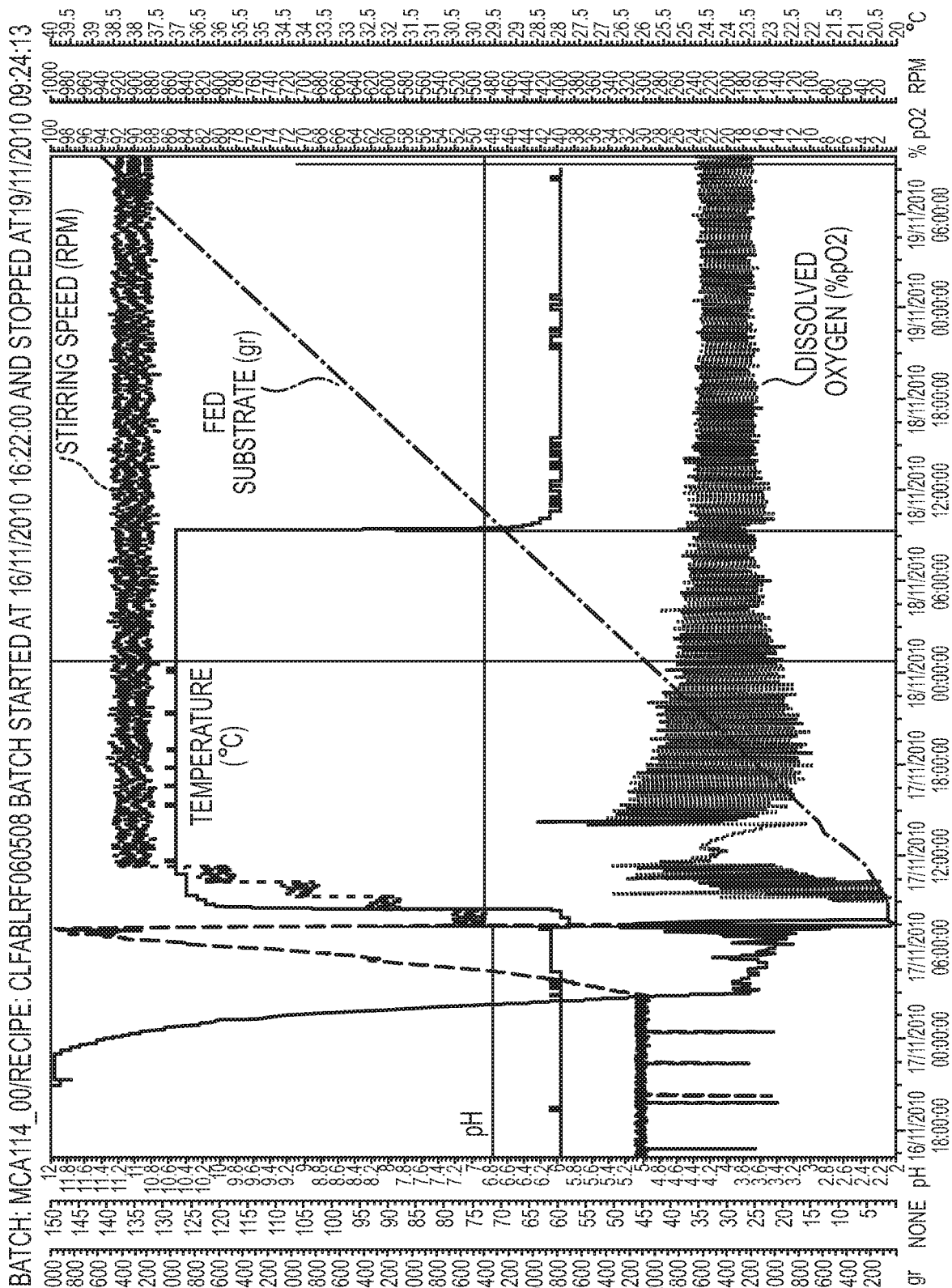


A TYPICAL FERMENTATION PROFILE WITH THE HIGH CELL DENSITY INDUCTION (HCDI) PROCESSES AND THE PARAMETERS MONITORED DURING 20L - SCALE FED-BATCH FERMENTATION.

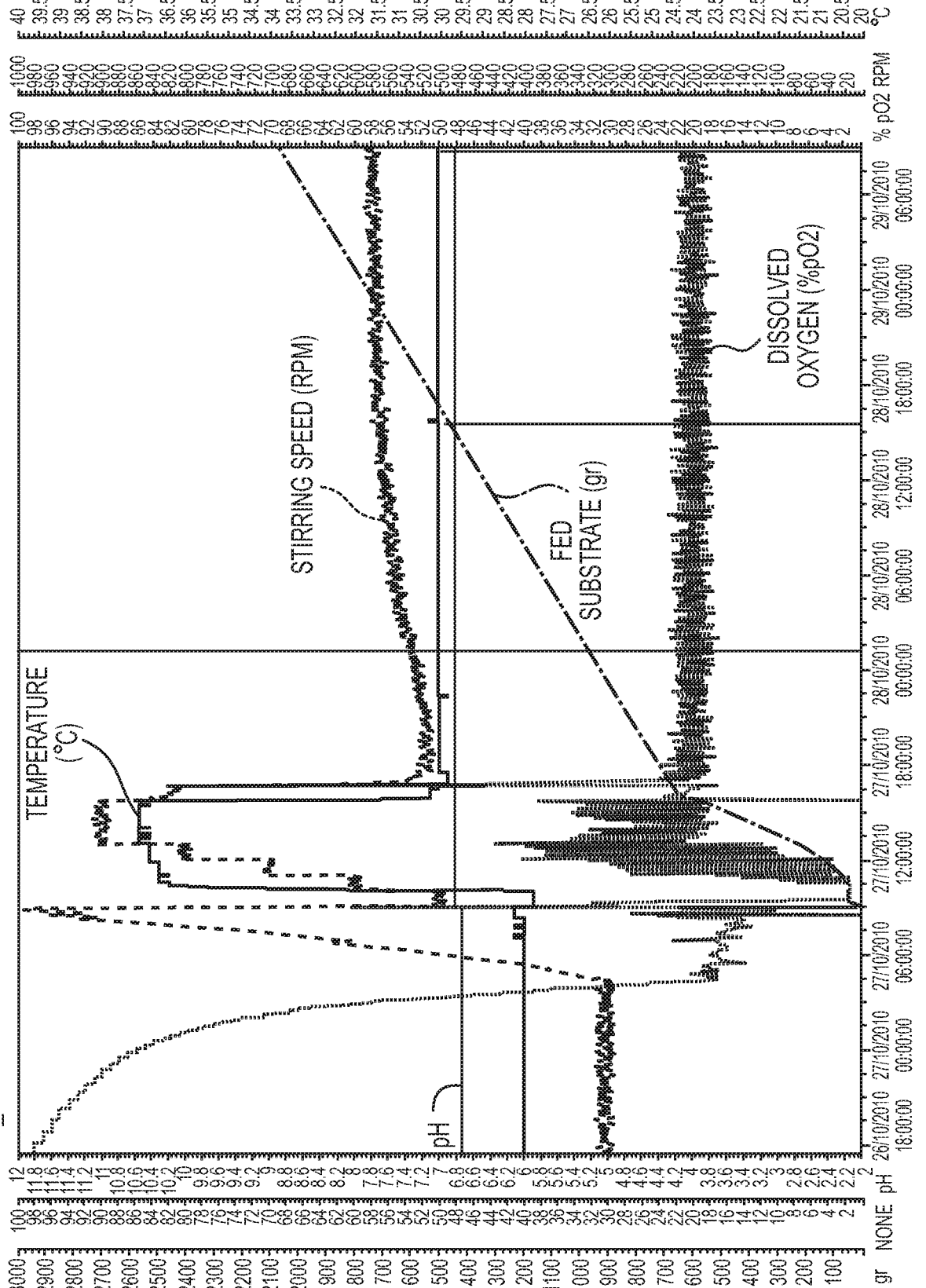


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FIG. 2

A TYPICAL FERMENTATION PROFILE WITH THE LOW CELL DENSITY INDUCTION (LDCI) PROCESSES AND THE PARAMETERS MONITERED DURING 20L - SCALE FED-BATCH FERMENTATION.

BATCH: MCA109_00/RECIPE: CLFABLR060508 BATCH STARTED AT 26/10/2010 17:30:31 AND STOPPED AT 29/10/2010 09:02:11



UspA2 YIELD FROM PROTEIN CONSTRUCTS MC-001, MC-002, MC-004, MC-005, MC-006, MC-007, MC-008 AND MC-010 EVALUATED IN FERMENTER; DATA FROM TABLE 4. His = HISTIDINE.

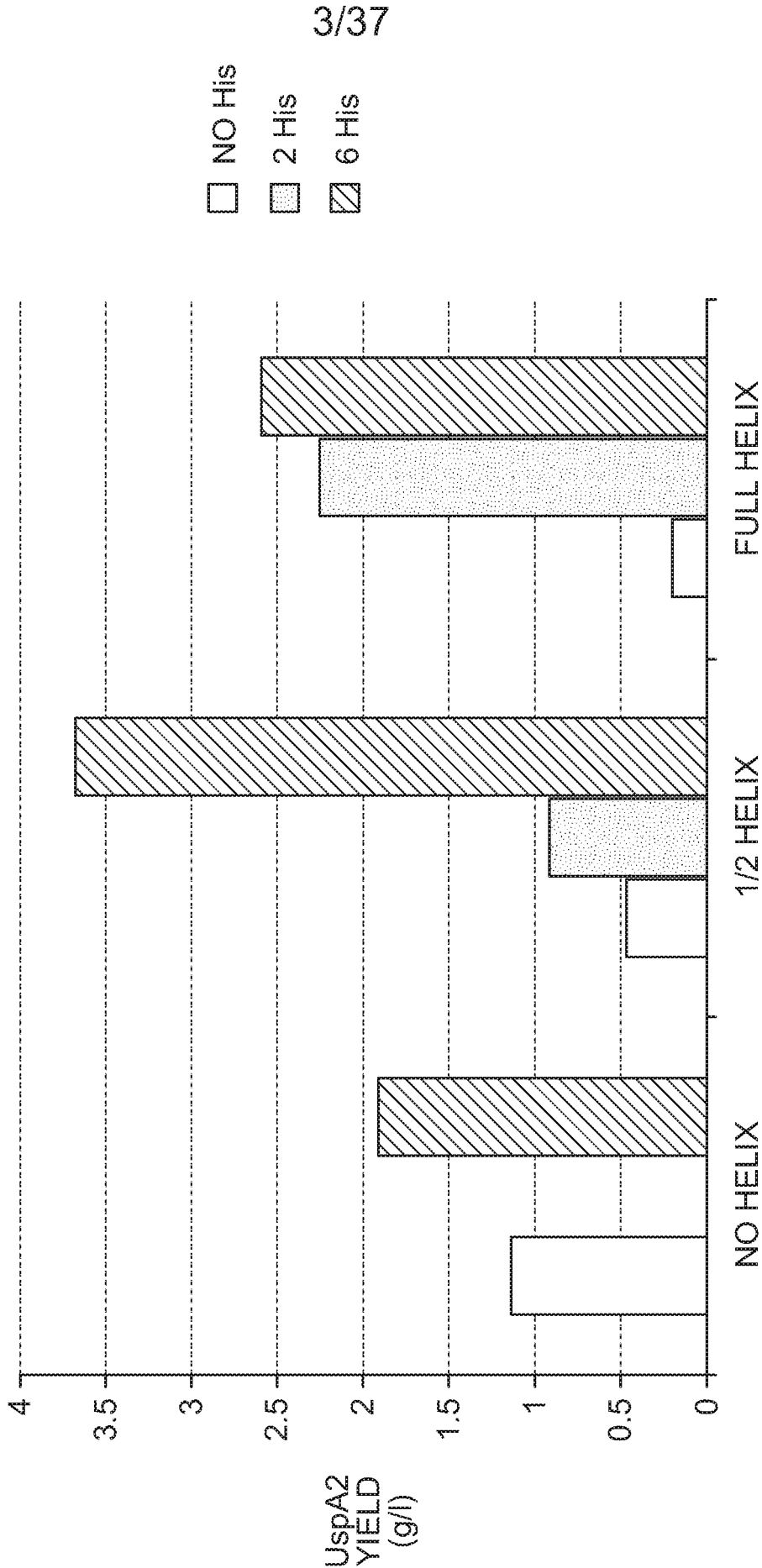


FIG. 3

MOLECULAR WEIGHT DISTRIBUTION OF PURIFIED MC-005 DETERMINED BY SEDIMENTATION VELOCITY ANALYTICAL ULTRACENTRIFUGATION. THE MAJORITY OF PROTEIN IS FOUND AS A TRIMER, WITH A SMALL PROPORTION OF A HIGHER MOLECULAR WEIGHT OLIGMER THAT MAY CORRESPOND TO DIMER OF TRIMER. MW = MOLECULAR WEIGHT. kDa = KILODALTON.

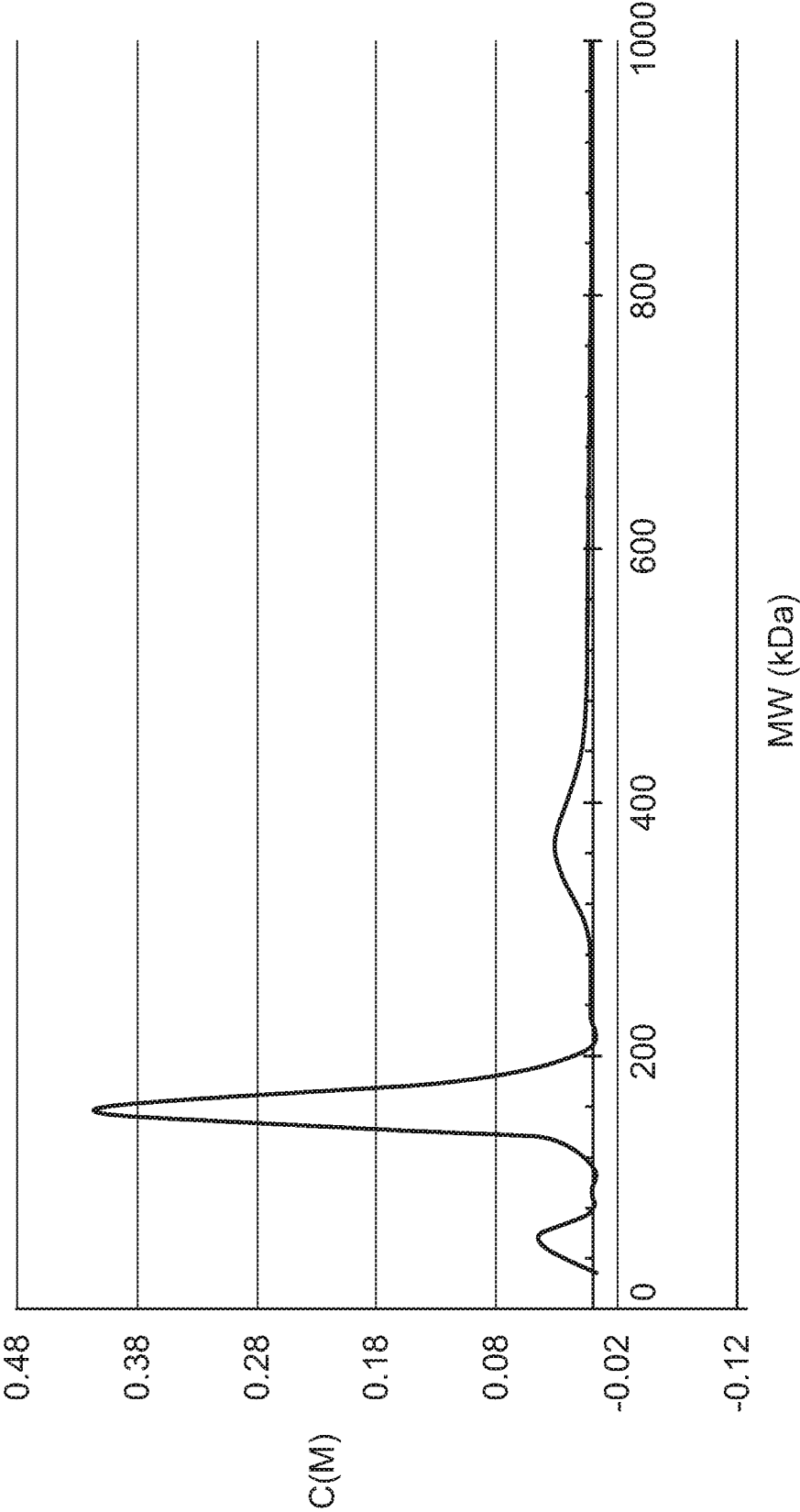


FIG. 4

MOLECULAR WEIGHT DISTRIBUTION OF PURIFIED MC-001 DETERMINED
BY SEDIMENTATION VELOCITY ANALYTICAL ULTRACENTRIFUGATION.
MAJORITY OF PROTEIN IS FOUND AS A TRIMER.

