On the Use of Bayes Techniques to Bridge from an In Vitro System to a Clinical Trial

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Abstract
The MIMIC system allows for human experiments to be done in vitro and subjects to be treated with multiple treatments which are not possible in clinical trials. Instead of the typical animal studies, the system can evaluate human immune responses during the pre-clinical period without safety concerns. The objective is to investigate a Bayesian modeling which uses the study information from MIMIC as prior information to the analyses of a clinical trial from the same vaccine candidate. Benefits from MIMIC technology are fully described and results present a potential usage of MIMIC study.

Key Words: MIMIC Technology, Bayesian Modeling, Clinical Trials, In Vitro Study, Pre-Clinical, Vaccine

1. Introduction

Clinical trials (CTs) are required for registering a vaccine candidate from regulatory point of view. The design and analysis of a clinical trial is not only important for the reliability of the conclusion but also highly relates to time or money consuming. Reducing sample size or in another word, increasing power of conclusion is a perpetual topic. A new technology, Modular Immune In vitro construct technology, MIMIC® (MIMIC; Warren, W., 2012) for short, was developed from Sanofi Pasteur VaxDesign®, which introduces an innovative point of view on evaluating immune responses of a vaccine candidate. In addition to responding to immune stimuli, other responses have been noticed. For example, the age related reduction in response to flu vaccines has been duplicated in MIMIC experiments (Dauner A., 2017).

The MIMIC technology is a process for removing immune system related blood cells from a donor and carrying out immunological assessments in vitro. An intuitive diagram in Figure 1 (Bharadwaj, 2012) shows how the system works. A special system is connected to a volunteer for collecting blood samples from the human. It filters out all anti-virus related cells in blood (such as PBMCs, T-cell, B-cells (Bharadwaj, 2012)) and those cells are stored under a lower temperature to keep them alive. All other blood cells with the plasma circulate back to the human body. The amount of collected blood samples is huge enough to be used for multiple studies. In each study, several identical blood samples from the same donor are mixed with different vaccines. It ensures that the immune responses obtained from multiple vaccine candidates are from the same person with ideally controlling for all other factors. The pure differences between vaccine effects could be evaluated through MIMIC technology.

The process is believed to approximate the workings of the human immune system in vivo. It has several advantages that can be exploited for vaccine development. From a data analytic view, the same blood sample from a single donor can be split and treated with multiple treatments. Since individual immune systems are known to be variable. The ability to block on donors substantially reduces the variability of comparisons. Most importantly, it does not provide any injection into the human bodies since all treatments are applied externally.

