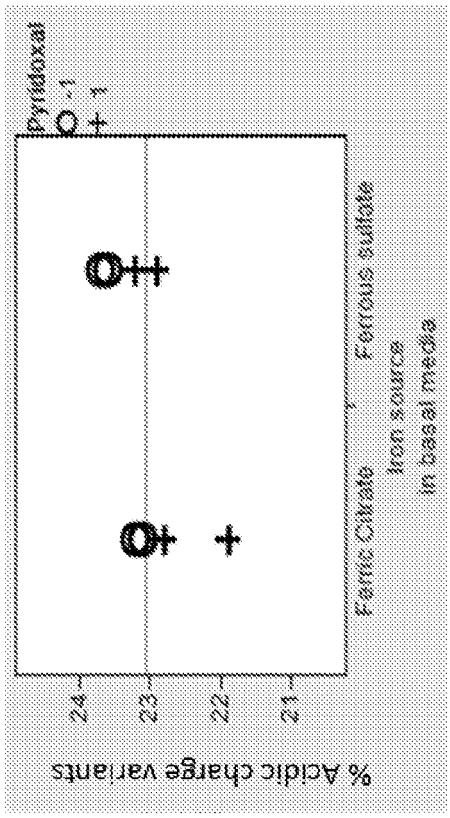
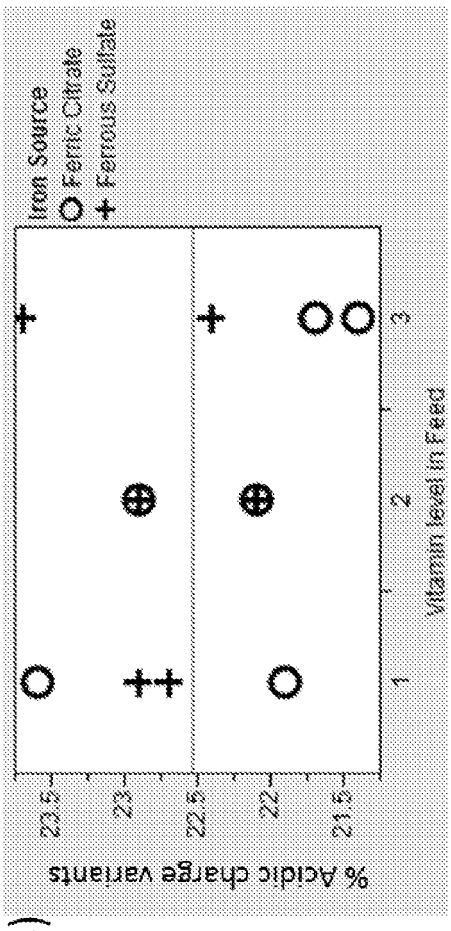

B)
% Acidic charge variants
24
23
22
21
Ferric Citrate
Ferrous sulfate
Iron source
in basal media
Pyridoxal
-1
1

Figure 11


B)
% Acidic charge variants
23.5
23
22.5
22
21.5
1
2
3
Vitamin level in Feed
Iron Source
Ferric Citrate
Ferrous Sulfate