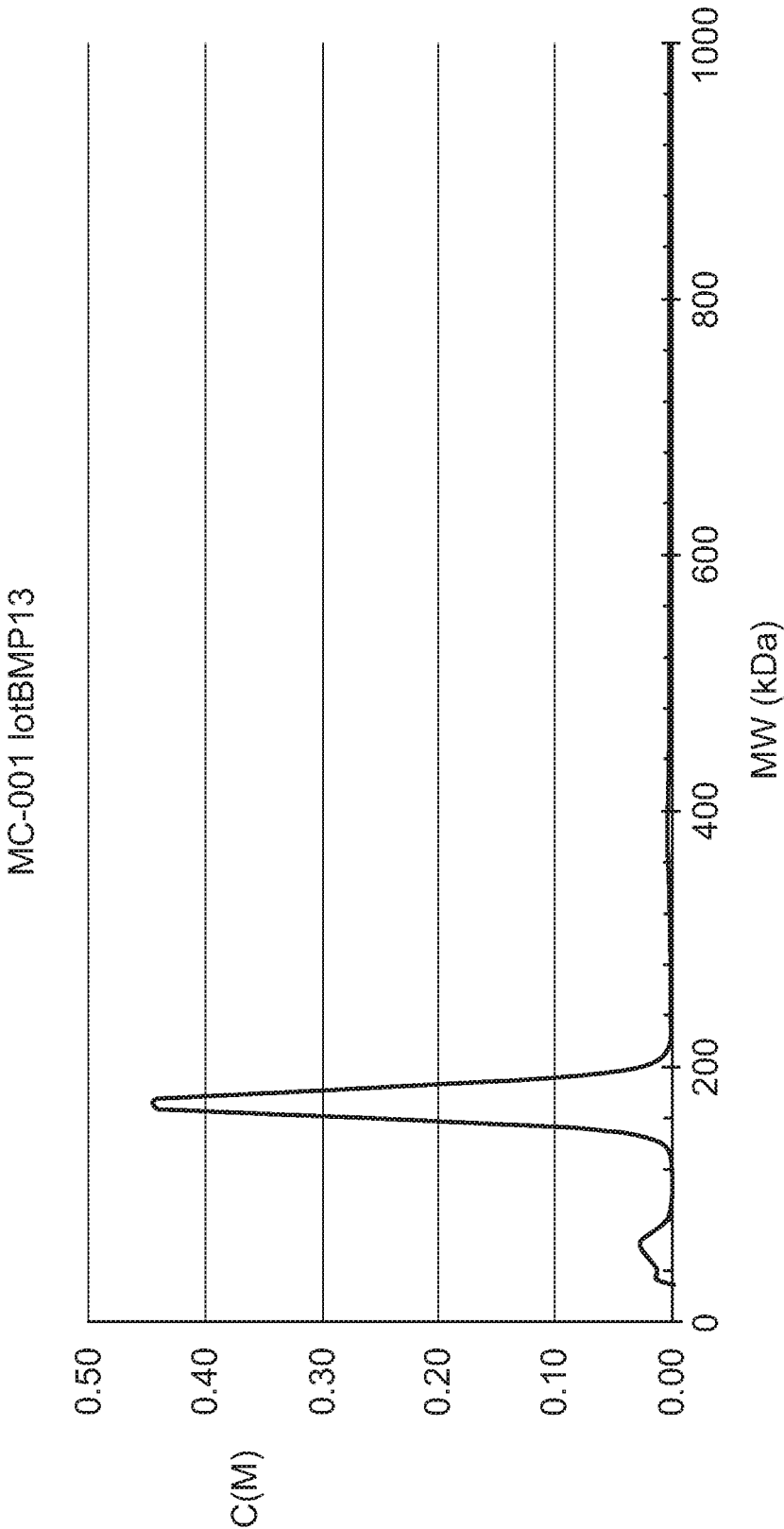


FIG. 5

MOLECULAR WEIGHT DISTRIBUTION OF PURIFIED MC-001 DETERMINED BY SEDIMENTATION VELOCITY ANALYTICAL ULTRACENTRIFUGATION. THE SAMPLE PRESENTS MULTIPLE SPECIES AND IS HIGHLY POLYDISPERSE. THE SEDIMENTATION COEFFICIENT OF THE MAJOR SPECIES DETECTED DOESN'T CORRESPOND TO THE ONE OF THE TRIMERS NORMALLY DETECTED IN THE OTHER LOTS.

MC-001 lotBMP14

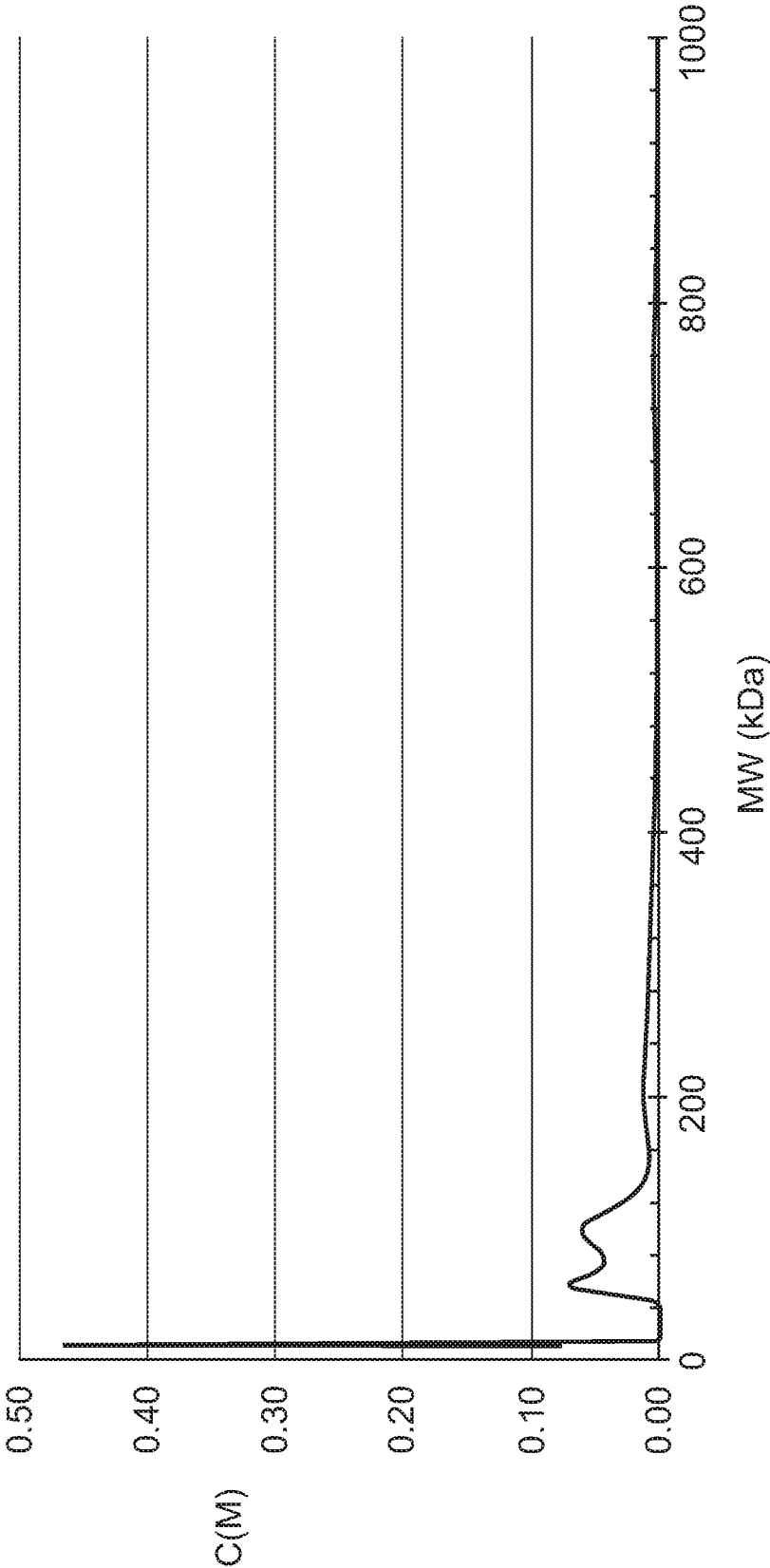


FIG. 6

MOLECULAR WEIGHT DISTRIBUTION OF PURIFIED MC-001 DETERMINED
BY SEDIMENTATION VELOCITY ANALYTICAL ULTRACENTRIFUGATION.
MAJORITY OF PROTEIN IS FOUND AS A TRIMER.

MC-001 lotBMP54

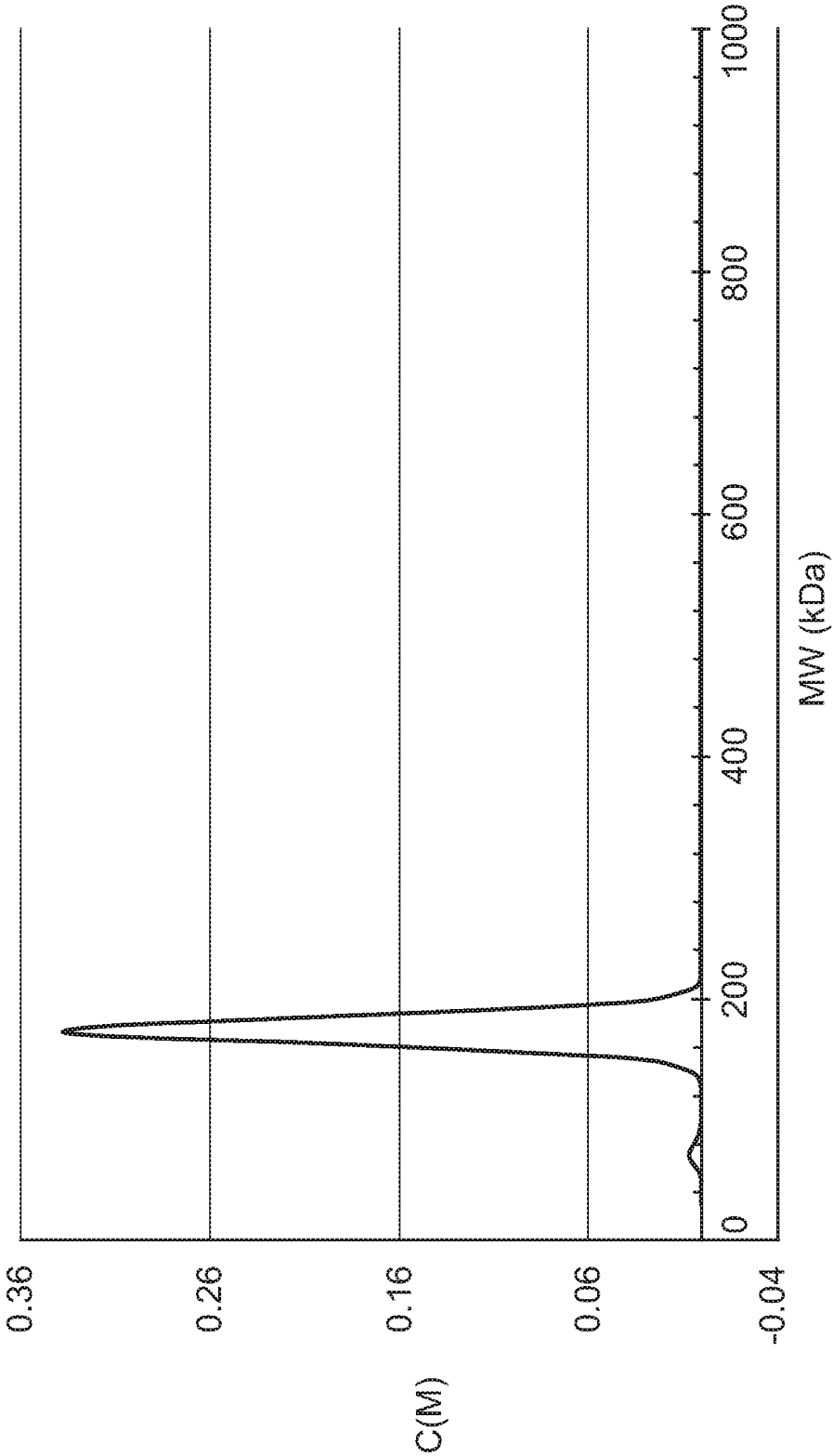


FIG. 7

MOLECULAR WEIGHT DISTRIBUTION OF PURIFIED MC-007 DETERMINED
BY SEDIMENTATION VELOCITY ANALYTICAL ULTRACENTRIFUGATION.
MAJORITY OF PROTEIN IS FOUND AS A TRIMER.

MC-007

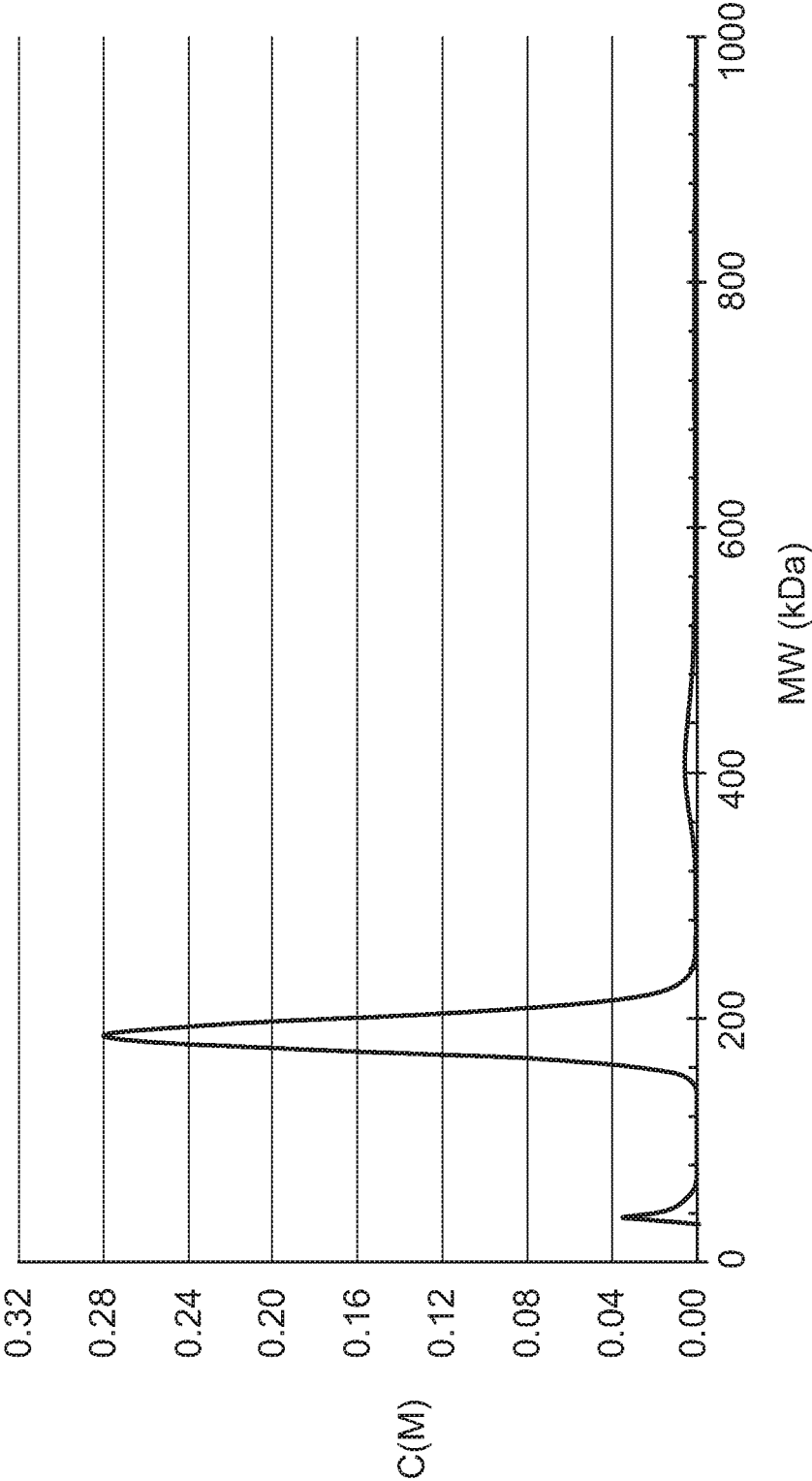


FIG. 8

FAR-UV CIRCULAR DICHROISM (CD) SPECTRA OF UspA2 CONSTRUCTS GIVING AN INDICATION OF PROTEIN SECONDARY STRUCTURES.

