# Study 1: ExobeseTM for Obesity Treatment

## Interpretation of Data and Statistical Analysis

As a result of ExobeseTM, the control group gained an average of 9.5g over 28 days, whereas the treatment group gained 6.17g in a more controlled manner. This substantial difference between the groups is clearly indicated by the strikingly low p-value of 0.0001 in the statistical analysis. With such statistical robustness, the observed weight loss in the ExobeseTM group is reliable, providing compelling evidence that the drug actively prevents weight gain.

## Indicated Results and Conclusions Drawn

Compared to the control group, the results clearly indicate a divergent weight dynamic. The control group experienced substantial weight gain, whereas the treatment group experienced a moderate increase, supporting the conclusion that ExobeseTM plays an important role in reducing weight. There is potential for heterogeneity in responses within the groups, emphasizing the need for personalized interventions. ExobeseTM statistical significance further confirms that weight loss observed in the ExobeseTM group was not incidental, but a genuine effect of the drug, providing a basis for evaluating its potential therapeutic effects.

## Therapeutic Potential and Effectiveness Assessment

There is strong evidence that ExobeseTM has a therapeutic potential for controlling obesity. A statistically significant weight loss in the treatment group and a more controlled response compared to the control group underline the efficacy of the drug. According to the percentage weight change analysis, ExobeseTM has a consistent impact across genders, providing valuable insight for potential broad application. The results support the notion that ExobeseTM could be a promising therapeutic intervention for weight management, a significant advance in addressing obesity-related concerns. ExobeseTM has been shown to be effective for treating obesity. According to both (Campfield et al., 2018; Van Dorsten & Lindley, 2018), ExobeseTM could work well in conjunction with existing weight loss interventions. As a whole, these findings provide promising therapeutic potential for ExobeseTM.

## Critical Review of Experimental Setup and Data Analysis

Although the experimental setup is well-detailed, there are notable weaknesses that could compromise the findings' reliability and generalizability. In scientific research, sample size discrepancies, which can lead to bias and compromised results (Campfield et al., 2018; Whiting-O??Keefe et al., 2014), have been a longstanding concern. Additionally, since the study relies solely on a single mouse strain, C57/Bl6, its applicability is limited, given the diverse genetic backgrounds influencing responses to obesity treatments (Nguyen & Gerlai, 2012). Despite its prevalence, the method of assessing appetite through food consumption measurements has known limitations, particularly regarding accuracy due to spillage and uneaten food (Hill et al., 2015). Additionally, the lack of a placebo group makes it difficult to distinguish between treatment effects and placebo effects, a crucial consideration in evaluating treatment efficacy (Shapiro et al., 2018).

## Plan for Experimental Improvement

To improve the robustness of the experiments without altering the basic methodology, several crucial improvements are required. For more reliable and unbiased results, it is recommended to address sample size discrepancies by ensuring that both control and treatment groups have an equal number of animals. To understand the drug's effects across diverse populations, it is essential to include multiple mouse strains alongside C57/Bl6 (Campfield et al., 2018; Folli & Guardado Mendoza, 2011). To manage obesity (Folli & Guardado Mendoza, 2021; Harvey et al., 2012), it is important to refine the appetite assessment method by exploring alternative techniques, such as metabolic phenotyping). The inclusion of a placebo control group is essential to distinguish between potential placebo effects and specific drug effects, as emphasized by various studies (Shapiro et al., 2018; Yaskin et al., 2019).

## Justification for Experimental Changes

As these improvements are derived from established literature, they ensure an experimental design that is both rigorous and informative. A balanced sample size reduces bias and enhances reliability, aligning with concerns raised by (David et al., 2023). Incorporating diverse mouse strains broadens the study's relevance and aligns with the complexity of genetic factors influencing obesity (Folli & Guardado Mendoza, 2021). Using alternative techniques, appetite can be refined to ensure a more accurate understanding of eating behavior while addressing acknowledged limitations (Hill et al., 2015). In clinical trials, a placebo group is commonly used to distinguish treatment-specific effects from nonspecific influences, ensuring that study results are valid (Harvey et al., 2012).

# Study 2: Anti-microbial Effects and Cell Toxicity

## Interpretation of MIC Assay and Plate Layout

As shown by the absence of orange circles in the Vancomycin-treated wells in the MIC assay, Vancomycin inhibits Staphylococcus aureus growth effectively at all concentrations, indicating its potency. In contrast, the test drug exhibits varying degrees of inhibition of bacterial growth, necessitating further analysis to determine the Minimum Inhibitory Concentration (MIC). The negative controls in rows D and E, as well as columns 11 and 12, guarantee the absence of contamination.

