Curation and Analysis of Host Gene Responses induced by Live Attenuated and Trivalent Inactivated Influenza Vaccines

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ABSTRACT

A vaccine is a biological preparation that is administered to produce or artificially increase immunity to disease. Vaccines aim to stimulate immune factors, bodily compounds which help trigger protective responses to pathogens. A vaccine-induced host immune factor (Vimmutor), therefore, is an immune factor that is either regulated by the administration of a vaccine (Figure 1a) or modulates and directs the vaccine to an appropriate immune response (Figure 1b). The study of Vimmutors is essential to predicting immune responses to vaccines and identifying these responses at quicker rate than antibody titers. One goal of this project is to expand the current Vimmutor Database (VimmutorDB), a program under the Vaccine Investigation and Online Information Network system (VIOLIN) (Reference 1), which includes vaccine information from peer reviewed articles in PubMed. This project focused on the Vimmutors associated with influenza vaccines. Specifically, 173 human genes induced by FluMist, a live attenuated influenza vaccine (LAIV) and Fluarix, a trivalent inactivated vaccine (TIV) have been compiled, both of which correlate with protection. Results of bioinformatics analyses have shown significant differences between LAIV and TIV Vimmutors in biological processes and molecular functions, such as their roles in antigen versus cell-receptor binding. Conversely, important similarities between the LAIV and TIV have also been shown, including correlations with the genes interleukin-6 signal transducer (IL-6ST), interleukin-6 receptor (IL-6R), and tumor necrosis factors (TNF). These findings provide useful information about early indications of vaccine-induced host gene responses, and can be used to further the progress of vaccine production and efficacy.

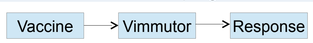

Figure 1a: Downstream vaccine response

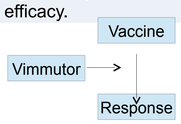

Figure 1b: Upstream vaccine response

OBJECTIVES

1. Gather gene response data that can be used to provide insight about protection more quickly than antibody titers
2. Provide evidence for LAIV gene correlate for standard measure of successful vaccination
3. Expand VimmutorDB to allow for large-scale analysis of influenza vaccine-induced host gene responses.

METHODS

Vimmutors were collected from five peer-reviewed articles from PubMed, including Nakaya et al, 2011 (Reference 2), and entered into VimmutorDB along with their host gene response type and a description of the response (Figure 3). The basic gene data was first compiled to observe patterns. Entrez Gene IDs were then compiled and the gene lists for both LAIV and TIV were uploaded to DAVID Bioinformatics Resources 6.7. In DAVID, genes were compared with terms from three parts of the Gene Ontology (GO): biological processes, cellular components, and molecular functions. Genes from both LAIV and TIV vaccines that correlated with the most significant terms from each ontology were recorded and then compared. These procedures are represented graphically in Figure 2 below.

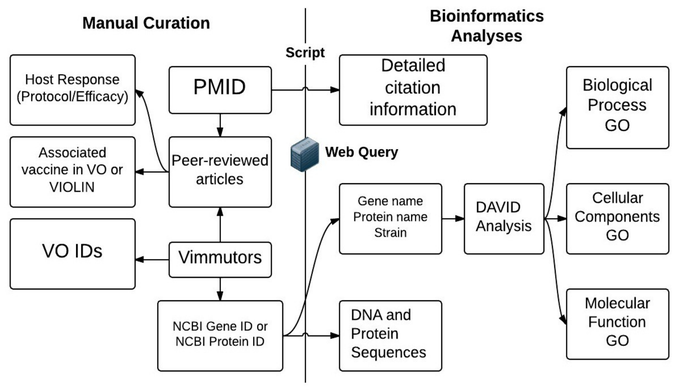

Figure 2: Workflow Diagram for Influenza Vimmutor collection and analysis. Vimmutors were collected from peer-reviewed PubMed articles, and an internally developed script used an input sequence from a NCBI Database (e.g., NCBI Entrez Gene database) to automatically retrieve different types of information. Note: The results from the Cellular Components part of GO were not included in the 'Results' sectiion because they were not relevant to the focus of this poster.