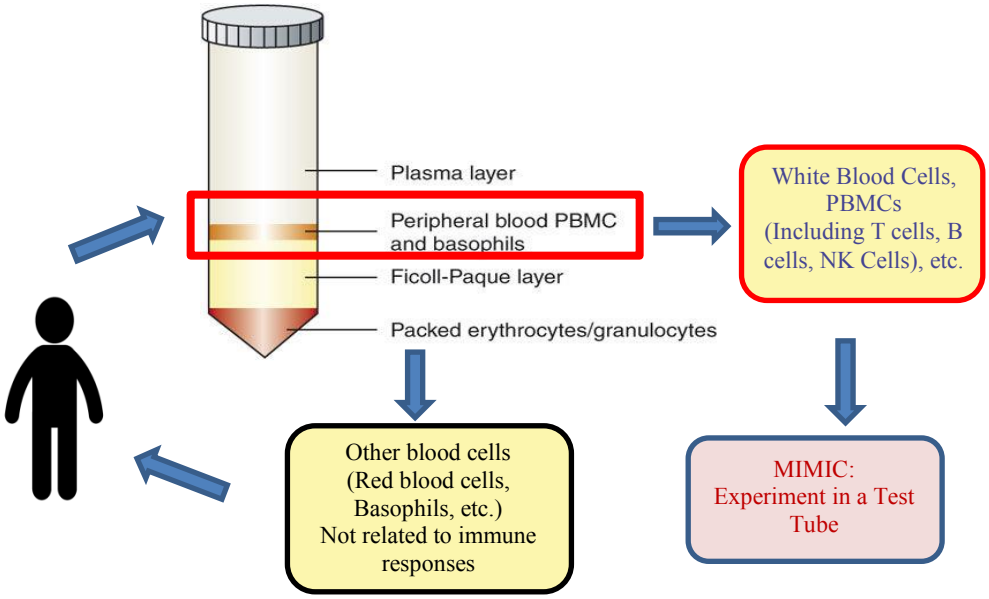

Figure 1. The diagram of Modular IMmune In vitro Construct Technology

The resultant experiment and data define a classical randomized complete block (RCBD) (Lawal B., 2014) experiment with donor as the block. There is usually enough whole blood for each donor to be treated with as many as 10 treatments. Since the studies can be run much faster than a clinical trial the MIMIC studies are a good candidate for a screening experiment used to eliminate weak or unacceptable formulations for example.

2. Objective

MIMIC studies have been carried out to assess formulations, screen different vaccines and give some measurements of different vaccines and antigens. We wanted to use the results of a MIMIC study to assist in the design of a following clinical trial. Not only the vaccine administration differs, but also the assay used to test the immune responses of a MIMIC study is related to but not the same as those used in clinical trials. It is hoped that the intensity of responses in both studies would be similar.

We investigated the idea of using Bayesian methods to bridge from a MIMIC study to a corresponding clinical trial. We had a MIMIC trial and a clinical trial run within the same age group and using the same manufactured batches of vaccine. We wanted to use a posterior from the MIMIC study to define a prior for the clinical trial and then use the resultant posterior of the clinical trial to evaluate the performance of the effort. We hoped that we might get some improvements in the width of confidence interval or the precision

of some estimates. If so that might lead to sample size reduction or other trial improvements.

There are reasons to believe that the result of the MIMIC would be similar to the results of a CT at some level. There are, nevertheless, challenges. The assays are not identical. The MIMIC is a randomized complete block design (RCBD) while the CT is a complete random design (CRD) (Addelman & Sidney, 1969) so that the error structures are different. In particular, the donor to donor variation is confounded with the random error in the CT but removed from comparisons in the MIMIC study. Though there is hope that the effects measured in the two studies are associated in some way they may have a complex non-linear relationship.

3. Data Sets

3.1 Data

Table 1 gives the details of the common or different characteristics between the datasets in each study. The datasets used for the research are the MIMIC study and clinical trial for investigating exactly same vaccines: Vaccines I II III IV where group III is used as the reference group. The interesting antigens are 4 virus strains denoted as A, B, C and D. The CT is a typical completed randomized design (CRD) whereas the MIMIC is randomized complete block design (RCBD) as introduced above. Over 100 subjects are enrolled in the CT in each group whereas 25 donors were enrolled in the MIMIC study. Although the sample size is smaller in MIMIC than in CT, because of the study design, MIMIC is still as powerful as CT to conclude on treatment effects.

Both studies measured the baseline and the post-vaccination immune response. Two different assays are used in the two studies. In CT, HAI assay is used as a typical laboratory assay to test the amount of antibodies stimulated by the vaccine. In MIMIC, SA-HAI assay is developed to test the amount of B-cells and T-cells which indirectly represent the level of immune responses.

Table 1: Comparisons on the Characteristics between MIMIC Study and CT

	Common to Both	CT Specific	MIMIC specific
4 Vaccines	Groups I II III IV		
Sample size per group		Over 100 in each Group	25 in Total
Virus strains	Strain A, B, C, D		
Baseline immunogenicity	Yes		
Post vaccination immunogenicity	Yes		
Assay for immunogenicity		HAI	SA-HAI
Study Design		CRD	RCBD

3.2 Descriptive Analyses

Figure 2 represents the distributions of immune responses for each strain in vaccine group I by histograms, box plots and estimated parametric distributions. It seems that consistently higher mean immune responses are observed in CT than those in MIMIC. Vaccine groups II, III and IV represent a very similar trend with those in vaccine group I. These data are the log10 transformation from the original test results. The endpoint

comparisons in these types of experiments are always the geometric mean titer ratios of the two treatments. The hope is that the relationship between groups is clearer or these ratios might be more similar than the raw titers.