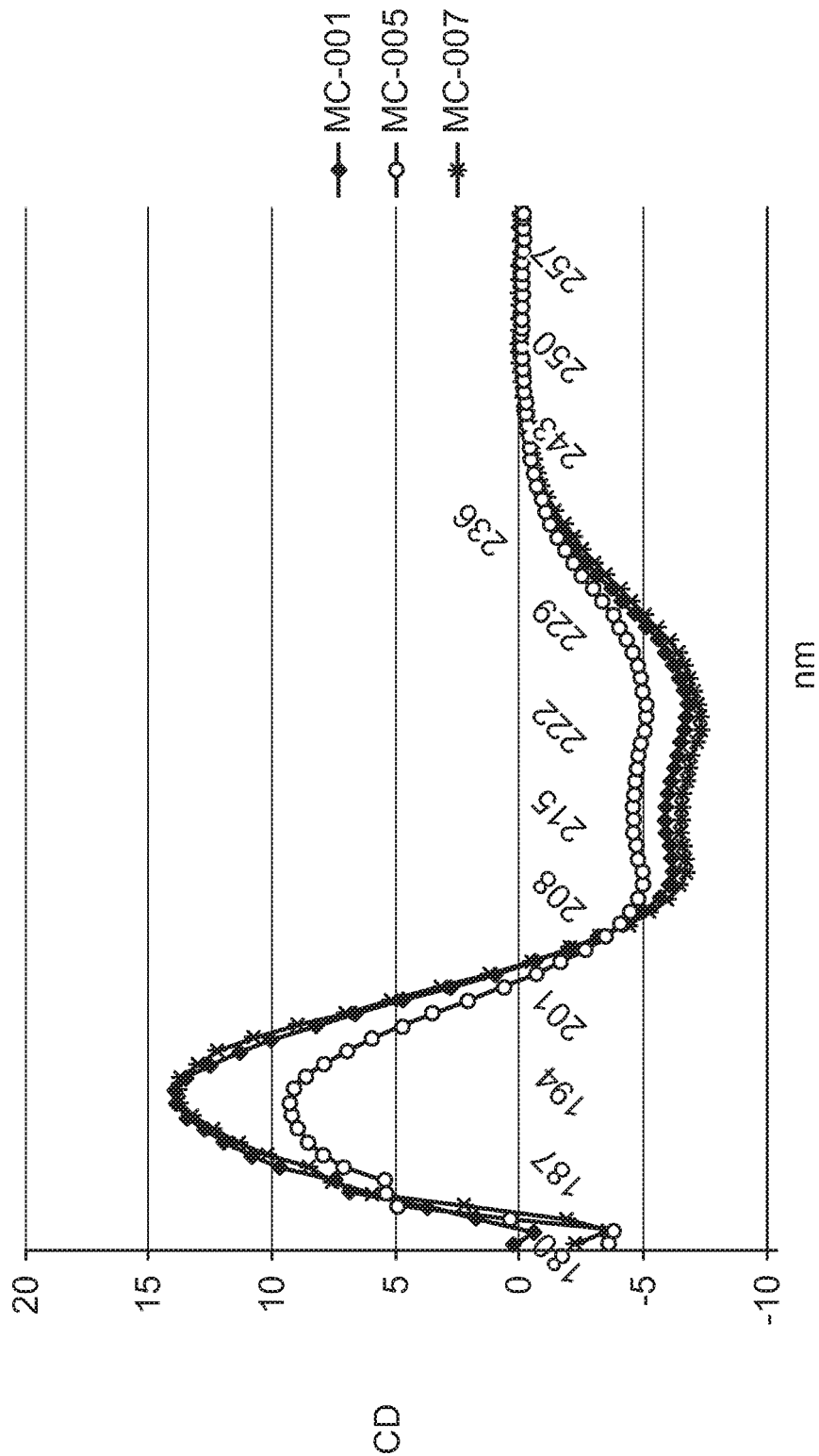


FIG. 9

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SECONDARY STRUCTURES MONITORING BY CIRCULAR DICHROISM (CD) DURING THERMAL UNFOLDING OF MC-005 (UspA2 Δ HELIX+6HIS). VISUAL ANALYSIS OF THE SPECTRA CLEARLY SHOWS THAT PROTEIN LOSES MOST OF ITS SECONDARY STRUCTURES AT 33 °C.

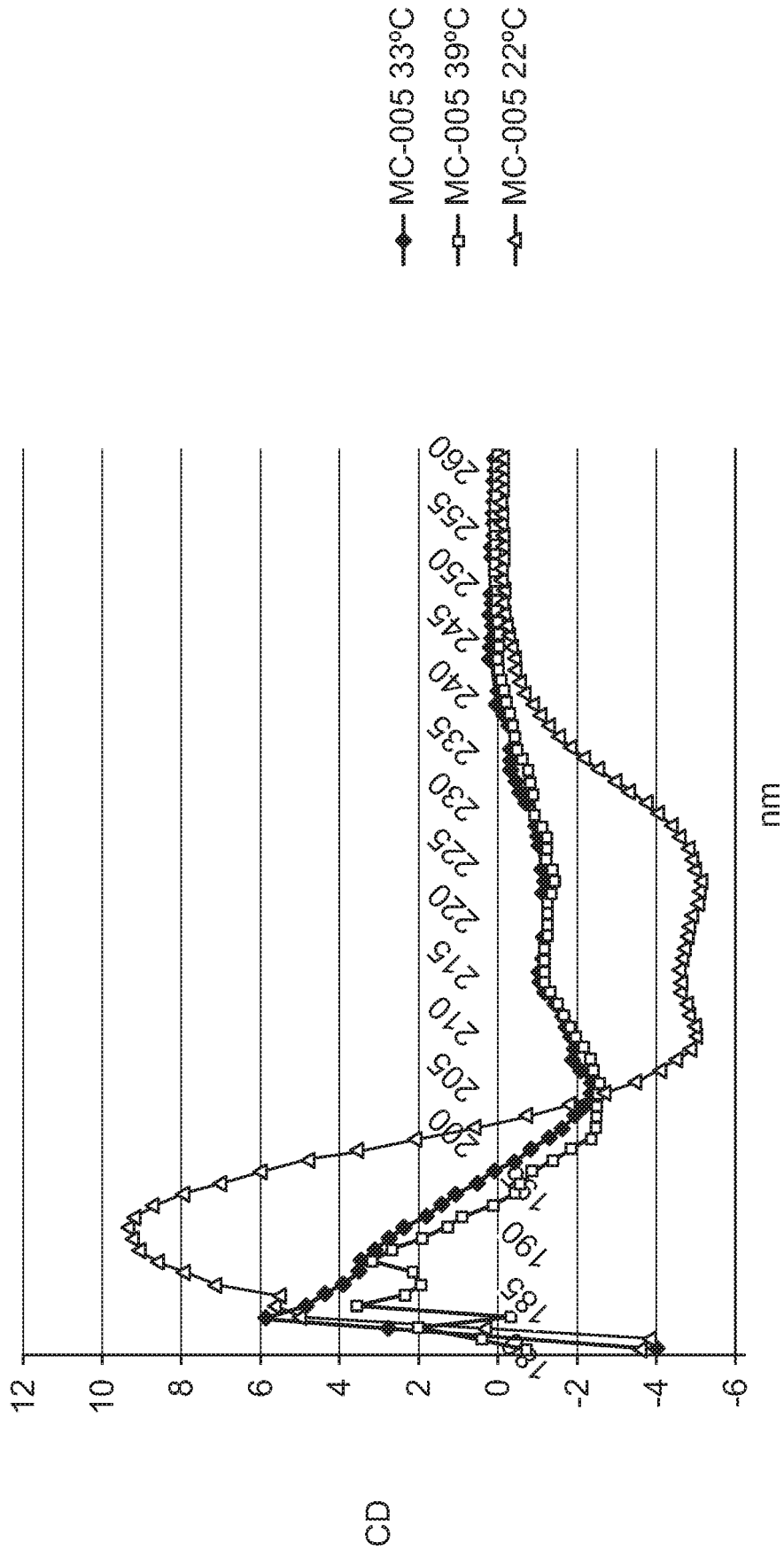


FIG. 10

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SECONDARY STRUCTURES MONITORING BY CIRCULAR DICHROISM (CD) DURING THERMAL UNFOLDING OF MC-007 (UspA2 FULL HELIX + 6HIS). VISUAL ANALYSIS OF THE SPECTRA SHOWS THAT LOSS OF SECONDARY STRUCTURE IS SLOWER COMPARED TO THE CONSTRUCT WITHOUT HELIX. STRUCTURAL CHANGES ARE DETECTABLE UPON HEATING TO 33°C, BUT COMPLETE UNFOLDING SEEMS TO OCCUR BETWEEN 35°C AND 37°C.

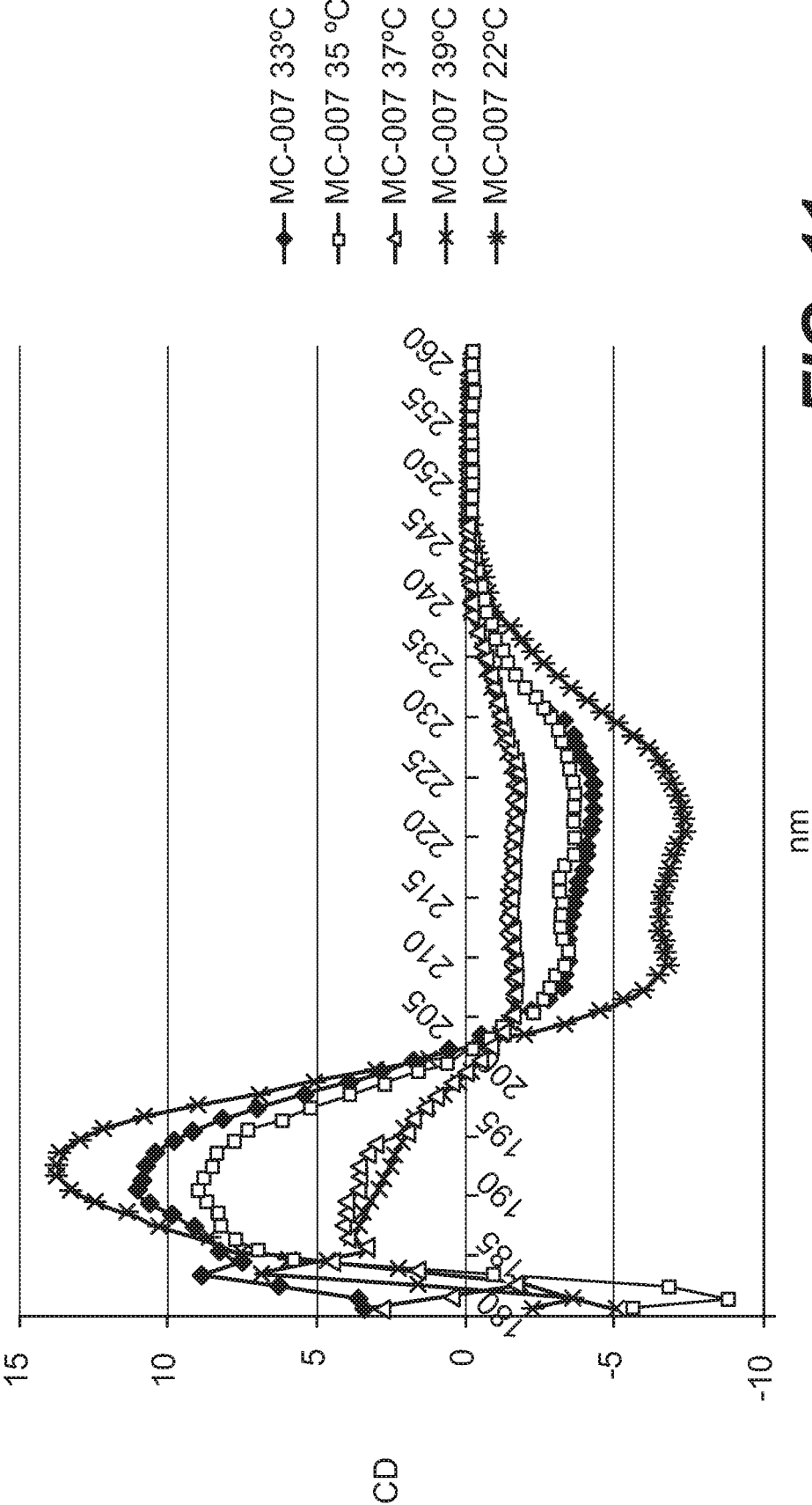
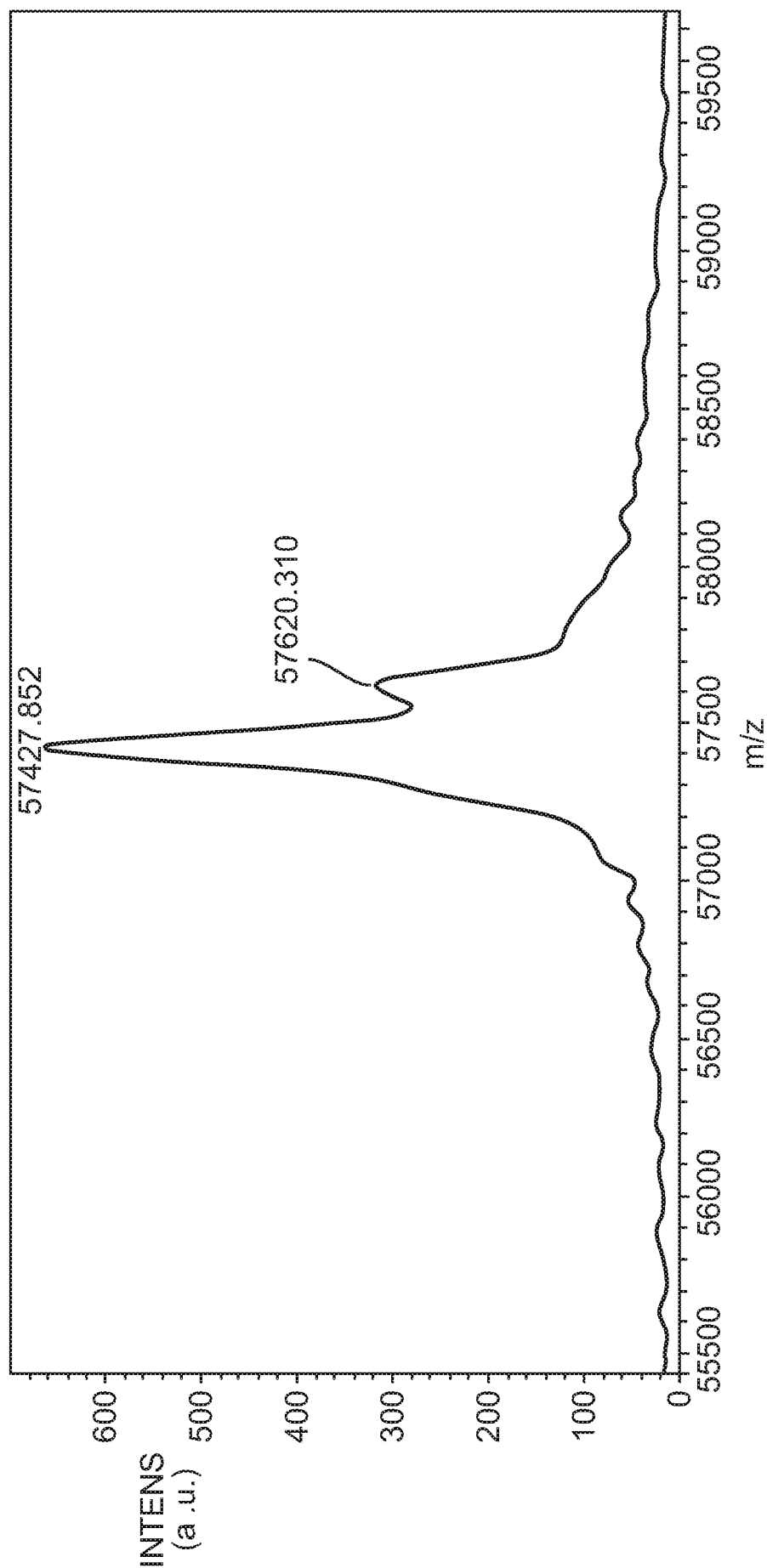


FIG. 11

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MALDI SPECTRUM OF MC-001 LOT opt-01. THE MASS OBSERVED AT 57427Da MAY BE COHERENT WITH THE DEMETHIONYLATED PROTEIN, WHILE THE PEAK AT 57620Da COULD CORRESPOND TO THE COMPLETE PROTEIN.

