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**Efficacy Evaluation of a Twelve PEG-Peptide Mixture and a Twelve Peptide Mixture
Against Established Mammary 25 Murine Mammary Carcinoma**



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Executive Digest

A twelve PEG-peptide mixture was administered intravenously either once daily for 14-17 consecutive days, every third day for six injections, or every second day for nine injections to mice bearing established Mammary 25 murine mammary carcinomas. Treatment was well tolerated but inactive regardless of dosage level or schedule.

A twelve peptide mixture (non-pegylated) was administered intravenously once daily for 14 consecutive days to mice bearing established Mammary 25 murine mammary carcinomas as a comparator. Treatment was well tolerated but inactive.

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Contents

<u>Section</u>	<u>Page</u>
A. Introduction	04
B. Materials and Methods	04
C. Results/Discussion	07
D. Glossary and Endpoint Details	08
E. References	11
F. Tables	
1. Group Toxicity and Response Summary	12
2. Individual Animal Toxicity and Response Summary	14
3. Group Statistics	18
G. Figures	
1. Group Comparisons with Std. Error by Mean	20
2. Median Group Comparisons	22
3. Individual Tumor Growth Curves	24
4. Body Weight Change Summary with Std. Error	27
H. Appendices	
1. Protocol Summary	29
2. Raw Data – Tumor Measurements and Body Weights	31
3. Raw Data – Daily Census	35
4. Raw Data – Clinical Signs, Observations, and Comments	37
5. Tumor Burdens	39
6. Consolidated Body Weights	43
7. Consolidated Percent Body Weight Change	47
8. T/C Values	51
9. Fold Growth Values	53
10. Peptide Prep Procedure	57

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Introduction

The purpose of this study was to evaluate the efficacy of a mixture of twelve pegylated-peptides against established Mammary 25 murine mammary carcinoma. The peptide mixture was administered intravenously either on a daily, every other day, or every third day schedule. A mixture of the same non-pegylated peptides served as a comparator.

Materials and Methods

Chemicals

Twelve PEG-peptides (labeled in consecutive order from G1-G12) were obtained from Sanare as white powders. The intent was to administer an amount of each peptide in the mix such that the final aggregate concentration in the mouse blood volume (~2ml) was 0.8mg/ml after each dose. The peptide mix was prepared by first adding 10ml of phosphate buffered saline (PBS 1X) to each vial of powder to achieve a concentration of 2mg/ml for each peptide. A further dilution was made to account for each peptide's molecular weight to provide equimolar concentrations of each peptide at 0.35 μ M. Equal amounts of each of the twelve pegylated peptides were then combined to form the top dose solution (Appendix 10). The top dose solution was aliquoted out for each dosing day and frozen at -20°C until use. On each day of treatment, one frozen vial of top dose solution was removed from the freezer and thawed at room temperature. The dosing solution was clear and colorless with a pH value of 7. Lower dosages were prepared by direct dilution of the top dose with the appropriate amount of PBS.

Twelve peptides (labeled in consecutive order from 25-36) were obtained from Sanare as white powders. The intent was to administer an amount of each peptide in the mix such that the final aggregate concentration in the mouse blood volume (~2ml) was 0.8mg/ml after each dose. The peptide mix was prepared by first adding 5ml of phosphate buffered saline (PBS) to each vial of powder to achieve a concentration of 2mg/ml for each peptide. A further dilution was made to account for each of the peptide's molecular weight to provide equimolar concentrations of each peptide at 0.35 μ M. Equal amounts of each of the twelve peptides were then combined to form the top dose solution (Appendix 10). The top dose solution was aliquoted out for each dosing day and frozen at -80°C until use. On each day of treatment, one frozen vial of top dose solution was removed from the freezer and thawed at room temperature. The dosing solution was clear and colorless with a pH value of 8. *NOTE: The original non-pegylated prep was performed for study SANA200802R1 (MIR948) and equal parts of the remaining equal molar stock solutions were mixed resulting in the dosing solutions for this study.*

Animals and Husbandry

Female BALB/c mice were obtained from Charles River Labs. They were 6-7 weeks old on Day 1 of the experiment. The animals were fed irradiated Rodent Diet 5053 (LabDiet™) and water *ad libitum*. Mice were housed in static cages with Bed-O'Cobs™ bedding inside Biobubble® Clean Rooms that provide H.E.P.A filtered air into the bubble environment at 100 complete air changes per hour. All treatments, body weight determinations, and tumor measurements were carried out in the bubble environment. The environment was controlled to a temperature range of 70° \pm 2°F and a humidity range of 30-70%.

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Test animals were implanted subcutaneously on Day 0 with 30 to 60mg tumor fragments using an 11-gauge trocar needle. All animals were observed for clinical signs at least once daily. Animals with tumors in excess of 1g or with ulcerated tumors were euthanized, as were those found in obvious distress or in a moribund condition. All procedures carried out in this experiment were conducted in compliance with all the laws, regulations and guidelines of the National Institutes of Health (NIH) and with the approval of MIR's Animal Care and Use Committee.

Treatment

Treatments began on Day 9, when the mean estimated tumor mass across all groups in the experiment was 97mg (range of group means, 92-103mg). All animals weighed ≥ 15.9 g at the initiation of therapy. Mean group body weights at first treatment were well-matched (range of group means, 17-18g). All animals were dosed according to individual body weight on the day of treatment (0.2ml/20g) as indicated in the protocol (Appendix 1).

Measurements and Endpoints

Testing in this experiment was generally carried out adhering to the general principles established by the groups of Schabel, Skipper, Griswold, Corbett, Leopold, Ross and the NCI (1-7). Body weights and tumor measurements were recorded twice weekly. Tumor burden (mg) was estimated from caliper measurements by the formula for the volume of a prolate ellipsoid assuming unit density as: Tumor burden (mg) = $(L \times W^2)/2$, where L and W are the respective orthogonal tumor length and width measurements (mm).

The primary endpoints used to evaluate efficacy were: tumor growth delay, complete and partial tumor response, and the number of tumor-free survivors at the end of the study. A complete response (CR) is defined as a decrease in tumor mass to an undetectable size (<50 mg), and a partial response (PR) is defined as a $\geq 50\%$ decrease in tumor mass from that at first treatment. PRs are exclusive of CRs, as are Tumor-Free Survivors (TFS). Tumor Growth Delay (T-C) was also used to quantify efficacy. Tumor growth delay for this experiment was expressed as a T-C value, where T and C are the median times in days required for the treatment and control group tumors, respectively, to grow to a selected evaluation size, 750mg.

Net Log₁₀ Tumor Cell Kill (Net Kill) is used as a secondary efficacy endpoint. Net Kill is the change in tumor burden (logs) over the treatment period. Traditionally Net Kill is estimated by the method of Schabel et al (1,3) but this method requires that initial and post treatment regrowth rates be equal, a condition that may not be met by certain differentiation therapies or those that induce significant tumor bed effects. We instead use a modification of the method of Ross et al (7), that can accommodate any regrowth rate and which simply assumes that the tumor regrowth rate between dosage levels are similar to that observed post treatment. This method simply extrapolates (or interpolates) the regrowth curve back to the last day of treatment for the calculation of Net Kill. The two methods converge to the same result if initial and regrowth rates are similar. The Net Kill value allows quantitative comparison of efficacy across multiple experimental protocols and across models by normalizing the efficacy data for treatment regimens of varied duration and differences in tumor growth rates between experiments or models. Positive values indicate that an actual reduction of tumor burden had occurred at the end of therapy relative to the pretreatment burden. Negative values indicate the tumor grew (although possibly more slowly than the control tumors) during treatment. Thus negative Net Kill values do not necessarily imply a complete lack of

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activity. Tumor-free survivors and tumor-bearing survivors whose tumors had not reached the Evaluation Size by the last day of the experiment were assigned a Time to Evaluation Size of > the last day of the experiment and were included in calculations of Tumor Growth Delay and Net Kill.

The calculation of Net Kill by either method requires the establishment of the tumor doubling time (Td), which is the time in days for the tumor burden to double. Doubling time (Td) was estimated from the least squares best-fit straight line from a log-linear plot of tumor burden vs. time over the period of exponential growth (~200 to ~800mg range). This parameter was calculated and tabulated in Table 2 for every animal in the experiment.

When control group and treated group doubling times are not similar, the relative contribution of direct tumor cell killing and hindered tumor regrowth to the overall therapeutic effect are estimated by a modification of the method of Ross et al. (7)

Assessment of Side Effects

All animals were observed for clinical signs at least once daily. Animals were weighed on each day of treatment and at least twice weekly thereafter. Individual body weights were recorded twice weekly.

Treatment-related weight loss in excess of 20% is generally considered unacceptably toxic. In this report, a dosage level is described as tolerated if treatment-related weight loss (during and two weeks after treatment) is <20% and mortality during this period in the absence of potentially lethal tumor burdens is ≤10%.

Upon death or euthanasia, all animals were necropsied to provide a general assessment of potential cause of death and perhaps target organs for toxicity. The presence or absence of metastases was also noted. Remarkable observations of clinical signs and necropsy findings have been tabulated in Appendix 4. Individual and group toxicity findings have been summarized in Tables 1-3 and Figure 3.

Statistics

The median times to evaluation size for all study groups were first analyzed by application of the Kruskal-Wallis rank sum test analysis to determine if any significant differences existed between all study groups. Upon identification that significant differences exist, post-hoc multiple comparisons to a specified control group were conducted using an additional Kruskal-Wallis test followed by the multiple comparison procedure of Dunn.

Statistical significance was determined using built-in Microsoft Excel data analysis tools, SigmaStat 3.0, and the R-Project (8).

Data Retrieval

MIR Preclinical Services retains permanent “active” copies (on CD) of all experiments unless instructed otherwise, including active graphics applications.

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Results/Discussion

Tumor Growth/General Observations/Controls

The mean estimated tumor burden across all groups in the experiment on the first day of treatment was 97mg and all of the groups in the experiment were well-matched (range, 92-103mg). All animals weighed ≥ 15.9 g at the initiation of therapy. Mean group body weights at first treatment were also well-matched (range, 17-18g). A tumor burden of 750mg was chosen for evaluation of efficacy by tumor growth delay. The Median Control Tumor reached evaluation size on Day 18, and the tumor volume doubling time for the Control Group was 2.7 days (Table 2). Control animals experienced a 1.5g mean weight gain during the treatment regimen. There were no spontaneous regressions in the Control Group. Thioglycolate cultures of all 5 donor tumors used for implantation of this study were negative for gross bacterial contamination. Based on historical data for this model, the biology of the Control Group was judged to be within the normal range.

Toxicity

Treatment with the PEG-peptide mixture was well tolerated at all tested dosage levels (80, 40, and 20mg/kg) and schedules, producing no treatment-related mortality or treatment-related weight loss (Table 1 and 2, Figure 3). One animal receiving the 80mg/kg PEG-peptide mixture for a single dose (Group 5) was removed from study due to tumor ulceration. Upon necropsy all animals had enlarged spleens, including control animals, indicative of tumor progression (Appendix 4).

Treatment with the peptide mix (non-PEG) at 80mg/kg dosed intravenously daily for fourteen consecutive days was well tolerated resulting in no treatment-related mortality or weight loss (Tables 1 and 2, Figure 3). Upon necropsy all animals had enlarged spleens, including control animals, indicative of tumor progression (Appendix 4).

Efficacy

Intravenous treatment with the twelve PEG-peptide mixture was ineffective at all dosage levels (80, 40, and 20mg/kg) and schedules (QD, Q3D, Q2D), producing insignificant and minimal tumor growth delays ranging from 2.1 to 0.0 days and net tumor cell kill values ranging from 0.0 to -1.80 logs, which indicate tumor progression during treatment. Treatment produced no tumor regressions or tumor-free survivors (Tables 1, 2 and 3).

Daily intravenous treatment with the twelve peptide mixture (non-PEG) at 80mg/kg was also ineffective, producing an insignificant tumor growth delay of 1.2 days and a negative net tumor cell kill value of -1.27 logs. Treatment produced no tumor regressions or tumor-free survivors (Tables 1, 2 and 3).

Based on the total lack of treatment-related toxicity seen in this study, it is possible that higher dosage levels could produce meaningful anti-cancer activity at tolerated dosage levels. However, the relatively low solubility of the peptides in aqueous media may preclude the use of higher dose concentrations.