Table 2 shows the summary statistics of the treatment effects of vaccine group I, II, III, IV for each strain. Overall the observations are consistent with those observed in Figure 2. The mean immune responses in a log10 scale are higher in CT than in MIMIC study for all strains and all vaccine groups. However, the directions of quantitative increasing are not consistent between two studies among different strains within the same vaccine group or among different vaccines within the same treatment group. For example in vaccine I, the mean immune response in CT is 2.3 for strain A which is lower than 2.57 for strain C whereas it is 1.33 in MIMIC for strain A which is higher in 0.99 for strain C.

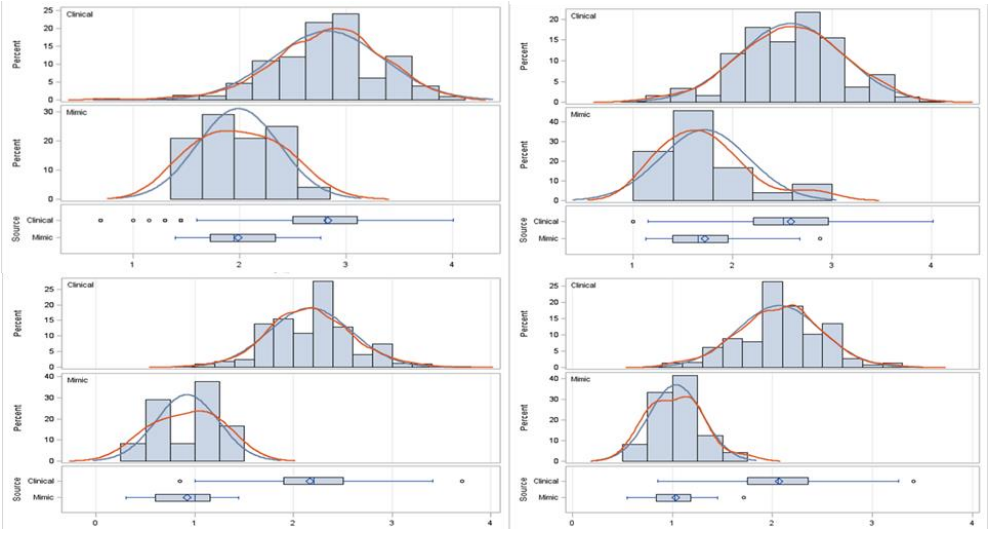

Figure 2: Distribution of post-vaccination immune responses for 4 virus strains for vaccine group I comparing CT (upper) to MIMIC (lower)

Table 2: A Summary Statistics of the Post-vaccination Immune Responses in MIMIC and CT for Each Vaccine Group for Each Strain

		Vaccine I		Vaccine II		Vaccine III		Vaccine IV	
Strain	**Study**	**Mean**	**Std**	**Mean**	**Std**	**Mean**	**Std**	**Mean**	**Std**
A	**MIMIC**	1.33	0.94	1.40	1.04	1.23	0.91	1.24	0.96
	CT	2.30	0.46	2.27	0.48	2.49	0.43	2.31	0.47
B	**MIMIC**	1.08	0.84	1.15	0.95	1.06	0.79	1.00	0.71
	CT	2.84	0.59	2.80	0.60	3.07	0.57	2.83	0.62
C	**MIMIC**	0.99	0.76	1.07	0.97	1.09	0.86	1.12	0.76
	CT	2.57	0.55	2.62	0.45	2.66	0.43	2.64	0.38
D	**MIMIC**	0.46	0.52	0.53	0.58	0.67	0.67	0.53	0.59
	CT	2.50	0.44	2.54	0.44	2.32	0.44	2.28	0.48

