





#### Can Mice Legated Ilea Loop (MLIL) Model Replace Rabbit Ilea Loop (RIL) for studying Bacterial Diarrhea?



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- Shigellosis is a diarrheal disease and still a big problem in developing countries (Agtini et all. 2007, Herwana et all 2010)
- The best prevention is by using a vaccine but so far there has not be any suitable vaccine (Kweon 2008, Levine 2007)
- Molecule adhesion of bacteria can serve as a basic component of the Pertussis vaccine (Poolman and Hallander 2007).
- S. dysenteriae sub-unit pili protein which has MW 7,9 and 48,9 kDa molecule is adhesive (IJMR major correction)

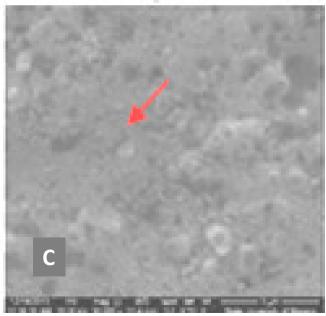
### The aims of the study

"are to clarify that *S. dysenteriae* sub-unit pili proteins which have MW 7,9 and 48,9 kDa as an adhesive molecule can show diarrhea protection which was confirmed by using MLIL methode"

#### Method

- To know the protectively antigen the study was conducted by post control study design
- The method of study was MLIL.
- Mice were divided into four groups.
  - 1. The first group without immunization was used as a control group.
  - 2. The second group was immunized with *S. dysenteriae* subunit pili protein which has MW 7,9 kDa.
  - 3. Third group was immunized with *S. dysenteriae* sub-unit pili protein which has MW 48,9 kDa.
  - 4. The last group was immunized with combination of *S. dysenteriae* sub-unit pili proteins which has MW 7,9 and 48,9 kDa. The chosen adjuvant immunogen was ISCOM.
- Data were analized using ANOVA and Tuckey test

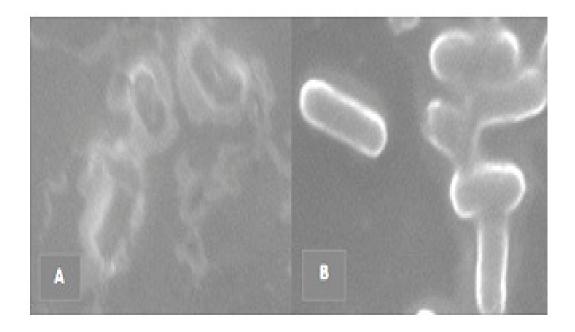


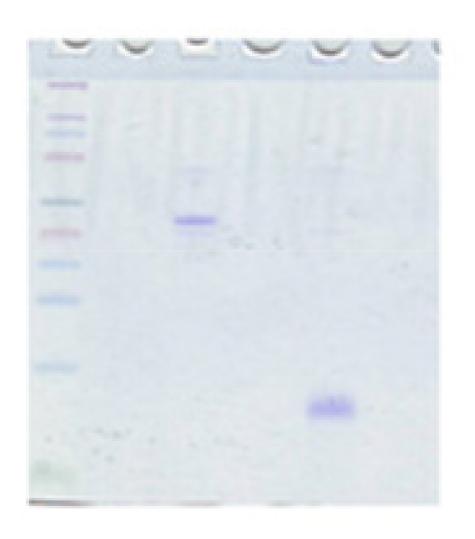


#### **RESULTS**

#### **Bacterial Pillus Cutter**

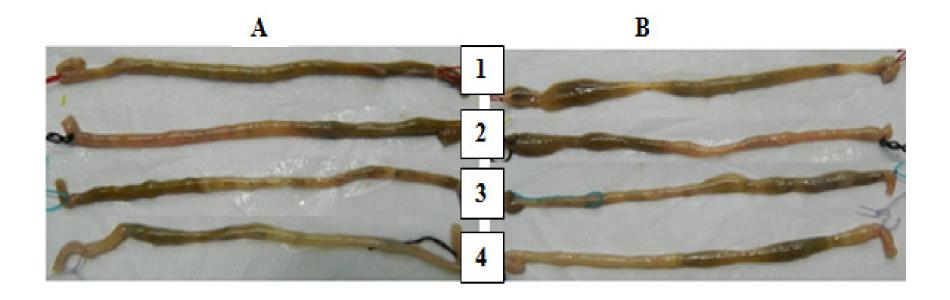
- A Whole cells S. dysenteriae
- **B** S. dysenteriae after cutting
- C Protein pili S. dysenteriae