COMMENT 1 UspA2 opt-His lot opt 01 bm_CHCl3/MeOH_Sin
COMMENT 2

**FIG. 12**

MALDI SPECTRUM OF MC-011 LOT BMP37. THE MASS OBSERVED MAY BE
COHERENT WITH THE DEMETHIONYLATED PROTEIN. THE TWO OTHER PEAKS
AT +186Da AND +366Da ARE UNIDENTIFIED.

COMMENT 1 Uspa2_BMP37
COMMENT 2

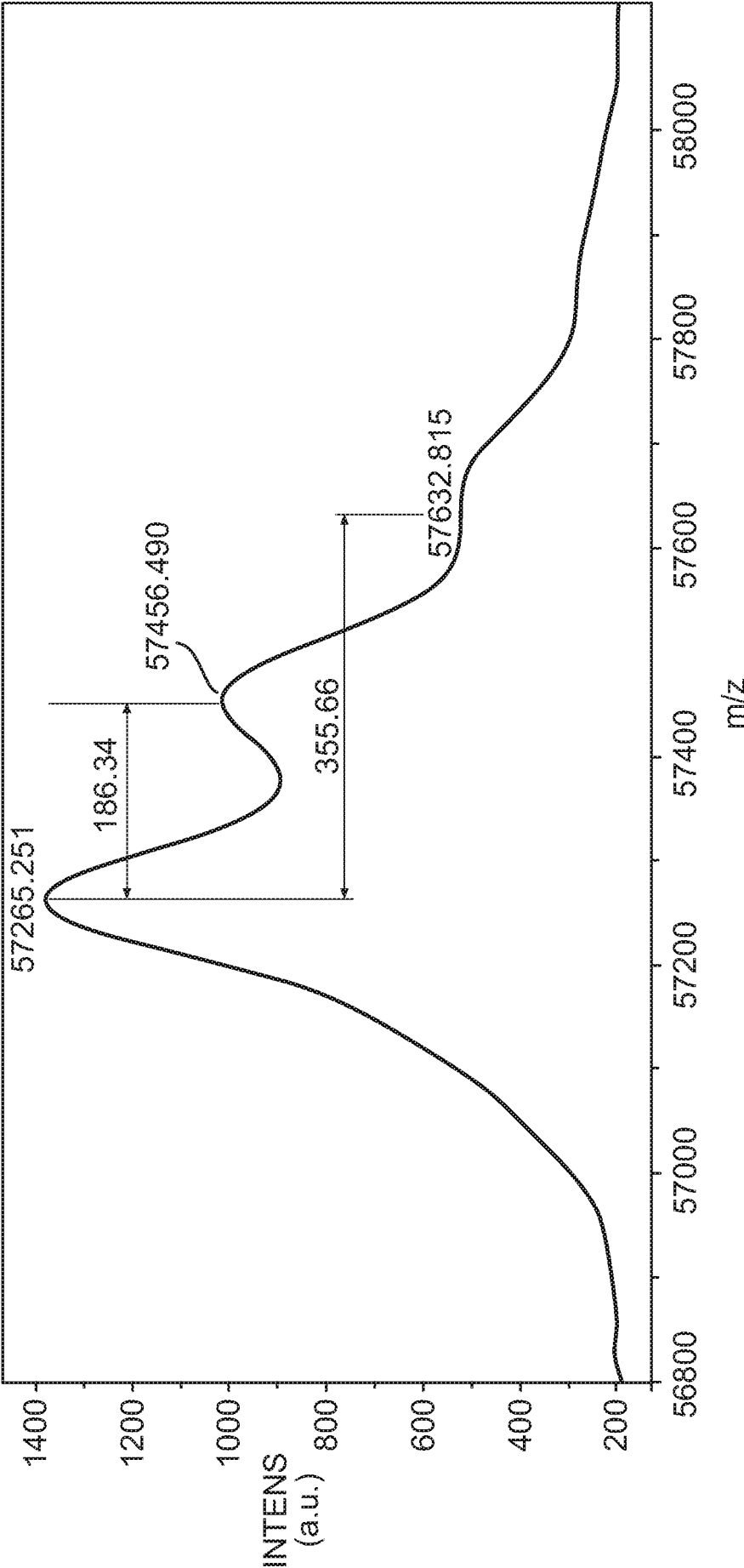


FIG. 13

PROTECTIVE EFFICACY OF MC-001 AND MC-007 IN A MOUSE MODEL OF LUNG COLONIZATION.

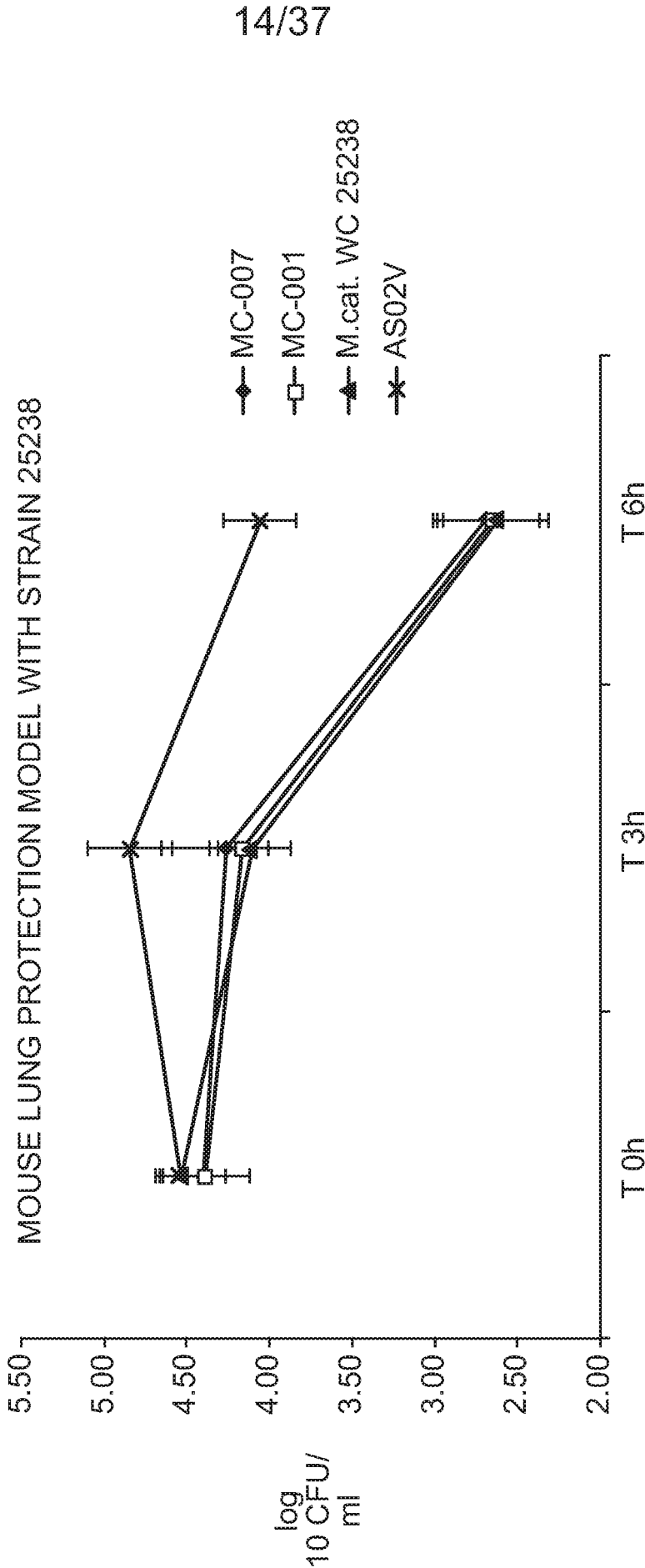


FIG. 14

ANTIBODY RESPONSE DIRECTED AGAINST UspA2 INDUCED AFTER INTRAMUSCULAR ADMINISTRATION IN MICE, WHERE PII AND PIII INDICATE, RESPECTIVELY, ANTI-IgG LEVELS IN SERA COLLECTED AT DAY 28 (POST II) AND DAY 42 (POST III).

IMMUNOGENICITY OF VARIOUS UspA2 FORMULATIONS IN MICE

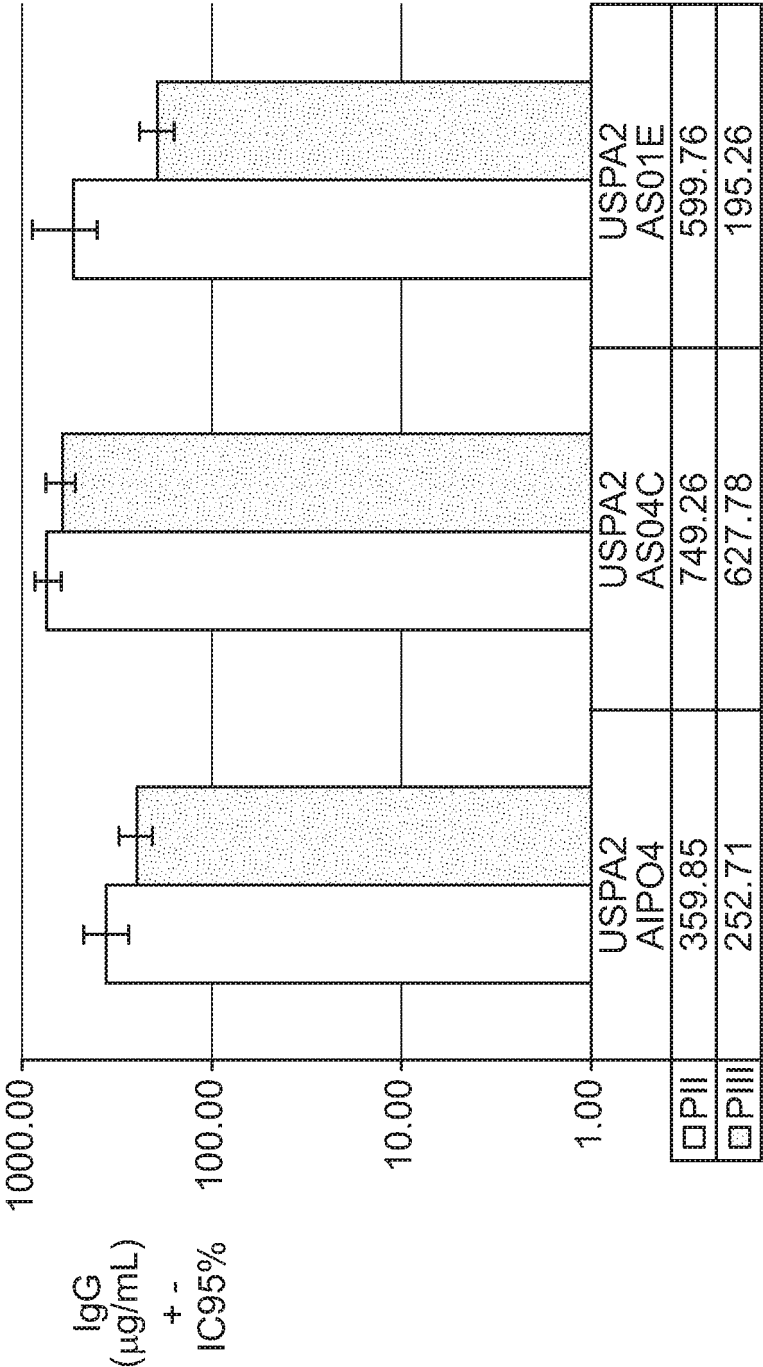


FIG. 15

BACTERICIDAL TITERS INDUCED BY UspA2 AGAINST A HOMOLOGOUS STRAIN, FORMULATED WITH DIFFERENT ADJUVANTS (AS01E, AS04C AND AIPO4). **= FIVE POOLS TESTED (5 POOLS OF FIVE SERA WERE TESTED).

BACTERICIDAL RESPONSES INDUCED BY UspA2 FORMULATED IN AS01E, AS04C, AND AIPO4

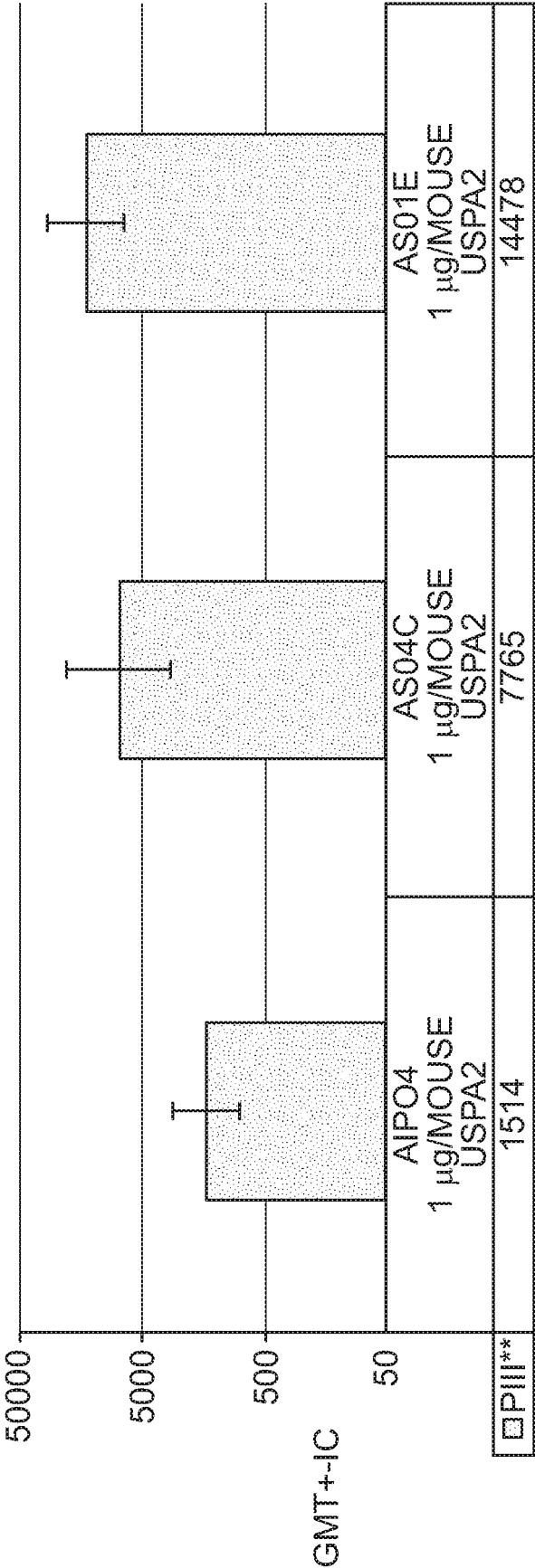


FIG. 16

ANTIBODY RESPONSE DIRECTED AGAINST UspA2 INDUCED AFTER INTRAMUSCULAR ADMINISTRATION IN MICE, USING DIFFERENT FORMULATIONS OF ANTIGENS AND ADJUVANTS.