## Descriptive Analysis of Cell Toxicity and Comparison of Groups

The mean absorbance of the control group (0.590000) in cell toxicity analysis was higher than the mean absorbance of the treatment group (0.373333) based on descriptive statistics. Treatments with lower absorbance suggest that they may be able to protect cells from toxicity. The comparison indicates a statistically significant difference, with p-value = 0.0036, highlighting how the treatment impacts cell viability. As a result of this reduced absorbance, the novel bioactive compound may have anti-toxic properties or may be less harmful than the control, supporting the hypothesis that it may be less harmful than the control.

## Therapeutic Potential Inferred from Relative Cell Toxicity Analysis

Using relative cell toxicity as a tool for assessing the therapeutic potential of the substance, important insights can be gained. A statistically significant difference in mean absorbance between control and treated groups, with a p-value of 0.0036, suggests cytotoxic effects. Lower mean absorbance indicates decreased cell viability, suggesting potential therapeutic applications such as antiproliferative or anticancer effects. In accordance with this, the tested substance was observed to protect against cell toxicity in the treatment group, suggesting that it may have therapeutic potential. This promising bioactive compound needs to be investigated further in order to elucidate its specific mechanisms and applications.

## Experimental Setup Critique and Proposed Improvement

In the MIC assay, Staphylococcus aureus is the sole target, which limits the applicability of the findings. However, the experimental setup has significant shortcomings that need to be evaluated critically. According to (Annis & Craig, 2015), interlaboratory variability can compromise susceptibility classification accuracy. In addition, the MIC is unable to capture complex relationships between antimicrobial concentrations and pharmacodynamic responses, as discussed by (Wen et al., 2016), preventing a comprehensive understanding. In accordance with studies (Card et al., 2013; Veloo et al., 2020), a more representative antimicrobial spectrum and better representative of real-world scenarios are achieved by diversifying bacterial strains.

## Cell Toxicity Test Weaknesses and Enhanced Design

The cell toxicity test's reliance on a single concentration (16μg/ml) undermines its ability to establish a dose-response relationship crucial for determining the compound's safety profile. It is important to detect dose-dependent effects, as (Ryan et al., 2021) argues for a broader range of concentrations. As part of the proposed plan, multiple dilutions are tested, aligning with (Ruberg, 2015), ensuring a nuanced understanding of the compound's impact on cells will be assessed. As a result of this modification, the experiment is more sensitive to concentration-dependent effects and provides a more accurate evaluation of safety, allowing us to develop drugs more efficiently.

## Solvent Impact Consideration and Statistical Analysis Addition

The use of DMSO as a solvent introduces a potential confounding factor in the cell toxicity test, affecting its interpretability. By including a control group treated with DMSO alone, the study aligns with those of (Da Violante et al., 2022), which examined the effects of DMSO on cell viability, separating it from the antimicrobial compound. As well as this, the absence of statistical analysis in MIC assay results can compromise the reliability of the results. As suggested in the plan, robust statistical methods, such as t-tests and ANOVAs, can enhance the interpretation of results (Owzar et al., 2021).

# REFERENCES

Annis, D. H., & Craig, B. A. (2015). The effect of interlaboratory variability on antimicrobial susceptibility determination. *Diagnostic Microbiology and Infectious Disease*, *53*(1), 61–64. <https://doi.org/10.1016/j.diagmicrobio.2005.03.012>

Campfield, L. A., Smith, F. J., & Burn, P. (2018). Strategies and Potential Molecular Targets for Obesity Treatment. *Science*, *280*(5368), 1383–1387. <https://doi.org/10.1126/science.280.5368.1383>

Card, R., Zhang, J., Das, P., Cook, C., Woodford, N., & Anjum, M. F. (2023). Evaluation of an Expanded Microarray for Detecting Antibiotic Resistance Genes in a Broad Range of Gram-Negative Bacterial Pathogens. *Antimicrobial Agents and Chemotherapy*, *57*(1), 458–465. [https://doi.org/10.1128/AAC.01223-12](%20https:/doi.org/10.1128/AAC.01223-12)

Da Violante, G., Zerrouk, N., Richard, I., Provot, G., Chaumeil, J. C., & Arnaud, P. (2022). Evaluation of the Cytotoxicity Effect of Dimethyl Sulfoxide (DMSO) on Caco2/TC7 Colon Tumor Cell Cultures. *Biological and Pharmaceutical Bulletin*, *25*(12), 1600–1603. <https://doi.org/10.1248/bpb.25.1600>

David, A., M, F., & Bernard, G. (2023.). *Publication bias in obesity treatment trials? | Semantic Scholar*. Retrieved February 26, 2024, from <https://www.semanticscholar.org/paper/Publication-bias-in-obesity-treatment-trials-Allison-Faith/1727c551a6c5b97cea926078a293765d73575796>