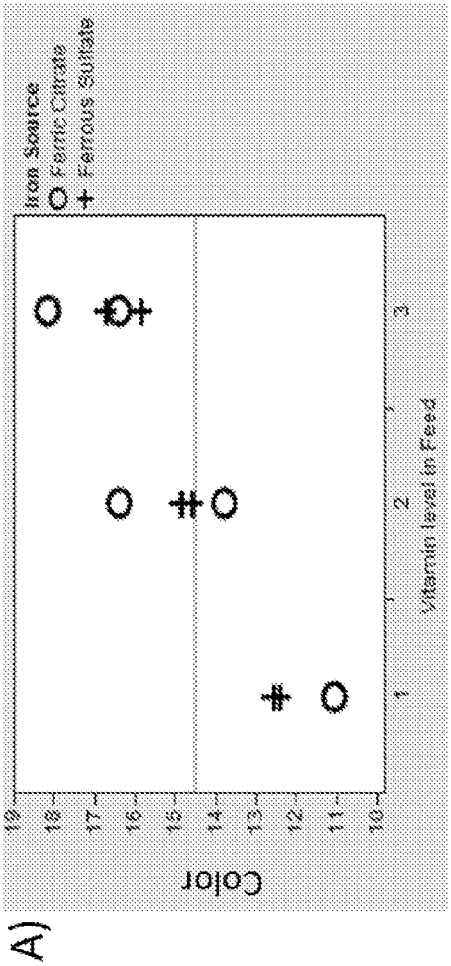

A)
Color
19
18
17
16
15
14
13
12
11
10
1
2
3
Vitamin level in Feed
Iron Source
Ferric Citrate
Ferrous Sulfate

Figure 12

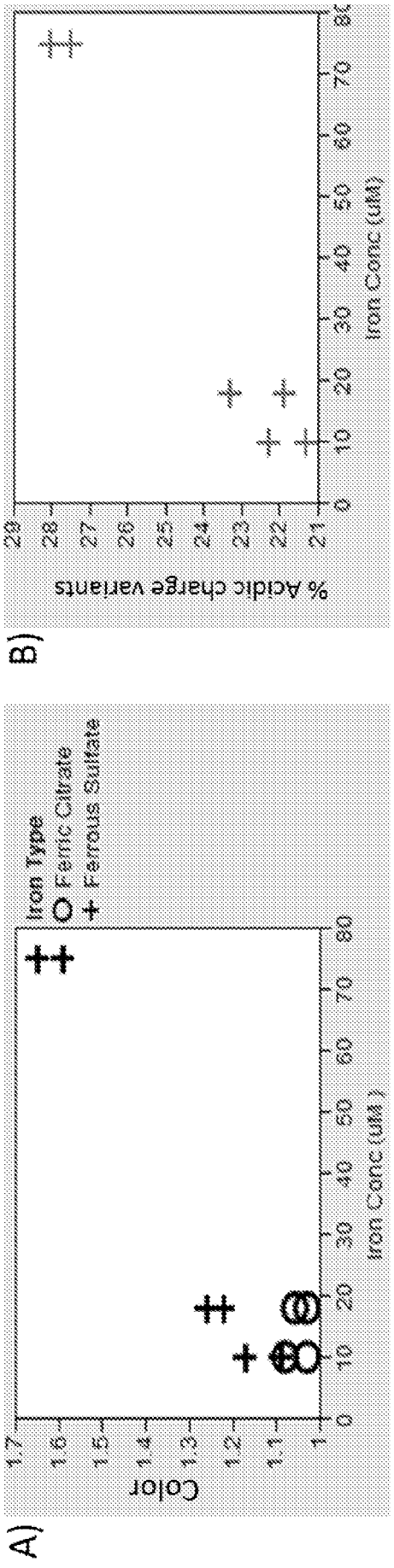

A)
Color
Iron Conc (uM)
Iron Type
Ferric Citrate
Ferrous Sulfate
B)
% Acidic charge variants
Iron Conc (uM)