IMMUNOGENICITY OF VARIOUS UspA2 FORMULATIONS IN MICE

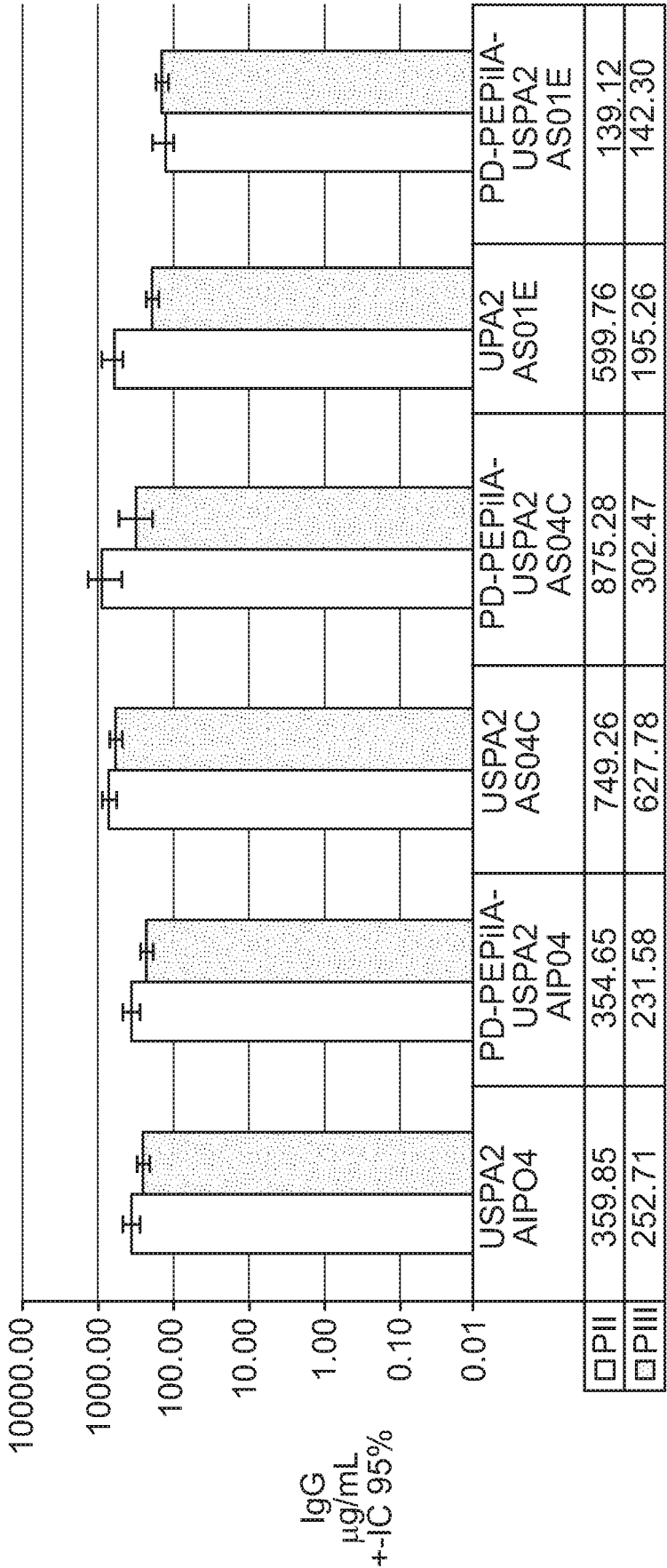


FIG. 17

BACTERICIDAL TITERS INDUCED BY UspA2 AGAINST A HOMOLOGOUS STRAIN, USING
DIFFERENT FORMULATIONS OF ANTIGENS AND ADJUVANTS. **= FIVE POOLS TESTED
(5 POOLS OF FIVE SERA WERE TESTED)

BACTERICIDAL RESPONSES INDUCED BY UspA2 FORMULATED IN AS01E, AS04C, AND AIPO4

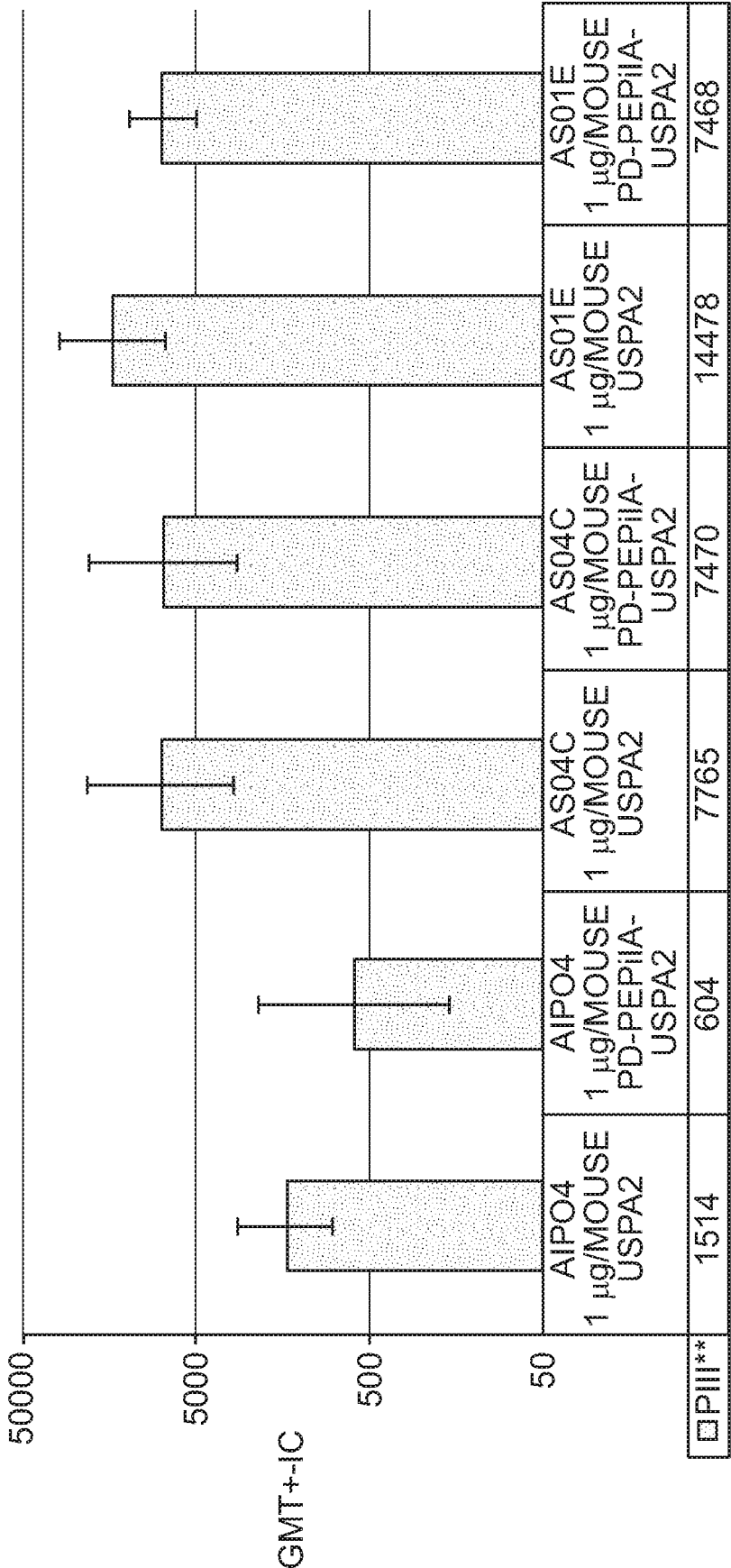


FIG. 18

IgG RESPONSE INDUCED AGAINST PD IN MICE BY PD-PEPiiA-USpA2 VACCINE
(A TRIVALENT NTHi-M.cat. VACCINE), FORMULATED WITH DIFFERENT ADJUVANTS.

IgG RESPONSE INDUCED AGAINST PD IN MICE BY TRIVALENT
NTHi-M.cat.VACCINE FORMULATIONS

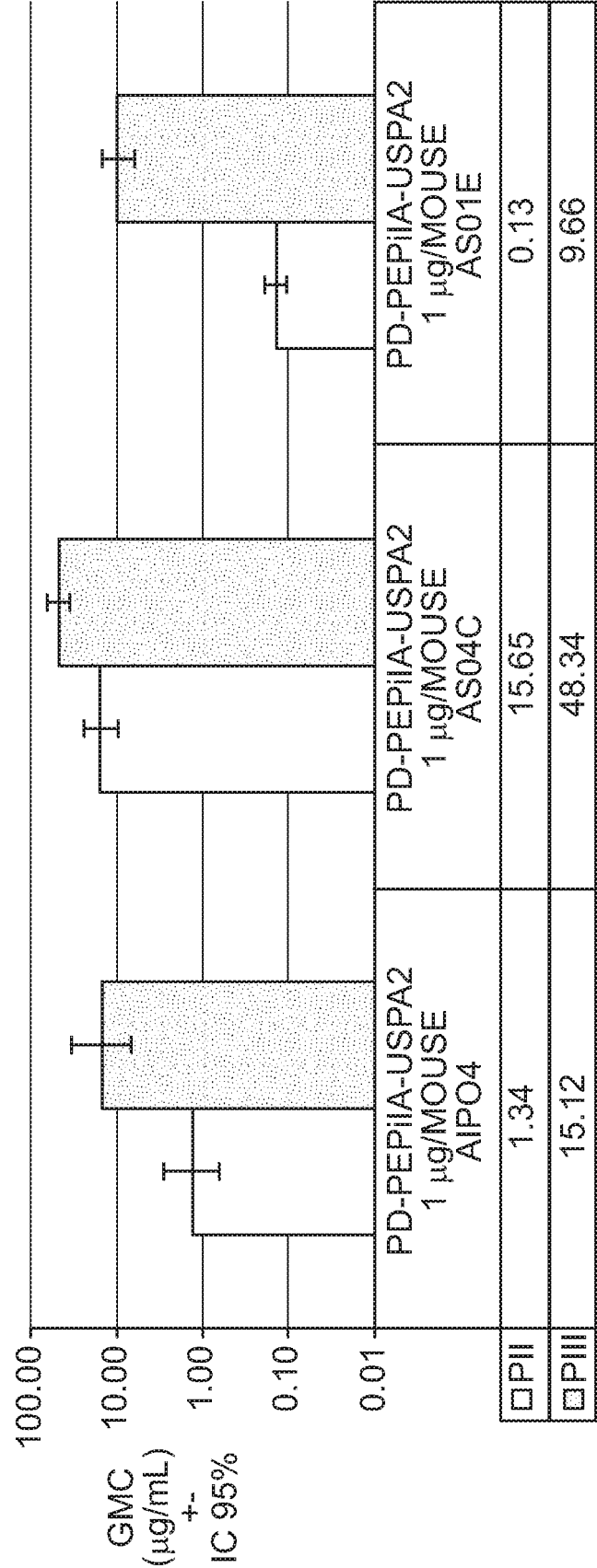


FIG. 19

IgG RESPONSE INDUCED AGAINST PE IN MICE BY PD-PEPIA-USpA2 VACCINE,
(A TRIVALENT NTHi-M.cat. VACCINE), FORMULATED WITH DIFFERENT ADJUVANTS.

IgG RESPONSE INDUCED AGAINST PE IN MICE BY TRIVALENT
NTHi-M.cat. VACCINE FORMULATIONS

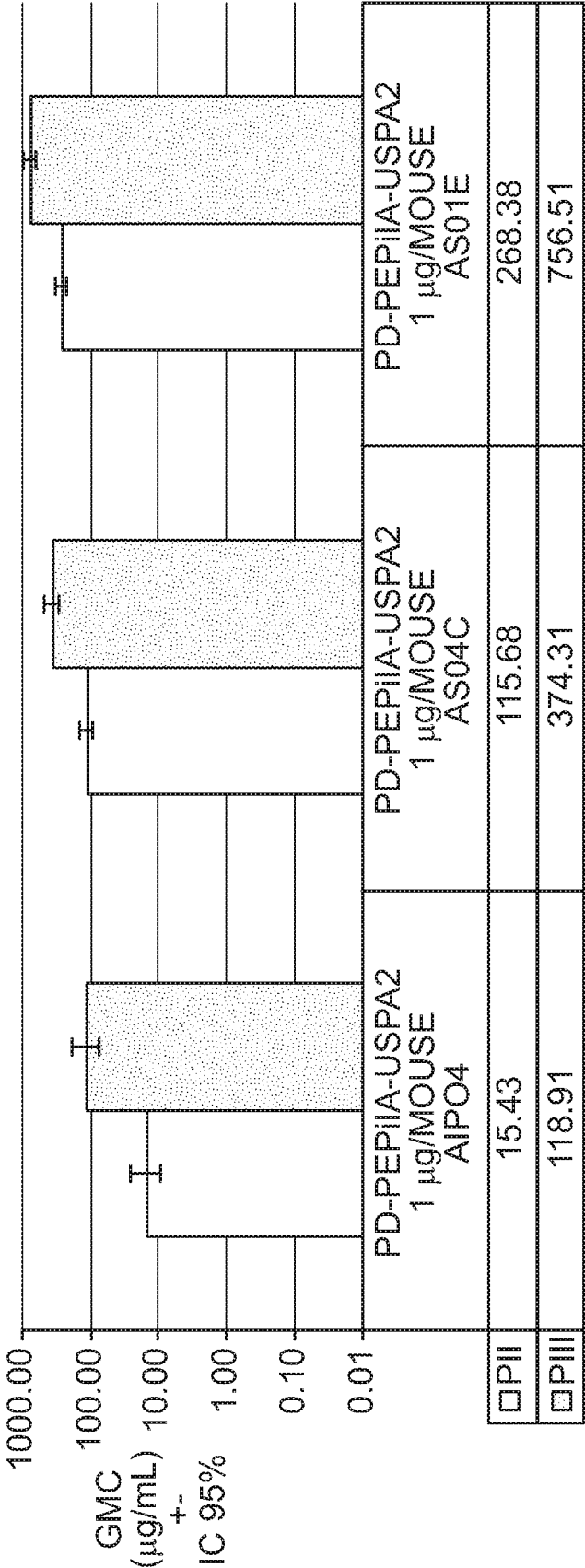


FIG. 20

IgG RESPONSE INDUCED AGAINST PIIA IN MICE BY PD-PEPIIA-USpA2 VACCINE
(A TRIVALENT NTHi-M.cat. VACCINE), FORMULATED WITH DIFFERENT ADJUVANTS.

IgG RESPONSE INDUCED AGAINST PIIA IN MICE BY TRIVALENT
NTHi-Mcat VACCINE FORMULATIONS

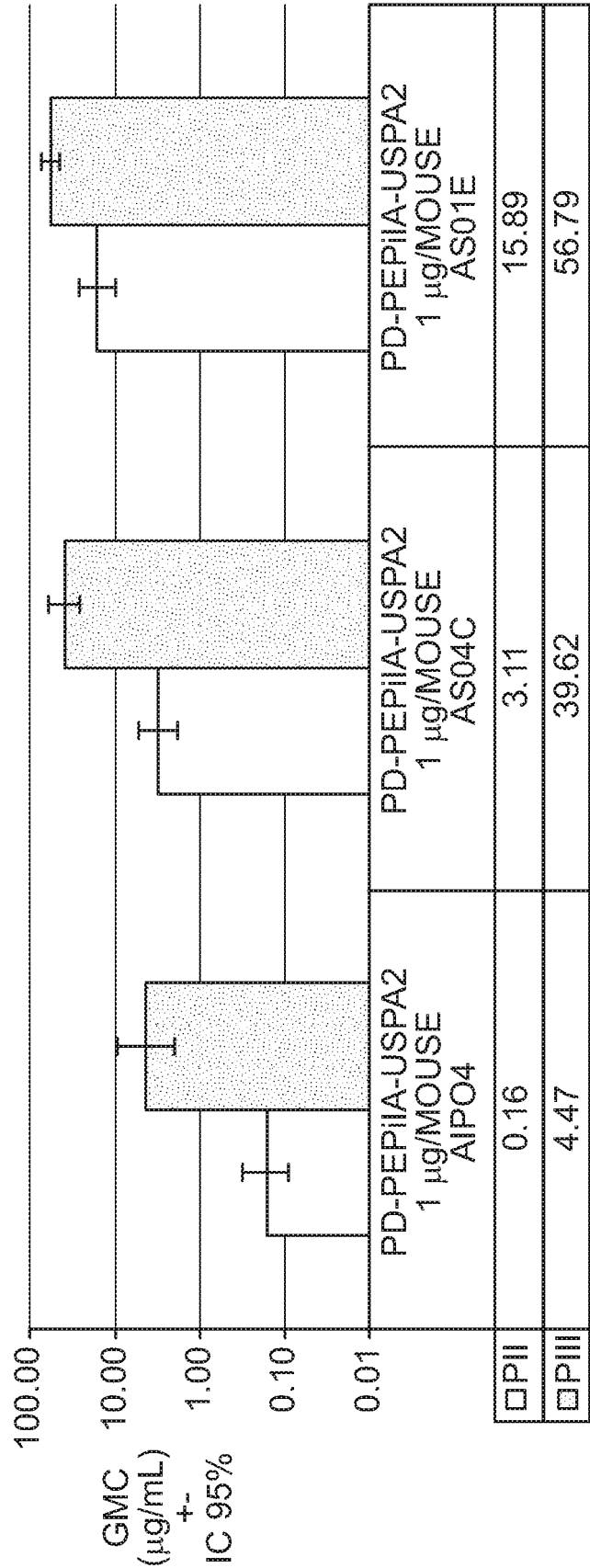


FIG. 21

IMMUNGENICITY OF PE IN THE BIVALENT NTHi PD-PEPiA AND TRIVALENT NTHi-M.cat. PD-PEPiA-USpA2 FORMULATIONS WITH AS01E.