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Glossary

Apparent Net Tumor Cell Kill – The net change in tumor burden (measured in logs) between the first and last treatments. An equation is derived for the tumor growth of each mouse in the experiment from a least squares best fit of the exponential portion of the tumor growth curve post treatment. This equation is solved to give the apparent tumor burden surviving the last day of treatment for each animal. Median values are used for comparison of treatment groups to the controls to derive the net reduction in tumor burden during treatment (Net log₁₀ Kill, or NK_{apparent}). This modification of the methods of Schabel et al. (1, 3) and Ross et al (7) allows an estimate of reduction in tumor burden even in the circumstance of tumor regrowth rates that are significantly different from the control growth rates. In general the method of Schabel overestimates net reduction in tumor burden (Net Kill) values. NK_{apparent} values faithfully estimate the tumor burden at last treatment, but they don't really estimate how much of the effect was due to cell killing and how much was due to hindered regrowth during treatment if the regrowth rates are dissimilar to initial or control growth rates. The term Net Kill is actually a misnomer for the combined effects of cell killing and hindered tumor growth between treatments. With the long treatment regimens in common use today, the relative contribution of hindered regrowth to the NK_{apparent} can be large. However the relative contributions of cell killing and hindered regrowth can be estimated. These questions can be important to those designing cytotoxic therapies or those dealing with compounds of mixed mechanisms. For this reason we also report fractional effect values that estimate the relative contributions of cell killing and slowed regrowth to the net change in tumor burden during therapy (NK_{apparent}). As always NK_{apparent} values normalize the data for differences in duration of treatment and also tumor doubling times across tumor models and experiments, allowing quantitative comparison between experiments. Two underlying assumptions are required for our calculations: (1) all treatment fractions produce equivalent cell killing, and (2) the regrowth rate between fractions is identical to the post treatment regrowth rate. These assumptions are also required for the traditional Schabel calculations, but those carry the additional assumption that the regrowth and initial growth rates are equal. (*Group efficacy parameter*)

Complete Regression (CR) – An animal is credited a complete regression if the tumor burden is reduced to an immeasurable volume at any point after the first treatment. Our convention is to record any tumor measurement less than 3 mm as a "0". This is in keeping with the convention of the NCI and reflects the inherent and unacceptably high mechanical error in such measurements in addition to the uncertain biology of what is measured at those small sizes (4). (*Individual efficacy parameter*)

Day 0 – The day tumors are implanted into the animals (Not to be confused with the first day of treatment which is always indicated relative to Day 0).

Day of Max. Body Wt. Loss – The day of body weight nadir (if any) between the first day of treatment and 2 weeks after the last day of treatment. Reported as "NA" if no weight loss occurred. (*Group toxicity parameter*)

Evaluation Size – The tumor burden (mg) selected for calculation of tumor growth delay. The Evaluation Size is selected from the exponential portion of the control tumor growth curve where the error of measurement tends to be minimal (usually between 500 and 1000mg).

Fractional Effect - When initial and post-treatment regrowth rates are different it is possible to estimate the relative contributions of hindered regrowth rate and tumor cell killing to the observed net reduction in tumor burden (NK_{apparent}). This is accomplished by first calculating the Gross Kill for the group (Schabel (1)) based on the median value calculated from best fit equations for the actual regrowth data for each mouse in the experiment. The Gross Kill is then used to calculate a theoretical Net Kill value (NK_{corrected}) that would have been obtained if the tumors had re-grown at their original (pretreatment) growth rates. The ratio of the NK_{corrected} and NK_{apparent} values provides an estimate of the percentage of the therapeutic effect that was due to actual reductions in cell number (Tumor cell kill (%)). The rest of the effect (Hindered Regrowth (%)) is due to hindered post treatment regrowth rates during the treatment regimen. In studies with long durations of treatment, the relative contribution of hindered regrowth to NK_{apparent}

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can be large, even for small differences between growth and regrowth rates. (*Group efficacy parameter*)

Maximum Rx-Related Body Wt. Loss – This is meant to reflect treatment-related weight change. Calculated as the maximum weight loss occurring anytime between the first day of treatment and two weeks from the last day of treatment, expressed as a percentage of the weight on the first day of treatment. (*Group toxicity parameter*)

Partial Regression (PR) – An animal is credited with a partial regression if its tumor burden decreases to less than half of the tumor burden at first treatment. PRs are tabulated exclusive of CRs. (*Individual efficacy parameter*)

Percent Body Wt. Change – Change in body weight between first and last days of treatment expressed as a percentage of the body weight on the first day of treatment. If actual body weight measurements were not recorded on precisely the first and last days of treatment, these values are calculated by interpolation between the closest measurements on either side of the target date. (*Individual toxicity parameter*)

% Rx Related Deaths – The percentage of animals presumed to have succumbed to treatment-related toxicity expressed as a percentage of the number of evaluable animals in the group. An animal is presumed to experience a treatment-related death if it is found dead or is euthanized in moribund condition within 2 weeks of the last treatment with a tumor burden less than half that of the smallest lethal tumor in the control group and shows no evidence of infection, mechanical dosing trauma, or other obvious causes of morbidity at necropsy. Animals dying from non-treatment-related causes prior to reaching an evaluable efficacy endpoint are excluded from this evaluation and the number of animals in the group is reduced accordingly. (*Group toxicity parameter*)

% Tumor Free Survivors (TFS) – Any animal with no measurable evidence of disease on the last day of the experiment. This value is exclusive of CRs. These animals may or may not represent “cures”, depending on when the experiment was terminated. We recommend that no animal be assumed cured unless the animal has been held for a period of time past the last treatment equal to twice the time required to allow a single surviving cell to grow to a tumor burden of 500mg. That time in days can be calculated as $(57.6 \times Td)$.

Recovery Time – The length of time in days for treatment-related weight loss to be recovered. Measured as the time from the nadir of body weight to return to the pretreatment value. This parameter is an important monitor of lingering or delayed toxicity. (*Group toxicity parameter*)

Rx Related Death – An animal is presumed to experience a treatment-related death if it is found dead or is euthanized in moribund condition within 2 weeks of the last treatment with a tumor burden less than half that of the smallest lethal tumor in the control group and shows no evidence of infection, mechanical dosing trauma, or other obvious causes of morbidity at necropsy. (*Individual toxicity parameter*)

Td (Tumor Doubling Time) – The growth rate of the tumor expressed as the volume doubling time (days). Calculated from a log-linear least squares regression of the exponential portion of the tumor growth curve. These values are used to compute tumor cell kill, fractional effect, and surviving fraction estimates. They are also used to assess the appropriateness of the biology of the tumor in this experiment against historical values.

Therapeutic Index – We define therapeutic index as simply the range of tolerated dosage levels that produce substantial anticancer activity. Substantial activity for this purpose is defined as a tumor growth delay that is \geq the duration of treatment and that is also statistically different from the control at the $P \leq 0.05$ level.

Time to Evaluation Size – The time (days) it takes a tumor to reach the specified Evaluation Size. Calculated from a log-linear least squares best fit of tumor burden versus time for the exponential portion of the final (post-treatment) tumor growth curve. This value is calculated for every animal in the experiment. The group medians are then used to calculate the Tumor Growth Delay. (*Individual efficacy parameter*)

Time to Fold Growth End Point – Occasionally (usually when initial mean tumor burdens across all groups are not well-matched) it is advantageous to display efficacy parameters in terms of fold growth, where the selected endpoint is the time it takes to reach a selected multiple of

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initial tumor burden. This increases the probability of uncovering a statistically significant therapeutic effect by eliminating the confounding effects of disparity in initial tumor sizes. Calculated as described for Time to Evaluation Size from the fold growth data (Appendix 9). (*Individual efficacy parameter*)

Tumor Burden at Last Rx – The tumor burden on the last day of treatment. This value is calculated from a log-linear least squares best fit of tumor burden versus time for the exponential portion of the final (post-treatment) tumor growth curve. (Presented to facilitate T/C comparisons. Additional T/C information is presented in Appendix 8. T/C values presented in Appendix 8 are simple ratios, not the NCI convention)

Tumor Growth Delay – Tumor Growth Delay (T-C) is the difference between the median times it takes the treated group and the control group (always the first group in Table 2) to reach the stated evaluation size. This is calculated from the median times to evaluation size for each animal in the group, not from interpolation of the median growth curve. Net kill values are not presented when the T-C is negative. (*Group efficacy parameter*)

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References

- 1) Schabel F, Griswold D, Laster W, Corbett T, Lloyd H. Quantitative evaluation of anticancer agent activity in experimental animals. *Pharmac. Ther. A.* (1) 411-435, 1977.
- 2) Corbett, T, Griswold D, Roberts B, Peckham J, Schabel F Evaluation of single agents and combinations of chemotherapeutic agents in mouse colon carcinomas. *Cancer* 1977; 40(5); 2660-2690.
- 3) Schabel F, Griswold D, Corbett T, Laster R, Mayo J, Lloyd H. Testing therapeutic hypotheses in mice and man: Observations on the therapeutic activity against advanced solid tumors of mice treated with anticancer drugs that have demonstrated or potential clinical utility for treatment of advanced solid tumors of man. *Methods in Cancer Research* (17) 3-51, 1979.
- 4) Plowman J, Dykes D, Hollingshead M, Simpson-Herren L, and Alley M. Human tumor xenograft models in NCI drug development. In: *Anticancer drug development guide: preclinical screening, clinical trials, and approval*. Teicher (ed) Humana Press Inc. 1993.
- 5) Corbett T, Valeriote F, LoRusso P, Polin L, Panchapor C, Pugh S, White K, Knight J, Demchik L, Jones J, Jones L, Lowichik N, Biernat L, Foster B, Wozniak A, Lisow L, Valdivieso M, Baker L, Leopold W, Sebolt J, Bissery M, Mattes K, Dzubow J, Rake J, Perni R, Wentland M, Coughlin S, Shaw JM, Liversidge G, Liversidge E, Bruno J, Sarpotdar P, Moore R, Patterson G. Tumor models and the discovery and secondary evaluation of solid tumor active agents. *Int J Pharmacognosy* 1995; 33(supplement): 102-122.
- 6) Corbett T, Roberts BJ, Lawson AJ, Leopold WR, et al. Transplantable Syngeneic Rodent Tumors: Solid Tumors of Mice. In: *Tumor Models in Cancer Research* (BA Teicher ed). Humana Press, Totowa, NJ. pp. 41-71, 2002.
- 7) Ross B, Zhao Y, Neal E, Stegman L, Ercolani M, Ben-Yoseph O, Chenevert T. Contributions of cell kill and post-treatment tumor growth rates to the repopulation of intracerebral 9L tumors after chemotherapy: An MRI study. *PNAS* (95) 7012-7017, 1998.
- 8) R Development Core Team. *R: A Language and Environment for Statistical Computing*. 2005, R Foundation for Statistical Computing: Vienna, Austria. R, D.C.T., *R: A Language and Environment for Statistical Computing*. 2005, R Foundation for Statistical Computing: Vienna, Austria. <http://www.R-project.org>.

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Table 1 – Group Toxicity and Response Summary

Group Summary

Group #	Treatment	Dose (mg/kg/inj)	Schedule	Route	Toxicity				Efficacy				
					Maximum Treatment-related Weight Loss (%)	Day of Max. Treatment-related Weight Loss	Recovery Time (days)	% Rx Related Deaths	Tumor Growth Delay (days)	Apparent Net Tumor Cell Kill (logs)	% CR	% PR	% TFS
1	Vehicle Treated Control	0.2ml/20g	QDx14**	IV	GAIN	NA	NA	0	NA	NA	0	0	0
2	Peg-Peptide Mixture	80.00	QDx17**	IV	GAIN	NA	NA	0	1.8	-1.59	0	0	0
3	Peg-Peptide Mixture	40.00	QDx17**	IV	GAIN	NA	NA	0	1.4	-1.50	0	0	0
4	Peg-Peptide Mixture	20.00	QDx14**	IV	GAIN	NA	NA	0	2.1	-1.15	0	0	0
5	Peg-Peptide Mixture	80.00	Single Dose	IV	GAIN	NA	NA	0	2.1	0.00	0	0	0
6	Peg-Peptide Mixture	80.00	Q3Dx6^	IV	GAIN	NA	NA	0	0.5	-1.59	0	0	0
7	Peg-Peptide Mixture	80.00	Q2Dx9*	IV	GAIN	NA	NA	0	0.0	-1.80	0	0	0
8	Peptide Mixture	80.00	QDx14**	IV	GAIN	NA	NA	0	1.2	-1.27	0	0	0

*= Original protocol quoted for 10 injections. Schedule reflects actual number of injections given due to removal of animals for excess in tumor burden.