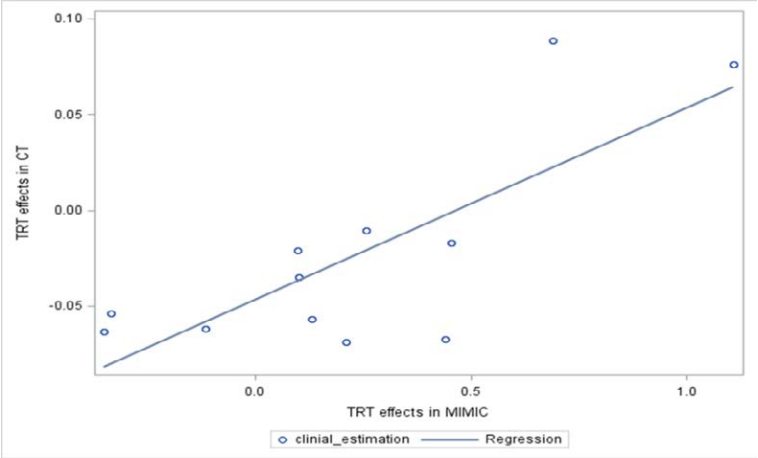

Figure 3: The regression for treatment effects of each investigational vaccine group (vs. group III) in CT (y-axis) vs. MIMIC (x-axis) for all strains

There is some empirical support for these views in the results of the two experiments when they were run classically. Figure 3 gives the log-differences in means for each strain and each vaccine from the two studies plotted against each other. Though there are variations between them there is a clear positive relationship between the corresponding treatment effects in the two experiments. The variability is large even if the trend seems mainly linear. That indicates there may be other unaccounted sources of variability. The variability represented here will probably be reflected in the translation from the MIMIC study to the clinical trial when we build a prior for the CT using the posterior from the MIMIC. We may need a more complicated transformation or the use of other variables not yet considered.

4. Statistical Methods

We use Bayesian structure to build models for the posterior from the MIMIC and the prior for the clinical trial based on above observations. The following are the details of Bayesian two-steps model. The model for MIMIC study is given by the following equations which describe a mixed model with a random factor for donors for each strain.

$$
\begin{aligned}
&Y_{Mi} \sim \log\text{-}N(\mu_i, Var_M);\\
&\mu_i = X_M \beta_M + B_i;\\
&X_M = (\mathbf{1}, X^M_{GroupI}, X^M_{GroupII}, X^M_{GroupIII});\\
&\beta_M = (\beta^M_0, \beta^M_{GroupI}, \beta^M_{GroupII}, \beta^M_{GroupIII});\\
&B_i \sim N(0, Var_B);
\end{aligned}
$$

The immune responses Y_{Mi} in MIMIC for each subject follows a log-Normal distribution with mean μ_i and variance Var_M where $i = 1,...,25$ indicating block numbers. μ_i is a linear function of the fixed effect of the group indicator and a random effect of a block factor B_i. Baseline immune responses of MIMIC is not embeded as a factor of the mean responses as the the block effect has seperated out the subject-specific impacts so the interested treatment effects β_M is not influenced by baseline in a RCBD.

All of the parameters have conjugate priors (Raiffa & Schlaifer, 1961) for the MIMIC study. The beta coefficients have a normal distribution with mean 0 and sufficiently large variances. The two variances follow inverse gamma distributions.

The Bayesian model for the CT for each strain is given by the following:

$$
\begin{aligned}
&Y_{Cj} \sim \log\text{–}N(\mu_C, Var_C);\\
&\mu_C = X_C\beta_C;\\
&X_C = (\mathbf{1}, X_{GroupI}, X_{GroupII}, X_{GroupIII}, X_{Baseline});\\
&\beta_C = (\beta_{Intercept}, \beta_{GroupI}, \beta_{GroupII}, \beta_{GroupIII}, \beta_{Baseline});
\end{aligned}
$$

The immune responses Y_{Cj} follow log-Normal distribution with mean μ_C and variance Var_C where the $j = 1,\ldots,$ # of subjects in total in CT. The mean immune responses are a linear function with variables of group indicators and baseline immune responses. Similarly with MIMIC model, the priors for each of the parameters β_C follows Normal distribution whereas Var_C follows inverse Gamma distribution.