IMPACT OF THE ADDITION OF UspA2 ON THE IMMUNGENICITY OF PD, PE AND P*i*A. ANTI-PE TITERS, 14P*II* - P*III*

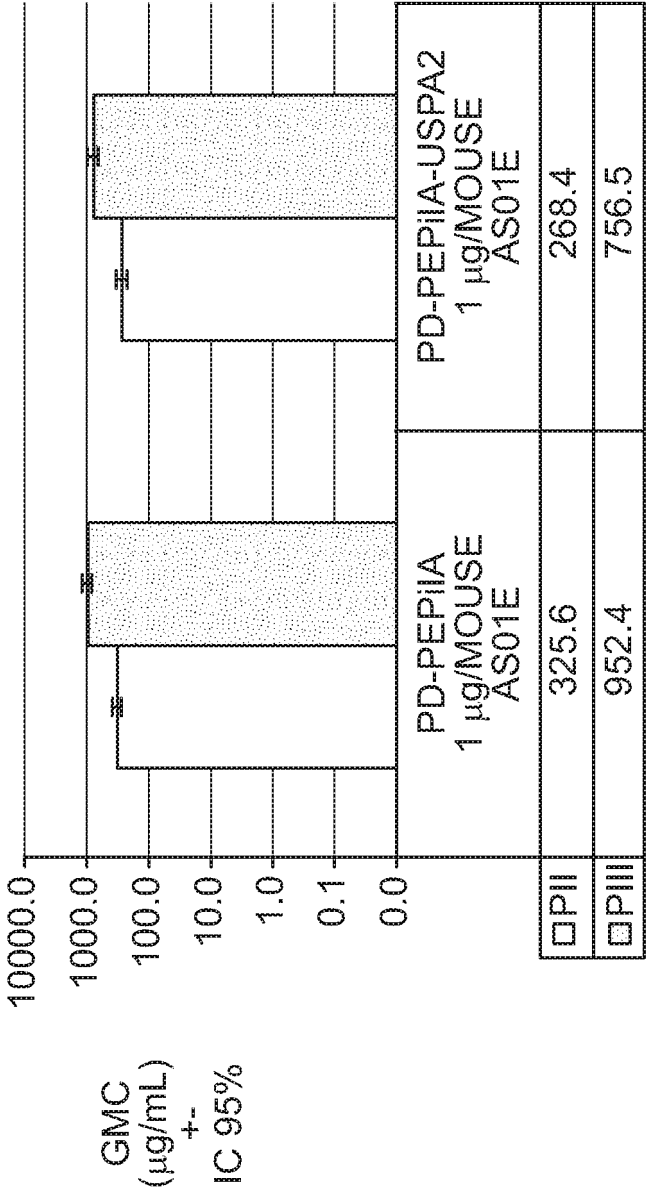


FIG. 22

IMMUNOGENICITY OF PIIA IN THE BIVALENT NTHi PD-PEPIA AND TRIVALENT NTHi-M.cat. PD-PEPIA-USpA2 FORMULATIONS WITH AS01E.

IMPACT OF THE ADDITION OF UspA2 ON THE IMMUNOGENICITY OF PD, PE AND PIIA. ANTI-PIIA TITERS, 14PII - PIII

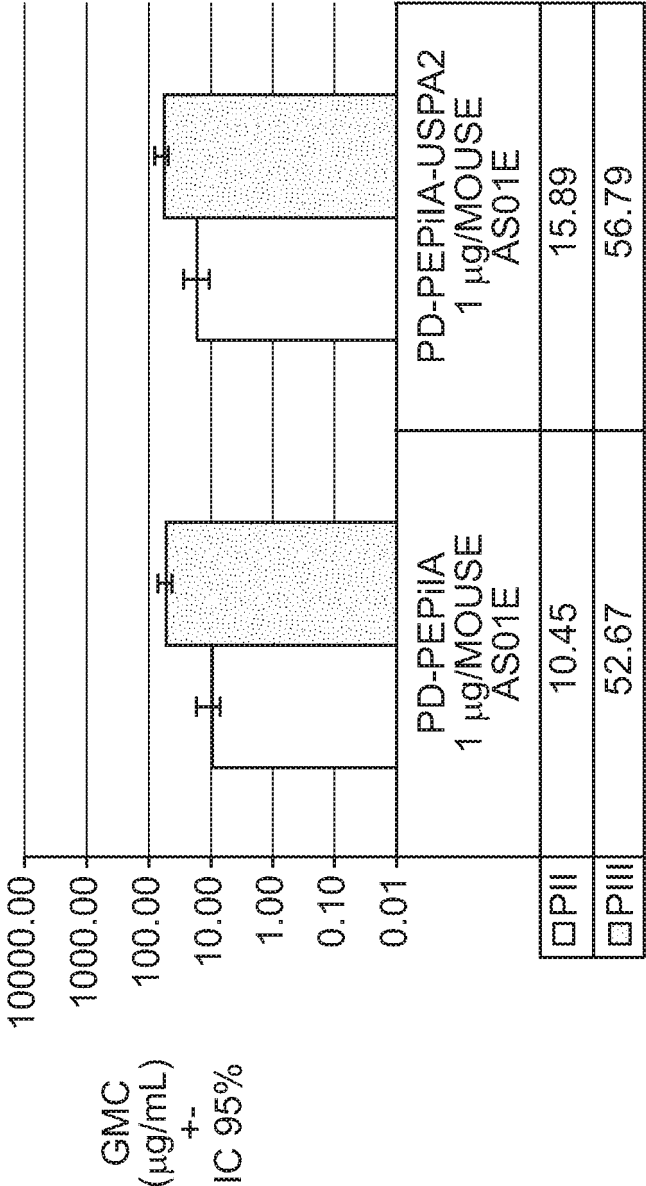


FIG. 23

IMMUNOGENICITY OF PD IN THE BIVALENT NTHi PD-PEPIIA AND TRIVALENT NTHi-M.cat. PD-PEPIIA-UspA2 FORMULATIONS WITH AS01E.

IMPACT OF THE ADDITION OF UspA2 ON THE IMMUNOGENICITY OF PD, PE AND PIIA. ANTI-PD TITERS, 14PII - PIII

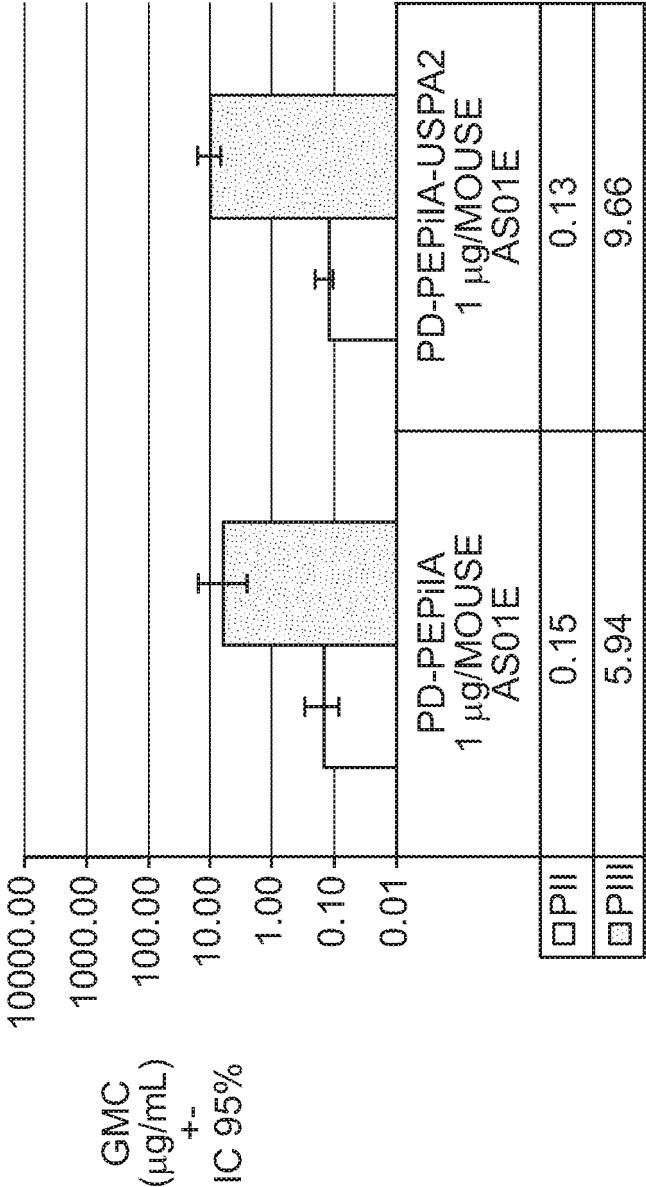
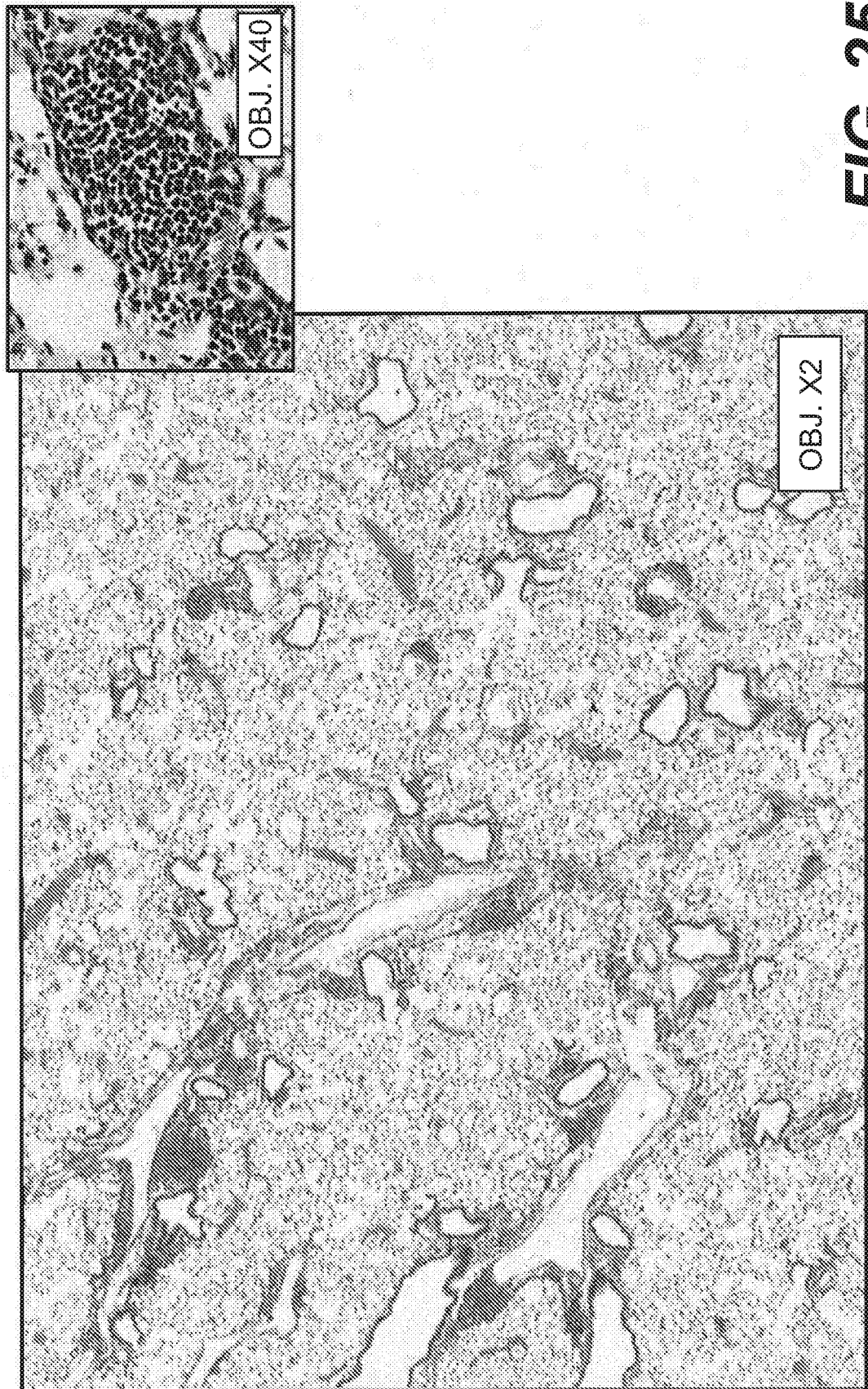


FIG. 24

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EFFECT OF THE TETRAVALENT PD/PEP1IA/ UspA2/ AS01E VACCINE FORMULATION ON MOUSE LUNGS
PRE-SENSITIZED WITH HEAT INACTIVATED M. cat. - PERIVASCULARITIS AND PERIBRONCHIOLITIS
IN PBS IMMUNIZED MICE.

**FIG. 25**

EFFECT OF THE TETRAVALENT PD/ PEPIIA/ UspA2/ AS01E VACCINE FORMULATION ON MOUSE LUNGS PRE-SENSITIZED WITH HEAT INACTIVATED M. cat. - DAY 2 POST-IMMUNIZATION

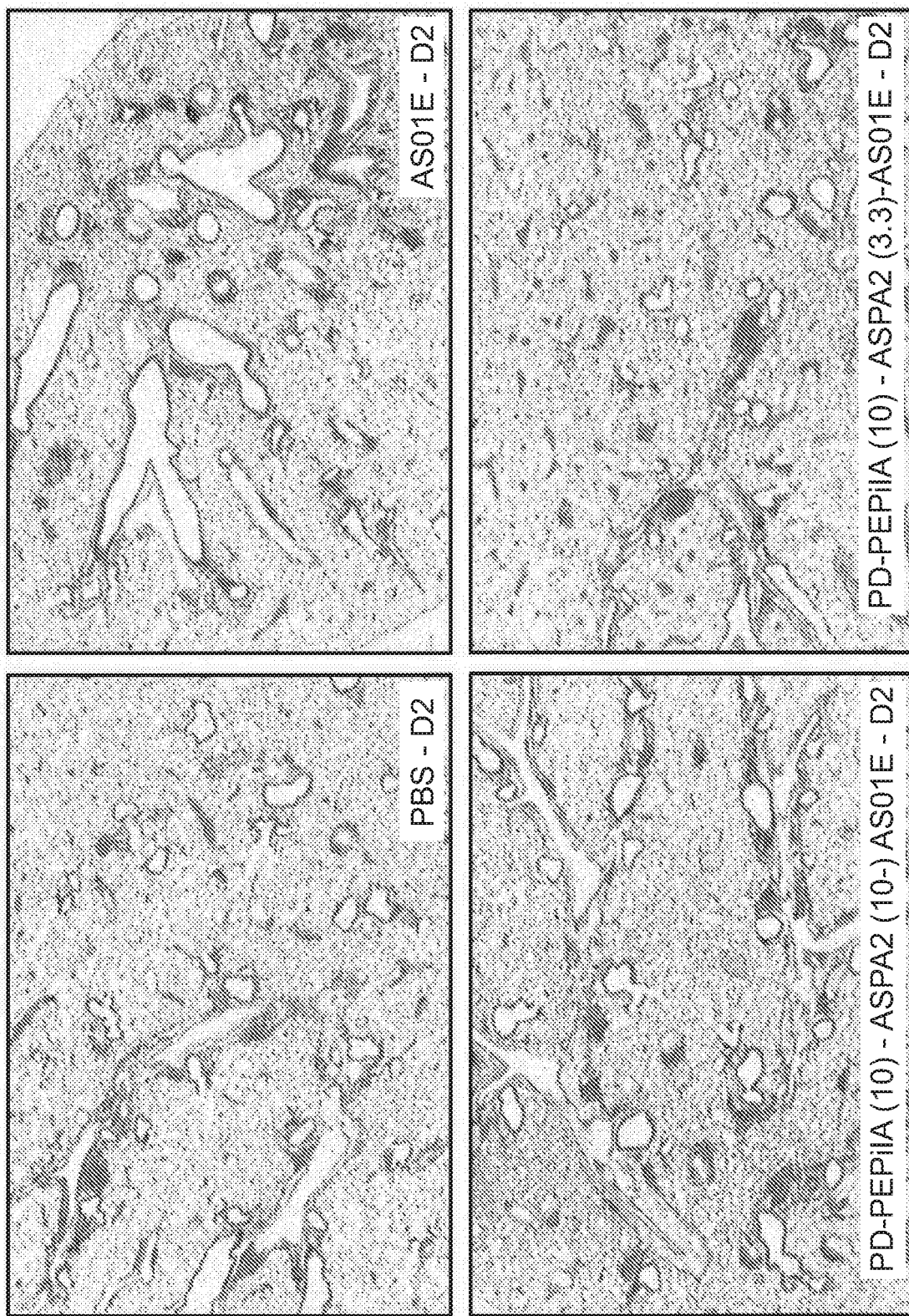


FIG. 26

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EFFECT OF THE TETRAVALENT PD/PEPIA/ UspA2/ AS01E VACCINE FORMULATION ON MOUSE LUNGS PRE-SENSITIZED WITH HEAT INACTIVATED M. cat. - DAY 7 POST-IMMUNIZATION