**= Original protocol quoted for 21 injections. Schedule reflects actual number of injections given due to removal of animals for excess in tumor burden.

= Original protocol quoted for 7 injections. Schedule reflects actual number of injections given due to removal of animals for excess in tumor burden.

GROUP SUMMARY ENDPOINT DEFINITIONS AND CALCULATION METHODS

Maximum Mean BW Loss (%)	Calculated from the minimum of the mean BW curve (while there are greater than half the animals still surviving) for each group within the Rx period and out to 2 weeks after the end of Rx.
Day of Maximum Mean BW Loss	Calculated from the minimum of the mean BW curve (while there are greater than half the animals still surviving) for each group within the Rx period and out to 2 weeks after the end of Rx.
Recovery Time (days)	The number of days from the time of the minimum mean BW to recover the lost BW. Calculated only for animals which lost BW and later recovered the lost BW.
% Rx Related Deaths	% of mice in each group with treatment-related deaths.
Tumor Growth Delay (days)	The median time to evaluation size is calculated for each group (see Table 1). The tumor growth delay (TGD) is calculated by subtracting the median TGD for the control group from the median TGD for each treatment group.
Apparent Net Tumor Cell Kill (logs)	By interpolation, the log-linear regression line for each animal is used to calculate the tumor weight (TW) at the start of treatment, and the TW at the end of treatment, for control and treatment groups, respectively. For each treatment group, the median TW (end of treatment) is subtracted from the median control TW (start of treatment) to calculate the apparent net cell kill (in log units).
% CR	% of mice in each group that experienced complete regressions.
% PR	% of mice in each group that experienced partial regressions.
% TFS	% of mice in each group that were tumor free survivors.

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Table 2 – Individual Animal Toxicity and Response Summary

Calculated End Points

Evaluation Size		750 mg	Last Day of Expt.		26						
Fold Growth End Point		4									
Group # 1		Treatment Vehicle Treated Control			Growth Endpoints						
Dose 0.2ml/20g											
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	NA	BW GAIN	no	2.8	NA	17.3	15.7	no	no	no	
2	NA	BW GAIN	no	3.1	NA	16.9	14.4	no	no	no	
3	5.1	0.6	no	2.0	1223	20.4	17.7	no	no	no	
4	NA	BW GAIN	no	2.9	NA	18.6	14.7	no	no	no	
5	NA	BW GAIN	no	2.5	NA	17.3	15.3	no	no	no	
6	14.4	BW GAIN	no	2.1	1129	19.9	17.2	no	no	no	
7	NA	BW GAIN	no	2.7	NA	17.2	15.1	no	no	no	
8	8.5	3.8	no	3.7	940	21.1	17.0	no	no	no	
Mean			Total	2.7	NA	18.6	15.9	Total	Total	Total	
SD			Rx Deaths:	0.6	NA	1.6	1.2	CR:	PR:	TFS:	
Median			0	2.7	NA	18.0	15.5	0	0	0	
Group # 2		Treatment Peg-Peptide Mixture			Growth Endpoints						
Dose 80.00											
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	16.9	BW GAIN	no	3.1	1375	21.9	17.9	no	no	no	
2	NA	BW GAIN	no	2.9	NA	17.4	15.1	no	no	no	
3	NA	BW GAIN	no	2.6	NA	19.8	16.9	no	no	no	
4	NA	BW GAIN	no	1.9	NA	19.9	18.3	no	no	no	
5	13.1	BW GAIN	no	2.8	1661	22.1	18.4	no	no	no	
6	NA	BW GAIN	no	3.2	NA	19.7	17.1	no	no	no	
7	NA	BW GAIN	no	2.1	NA	18.4	15.6	no	no	no	
8	NA	BW GAIN	no	3.6	NA	17.6	16.2	no	no	no	
Mean			Total	2.8	NA	19.6	16.9	Total	Total	Total	
SD			Rx Deaths:	0.6	NA	1.8	1.2	CR:	PR:	TFS:	
Median			0	2.9	NA	19.8	17.0	0	0	0	
Group # 3		Treatment Peg-Peptide Mixture			Growth Endpoints						
Dose 40.00											
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	NA	BW GAIN	no	3.5	NA	19.5	17.5	no	no	no	
2	NA	BW GAIN	no	3.3	NA	19.8	15.5	no	no	no	
3	NA	3.3	no	3.5	NA	19.1	16.4	no	no	no	
4	NA	BW GAIN	no	3.3	NA	18.4	15.8	no	no	no	
5	NA	BW GAIN	no	2.7	NA	17.8	14.2	no	no	no	
6	NA	BW GAIN	no	3.0	NA	17.8	15.4	no	no	no	
7	12.6	BW GAIN	no	3.8	1082	22.6	16.7	no	no	no	
8	NA	BW GAIN	no	3.1	NA	19.6	15.6	no	no	no	
Mean			Total	3.3	NA	19.3	15.9	Total	Total	Total	
SD			Rx Deaths:	0.3	NA	1.6	1.0	CR:	PR:	TFS:	
Median			0	3.3	NA	19.3	15.7	0	0	0	
Group # 4		Treatment Peg-Peptide Mixture			Growth Endpoints						
Dose 20.00											
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	NA	BW GAIN	no	2.4	NA	16.2	13.6	no	no	no	
2	10.1	BW GAIN	no	2.8	1596	19.8	16.7	no	no	no	
3	NA	BW GAIN	no	3.6	NA	18.4	16.3	no	no	no	
4	0.8	BW GAIN	no	3.7	1120	20.1	17.1	no	no	no	
5	8.2	BW GAIN	no	3.8	1090	20.6	18.5	no	no	no	
6	8.4	BW GAIN	no	4.0	951	21.2	18.0	no	no	no	
7	5.5	BW GAIN	no	3.2	1366	19.9	17.4	no	no	no	
8	7.6	BW GAIN	no	3.1	1064	20.5	16.5	no	no	no	
Mean			Total	3.3	1198	19.6	16.8	Total	Total	Total	
SD			Rx Deaths:	0.6	238	1.6	1.5	CR:	PR:	TFS:	
Median			0	3.4	1105	20.0	16.9	0	0	0	

Group #		5									
Treatment		Peg-Peptide Mixture									
Dose		80.00									
		Growth Endpoints									
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	0.0	BW GAIN	no	3.4	108	20.5	17.8	no	no	no	
2	0.0	BW GAIN	no	2.6	108	17.1	15.0	no	no	no	
3	0.0	BW GAIN	no	2.7	108	18.0	15.9	no	no	no	
4	0.0	BW GAIN	no	3.1	100	18.9	16.1	no	no	no	
5	0.0	BW GAIN	no	3.7	88	20.0	16.0	no	no	no	
6	0.0	BW GAIN	no	3.8	108	21.1	18.1	no	no	no	
7	0.0	BW GAIN	no	3.3	75	NA	16.9	no	no	no	
8	0.0	BW GAIN	no	3.4	108	20.7	18.0	no	no	no	
Mean			Total	3.3	100	19.5	16.7	Total	Total	Total	
SD			Rx Deaths:	0.4	12	1.5	1.1	CR:	PR:	TFS:	
Median			0	3.4	108	20.0	16.5	0	0	0	
Group #		6									
Treatment		Peg-Peptide Mixture									
Dose		80.00									
		Growth Endpoints									
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	NA	BW GAIN	no	3.6	NA	17.8	16.5	no	no	no	
2	NA	BW GAIN	no	3.0	NA	20.3	15.6	no	no	no	
3	NA	BW GAIN	no	2.7	NA	15.4	14.4	no	no	no	
4	NA	BW GAIN	no	3.0	NA	21.2	17.2	no	no	no	
5	11.1	BW GAIN	no	3.7	1400	21.1	16.2	no	no	no	
6	NA	BW GAIN	no	2.0	NA	16.2	13.6	no	no	no	
7	NA	BW GAIN	no	2.9	NA	18.4	15.2	no	no	no	
8	NA	6.6	no	3.2	NA	18.5	15.9	no	no	no	
Mean			Total	3.0	NA	18.6	15.6	Total	Total	Total	
SD			Rx Deaths:	0.5	NA	2.2	1.2	CR:	PR:	TFS:	
Median			0	3.0	NA	18.4	15.7	0	0	0	
Group #		7									
Treatment		Peg-Peptide Mixture									
Dose		80.00									
		Growth Endpoints									
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	NA	BW GAIN	no	3.2	NA	19.4	16.9	no	no	no	
2	NA	BW GAIN	no	2.4	NA	17.3	14.6	no	no	no	
3	NA	BW GAIN	no	2.2	NA	18.3	14.8	no	no	no	
4	14.2	BW GAIN	no	4.7	1182	22.6	19.9	no	no	no	
5	NA	BW GAIN	no	2.8	NA	17.7	16.1	no	no	no	
6	NA	BW GAIN	no	2.8	NA	19.2	15.6	no	no	no	
7	NA	BW GAIN	no	2.6	NA	17.6	14.8	no	no	no	
8	NA	BW GAIN	no	2.7	NA	16.7	14.5	no	no	no	
Mean			Total	2.9	NA	18.6	15.9	Total	Total	Total	
SD			Rx Deaths:	0.8	NA	1.9	1.8	CR:	PR:	TFS:	
Median			0	2.7	NA	18.0	15.2	0	0	0	

Group #		8									
Treatment		Peptide Mixture									
Dose		80.00									
				Growth Endpoints							
Animal	BW Change During Rx (%)	Max. BW Loss (%)	Rx-related Death	Td (days)	Tumor Burden @ Last Rx	Time to Evaluation Size (days)	Time to Fold Growth EP (days)	Complete Regression	Partial Regression	Tumor Free Survivor	
1	NA	BW GAIN	no	2.9	NA	17.9	14.7	no	no	no	
2	10.7	BW GAIN	no	3.4	1094	19.9	15.4	no	no	no	
3	11.5	BW GAIN	no	4.3	923	21.6	19.1	no	no	no	
4	NA	BW GAIN	no	3.2	NA	20.1	17.5	no	no	no	
5	10.4	BW GAIN	no	3.2	1094	20.3	16.0	no	no	no	
6	NA	BW GAIN	no	2.9	NA	18.4	15.2	no	no	no	
7	NA	BW GAIN	no	2.8	NA	17.4	15.1	no	no	no	
8	NA	BW GAIN	no	2.8	NA	17.8	15.3	no	no	no	
Mean			Total	3.2	NA	19.2	16.0	Total	Total	Total	
SD			Rx Deaths:	0.5	NA	1.5	1.5	CR:	PR:	TFS:	
Median			0	3.1	NA	19.1	15.4	0	0	0	

ENDPOINT DEFINITIONS AND CALCULATION METHODS

BW Change During Rx (%)	Body weight change during treatment = (End Rx BW - Start Rx BW)
Max. BW Loss (%)	Maximum body weight loss during treatment, and out to 2 weeks following the end of treatment.
Rx-related Death	Treatment-related death, as determined by clinical signs observations (see Comments table), body weight measurements and behavior of other mice within the group.
Td (days)	Tumor doubling time calculated from log-linear regressions over growth data.
Tumor Burden @ Last Rx	Tumor size on day of last treatment as estimated by caliper measurement.
Time to Evaluation Size (days)	Calculated by interpolation of the log-linear regression line on the tumor growth data, to the evaluation size.
Time to Fold Growth EP (days)	Calculated by interpolation of the log-linear regression line on the fold growth data, to the evaluation size.
Complete Regression	Animals for which the tumor size decreased below 50mg.
Partial Regression	Animals for which the tumor size decreased to < 50% of the initial size.
Tumor Free Survivor	Animals for which a complete regression persisted to the end of the study.

