

Supplementary Materials: Polyacrylate-GnRH Peptide Conjugate as an Oral Contraceptive Vaccine Candidate

Mohammad Omer Faruck, Prashamsa Koirala, Jieru Yang, Michael J. D’Occhio, Mariusz Skwarczynski and Istvan Toth

Table S1. Element microanalysis of PMA, **1** and **2**.

Conjugates	Theorital Value			Experimental Value		
	N%	C%	N/C ratio	N%	C%	N/C ratio
PMA	1.53	55.00	0.028	1.51	54.80	0.028
PMA-P-GnRH (1)	10.27	49.81	0.206	10.19	49.80	0.204
PMA-PT-GnRH (2)	10.84	55.18	0.197	10.73	54.89	0.196

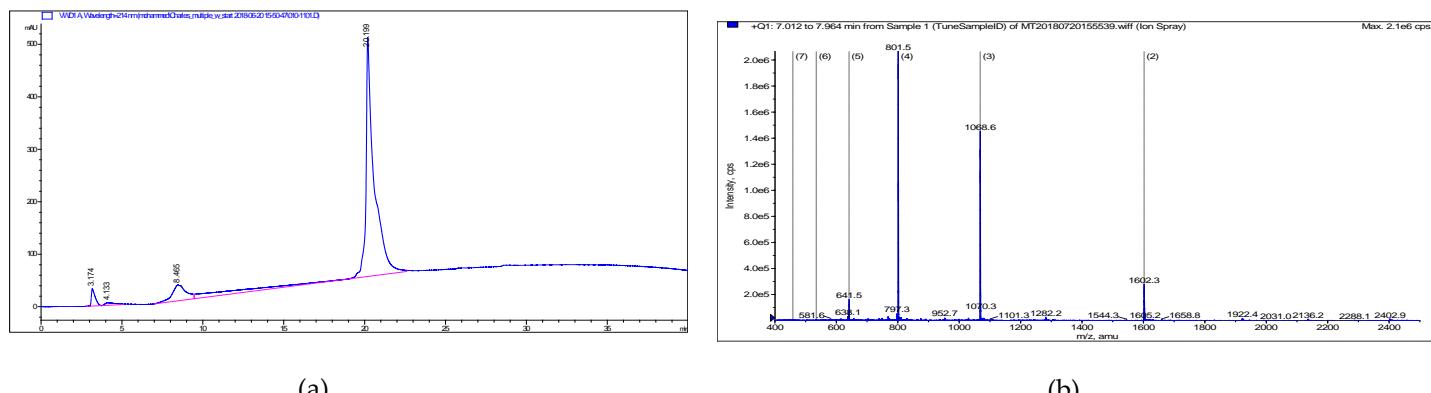


Figure S1. Analysis of purified compound PADRE-GnRH with alkyne moiety (a) HPLC (b)ESI-MS. HPLC, C18 column, t_r 20.1 min, purity 96%, yield 43%. Analytical RP-HPLC graphs show pure compounds in single peak. The mass from these peaks, matched to the desired.