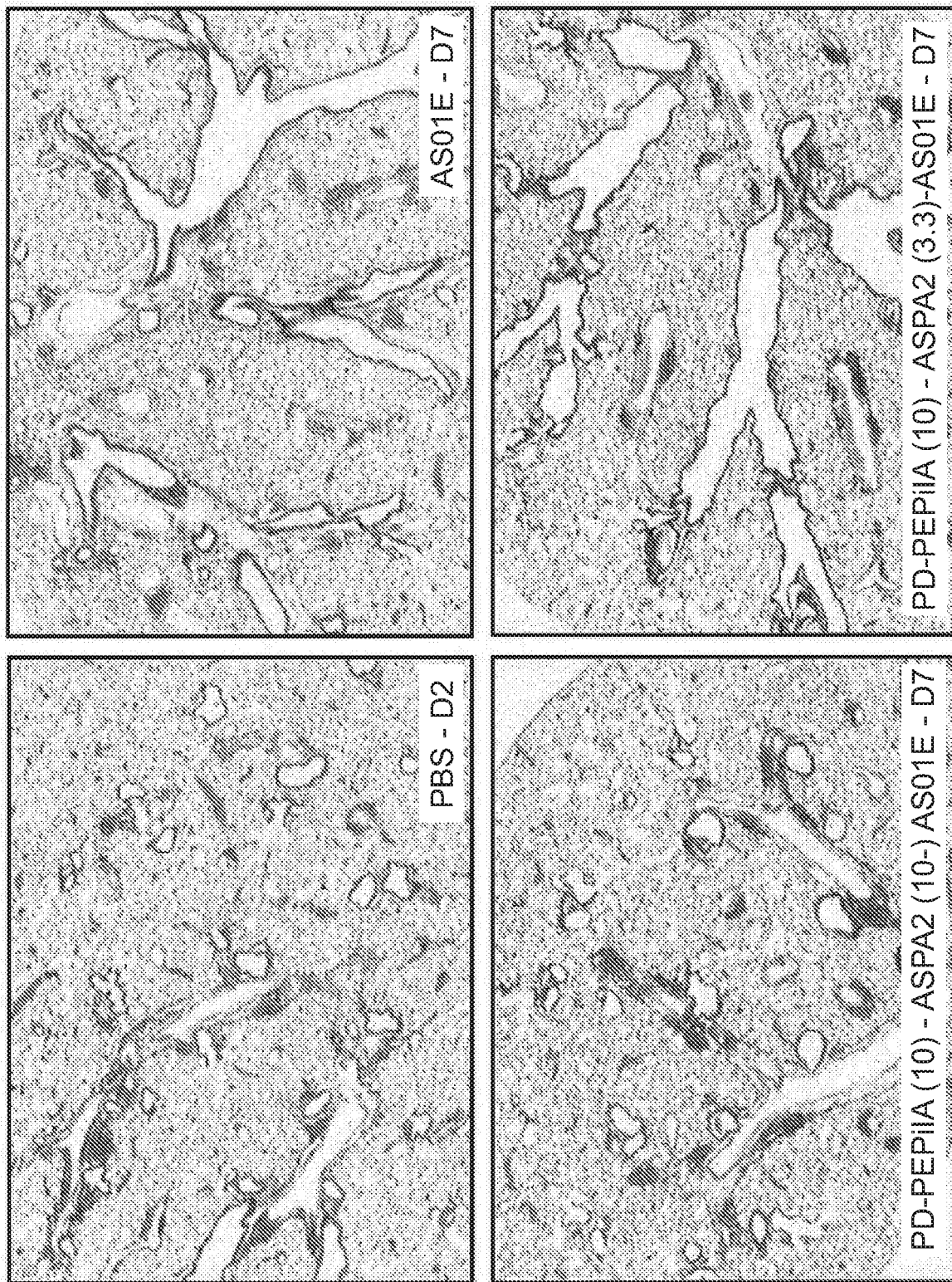


FIG. 27

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EFFECT OF THE TETRAVALENT PD/ PEPiIA/ UspA2/ AS01E VACCINE FORMULATION ON MOUSE LUNGS PRE-SENSITIZED WITH HEAT INACTIVATED M. cat. - DAY 14 POST-IMMUNIZATION

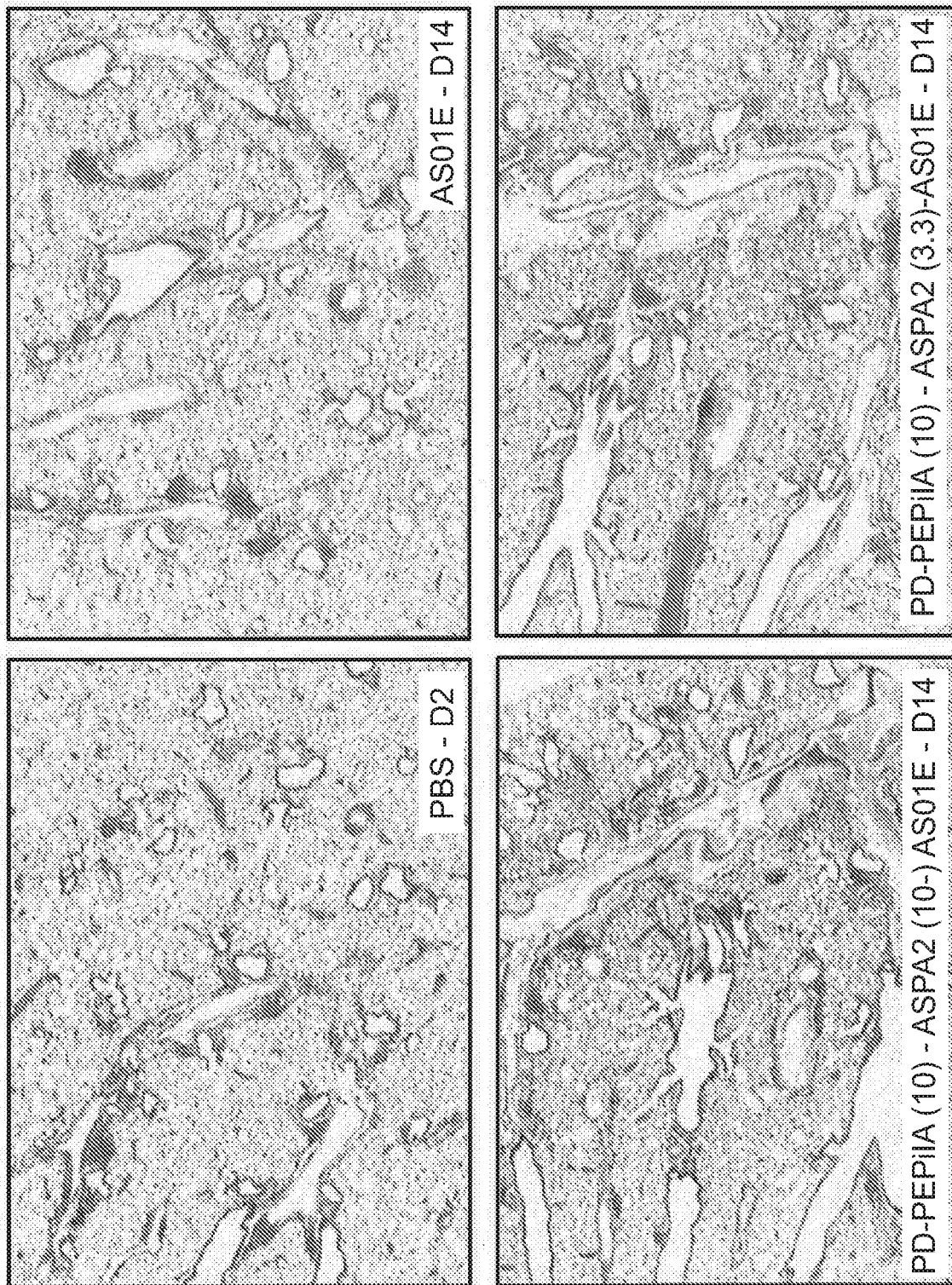


FIG. 28

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FIG. 29

EFFECT OF THE TETRAVALENT PD/ PEPIIA/ UspA2/ AS01E VACCINE FORMULATION ON
MOUSE LUNGS PRE-SENSITIZED WITH HEAT INACTIVATED M. cat. - DETAILED RESULTS

		PERIVASCULITIS/PERIBRONCHIOLITIS				
	AN. #	1	2	3	4	5
D2	PBS	3F L+M	3D L+M	3D L+M	3D L+M	A
	PD:PE-PiIA- USPA2 (10)	2D L+M	2-3D L+M	3D L+M	2-3D L+M	3D L+M
	PD:PE-PiIA- USPA2 (3.3)	2-3D L+M	2-3D L+M	2-3D L+M	3D L+M	3D L+M
	AS01E-3	3D L+M	2-3D L+M	2-3D L+M	2D L+M	3D L+M
D7	PD:PE-PiIA- USPA2 (10)	2-3D L+M	3D L+M	3D L+M	2-3D L+M	2-3D L+M
	PD:PE-PiIA- USPA2 (3.3)	3D L+M	2-3D L+M	2-3D L+M	2-3D L+M	2-3D L+M
	AS01E-3	3D L+M	2-3D L+M	2-3D L+M	2-3D L+M	2-3D L+M
	PD:PE-PiIA- USPA2 (10)	2D L+M	2-3D L+M	2-3D L+M	2-3D L+M	A
D14	PD:PE-PiIA- USPA2 (3.3)	2-3D L+M	2D L+M	2-3D L+M	2-3D L+M	A
	AS01E-3	2D L+M	2-3D L+M	2-3D L+M	2-3D L+M	2D L+M

LEGEND:

- THE FIRST LINE IN EACH BOX GIVES INFORMATION ABOUT THE INFLAMMATION SEVERITY: A = ABSENT; 1 = MINIMAL; 2 = SLIGHT; 3 = MODERATE; 4 = MARKED; 5 = SEVERE, AND THE ALTERATION DISTRIBUTION: F = FOCAL; M = MULTIFOCAL; D = DIFFUSE.
- THE SECOND LINE REFERS TO THE NATURE OF THE CELLS OBSERVED: L = LYMPHOCYTES; M = MACROPHAGES.

POST-VACCINATION LUNG CD4 T CELL RESPONSES UPON M. cat. WC RE-STIMULATION: LUNG CD4 CELLS EXPRESSING IL17. RESTIMULATED WITH HEAT-INACTIVATED M. cat. WHOLE CELLS (WC) OR MEDIUM.

20140268

LUNG CD4 CELLS EXPRESSING IL-17 STIM MEDIUM VS WC
DAYS 7 & 14 POST-VACCINATION

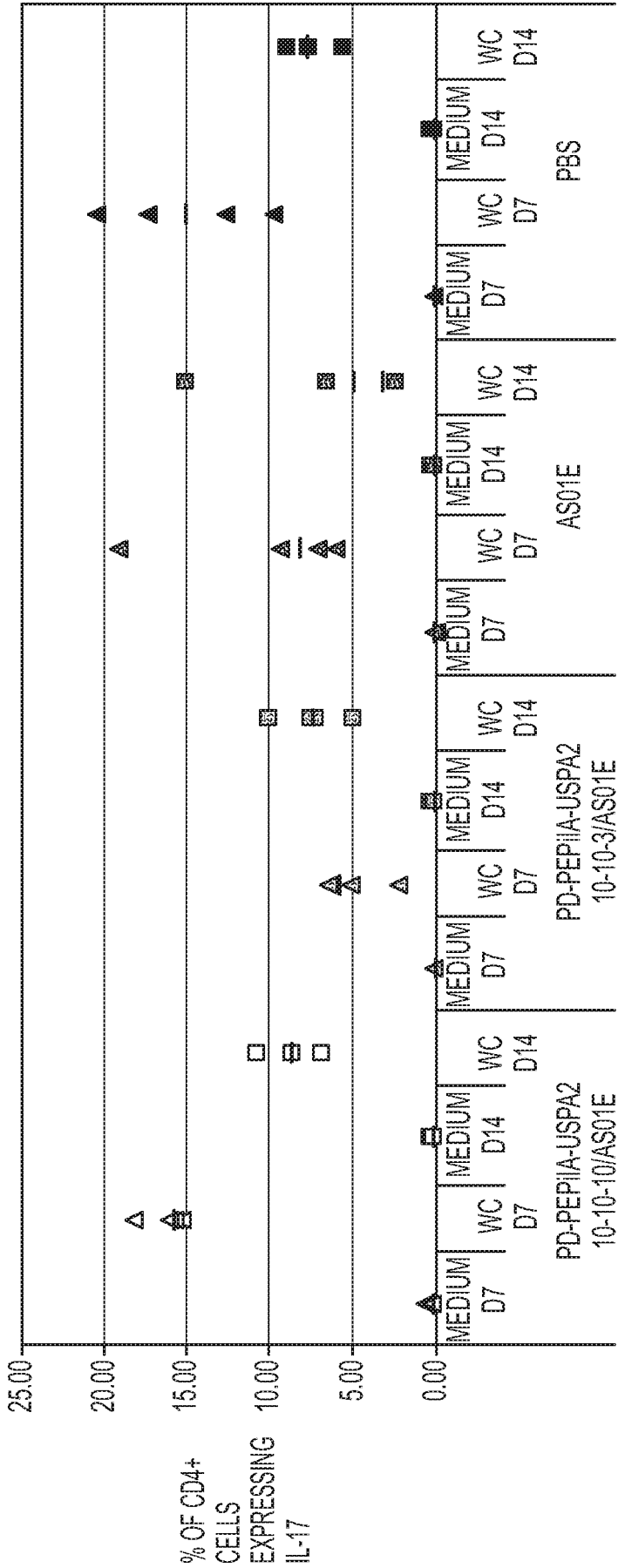


FIG. 30

POST-VACCINATION LUNG CD4 T CELL RESPONSES UPON M. cat. WC RE-STIMULATION. LUNG CD4 CELLS EXPRESSING TNF α . RESTIMULATED WITH HEAT-INACTIVATED M. cat. WHOLE CELLS (WC) OR MEDIUM.