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Table 3 – Group Statistics

Kruskal-Wallis One Way Analysis of Variance on Ranks

Data source: Time to 750mg Data MIR1052 Stats

Dependent Variable: Time to 750mg

Normality Test (Shapiro-Wilk) Passed (P = 0.646)

Equal Variance Test: Passed (P = 0.969)

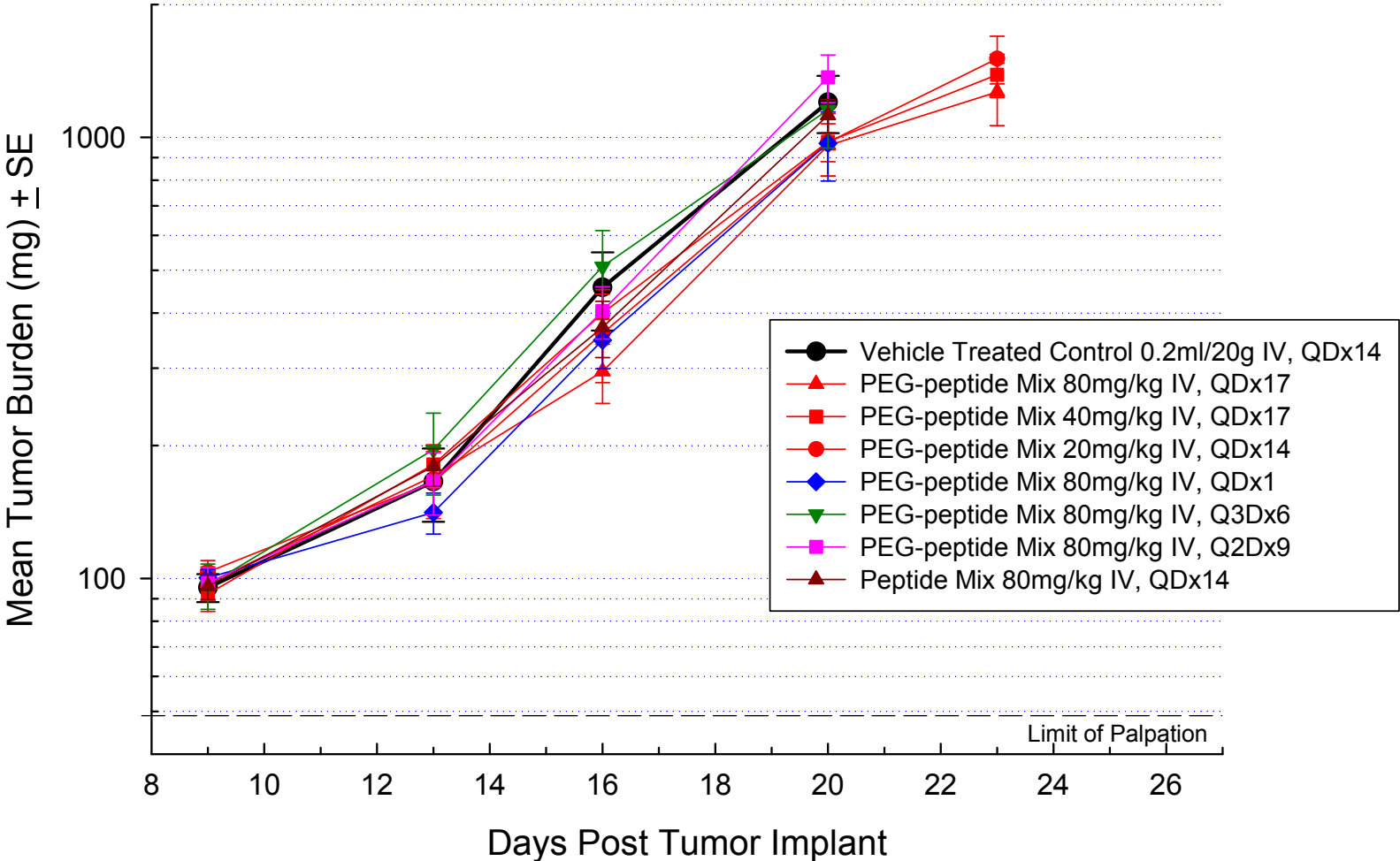
Group	N	Missing	Median	25%	75%
1.000	8	0	17.951	17.236	20.254
2.000	8	0	19.771	17.798	21.419
3.000	8	0	19.338	17.954	19.780
4.000	8	0	20.031	18.702	20.609
5.000	8	1	20.037	18.009	20.692
6.000	8	0	18.423	16.618	20.925
7.000	8	0	17.986	17.371	19.388
8.000	8	0	19.137	17.832	20.236

H = 4.752 with 7 degrees of freedom. (P = 0.690)

The differences in the median values among the treatment groups are not great enough to exclude the possibility that the difference is due to random sampling variability; there is not a statistically significant difference (P = 0.690)

Figure 1 – Group Comparisons with Std. Error by Mean

SANA200810 (MIR1052)
 Mam25 Tumor Burden
 Group Comparison with Std. Error



.....Figure &- ; fci d`7 ca dUf]gcb`VmA YX]Ub`

SANA200810 (MIR1052)
Mam25 Tumor Burden
Group Median Comparison

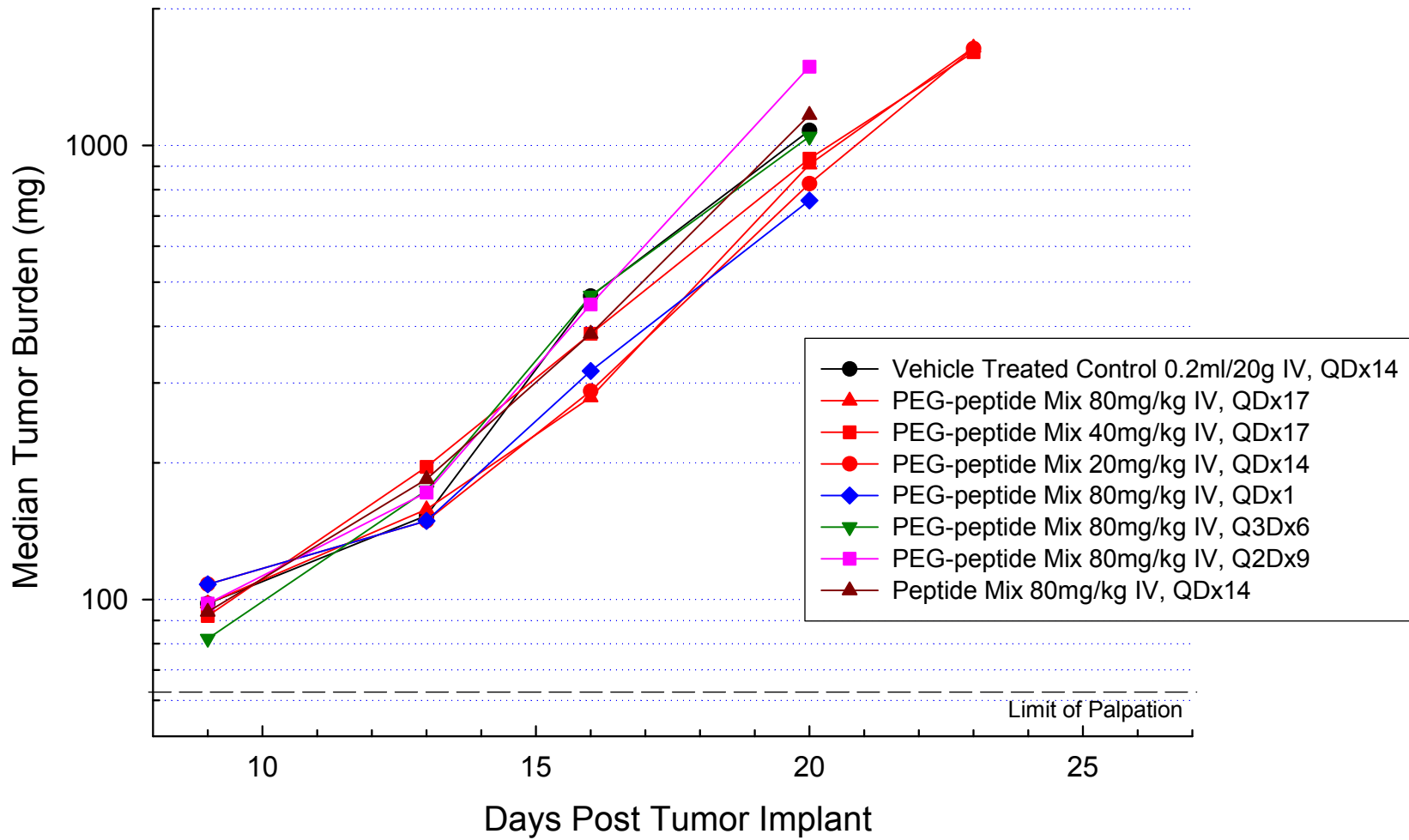
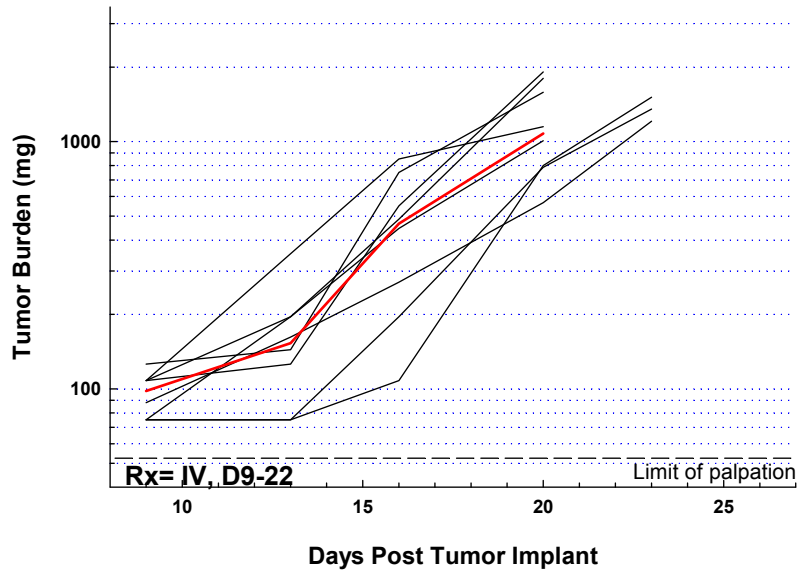
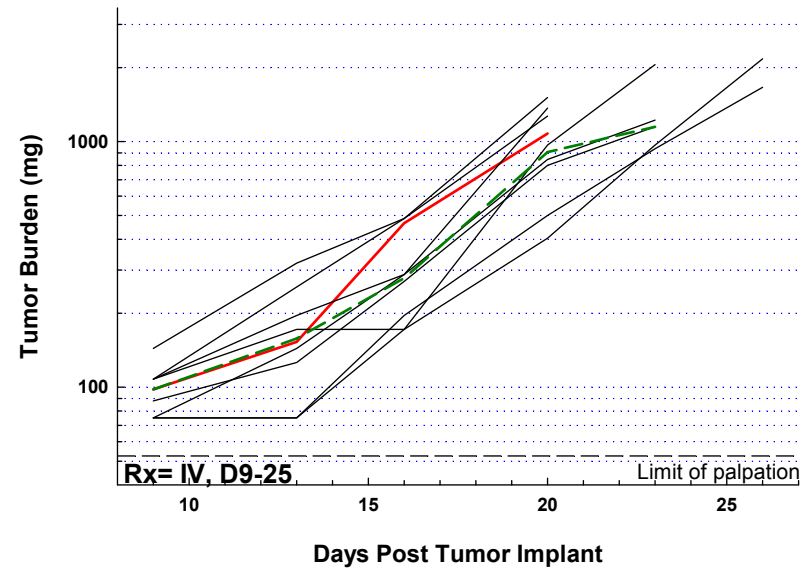


Figure 3 – Individual Tumor Growth Curves by Group

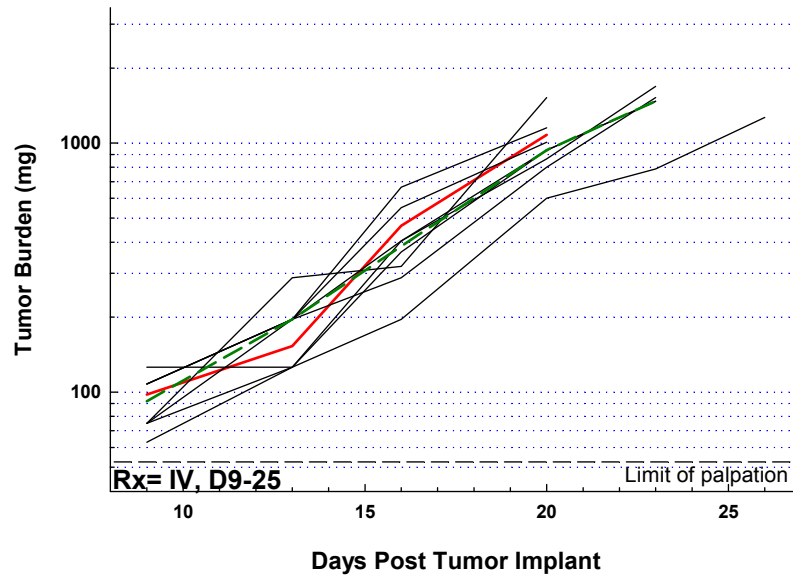
Group 1
Vehicle-Treated Control 0.2ml/20g