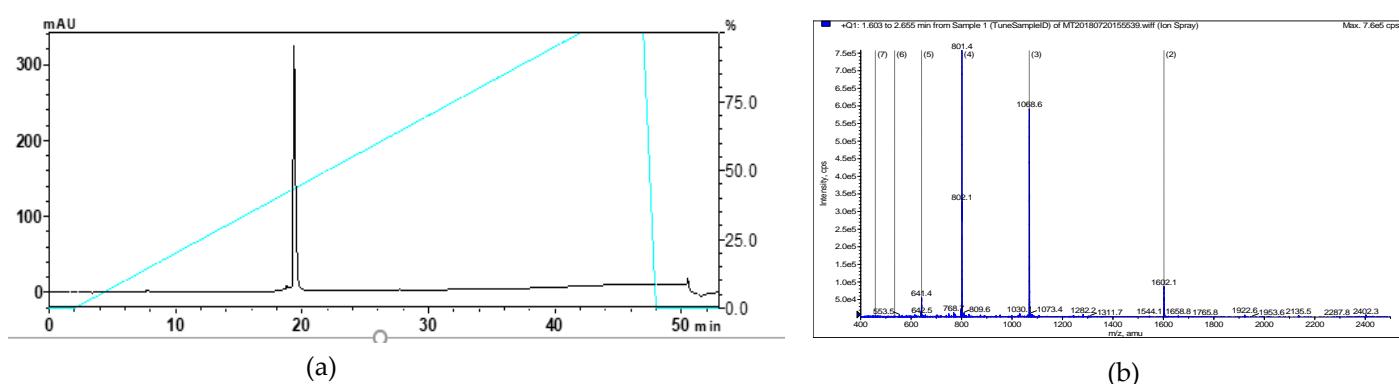


Figure S2. Analysis of purified compound pig T helper-GnRH with alkyne moiety (a) HPLC (b)ESI-MS. HPLC, C18 column, HPLC C18 column, t_r 19.8 min, purity 99%, yield 43% Analytical RP-HPLC graphs show pure compounds in single peak. The mass from these peaks, matched to the desired compounds in ESI-MS.

