# **EDA Report**

The Experimental Design Assistant (https://eda.nc3rs.org.uk) is an online tool which guides researchers through the design and analysis of in vivo experiments. Information is provided by the researcher to build an EDA diagram – see Annex. Depending on the information inputted specific prompts are triggered by the EDA which provide tailored advice and feedback on the experimental plan.

This report summarises the information provided by the researcher and the feedback from the EDA.

## **Section 1: Summary**

Title of EDA diagram	test3
Date report generated	25/07/2021

### Section 2: Information provided by the researcher

#### 1: Objectives

Null hypothesis	GATA3-siRNA inhibits the expression of GATA3 mRNA in Th2, TSLP-siRNA inhibits the expression of TSLP mRNA in epithelial cells and GATA3-siRNA and TSLP-siRNA co-effect can inhibits the expression of both GATA3 and TSLP mRNA in Th2 and epithelial cells
Alternative hypothesis	GATA3-siRNA cannot inhibits the expression of GATA3 mRNA in Th2, TSLP-siRNA cannot inhibits the expression of TSLP mRNA in epithelial cells and GATA3-siRNA and TSLP-siRNA co-effect can't inhibits the expression of both GATA3 and TSLP mRNA in Th2 and epithelial cells
Effect of interest	the relative expreesion of mRNA
Effect size	50% reduction in relative expression
Justification for effect size	the article Fu Z, Zhang X, Zhou X, Ur-Rehman U, Yu M, Liang H, Guo H, Guo X, Kong Y, Su Y, Ye Y, Hu X, Cheng W, Wu J, Wang Y, Gu Y, Lu SF, Wu D, Zen K, Li J, Yan C, Zhang CY, Chen X. In vivo self-assembled small RNAs as a new generation of RNAi therapeutics. Cell Res. 2021 Jun;31(6):631-648. doi: 10.1038/s41422-021-00491-z. Epub 2021 Mar 29. PMID: 33782530; PMCID: PMC8169669.



#### 2: Groups and sample size

Total number of animals in the experiment	21
Groups included in the primary analysis	7 groups:
• Group 1	role=test; n=3
• Group 2	role=test; n=3
• Group 3	role=test; n=3
• Group 4	role=test; n=3
• Group 5	role=test; n=3
• Group 6	role=test; n=3
• Group 7	role=control/comparator; n=3
Justification for sample size	use EDA power calculation

#### 3: Randomisation and blinding

Experimental unit	animal

There is one step in this experiment where experimental units are allocated to groups:

Allocation 1

Randomisation strategy	randomisation within blocks
Randomisation procedure	EDA spreadsheet
Allocation concealment	treatments coded for individual animals

There are six steps in this experiment where measurements are taken:

• Get Th2 and epithelial cells in tissue

Blinding during result assessment	animals individually coded

• look at the ratio of the activated Th2 cells and ILC-2 cells using FCM

Blinding during result assessment	animals individually coded
Difficulty during result assessment	i animais individually coded

· Measurement 1:culture

Blinding during result assessment	animals individually coded

• Measurement : get bronchoalveolar lavage fluid

Blinding during result assessment	animals individually coded

• measurement: Airway response value

Blinding during result assessment	animals individually coded
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Tissue sectioning and staining

Blinding during result assessment	measurements carried out by someone else
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There is one analysis in this experiment:

• Analysis 1? Physiological and biochemical indexes of type 2 inflammatory response

Blinding during analysis of the data	groups coded
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#### 4: Analysis

Details of the primary analysis (Analysis 1?Physiological and biochemical indexes of type 2 inflammatory response)

Statistical analysis method	unpaired t-test
Factor of interest	Independent variable of interest 1?Injectant, continuous, with 7 levels (Liposomes without plasmids, dexamethasone, gata3-si and tslp-si mcDNA, gata3-si minicircleDNA, minicircleDNA without siDNA fragment, pbs, tslp-si minicircleDNA)
Blocking factor	Nuisance variable : misoperation, categorical, with 1 level (1 day)
Covariate	NONE

#### Outcome measures

Outcome measures in the primary analysis	Outcome measure 1:Airway response value, treated as continuous; the different ratio of the activated Th2 cells and ILC-2 cells, treated as continuous; Outcome measure 3:Expression of inflammatory response related cytokines, treated as continuous; Outcome measure 5:TSLP mRNA and protein expressions in airway epithelial cell, treated as continuous; Outcome measure 4:tissue slice observe the inflammation, treated as categorical; Outcome measure 6:GATA3 mRNA and protein expressions in Th2 cell, treated as continuous
Other outcome measures	NONE

# 5: Characteristics of animals in this experiment

Animal:BALB/c mouse

Species	mouse
Strain	INFORMATION NOT PROVIDED
Sex	female
Age	mean=7, range=6-8, unit=week
Weight	INFORMATION NOT PROVIDED

#### Section 3: Summary of the feedback provided by the EDA

Critique (Table 6) and advice (Table 7) from the EDA is dependent on the quality, including accuracy and completeness, of the information inputted by the researcher. Where the researcher has not addressed issues detected by the EDA, it is important to consider whether this undermines the design of the study.

#### 6: Critique

Total number of issues	0
Issues related to the diagram structure, which might compromise the accuracy of this report	0
Issues related to internal consistency	0
Issues related to missing information	0
Issues suggesting improvements to the design	0

#### 7: Advice for the primary analysis

Suggestion for a method of analysis appropriate for the design	Complex design - categorical outcome measure
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