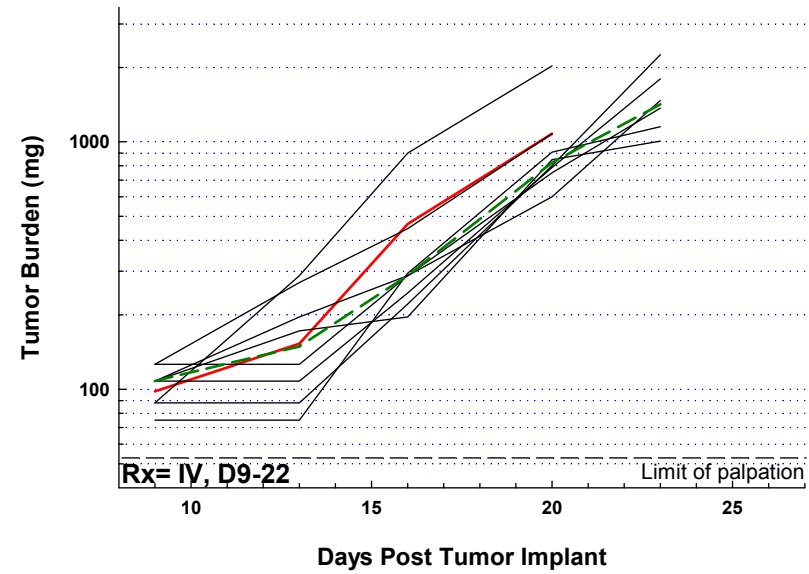
Group 2
PEG-Peptide Mixture 80mg/kg



Group 3
PEG-Peptide Mixture 40mg/kg



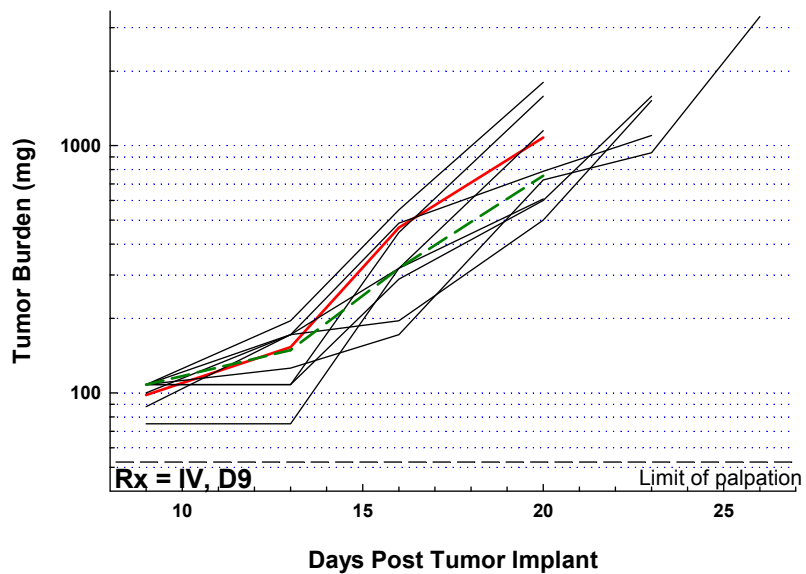
Group 4
PEG-Peptide Mixture 20mg/kg



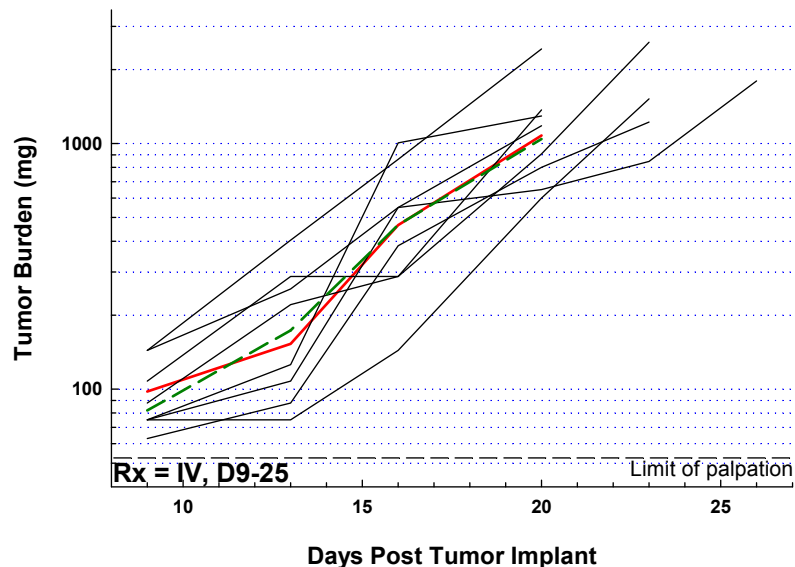
— Control Median

- - - Group Median

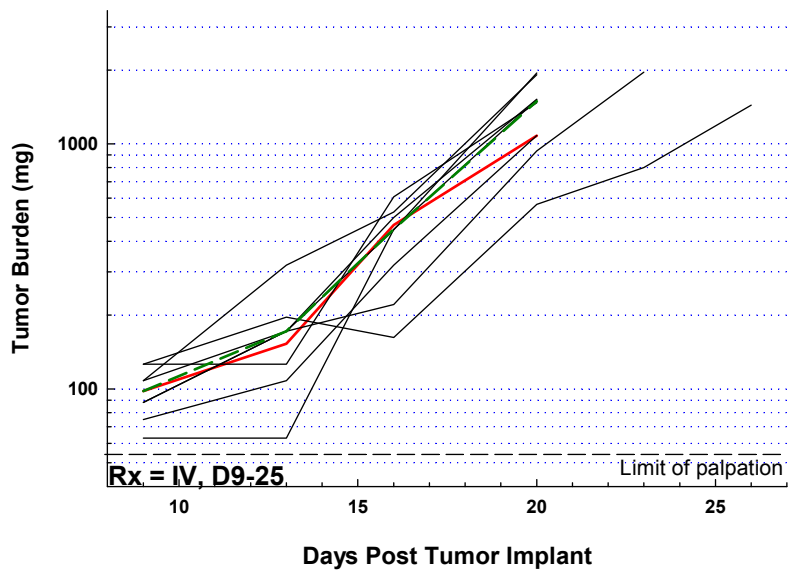
Group 5
PEG-Peptide Mixture 80mg/kg



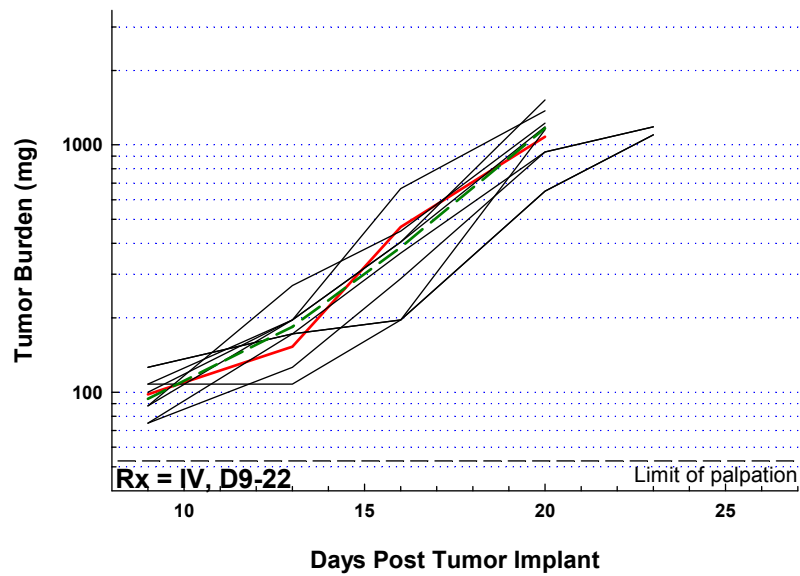
Group 6
PEG-Peptide Mixture 80mg/kg



Group 7
PEG-Peptide Mixture 80mg/kg



Group 8
Peptide Mixture 80mg/kg

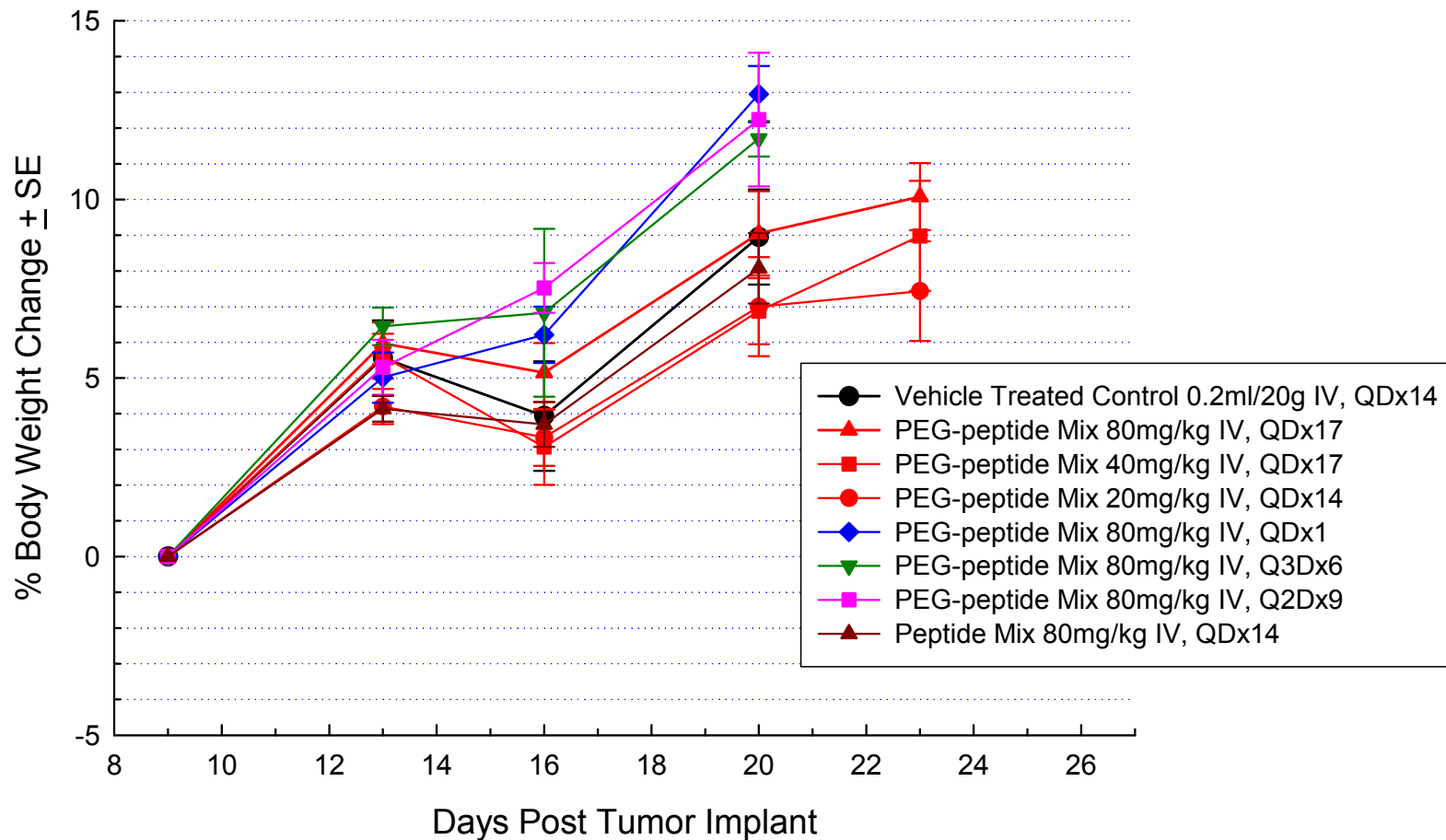


— Control Median

- - - Group Median

Figure 4 – % Body Weight Change Summary with Std. Error

SANA200810 (MIR1052)
 Mean Body Weight Change by Group with Std. Error



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Appendix 1 – Protocol Summary