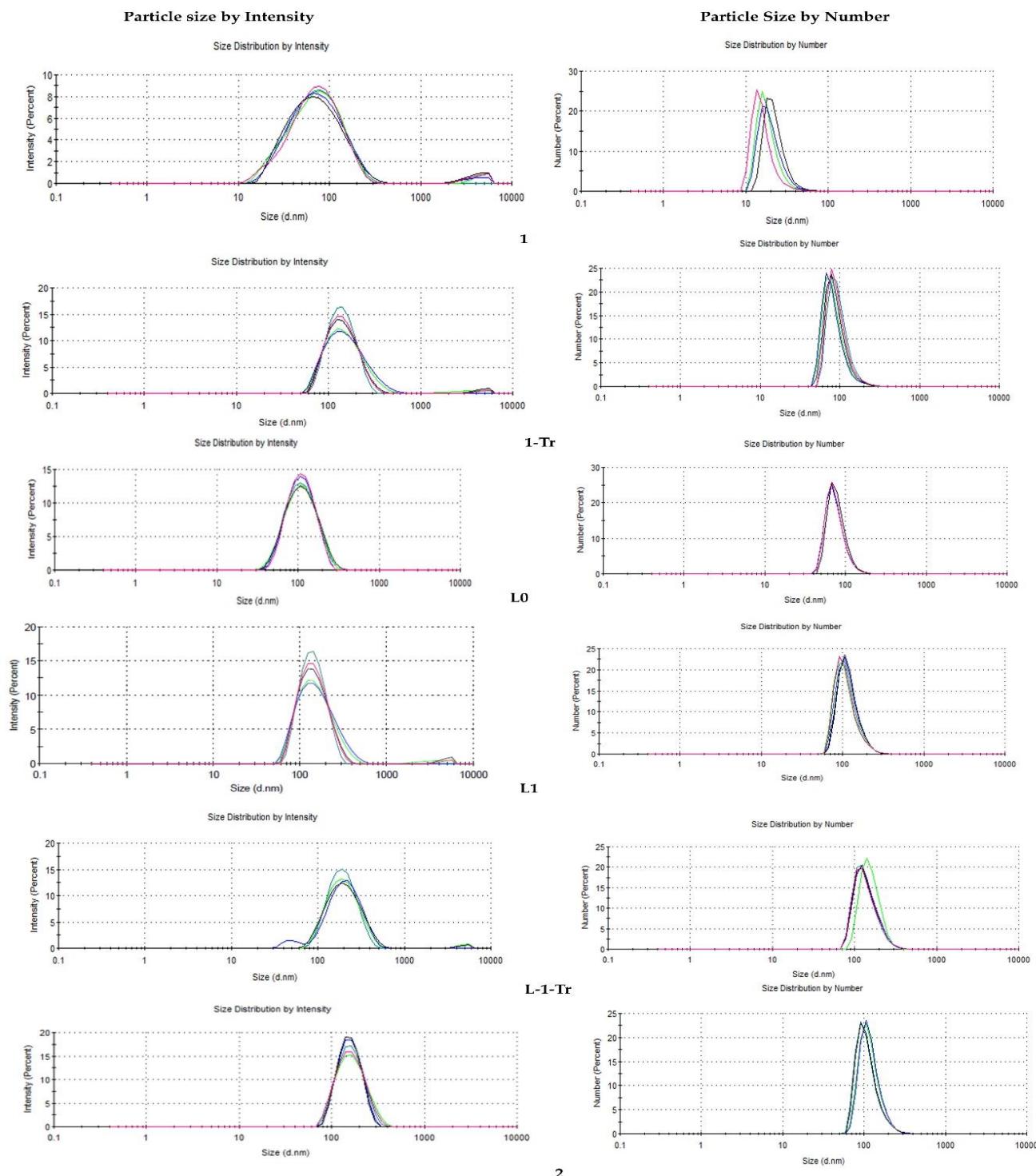


Figure S3. Particle size distribution by intensity and Particle size distribution by number of all vaccine compounds. **1** particle size 62 ± 1 nm PDI 0.36 ± 0.03 , compound **1-Tr** particle size 140 ± 4 nm, PDI 0.19 ± 0.07 , **L-0** particle size 99 ± 1 nm, PDI 0.14 ± 0.03 , **L-1** particle size 146 ± 2 , PDI 0.19 ± 0.04 , **L-1-Tr** particle size 171 ± 2 nm, PDI 0.22 ± 0.09 , Compound **2** particle size 141 ± 5 nm, PDI 0.08 ± 0.02 .