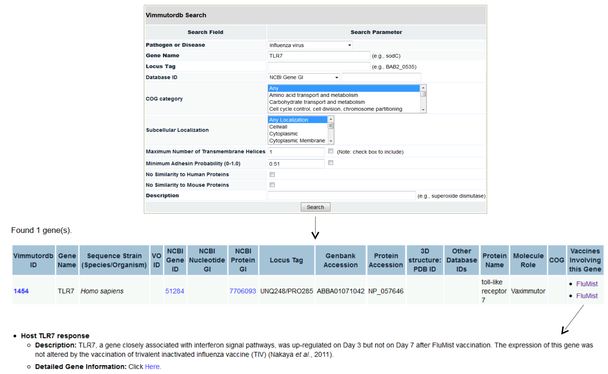

Figure 3: Adding Vimmutor to VimmutorDB. Host genes are accessed using the NCBI Gene database. Gene information is then queried and added under the host gene response in VimmutorDB.

RESULTS

The initial categorization of Vimmutors shows up and down regulation in various immune cell types or positive and negative correlation with antibody titers (for TIV). As evidenced by Figure 4, the vast majority of genes were up-regulated in cells or positively correlated with titers. The number of genes associated with various biological processes is also shown.

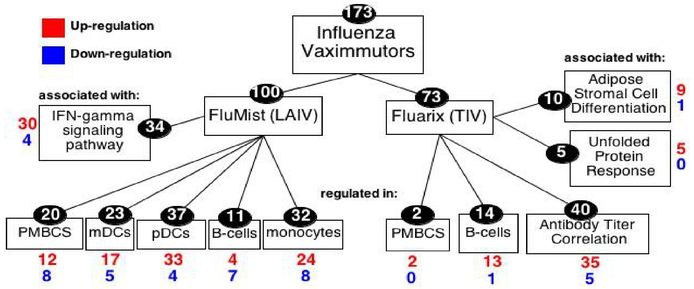

Figure 4: Categorization of influenza Vimmutors. The various cell types that Vimmutors were regulated in included peripheral blood mononuclear cells (PMBCs), myeloid dendritic cells (mDCs), plasmacytoid dendritic cells (pDCs), B-cells, and monocytes. Black numbers indicate totals, while red and blue numbers indicate up and down regulation, respectively.

Results from the Biological Processes and Molecular Functions part of the Gene Ontology are shown in Table 1a and 2a, respectively. The most significant genes and terms along with their adjusted p-value are included.

FLUARIX ASSOCIATED BIOLOGICAL PROCESSES

(#) NCBI ID	NAME	(#) Gene Ontology terms	BENJAMINI
(6) 3569	IL6	(8) positive regulation of cytokine production	1.90E-004
(6) 7124	TNF	(8) positive regulation of phosphate metabolic process	1.90E-004
(4) 64127	NOD2	(7) positive regulation of lymphocyte activation	8.00E-004
(4) 5663	PSEN1	(11) protein kinase cascade	9.60E-004
(4) 857	CAV1	(9) positive regulation of transport	9.70E-004
(4) 3572	IL6ST	(10) negative regulation of apoptosis	2.40E-003
		(4) positive regulation of interleukin-6 production	3.50E-003
		(8) blood vessel development	5.20E-003

FLUMIST ASSOCIATED BIOLOGICAL PROCESSES

(#) NCBI ID	NAME	(#) Gene Ontology terms	BENJAMINI
(4) 11213	IRAK3	(14) positive regulation of cytokine production	4.60E-012
(4) 3572	IL6ST	(19) regulation of protein kinase cascade	6.10E-012
(7) 3569	IL6	(13) positive regulation of defense response	6.40E-012
(5) 3329	HSPD1	(13) response to lipopolysaccharide	1.10E-011
(5) 7124	TNF	(13) positive regulation of lymphocyte activation	1.30E-010
(4) 3717	JAK2	(11) positive regulation of DNA binding	1.70E-009
(4) 3654	IRAK1	(9) positive regulation of interleukin-6 production	3.20E-009
(4) 3570	IL6R	(17) negative regulation of apoptosis	5.30E-008

Table 1a: Most significant terms from BP_GO_5 and their gene correlates. Gene lists analyzed using the most specific filter (level 5). Entries in black are those which are unique to the vaccine, while entries in blue are shared between the two vaccines. The number next to 'NCBI ID' and 'Gene Ontology terms' is the number of genes associated with that entry. Note: the genes and ontology terms in each graph do not correspond to one another. Terms are organized by lowest Benjamini value (adjusted p-value).