We assume a linear link for the treatment effects in the MIMIC trail and the CT for all strains for all vaccine groups. The posterior distribution from the MIMIC will be used as a prior with the linear relationship define in the following:

$$
\begin{aligned}
&\beta_X^{CT} \sim N(b_0 + b_1\beta_X^M, Var_X^{\varepsilon})\\
&\beta_X^{CT} \in (\beta_{Intercept}, \beta_{GroupI}, \beta_{GroupII}, \beta_{GroupIII});
\end{aligned}
$$

where the coefficients $(b_0, b_1, Var_X^{\varepsilon})$ are estimated intercept, slope and the errors from the linear regression displayed in Figure 3.

5. Analyses Results

If we use the identical link where the vaccine effects are same in MIMIC and CT, we get the results for strain A and strain B summarized in Table 3. It displays the results from Bayesian two-steps model using the identical link between the MIMIC posterior and the CT prior. Additional results are also provided by the typical Bayesian method applied on CT where the MIMIC is not used as prior information. Instead, non-informative priors (Jeffreys, H., 1946) are used as priors in the typical Bayesian model. Moreover, a generalized linear model is applied as well to the dataset for CT. We can see that the use of the Bayesian techniques does not improve the results over the classical analyses of the clinical trial given in the table. In fact, it seems not as good as the typical Bayesian method or the GLM. The mean estimates of treatment effects are far away from the typical Bayesian method and the GLM. The variances are also wider than the GLM results. Results for strain C and D are quite similar which show poor estimates from Bayesian two-steps modelling.

Table 4 gives the results from Bayesian two-steps analyses by using the linear link between the posteriors of MIMIC and the priors from the CT. The typical Bayesian modeling and GLM for CT are also provided in the same table. Compared to the

Bayesian two-steps analyses by using the identity link, the precision of estimates are much narrower of the mean estimates. It is clear that the linear link works better than the identical link. However, the results does not show a clear improvement compared to typical Bayesian modeling for CT using non-informative priors. Moreover, the results from Bayesian modeling shows bias compared to the classical analyses using GLM with even wider confidence intervals.

Table 3: Analyses Results of CT from Bayesian Two-steps Modeling Using Identical Link for Treatment Effects and Variance between MIMIC and CT (Strain A and B)

	Bayesian Two-Steps using Identical Link				Bayesian using Non-informative Prior				GLM			
Strain A	**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% CI**	
Intercept	0.413	0.03	0.357	0.472	0.413	0.025	0.367	0.464	0.438	0.025	0.39	0.487
Group I	-0.071	0.021	-0.111	-0.029	-0.07	0.018	-0.108	-0.037	-0.062	0.016	-0.093	-0.031
Group II	-0.077	0.021	-0.118	-0.035	-0.075	0.018	-0.107	-0.035	-0.063	0.016	-0.094	-0.033
Group IV	-0.063	0.024	-0.109	-0.016	-0.06	0.021	-0.102	-0.021	-0.054	0.018	-0.089	-0.018
Baseline	0.243	0.012	0.218	0.266	0.242	0.01	0.222	0.262	0.232	0.01	0.213	0.251
Strain B	**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% CI**	
Intercept	0.689	0.043	0.604	0.771	0.695	0.029	0.639	0.756	0.762	0.028	0.708	0.816
Group I	-0.069	0.03	-0.123	-0.007	-0.072	0.021	-0.113	-0.031	-0.069	0.019	-0.105	-0.033
Group II	-0.066	0.03	-0.125	-0.007	-0.069	0.022	-0.107	-0.024	-0.068	0.019	-0.104	-0.031
Group IV	-0.056	0.033	-0.124	0.005	-0.058	0.024	-0.102	-0.013	-0.057	0.022	-0.1	-0.015
Baseline	0.215	0.019	0.176	0.251	0.213	0.013	0.188	0.238	0.186	0.012	0.164	0.209