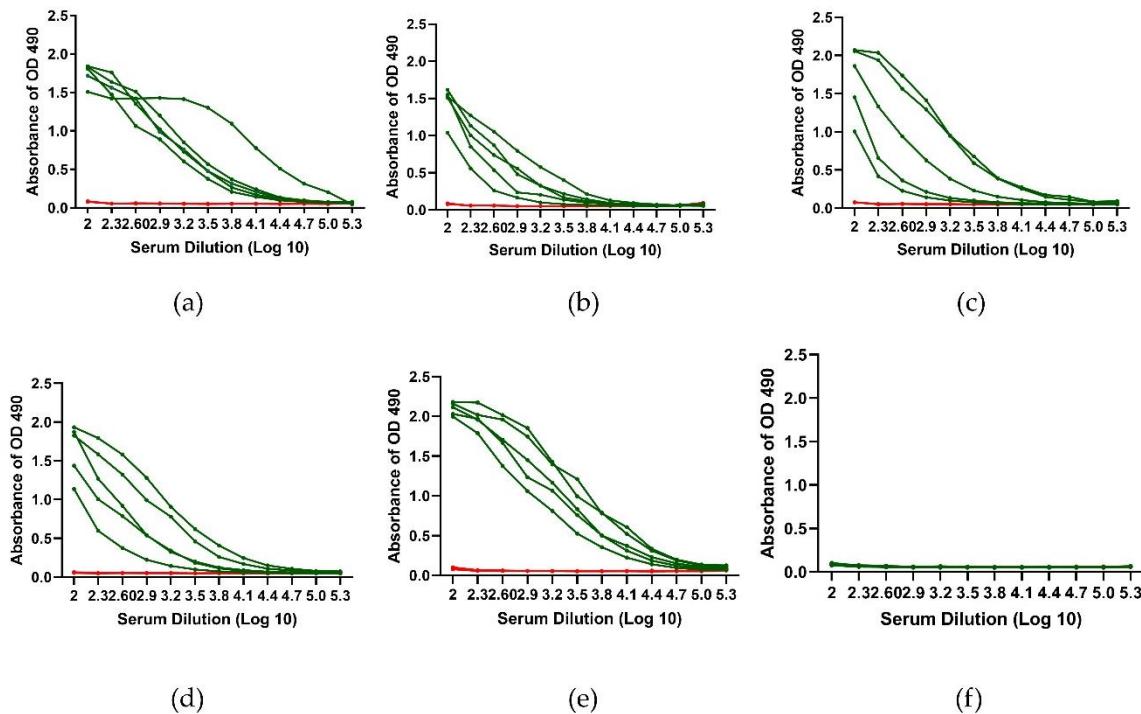


Figure S4. GnRH-Specific antibody (IgG) responses following subcutaneous administration of the vaccine candidates in mice. Optical density (OD) value from ELISA analysis of log serum dilution after single immunization (a) 1, (b) 1-Tr, (c) L-1, (d) L-1-Tr, (e) 1+CFA, (f) PBS.

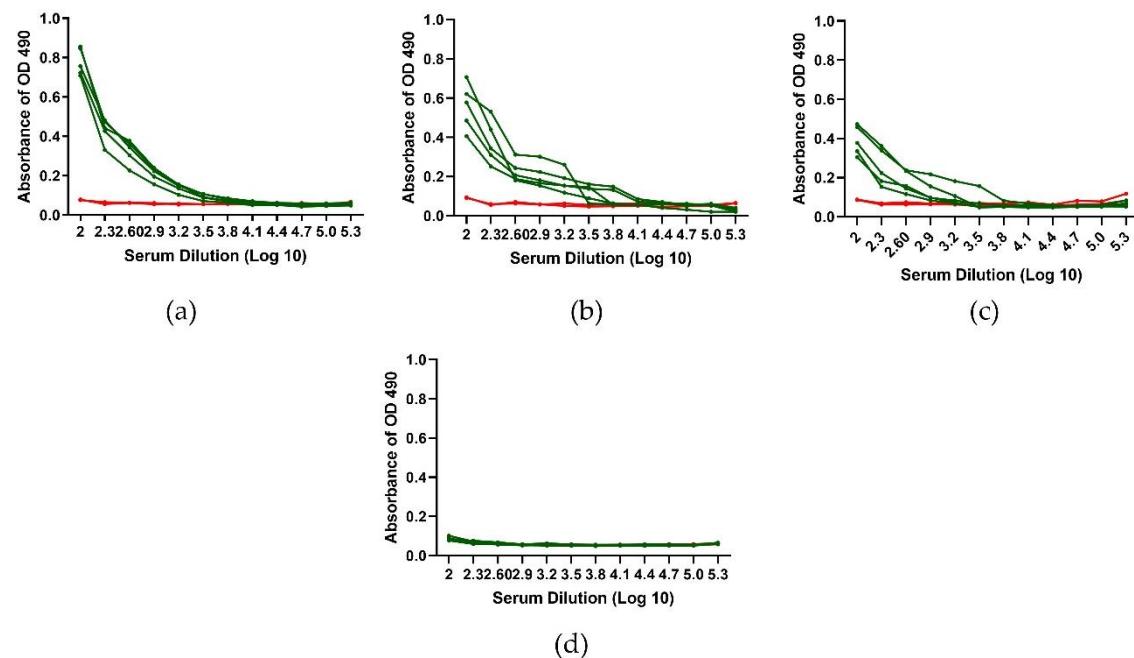


Figure S5. GnRH-Specific antibody (IgG) responses following subcutaneous administration of the vaccine candidates in mice. Optical density (OD) value from ELISA analysis of log serum dilution after single immunization (a) 1, (b) 1-Tr, (c) 1+CTB, (d) PBS.



Figure S6. Female Large White pigs were used for the injection and oral vaccine study (left panel). Oral vaccine was delivered by gavage using a Shoof Luer-Lock (206 285) drench nozzle (right panel).