20140268

LUNG CD4 CELLS EXPRESSING TNF α STIM MEDIUM VS WC
DAYS 7 & 14 POST-VACCINATION

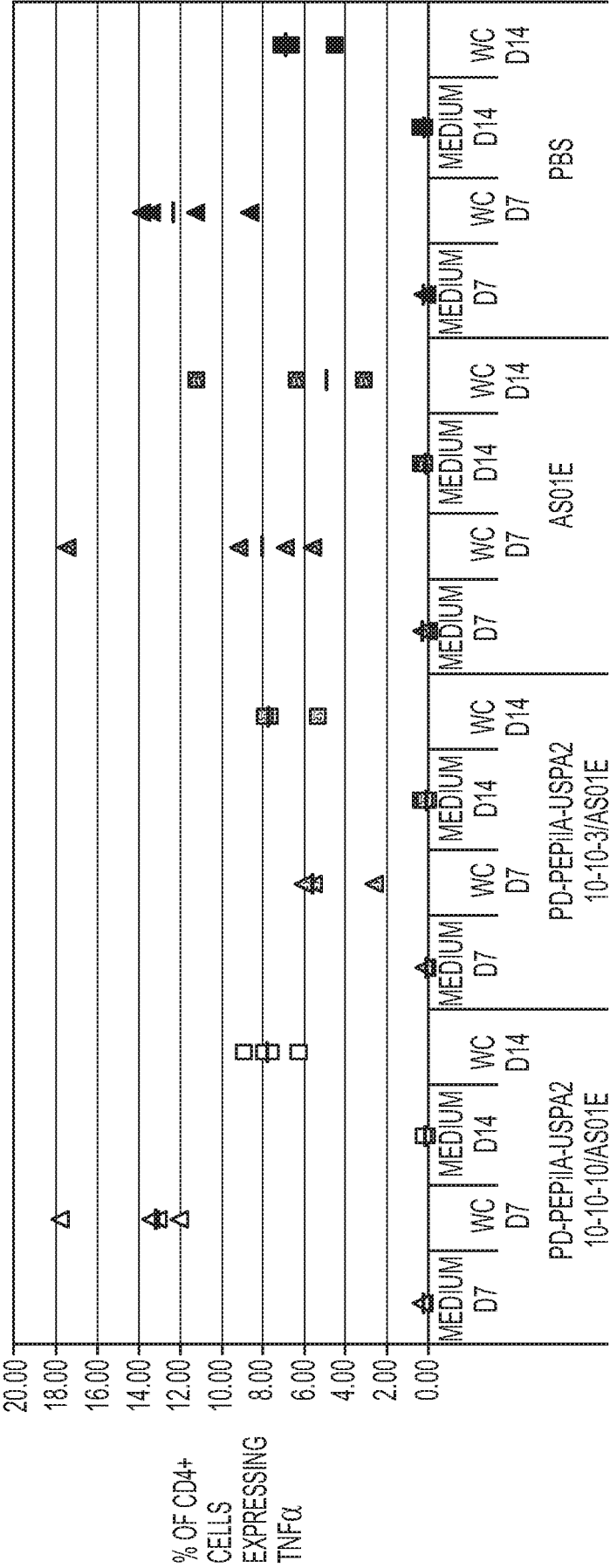


FIG. 31

POST-VACCINATION LUNG CD4 T CELL RESPONSES UPON M. cat. WC RE-STIMULATION. LUNG CD4 CELLS EXPRESSING INF γ . RESTIMULATED WITH HEAT-INACTIVATED M. cat. WHOLE CELLS (WC) OR MEDIUM.

20140268

LUNG CD4 CELLS EXPRESSING INF γ STIM MEDIUM VS WC
DAYS 7 & 14 POST-VACCINATION

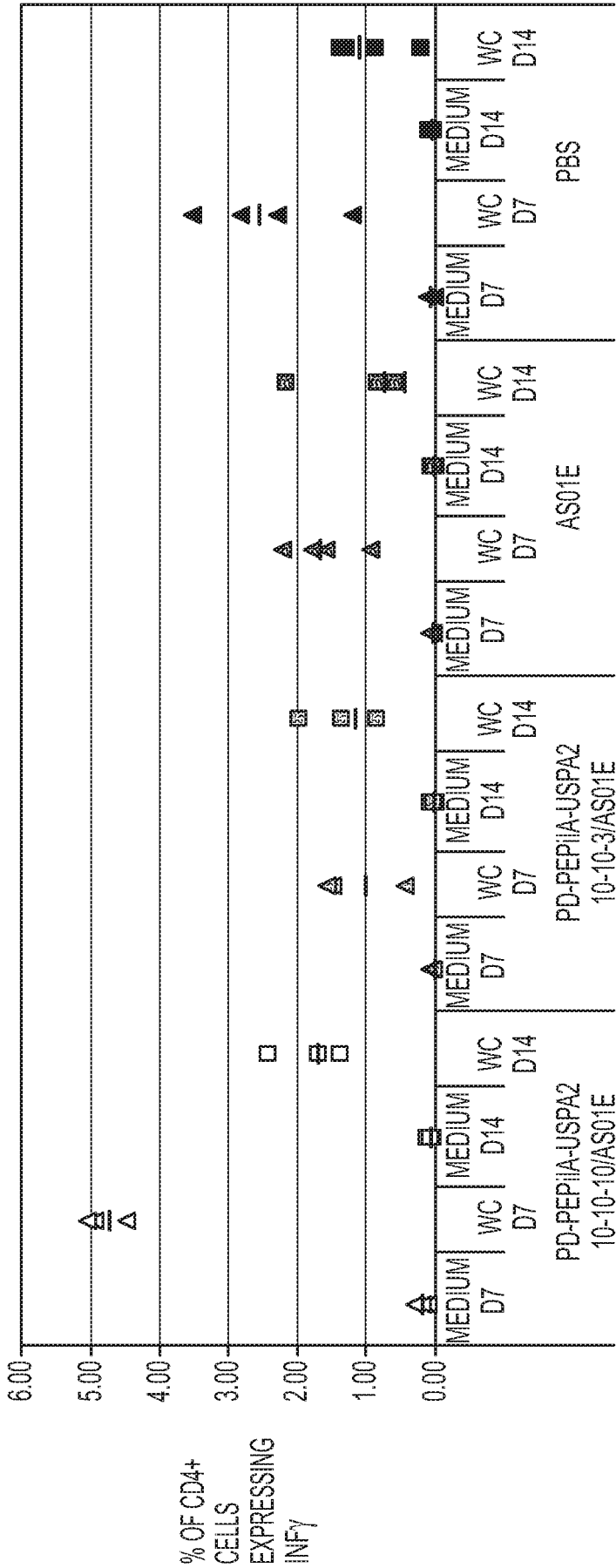


FIG. 32

POST-VACCINATION LUNG CD4 T CELL RESPONSES UPON M. cat. WC RE-STIMULATION. LUNG CD4
CELLS EXPRESSING IL-13. RESTIMULATED WITH HEAT-INACTIVATED M. cat. WHOLE CELLS (WC) OR MEDIUM.

20140268

LUNG CD4 CELLS EXPRESSING IL-13 STIM MEDIUM VS WC
DAYS 7 & 14 POST-VACCINATION

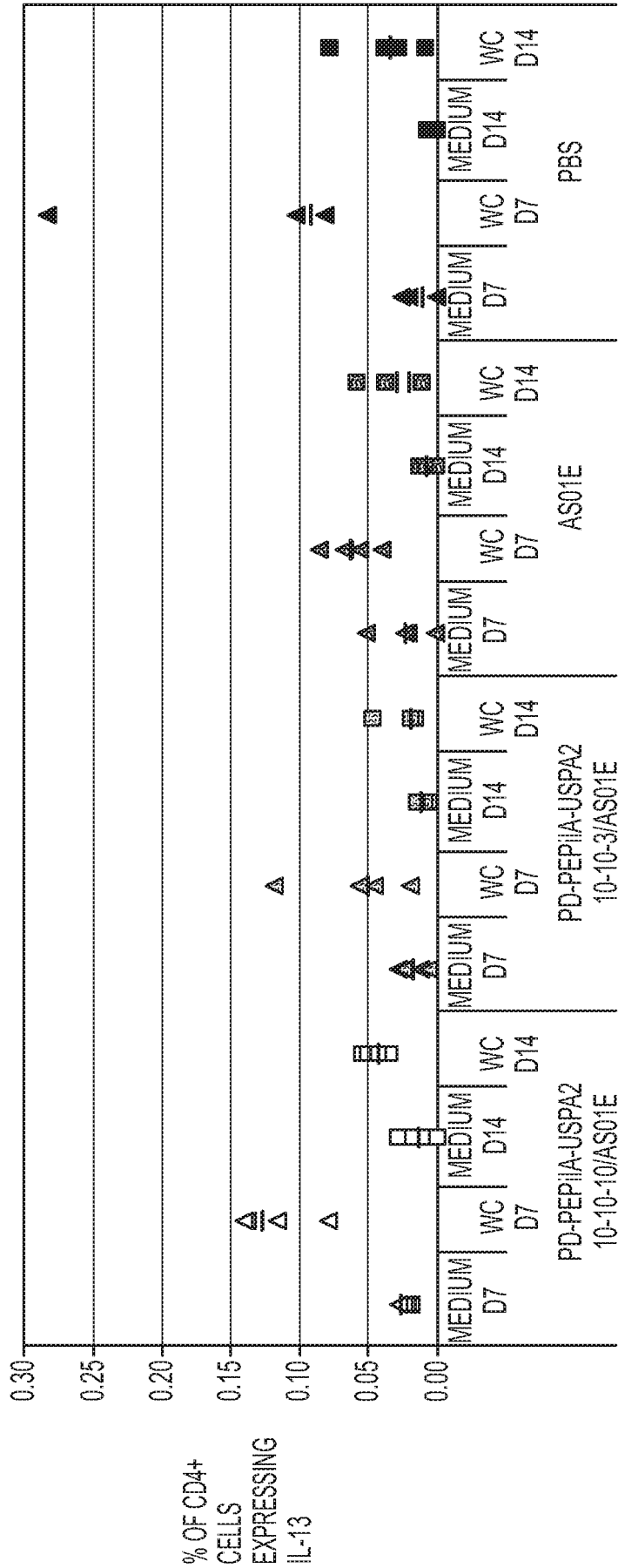


FIG. 33

POST-CHALLENGE LUNG CD4 T CELL RESPONSES UPON M. cat. WC RE-STIMULATION. LUNG CD4
CELLS EXPRESSING INF γ . RESTIMULATED WITH HEAT-INACTIVATED M. cat. WHOLE CELLS (WC) OR MEDIUM.

20140416

LUNG CD4 CELLS EXPRESSING INF γ STIM MEDIUM VS WC
DAYS 7 & 14 POST-CHALLENGE

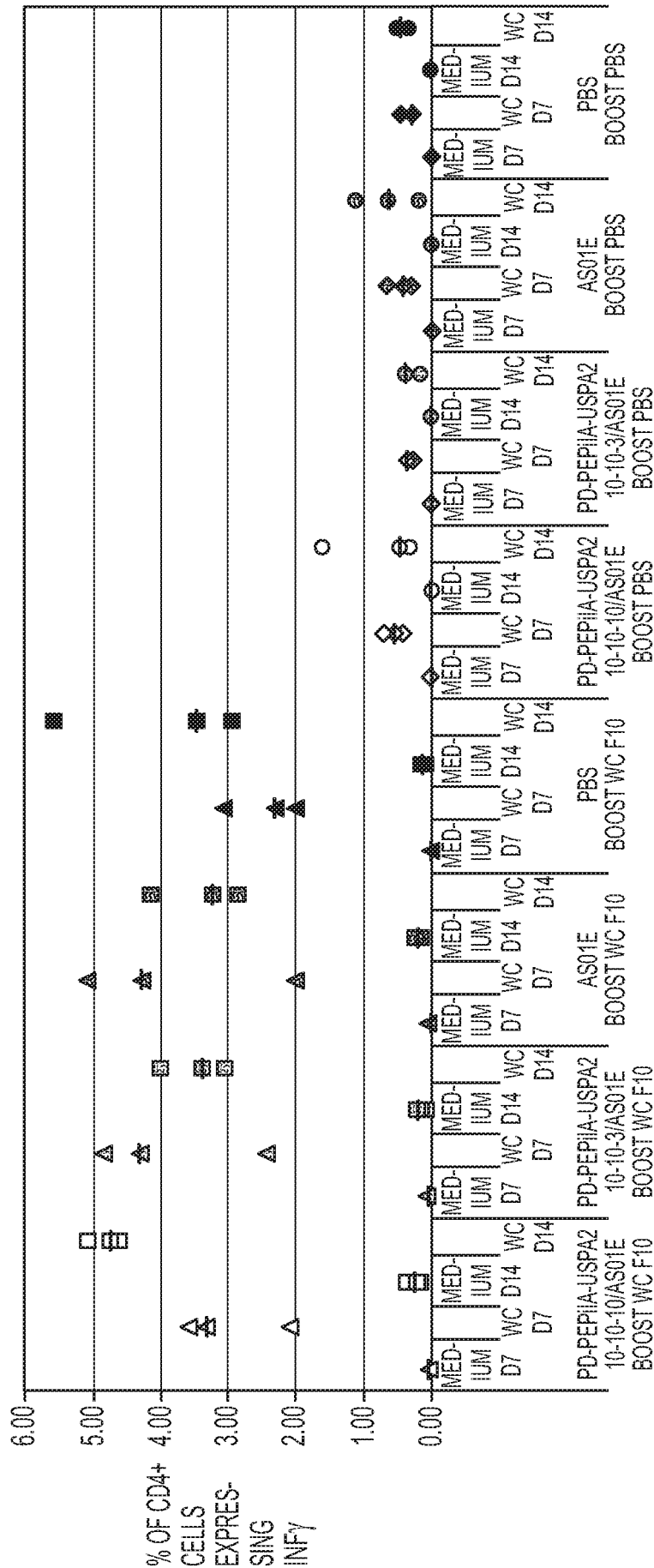


FIG. 36

POST-CHALLENGE LUNG CD4 T CELL RESPONSES UPON M. cat. WC RE-STIMULATION. LUNG CD4 CELLS EXPRESSING IL-13. RESTIMULATED WITH HEAT-INACTIVATED M. cat. WHOLE CELLS (WC) OR MEDIUM.

20140416

LUNG CD4 CELLS EXPRESSING IL-13 STIM MEDIUM VS WC
DAYS 7 & 14 POST-CHALLENGE

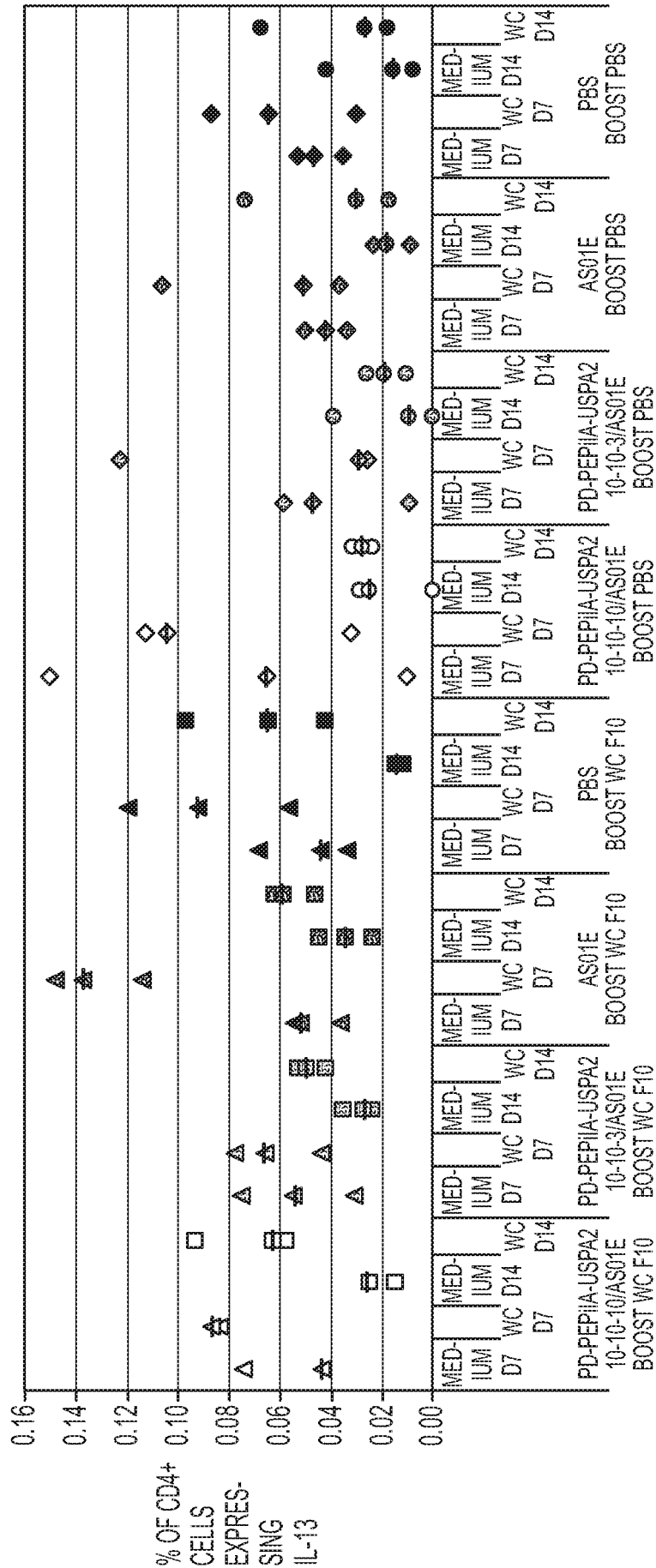


FIG. 37