Table 4: Analyses Results of CT from Bayesian Two-steps Modeling Using Linear Link for Treatment Effects and Variance between MIMIC and CT (Strain A and B)

	Bayesian Two-Steps using Linear Link				Bayesian using Non-informative Prior				GLM			
Strain A	**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% CI**	
Intercept	0.414	0.028	0.361	0.468	0.413	0.025	0.367	0.464	0.438	0.025	0.39	0.487
Group I	-0.074	0.017	-0.106	-0.043	-0.07	0.018	-0.108	-0.037	-0.062	0.016	-0.093	-0.031
Group II	-0.076	0.017	-0.109	-0.045	-0.075	0.018	-0.107	-0.035	-0.063	0.016	-0.094	-0.033
Group IV	-0.064	0.019	-0.1	-0.028	-0.06	0.021	-0.102	-0.021	-0.054	0.018	-0.089	-0.018
Baseline	0.242	0.013	0.218	0.267	0.242	0.01	0.222	0.262	0.232	0.01	0.213	0.251
Strain B	**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% HPD Interval**		**Mean**	**Std**	**95% CI**	
Intercept	0.689	0.037	0.621	0.767	0.695	0.029	0.639	0.756	0.762	0.028	0.708	0.816
Group I	-0.067	0.022	-0.11	-0.025	-0.072	0.021	-0.113	-0.031	-0.069	0.019	-0.105	-0.033
Group II	-0.06	0.022	-0.108	-0.019	-0.069	0.022	-0.107	-0.024	-0.068	0.019	-0.104	-0.031
Group IV	-0.055	0.025	-0.105	-0.008	-0.058	0.024	-0.102	-0.013	-0.057	0.022	-0.1	-0.015
Baseline	0.214	0.018	0.175	0.246	0.213	0.013	0.188	0.238	0.186	0.012	0.164	0.209

6. Conclusions

This research shows an initial step of understanding MIMIC system and investigating the usage on the following CT. We can easily summarize that the Bayesian two-steps modeling using linear link provide better estimates compared to the same model using the identical link. However, there is no improvement on the presicion of the estimates compared to the typical Bayesian model without using MIMIC study as the prior information or a generalized linear model. It seems that MIMIC study could provide a better estimates at some point of view. However, the relation between MIMIC and CT is not completely discovered to reduce the bias or improve the precision from the estimates.

Obviously, the linear link is not an accurate estimate of the relationship among treatment effects. From statistical point of view, more datasets would be required to get a more accurate estimation of the linear link. The links other than linear relation are also possible to better describe the relation. Moreover, links through treatment effects could already be biased no matter which type of link is considered since the fundenmantal relationship between MIMIC and CT could be better described through other characteristics. Futher investigations on the fundenmantal structure between MIMIC and CT are nessesary which could be very different from what we have obtained so far.

Some major issues are still unclear during the reseach. It is known that the vaccine administration is already different and in addition two different assays are used to test the laboratory immune responses from the two studies. However, the quantitative differences are unclear and not precisely evaluated. It is the truth that the blood samples used for CT is the complete blood sample from human beings whereas the blood samples used for MIMIC are filtered which only include anti-virus related cells (e.g. T-cells, B-cells, etc.). But no laboratory tests are performed to show the partial blood samples in vitro will have exactly the same immune responses with the complete blood samples in human beings. All above could lead to potential bias of the treatment effects which are not able to be solved only by statistical modelings.

Nevertherless, the benefits from MIMIC study cannot be ignored which has no harm to human beings and immune responses in humans could be evaluated in a early phase without the risk of any safety concern. The new technology is still under development and more studies will be conducted. With the further investigations on more datasets from MIMIC, the design and analyses of clinical trials could be greatly improved in future.

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