Study #SANA200810
MIR # 1052

Study Title Efficacy Evaluation of Peptide and PEG-Peptide Mixtures Against Mam25 Murine Mammary Carcinoma
Principal Scientist M. Baugher

Tumor = Mam25
Location = SC
Animal = Mouse
Strain = Balb/c

Gender = F
Test Animals = 64
Implanted Animals = 96
Implant Date = 2/11/2009

Evaluation Size = 750 mg
Fold Growth End Point = 4 x

									Treatment Days	
Group	# Animals	Compound	Route	Schedule	Dose (mg/kg/inj)	Rx Start	Rx End	Start	End	
1	8	Vehicle Treated Control	IV	QDx14**	0.2ml/20g	20-Feb-09	5-Mar-09	9	22	
2	8	Peg-Peptide Mixture	IV	QDx17**	80.00	20-Feb-09	8-Mar-09	9	25	
3	8	Peg-Peptide Mixture	IV	QDx17**	40.00	20-Feb-09	8-Mar-09	9	25	
4	8	Peg-Peptide Mixture	IV	QDx14**	20.00	20-Feb-09	5-Mar-09	9	22	
5	8	Peg-Peptide Mixture	IV	Single Dose	80.00	20-Feb-09	20-Feb-09	9	9	
6	8	Peg-Peptide Mixture	IV	Q3Dx6^	80.00	20-Feb-09	8-Mar-09	9	25	
7	8	Peg-Peptide Mixture	IV	Q2Dx9*	80.00	20-Feb-09	8-Mar-09	9	25	
8	8	Peptide Mixture	IV	QDx14**	80.00	20-Feb-09	5-Mar-09	9	22	

Drugs					
Drug Name	Vehicle	Source	Lot #	Amount Needed (mg)	Stability
PEG-Peptide mixture	PBS	Sanare		20	
Peptide mixture	PBS	Sanare		5	

Measurement Frequencies	General Procedures	
Body Weights: 2x/wk	Allow animals to acclimate for 5 days/ Implant Mam25 tumor fragments (30 to 70mg) into mice high in the right axilla (just under arm). Triage mice into treatment groups on Day 3. Distribute animals to treatment groups such that the mean body weight in each group is within 10% of the overall mean. Body weights and tumor measures are to be recorded 2x/week, clinical signs daily. Mice dosed individually by body weight on the day of treatment as described above. Animals with tumor burdens greater than 1g or found in a moribund condition will be euthanized. Hold animals for Tumor Growth Delay Endpoint and complete regression/partial regression/ tumor free survivor determination. Provide weekly interim reports. Full report will be provided to the client with detailed methods, statistical multi-endpoint analysis, publication quality graphics, discussion of results and all raw data.	
Tumor Weights: 2x/wk		
Sac. Tumor Weight (g): >1g		

General Comments

The purpose of this experiment is to evaluate the efficacy of the Peg-Peptide mixture against established Mam25 in Balb/c mice. The cost for this experiment as written is \$18,200 USD.
PD sampling: Take tumor samples of two mice from the group with the strongest response.

*= Original protocol quoted for 10 injections. Schedule reflects actual number of injections given due to removal of animals for excess in tumor burden.
 **= Original protocol quoted for 21 injections. Schedule reflects actual number of injections given due to removal of animals for excess in tumor burden.
 ^= Original protocol quoted for 7 injections. Schedule reflects actual number of injections given due to removal of animals for excess in tumor burden.

Inoculum			Thioglycolate		
Trocar	% Brei	Number of Cells Injected	Tumor Source Code	Number of positives	Total tubes
<input checked="" type="checkbox"/>			Mam25/35 (T,1-20-09)	0	5

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Appendix 2 – Raw Data (Tumor Measurements and Body Weights)

Group #1 Treatment: **Vehicle Treated Control** Dose: **0.2ml/20g**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	6	7	17.3	6	8	17.9	10	15	18	12	22	19.7																		
2	6	6	18.3	8	11	19.5	10	17	19.9	11	19	19.5																		
3	5	6	17.1	5	6	17.4	6	6	17	10	16	17.5	12	21	18.2															
4	5	6	16.2	7	8	17	9	11	16.7	12	14	17.9																		
5	6	6	16.4	6	7	17.5	10	11	17.6	15	17	18.3																		
6	5	6	16.4	5	6	17.9	7	8	17.2	11	13	18.3	13	16	19															
7	6	6	16.5	7	8	18.1	9	12	17.8	15	16	18																		
8	5	7	18.4	6	9	18.8	7	11	17.7	9	14	19.5	11	20	20.2															

Group #2 Treatment: **Peg-Peptide Mixture** Dose: **80**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	5	6	16	5	6	16.6	7	8	16.6	10	10	17	12	13	17.9	14	17	19.1												
2	6	6	17	8	8	18.1	9	12	18.3	12	21	19.4																		
3	5	7	17.1	6	7	18.1	7	11	18	10	16	17.9	12	16	18.3															
4	6	6	18.3	7	7	18.9	7	7	18.4	11	16	19.5	14	21	20.2															
5	5	6	16.5	5	6	17.8	7	7	17.2	9	10	17.7	11	16	18	16	17	19												
6	6	6	16.7	7	8	17.7	8	9	17.9	11	14	18.3	12	17	18.7															
7	5	6	16.3	6	8	17.5	8	9	17.2	12	19	18.3																		
8	6	8	18.2	8	10	19.5	9	12	19.5	11	21	20.3																		

Group #3 Treatment: **Peg-Peptide Mixture** Dose: **40**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	6	7	16.7	6	7	17.9	9	10	17.6	12	13	18.2	14	15	19.1															
2	5	6	17.9	7	8	18.7	8	9	18.6	10	16	19	13	18	19.6															
3	6	6	18.4	7	8	19.1	9	10	17.8	12	12	19	15	15	19.7															
4	6	6	16.2	7	8	17.7	10	11	17	12	14	17.6																		
5	5	6	17.5	8	9	18.3	8	10	18.2	13	18	19																		
6	6	6	17.6	7	8	18.5	11	11	18.6	12	16	19.4																		
7	5	5	17.2	6	7	18.1	7	8	17.7	10	12	17.8	11	13	18.7	13	15	19.7												
8	5	6	19.4	6	7	20.4	9	9	19.6	12	13	20.5	14	15	20.4															

Group #4 Treatment: **Peg-Peptide Mixture** Dose: **20**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	5	7	17.8	8	9	18.9	10	18	19.1	13	24	20.1																		
2	5	7	18.5	5	7	19.1	7	9	18.8	10	16	20.1	14	23	20.5															
3	6	7	18.2	7	11	19.2	9	11	19	12	15	20.1																		
4	6	6	17	7	8	17.7	8	9	17	10	15	17	12	19	17.2															
5	6	7	19.4	6	7	20.2	8	9	20.1	10	12	20.6	14	15	21.2															
6	6	6	16.3	7	7	17.1	7	8	16.7	11	14	17.4	12	14	17.8															
7	6	6	17.5	6	6	17.8	7	10	17.9	11	13	18.2	15	16	18.6															
8	5	6	18	5	6	18.7	7	12	18.9	11	15	19.3	12	16	19.4															

Group #5 Treatment: **Peg-Peptide Mixture** Dose: **80**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	6	6	18	6	6	18.4	8	9	18.6	10	12	20.1	12	22	20.3															
2	6	6	18.2	7	8	19.1	10	11	19.2	15	16	20.6																		
3	6	6	17.7	6	6	18.6	9	11	18.9	12	22	20																		
4	5	8	17.7	7	7	18.3	7	13	18.4	11	19	19.4																		
5	5	7	18.2	7	7	19.3	9	12	19.5	11	13	20.5	13	13	20.9															
6	6	6	19	7	7	20.7	7	8	21	10	10	22.3	13	18	23															
7	5	6	17.1	5	6	17.8	7	13	18.3	9	15	19.5																		
8	6	6	18.2	6	7	19.2	7	7	19.2	11	12	20.4	12	13	20.8	17	23	22.5												

Group #6 Treatment: **Peg-Peptide Mixture** Dose: **80**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	6	8	16.3	8	8	17.1	10	11	17.1	13	14	18.2																		
2	5	5	16.6	5	7	18	8	12	17.8	10	16	18.4	12	17	18.1															
3	6	8	17.4	9	10	18.2	12	12	18.6	16	19	19.7																		
4	5	6	15.9	5	6	17.1	6	8	18.5	10	12	17.7	13	18	17.9															
5	5	6	17.7	6	6	18.8	10	11	19.9	10	13	19.4	11	14	19	15	16	20												
6	5	6	16.5	6	7	17.8	12	14	17.8	12	18	18.7																		
7	5	7	18.1	7	9	19	8	9	19.1	14	14	20																		
8	6	6	18.2	8	9	19.5	8	9	17	11	15	20.6	15	23	20.8															

Group #7

Treatment: **Peg-Peptide Mixture** Dose: **80**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day			
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	
1	6	6	17.3	7	7	18.4	7	9	18.7	12	13	19.4	14	20	20.3																
2	5	7	16.2	7	7	17.7	9	11	18	13	23	18.6																			
3	5	5	17.4	5	5	17.9	8	14	18.4	12	21	18.5																			
4	6	7	17.8	7	8	18.9	6	9	19.4	9	14	20.1	10	16	20	13	17	20.5													
5	6	7	18.4	6	7	19.4	9	15	19.9	14	15	19.1																			
6	5	6	18.7	6	6	19.2	8	10	20	12	15	21.5																			
7	5	7	17.2	7	7	18.2	10	10	18.2	13	18	20.8																			
8	6	6	19	8	10	19.7	9	13	20	15	17	21.3																			

Group #8

Treatment: **Peptide Mixture** Dose: **80**

Animal	Date 2/20/09 Day 9			Date 2/24/09 Day 13			Date 2/27/09 Day 16			Date 3/3/09 Day 20			Date 3/6/09 Day 23			Date 3/9/09 Day 26			Date Day			Date Day			Date Day			Date Day		
	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)	Width (mm)	Length (mm)	Body Weight (g)
1	5	7	18.4	7	11	19	8	14	18.5	12	17	18.7																		
2	5	6	17.7	7	7	18.6	9	9	18.6	12	13	19.2	13	14	19.8															
3	6	7	17.4	7	7	18.2	7	8	17.8	10	13	18.6	13	13	19.8															
4	6	6	17.5	6	6	18.3	7	8	18.5	12	16	19.2																		
5	5	6	17.6	6	7	18.5	8	9	18.5	12	13	19.3	13	14	19.5															
6	5	7	17.8	7	8	18.5	9	10	18.6	13	14	19.5																		
7	6	6	18.3	7	8	19.1	11	11	19	14	14	20.1																		
8	5	8	19.4	7	8	19.8	9	10	19.9	13	18	21.1																		

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Appendix 3 – Raw Data (Daily Census)

LEGEND: Sacrificed (s) Dead in cage (d) Missing (m) Accidental death (a)

Group	Date 2/20/09		Date 2/21/09		Date 2/22/09		Date 2/23/09		Date 2/24/09		Date 2/25/09		Date 2/26/09		Date 2/27/09		Date 2/28/09		Date 3/1/09		Date 3/2/09		Date 3/3/09		Date 3/4/09			
	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive		
1		8		8		8		8		8		8		8		8		8		8		8		8	s5	3		3
2		8		8		8		8		8		8		8		8		8		8		8		8	s3	5		5
3		8		8		8		8		8		8		8		8		8		8		8		8	s3	5		5
4		8		8		8		8		8		8		8		8		8		8		8		8	s2	6		6
5		8		8		8		8		8		8		8		8		8		8		8		8	s4	4		4
6		8		8		8		8		8		8		8		8		8		8		8		8	s4	4		4
7		8		8		8		8		8		8		8		8		8		8		8		8	s6	2		2
8		8		8		8		8		8		8		8		8		8		8		8		8	s5	3		3

Group	Date 3/5/09		Date 3/6/09		Date 3/7/09		Date 3/8/09		Date 3/9/09		Date 3/10/09		Date 3/11/09		Date 3/12/09		Date 3/13/09		Date 3/14/09		Date 3/15/09		Date 3/16/09		Date 3/17/09			
	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive	Change	Alive
1		3	s3	0		0		0		0																		
2		5	s3	2		2		2	S2	0																		
3		5	s4	1		1		1	S1	0																		
4		6	s6	0		0		0		0																		
5		4	s3	1		1		1	S1	0																		
6		4	s3	1		1		1	S1	0																		
7		2	s1	1		1		1	S1	0																		
8		3	s3	0		0		0		0																		

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Appendix 4 – Raw Data (Clinical Signs, Observations, and Comments)

Date	Day	Group	Animal	Tech	Comment
3-Mar-09	20	1	1,2,4,5,7	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
6-Mar-09	23	1	3,6,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
9-Mar-09	26	2	1,5	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
3-Mar-09	20	2	2,7,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
6-Mar-09	23	2	3,4,6	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
9-Mar-09	26	3	7	MB	Animal euthanized due to tumor burden >1g. Necropsy performed, enlarged spleens.
6-Mar-09	23	3	1,2,3,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
3-Mar-09	20	3	4,5,6	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
3-Mar-09	20	4	1,3	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
6-Mar-09	23	4	2,4,5,6,7,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
3-Mar-09	20	5	7	MB	Animal euthanized due to ulcerated tumor. Necropsy performed, no remarkable findings.
9-Mar-09	26	5	8	MB	Animal euthanized due to tumor burden >1g. Necropsy performed, enlarged spleens.
6-Mar-09	23	5	1,5,6	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
3-Mar-09	20	5	2,3,4	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
9-Mar-09	26	6	5	MB	Animal euthanized due to tumor burden >1g. Necropsy performed, enlarged spleens.
3-Mar-09	20	6	1,3,6,7	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
6-Mar-09	23	6	2,4,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
6-Mar-09	23	7	1	MB	Animal euthanized due to tumor burden >1g. Necropsy performed, enlarged spleens.
9-Mar-09	26	7	4	MB	Animal euthanized due to tumor burden >1g. Necropsy performed, enlarged spleens.
3-Mar-09	20	7	2,3,5,6,7,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
3-Mar-09	20	8	1,4,6,7,8	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.
6-Mar-09	23	8	2,3,5	MB	Animals euthanized due to tumor burden >1g. Necropsies performed, enlarged spleens.