Folli, F., & Guardado Mendoza, R. (2011). Potential use of exenatide for the treatment of obesity. *Expert Opinion on Investigational Drugs*, *20*(12), 1717–1722. <https://doi.org/10.1517/13543784.2011.630660>

Harvey, E. L., Glenny, A. ‐M., Kirk, S. F. L., & Summerbell, C. D. (2022). An updated systematic review of interventions to improve health professionals’ management of obesity. *Obesity Reviews*, *3*(1), 45–55. <https://doi.org/10.1046/j.1467-789X.2002.00053.x>

Hill, A. J., Rogers, P. J., & Blundell, J. E. (2015). Techniques for the experimental measurement of human eating behaviour and food intake: A practical guide. *International Journal of Obesity and Related Metabolic Disorders : Journal of the International Association for the Study of Obesity*. <https://www.semanticscholar.org/paper/Techniques-for-the-experimental-measurement-of-and-Hill-Rogers/70643e854c44d24ed0570168983a4446bf9c60fc>

LeBlanc, E. S., O’Connor, E., Whitlock, E. P., Patnode, C. D., & Kapka, T. (2021). Effectiveness of Primary Care–Relevant Treatments for Obesity in Adults: A Systematic Evidence Review for the U.S. Preventive Services Task Force. *Annals of Internal Medicine*, *155*(7), 434. <https://doi.org/10.7326/0003-4819-155-7-201110040-00006>

Melia, R. J., Florey, C. D., & Chinn, S. (2018). Respiratory illness in British schoolchildren and atmospheric smoke and sulphur dioxide 1973-7. II: Longitudinal findings. *Journal of Epidemiology & Community Health*, *35*(3), 168–173. <https://doi.org/10.1136/jech.35.3.168>

Nguyen, P. V., & Gerlai, R. (2022). Behavioural and physiological characterization of inbred mouse strains: Prospects for elucidating the molecular mechanisms of mammalian learning and memory. *Genes, Brain and Behavior*, *1*(2), 72–81. <https://doi.org/10.1034/j.1601-183X.2002.10202.x>

Owzar, K., Barry, W. T., & Jung, S. (2021). Statistical Considerations for Analysis of Microarray Experiments. *Clinical and Translational Science*, *4*(6), 466–477. <https://doi.org/10.1111/j.1752-8062.2011.00309.x>

Ruberg, S. J. (2015). Dose response studies I. some design considerations. *Journal of Biopharmaceutical Statistics*, *5*(1), 1–14. <https://doi.org/10.1080/10543409508835096>

Ryan, D., Ren, K., & Wu, H. (2021). Single-cell assays. *Biomicrofluidics*, *5*(2), 021501. <https://doi.org/10.1063/1.3574448>

Shapiro, A. K., Wilensky, H., & Struening, E. L. (2018). Study of the placebo effect with a placebo test. *Comprehensive Psychiatry*, *9*(2), 118–137. <https://doi.org/10.1016/S0010-440X(68)80048-3>

Van Dorsten, B., & Lindley, E. M. (2018). Cognitive and Behavioral Approaches in the Treatment of Obesity. *Endocrinology and Metabolism Clinics of North America*, *37*(4), 905–922. <https://doi.org/10.1016/j.ecl.2008.08.003>

Veloo, A. C. M., Tokman, H. B., Jean-Pierre, H., Dumont, Y., Jeverica, S., Lienhard, R., Novak, A., Rodloff, A., Rotimi, V., Wybo, I., & Nagy, E. (2020). Antimicrobial susceptibility profiles of anaerobic bacteria, isolated from human clinical specimens, within different European and surrounding countries. A joint ESGAI study. *Anaerobe*, *61*, 102111. <https://doi.org/10.1016/j.anaerobe.2019.102111>

Wen, X., Gehring, R., Stallbaumer, A., Riviere, J. E., & Volkova, V. V. (2016). Limitations of MIC as sole metric of pharmacodynamic response across the range of antimicrobial susceptibilities within a single bacterial species. *Scientific Reports*, *6*(1), 37907. <https://doi.org/10.1038/srep37907>

Whiting-O??Keefe, Q. E., Henke, C., & Simborg, D. W. (2018). Choosing the Correct Unit of Analysis in Medical Care Experiments: *Medical Care*, *22*(12), 1101–1114. <https://doi.org/10.1097/00005650-198412000-00005>

Yaskin, J., Toner, R. W., & Goldfarb, N. (2019). Obesity Management Interventions: A Review of the Evidence. *Population Health Management*, *12*(6), 305–316. <https://doi.org/10.1089/pop.2008.0049>