FLUARIX ASSOCIATED MOLECULAR FUNCTIONS

(#) NCBI ID	NAME	(#) Gene Ontology terms	BENJAMINI
(2) 3460	IFNGR2	(6) cytokine receptor activity	6.10E-004
(2) 3588	IL10RB	(7) cytokine binding	8.20E-004
(2) 3570	IL6R	(5) antigen binding	7.40E-003
(2) 3572	IL6ST		
(2) 8809	IL18R1		

FLUMIST ASSOCIATED MOLECULAR FUNCTIONS

(#) NCBI ID	NAME	(#) Gene Ontology terms	BENJAMINI
(4) 7048	TGFBR2	(7) cytokine receptor activity	1.80E-004
(4) 3717	JAK2	(17) protein kinase activity	1.80E-004
(4) 6885	MAP3K7	(4) interleukin-6 receptor binding	3.00E-004
(4) 3654	IRAK1	(24) adenyl ribonucleotide binding	4.20E-003
(4) 2322	FLT3	(5) cytokine binding	6.10E-003
(4) 3551	IKBKB	(4) tumor necrosis factor receptor binding	5.30E-003
(4) 3570	IL6R		
(4) 3572	IL6ST		

Table 1b: Most significant terms from MF_GO_5 and their gene correlates. Gene lists analyzed using the most specific filter (level 5). Entries in black are those which are unique to the vaccine, while entries in blue are shared between the two vaccines. The number next to 'NCBI ID' and 'Gene Ontology terms' is the number of genes associated with that entry. Note: the genes and ontology terms in each graph do not correspond to one another. Terms are organized by lowest Benjamini value (adjusted p-value).

DISCUSSION

Fluarix Vimmutors	Both	FluMist Vimmutors
· Phosphate metabolism · Transport · Blood vessel development · Antigen binding · NOD2 · PSEN1 · CAV1 · IFNGR2 · IL10RB	· Cytokine production · Lymphocyte activity · Apoptosis · IL-6 production · Cytokine binding · IL6R · IL6ST · TNF	· Defense response · DNA binding · Protein kinase activity · Interleukin-6 binding · Adenyl ribonucleotide binding · TNF receptor binding · IRAK 1, 3 · HSPD1 · JAK2 · TGFBR2 · MAP3K7

Some key differences exist between FluMist and Fluarix Vimmutors. Fluarix-associated Vimmutors show associations with antigen binding, thus providing a reason for the large amount of Fluarix genes that were positively correlated with antibody titers. FluMist-associated Vimmutors were regulated in certain immune cell types, and thus showed associations with cell-signaling processes, such as interleukin-6 and TNF receptor binding. FluMist Vimmutors were also associated with protein kinase activity, another important mechanism of cell communication.

Significant similarities also existed between the two sets of Vimmutors. Two subunits of the interleukin-6 protein complex were correlated with both vaccines in high amounts. The first subunit, IL6ST, encodes a protein that is a signal transducer in the cytokine receptor complex. Studies have shown that this gene plays a critical role in apoptosis. The second, IL6R, codes for the interleukin-6 receptor protein, which regulates cell growth and differentiation. TNF, or tumor necrosis factor was also a common Vimmutor between the two vaccines. TNF refers to a group of cytokines which can cause apoptosis and are implicated in tumor regression.

CONCLUSION

The study of Vimmutors is critical to the prediction and development of novel vaccines, and especially important in the realm of influenza. Analyzing vaccine induced host gene responses allows researchers to gain early insight into the efficacy of a vaccine. The compilation of Vimmutors in a database such as VimmutorDB is essential because it allows large scale analysis of host genes and their interactions with pathogens.

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