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Appendix 5 – Consolidated Tumor Burdens

Estimated Tumor Weights (mg)

Group #		Treatment Vehicle Treated Control				Route IV		Rx Start Day	Rx End Day
		Dose 0.2ml/20g				Schedule QDx14		9	22
Animal	Day	9	13	16	20	23	26		
1	126	144	750	1584					
2	108	352	850	1150					
3	75	75	108	800	1512				
4	75	196	446	1008					
5	108	126	550	1913					
6	75	75	196	787	1352				
7	108	196	486	1800					
8	88	162	270	567	1210				
Mean	95	166	457	1201	-	-	-	-	-
SD	20	89	260	505	-	-	-	-	-
Median	98	153	466	1079	-	-	-	-	-
Group #		Treatment Peg-Peptide Mixture				Route IV		Rx Start Day	Rx End Day
		Dose 80.00				Schedule QDx17		9	25
Animal	Day	9	13	16	20	23	26		
1	75	75	196	500	936	1666			
2	108	256	486	1512					
3	88	126	270	800	1152				
4	108	172	172	968	2058				
5	75	75	172	405	968	2176			
6	108	196	288	847	1224				
7	75	144	288	1368					
8	144	320	486	1271					
Mean	98	171	295	959	1268	-	-	-	-
SD	24	86	127	401	458	-	-	-	-
Median	98	158	279	908	1152	-	-	-	-
Group #		Treatment Peg-Peptide Mixture				Route IV		Rx Start Day	Rx End Day
		Dose 40.00				Schedule QDx17		9	25
Animal	Day	9	13	16	20	23	26		
1	126	126	405	936	1470				
2	75	196	288	800	1521				
3	108	196	405	864	1688				
4	108	196	550	1008					
5	75	288	320	1521					
6	108	196	666	1152					
7	63	126	196	600	787	1268			
8	75	126	365	936	1470				
Mean	92	181	399	977	1387	-	-	-	-
SD	23	55	149	272	347	-	-	-	-
Median	92	196	385	936	1470	-	-	-	-

Group #		Treatment Peg-Peptide Mixture					Route IV		Rx Start Day	Rx End Day
		Dose 20.00					Schedule QDx14		9	22
Animal\Day	9	13	16	20	23	26				
1	88	288	900	2028						
2	88	88	221	800	2254					
3	126	270	446	1080						
4	108	196	288	750	1368					
5	126	126	288	600	1470					
6	108	172	196	847	1008					
7	108	108	245	787	1800					
8	75	75	294	908	1152					
Mean	103	165	360	975	1509	-	-	-	-	
SD	18	81	231	447	456	-	-	-	-	
Median	108	149	288	824	1419	-	-	-	-	

Group #		Treatment Peg-Peptide Mixture					Route IV		Rx Start Day	Rx End Day
		Dose 80.00					Schedule Single Dose		9	9
Animal\Day	9	13	16	20	23	26				
1	108	108	288	600	1584					
2	108	196	550	1800						
3	108	108	446	1584						
4	100	172	319	1150						
5	88	172	486	787	1099					
6	108	172	196	500	1521					
7	75	75	319	608						
8	108	126	172	726	936	3324				
Mean	100	141	347	969	-	-	-	-	-	
SD	12	43	136	490	-	-	-	-	-	
Median	108	149	319	757	-	-	-	-	-	

Group #		Treatment Peg-Peptide Mixture					Route IV		Rx Start Day	Rx End Day
		Dose 80.00					Schedule Q3Dx6		9	25
Animal\Day	9	13	16	20	23	26				
1	144	256	550	1183						
2	63	88	384	800	1224					
3	144	405	864	2432						
4	75	75	144	600	1521					
5	75	108	550	650	847	1800				
6	75	126	1008	1296						
7	88	221	288	1372						
8	108	288	288	908	2588					
Mean	97	196	510	1155	-	-	-	-	-	
SD	32	117	299	591	-	-	-	-	-	
Median	82	174	467	1046	-	-	-	-	-	

Group #		Treatment Peg-Peptide Mixture					Route IV		Rx Start Day 9		Rx End Day 25	
		Dose 80.00					Schedule Q2Dx9					
Animal\Day	9	13	16	20	23	26						
1	108	172	221	936	1960							
2	88	172	446	1944								
3	63	63	448	1512								
4	126	196	162	567	800	1437						
5	126	126	608	1470								
6	75	108	320	1080								
7	88	172	500	1521								
8	108	320	527	1913								
Mean	98	166	404	1368	-	-	-	-	-	-	-	
SD	23	76	155	477	-	-	-	-	-	-	-	
Median	98	172	447	1491	-	-	-	-	-	-	-	

Group #		Treatment Peptide Mixture					Route IV		Rx Start Day 9		Rx End Day 22	
		Dose 80.00					Schedule QDx14					
Animal\Day	9	13	16	20	23	26						
1	88	270	448	1224								
2	75	172	365	936	1183							
3	126	172	196	650	1099							
4	108	108	196	1152								
5	75	126	288	936	1183							
6	88	196	405	1183								
7	108	196	666	1372								
8	100	196	405	1521								
Mean	96	180	371	1122	-	-	-	-	-	-	-	
SD	18	49	153	275	-	-	-	-	-	-	-	
Median	94	184	385	1168	-	-	-	-	-	-	-	

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Appendix 6 – Consolidated Body Weights

Group #		Treatment Peg-Peptide Mixture						Route IV		Rx Start Day 9		Rx End Day 25	
		Dose 80.00						Schedule Q2Dx9					
Animal\Day	9	13	16	20	23	26							
1	17.3	18.4	18.7	19.4	20.3								
2	16.2	17.7	18.0	18.6									
3	17.4	17.9	18.4	18.5									
4	17.8	18.9	19.4	20.1	20.0	20.5							
5	18.4	19.4	19.9	19.1									
6	18.7	19.2	20.0	21.5									
7	17.2	18.2	18.2	20.8									
8	19.0	19.7	20.0	21.3									
Mean	17.8	18.7	19.1	19.9	-	-	-	-	-	-	-	-	
SD	0.9	0.7	0.8	1.2	-	-	-	-	-	-	-	-	
Median	17.6	18.7	19.1	19.8	-	-	-	-	-	-	-	-	

Group #		Treatment Peptide Mixture						Route IV		Rx Start Day 9		Rx End Day 22	
		Dose 80.00						Schedule QDx14					
Animal\Day	9	13	16	20	23	26							
1	18.4	19.0	18.5	18.7									
2	17.7	18.6	18.6	19.2	19.8								
3	17.4	18.2	17.8	18.6	19.8								
4	17.5	18.3	18.5	19.2									
5	17.6	18.5	18.5	19.3	19.5								
6	17.8	18.5	18.6	19.5									
7	18.3	19.1	19.0	20.1									
8	19.4	19.8	19.9	21.1									
Mean	18.0	18.8	18.7	19.5	-	-	-	-	-	-	-	-	
SD	0.7	0.5	0.6	0.8	-	-	-	-	-	-	-	-	
Median	17.8	18.6	18.6	19.3	-	-	-	-	-	-	-	-	

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Appendix 7 – Consolidated % Body Weight Change

% Body Weight Change

Group # 1		Treatment Vehicle Treated Control						Route IV		Rx Start Day 9	Rx End Day 22
		Dose 0.2ml/20g						Schedule QDx14			
Animal\Day	9	13	16	20	23	26					
1	0.0	3.5	4.0	13.9							
2	0.0	6.6	8.7	6.6							
3	0.0	1.8	-0.6	2.3	6.4						
4	0.0	4.9	3.1	10.5							
5	0.0	6.7	7.3	11.6							
6	0.0	9.1	4.9	11.6	15.9						
7	0.0	9.7	7.9	9.1							
8	0.0	2.2	-3.8	6.0	9.8						
Mean	0.0	5.6	3.9	9.0	-	-	-	-	-	-	
SD	0.0	3.0	4.3	3.8	-	-	-	-	-	-	
Median	0.0	5.8	4.5	9.8	-	-	-	-	-	-	

Group # 2		Treatment Peg-Peptide Mixture						Route IV		Rx Start Day 9	Rx End Day 25
		Dose 80.00						Schedule QDx17			
Animal\Day	9	13	16	20	23	26					
1	0.0	3.8	3.8	6.3	11.9	19.4					
2	0.0	6.5	7.6	14.1							
3	0.0	5.8	5.3	4.7	7.0						
4	0.0	3.3	0.5	6.6	10.4						
5	0.0	7.9	4.2	7.3	9.1	15.2					
6	0.0	6.0	7.2	9.6	12.0						
7	0.0	7.4	5.5	12.3							
8	0.0	7.1	7.1	11.5							
Mean	0.0	6.0	5.2	9.1	10.1	-	-	-	-	-	
SD	0.0	1.7	2.3	3.3	2.1	-	-	-	-	-	
Median	0.0	6.3	5.4	8.5	10.4	-	-	-	-	-	

Group # 3		Treatment Peg-Peptide Mixture						Route IV		Rx Start Day 9	Rx End Day 25
		Dose 40.00						Schedule QDx17			
Animal\Day	9	13	16	20	23	26					
1	0.0	7.2	5.4	9.0	14.4						
2	0.0	4.5	3.9	6.1	9.5						
3	0.0	3.8	-3.3	3.3	7.1						
4	0.0	9.3	4.9	8.6							
5	0.0	4.6	4.0	8.6							
6	0.0	5.1	5.7	10.2							
7	0.0	5.2	2.9	3.5	8.7	14.5					
8	0.0	5.2	1.0	5.7	5.2						
Mean	0.0	5.6	3.1	6.9	9.0	-	-	-	-	-	
SD	0.0	1.8	3.0	2.6	3.4	-	-	-	-	-	
Median	0.0	5.2	4.0	7.4	8.7	-	-	-	-	-	

Group #		Treatment Peg-Peptide Mixture				Route IV		Rx Start Day 9		Rx End Day 25	
		Dose 80.00				Schedule Q2Dx9					
Animal\Day	9	13	16	20	23	26					
1	0.0	6.4	8.1	12.1	17.3						
2	0.0	9.3	11.1	14.8							
3	0.0	2.9	5.7	6.3							
4	0.0	6.2	9.0	12.9	12.4	15.2					
5	0.0	5.4	8.2	3.8							
6	0.0	2.7	7.0	15.0							
7	0.0	5.8	5.8	20.9							
8	0.0	3.7	5.3	12.1							
Mean	0.0	5.3	7.5	12.2	-	-	-	-	-	-	-
SD	0.0	2.2	2.0	5.3	-	-	-	-	-	-	-
Median	0.0	5.6	7.6	12.5	-	-	-	-	-	-	-