Figure 13

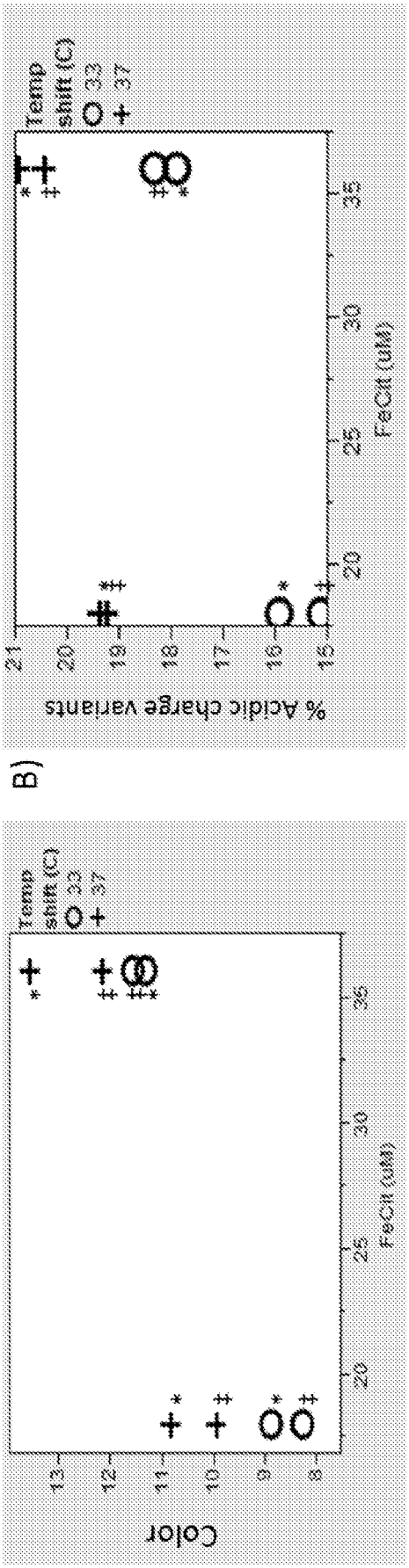
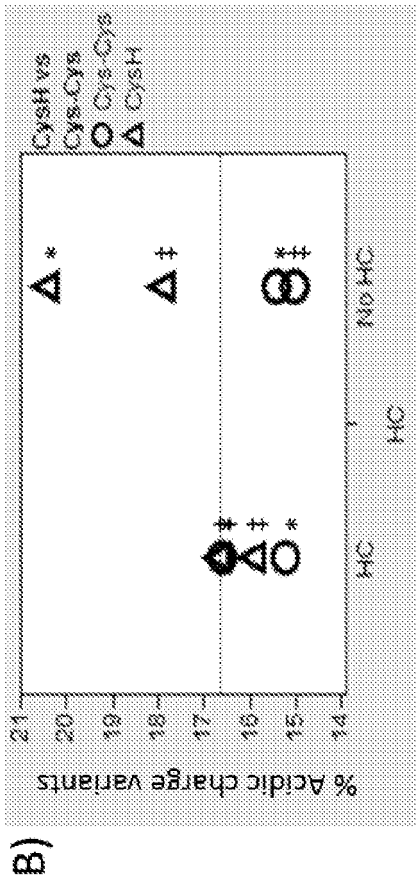
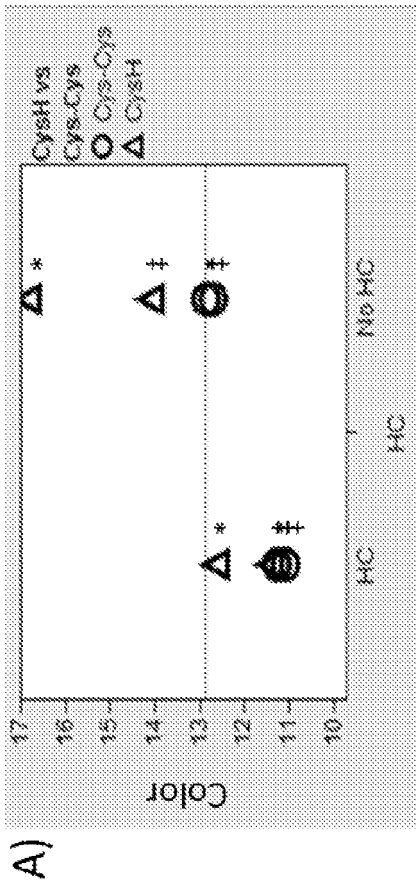

A)
Color
Temp shift (C)
33
37
FeCN (uM)
B)
% Acidic charge variants
Temp shift (C)
33
37
FeCN (uM)
* Modified Basal Media 1
‡ Modified Basal Media 3