Group #		Treatment Peptide Mixture				Route IV		Rx Start Day 9		Rx End Day 22	
		Dose 80.00				Schedule QDx14					
Animal\Day	9	13	16	20	23	26					
1	0.0	3.3	0.5	1.6							
2	0.0	5.1	5.1	8.5	11.9						
3	0.0	4.6	2.3	6.9	13.8						
4	0.0	4.6	5.7	9.7							
5	0.0	5.1	5.1	9.7	10.8						
6	0.0	3.9	4.5	9.6							
7	0.0	4.4	3.8	9.8							
8	0.0	2.1	2.6	8.8							
Mean	0.0	4.1	3.7	8.1	-	-	-	-	-	-	-
SD	0.0	1.0	1.8	2.8	-	-	-	-	-	-	-
Median	0.0	4.5	4.2	9.2	-	-	-	-	-	-	-

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Appendix 8 – Group T/C Values

Calculated T/C Values (%)

Group# \ Day	9	13	16	20	23	26
1	100	100	100	100		
2	100	103	60	84		
3	94	128	83	87		
4	110	97	62	76		
5	110	97	68	70		
6	84	114	100	97		
7	100	112	96	138		
8	96	120	83	108		

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Appendix 9 – Individual and Group Fold-Growth Values

Calculated Fold Growth

Group #	Treatment Vehicle Treated Control						Route IV						Rx Start Day	Rx End Day
	Dose 0.2ml/20g						Schedule QDx14						9	22
Animal\Day	9	13	16	20	23	26								
1	1.0	1.1	6.0	12.6										
2	1.0	3.3	7.9	10.6										
3	1.0	1.0	1.4	10.7	20.2									
4	1.0	2.6	5.9	13.4										
5	1.0	1.2	5.1	17.7										
6	1.0	1.0	2.6	10.5	18.0									
7	1.0	1.8	4.5	16.7										
8	1.0	1.8	3.1	6.4	13.8									
Mean	1.0	1.7	4.6	12.3	-	-	-	-	-	-	-	-	-	-
SD	0.0	0.8	2.1	3.6	-	-	-	-	-	-	-	-	-	-
Median	1.0	1.5	4.8	11.6	-	-	-	-	-	-	-	-	-	-
Group #	Treatment Peg-Peptide Mixture						Route IV						Rx Start Day	Rx End Day
	Dose 80.00						Schedule QDx17						9	25
Animal\Day	9	13	16	20	23	26								
1	1.0	1.0	2.6	6.7	12.5	22.2								
2	1.0	2.4	4.5	14.0										
3	1.0	1.4	3.1	9.1	13.1									
4	1.0	1.6	1.6	9.0	19.1									
5	1.0	1.0	2.3	5.4	12.9	29.0								
6	1.0	1.8	2.7	7.8	11.3									
7	1.0	1.9	3.8	18.2										
8	1.0	2.2	3.4	8.8										
Mean	1.0	1.7	3.0	9.9	13.8	-	-	-	-	-	-	-	-	-
SD	0.0	0.5	0.9	4.2	3.0	-	-	-	-	-	-	-	-	-
Median	1.0	1.7	2.9	8.9	12.9	-	-	-	-	-	-	-	-	-
Group #	Treatment Peg-Peptide Mixture						Route IV						Rx Start Day	Rx End Day
	Dose 40.00						Schedule QDx17						9	25
Animal\Day	9	13	16	20	23	26								
1	1.0	1.0	3.2	7.4	11.7									
2	1.0	2.6	3.8	10.7	20.3									
3	1.0	1.8	3.8	8.0	15.6									
4	1.0	1.8	5.1	9.3										
5	1.0	3.8	4.3	20.3										
6	1.0	1.8	6.2	10.7										
7	1.0	2.0	3.1	9.5	12.5	20.1								
8	1.0	1.7	4.9	12.5	19.6									
Mean	1.0	2.1	4.3	11.0	15.9	-	-	-	-	-	-	-	-	-
SD	0.0	0.8	1.0	4.1	4.0	-	-	-	-	-	-	-	-	-
Median	1.0	1.8	4.1	10.1	15.6	-	-	-	-	-	-	-	-	-

Group #		4				Treatment Peg-Peptide Mixture		Route IV		Rx Start Day	Rx End Day
		Dose 20.00				Schedule QDx14				9	22
Animal\Day	9	13	16	20	23	26					
1	1.0	3.3	10.2	23.0							
2	1.0	1.0	2.5	9.1	25.6						
3	1.0	2.1	3.5	8.6							
4	1.0	1.8	2.7	6.9	12.7						
5	1.0	1.0	2.3	4.8	11.7						
6	1.0	1.6	1.8	7.8	9.3						
7	1.0	1.0	2.3	7.3	16.7						
8	1.0	1.0	3.9	12.1	15.4						
Mean	1.0	1.6	3.7	10.0	15.2	-	-	-	-	-	-
SD	0.0	0.8	2.7	5.7	5.7	-	-	-	-	-	-
Median	1.0	1.3	2.6	8.2	14.0	-	-	-	-	-	-

Group #		5				Treatment Peg-Peptide Mixture		Route IV		Rx Start Day	Rx End Day
		Dose 80.00				Schedule Single Dose				9	9
Animal\Day	9	13	16	20	23	26					
1	1.0	1.0	2.7	5.6	14.7						
2	1.0	1.8	5.1	16.7							
3	1.0	1.0	4.1	14.7							
4	1.0	1.7	3.2	11.5							
5	1.0	2.0	5.5	8.9	12.5						
6	1.0	1.6	1.8	4.6	14.1						
7	1.0	1.0	4.3	8.1							
8	1.0	1.2	1.6	6.7	8.7	30.8					
Mean	1.0	1.4	3.5	9.6	-	-	-	-	-	-	-
SD	0.0	0.4	1.5	4.3	-	-	-	-	-	-	-
Median	1.0	1.4	3.7	8.5	-	-	-	-	-	-	-

Group #		6				Treatment Peg-Peptide Mixture		Route IV		Rx Start Day	Rx End Day
		Dose 80.00				Schedule Q3Dx6				9	25
Animal\Day	9	13	16	20	23	26					
1	1.0	1.8	3.8	8.2							
2	1.0	1.4	6.1	12.7	19.4						
3	1.0	2.8	6.0	16.9							
4	1.0	1.0	1.9	8.0	20.3						
5	1.0	1.4	7.3	8.7	11.3	24.0					
6	1.0	1.7	13.4	17.3							
7	1.0	2.5	3.3	15.6							
8	1.0	2.7	2.7	8.4	24.0						
Mean	1.0	1.9	5.6	12.0	-	-	-	-	-	-	-
SD	0.0	0.7	3.7	4.1	-	-	-	-	-	-	-
Median	1.0	1.7	4.9	10.7	-	-	-	-	-	-	-

Group #		Treatment Peg-Peptide Mixture						Route IV		Rx Start Day	Rx End Day
Dose 80.00		Schedule Q2Dx9								9	25
Animal	Day	9	13	16	20	23	26				
1	1.0	1.6	2.0	8.7	18.1						
2	1.0	2.0	5.1	22.1							
3	1.0	1.0	7.1	24.0							
4	1.0	1.6	1.3	4.5	6.3	11.4					
5	1.0	1.0	4.8	11.7							
6	1.0	1.4	4.3	14.4							
7	1.0	2.0	5.7	17.3							
8	1.0	3.0	4.9	17.7							
Mean	1.0	1.7	4.4	15.0	-	-	-	-	-	-	
SD	0.0	0.6	1.9	6.6	-	-	-	-	-	-	
Median	1.0	1.6	4.9	15.8	-	-	-	-	-	-	

Group #		Treatment Peptide Mixture						Route IV		Rx Start Day	Rx End Day
Dose 80.00		Schedule QDx14								9	22
Animal	Day	9	13	16	20	23	26				
1	1.0	3.1	5.1	13.9							
2	1.0	2.3	4.9	12.5	15.8						
3	1.0	1.4	1.6	5.2	8.7						
4	1.0	1.0	1.8	10.7							
5	1.0	1.7	3.8	12.5	15.8						
6	1.0	2.2	4.6	13.4							
7	1.0	1.8	6.2	12.7							
8	1.0	2.0	4.1	15.2							
Mean	1.0	1.9	4.0	12.0	-	-	-	-	-	-	
SD	0.0	0.6	1.6	3.1	-	-	-	-	-	-	
Median	1.0	1.9	4.3	12.6	-	-	-	-	-	-	

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Appendix 10 - Peptide Prep Procedure

PEG-peptide prep
2mg/ml dilution and Freeze Scheme

Peptide	MW	mg provided	ml PBS to add	conc mg/ml	conc needed for 0.000347 mmol/ml	volume of 2mg/ml needed	dilute XX ml to a final volume of X	ml of PBS to add	dosing days	withdraw volume (ml)	amt remaining	# 1.0 ml aliquots (equal molar conc)	remaining 2mg/ml solution	Remaining mg in 2mg/ml solution
G1	3462.89	20	10	2	1.201623	9	14.9797	5.9797	21	12.5	2.4797	2	1	2
G2	2036.25	20	10	2	0.706579	9	25.4749	16.4749	21	12.5	12.9749	13	1	2
G3	247.58	20	10	2	0.08591	1	23.2801	22.2801	21	12.5	10.7801	11	9	18
G4	2246.66	20	10	2	0.779591	9	23.089	14.089	21	12.5	10.589	11	1	2
G5	2736.31	20	10	2	0.9495	9	18.9574	9.9574	21	12.5	6.4574	6	1	2
G6	1466.63	20	10	2	0.508921	9	35.369	26.369	21	12.5	22.869	23	1	2
G7	2561.04	20	10	2	0.888681	9	20.2547	11.2547	21	12.5	7.7547	8	1	2
G8	2083.58	20	10	2	0.723002	9	24.8962	15.8962	21	12.5	12.3962	12	1	2
G9	2223.54	20	10	2	0.771568	9	23.3291	14.3291	21	12.5	10.8291	11	1	2
G10	1663.9	20	10	2	0.577373	9	31.1757	22.1757	21	12.5	18.6757	19	1	2
G11	2407.68	20	10	2	0.835465	9	21.5449	12.5449	21	12.5	9.0449	9	1	2
G12	108.22	20	10	2	0.037552	1	53.259	52.259	21	12.5	40.759	41	9	18
freeze remaining 2mg/ml soln in 1.0ml aliquots								5.9797	150 ml total					
peptide prep and freezing procedure.xls												freeze aliquot size =1.0 ml for the mix remaining		

2mg/ml dilution and Freeze Scheme

Peptide	MW	mg provided	ml PBS to add	conc mg/ml	conc needed for 0.000347mmol/ml	volume of 2mg/ml needed	dilute 2.0 ml to a final volume of X	ml of PBS to add	dosing days	withdraw volume (ml)	amt remainin g	# 1.0 ml aliquots
59	3125.5	10	5	2	1.0845485	2.2	4.057	1.857	7	3.9	0.157	0
60	1735.9	10	5	2	0.6023573	2.2	7.3046	5.1046	7	3.9	3.4046	3
61	1910.2	10	5	2	0.6628394	2.2	6.6381	4.4381	7	3.9	2.7381	3
62	1909.3	10	5	2	0.6625271	2.2	6.6412	4.4412	7	3.9	2.7412	3
63	2399	10	5	2	0.832453	2.2	5.2856	3.0856	7	3.9	1.3856	1
64	1326.5	10	5	2	0.4602955	2.2	9.5591	7.3591	7	3.9	5.6591	6
65	1129.3	10	5	2	0.3918671	2.2	11.2283	9.0283	7	3.9	7.3283	7
66	2223.7	10	5	2	0.7716239	2.2	5.7023	3.5023	7	3.9	1.8023	2
67	1746.2	10	5	2	0.6059314	2.2	7.2615	5.0615	7	3.9	3.3615	3
68	2070.3	10	5	2	0.7183941	2.2	6.1248	3.9248	7	3.9	2.2248	2
69	1570.8	10	5	2	0.5450676	2.2	8.0724	5.8724	7	3.9	4.1724	4
70	1886.2	10	5	2	0.6545114	2.2	6.7226	4.5226	7	3.9	2.8226	3
freeze remaining 2mg/ml soln in 1.0ml aliquots= 3 each								1.857	46.8 ml total 6.685714 freeze volume			
										freeze aliquot size =1.0 ml for the mix remaining		

peptide prep and freezing procedure.xls