Figure 14





* 100% vitamin level in basal media

‡ 50% vitamin level in basal media

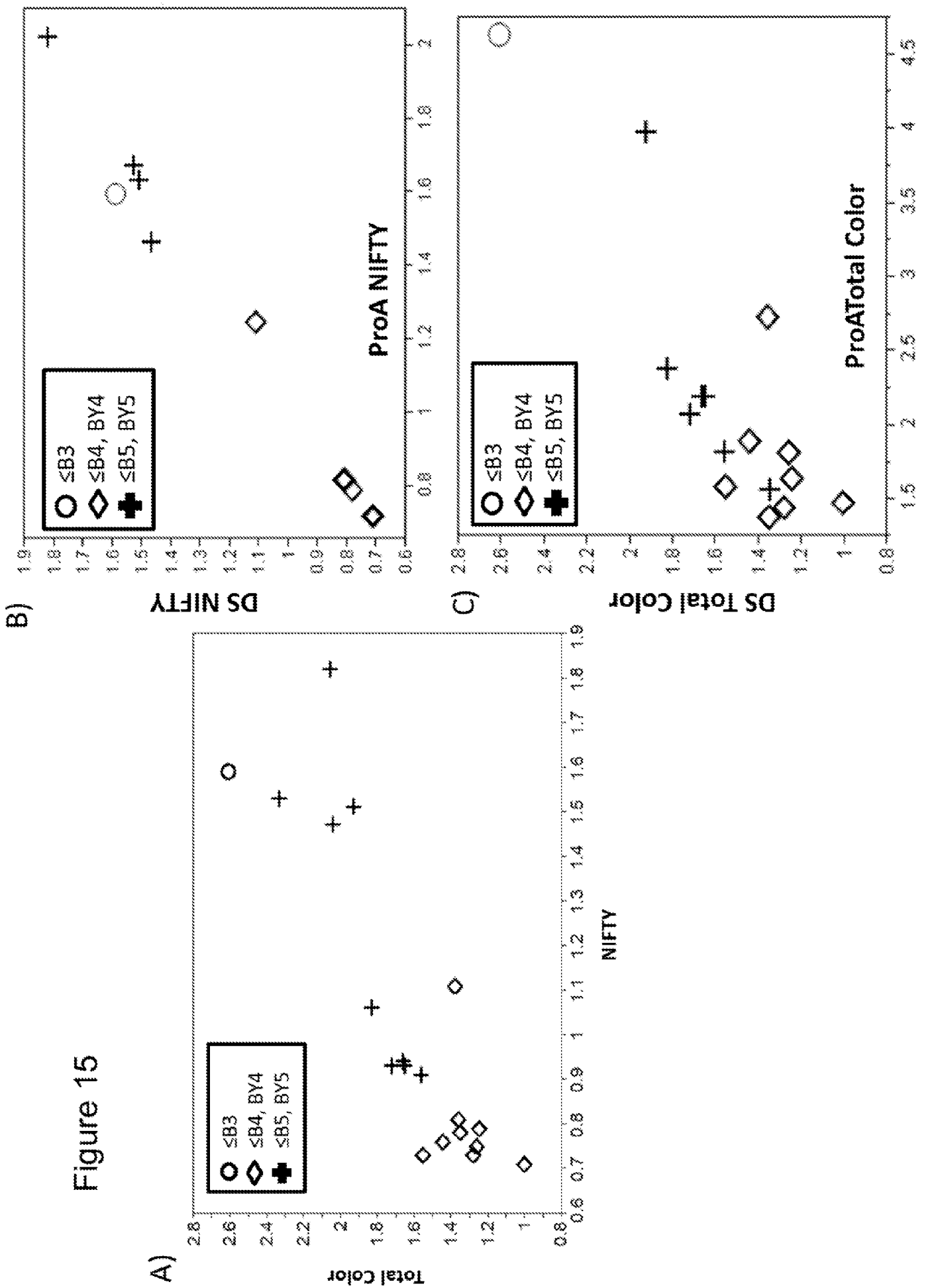

A)
Total Color
NIFTY
≤B3
≤B4, BY4
≤B5, BY5
B)
DS NIFTY
ProA NIFTY
C)
DS Total Color
ProATotal Color

Figure 15