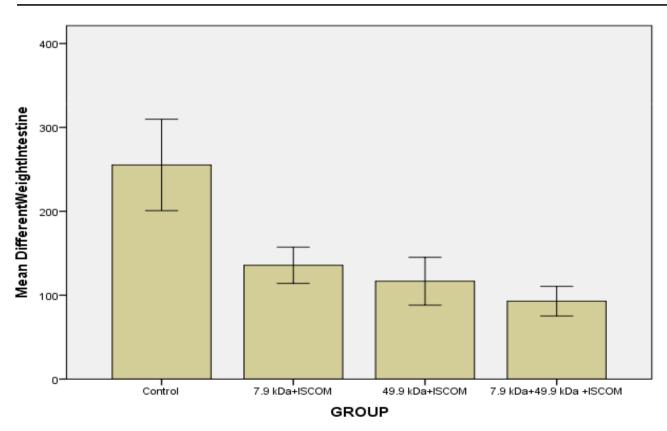
49.8 kDa

7.9 kDa



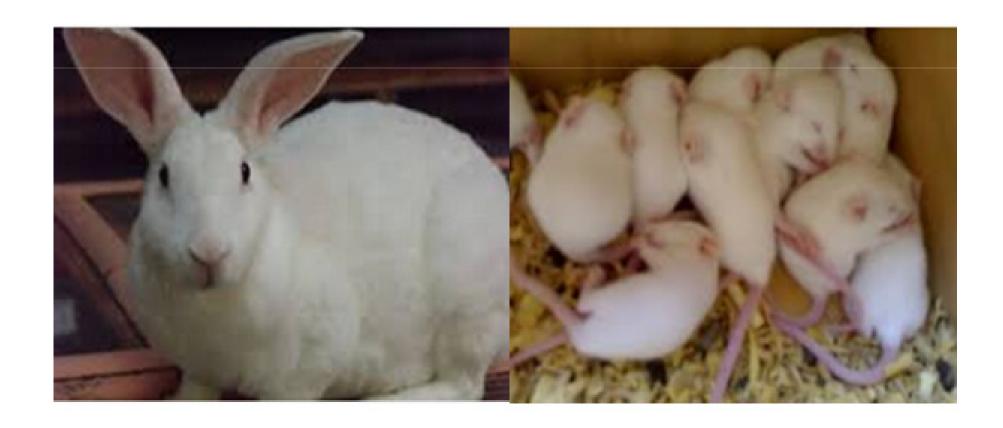
- A. MLIL before exposed with S. dysenteriae
- B. MLIL after exposed with S. dysenteriae
  - 1. MLIL control
  - 2. MLIL immunized with protein 7.9 kDa+ISCOM
  - 3. MLIL immunized with protein 49.9 kDa+ISCOM
  - 4. MLIL combine immunized protein 7.9 kDa with 48.9 kDa
    - + ISCOM

Sample	Mean ±SE	Siq
Control	255.20 ±19.615	a
7.9 kDa+ISCOM	135.60 ±7,782	ь
49.9 kDa+ISCOM	116.60 ±10.297	bc
7.9 kDa+49.9 kDa+ISCOM	$92.80 \pm 6.367$	bcd



Error bars: 95% CI

# Rabbit Ilea Loop (RIL) Model VS Mice Legated Ilea Loop (MLIL) Model

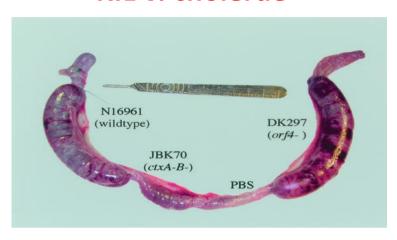


## Comparation RIL Vs MILL

No		RIL	MILL
1	Degrees of Animal	Upper than MILL	Lower than RIL
2	How two kill	* Given the opportunity to live 6 until 8 hours after the abdominal incision closed. * At this time the rabbit may be pain after narcotics effect lost	Animals were killed directly
3	Model	In -Vivo	Ex-Vivo

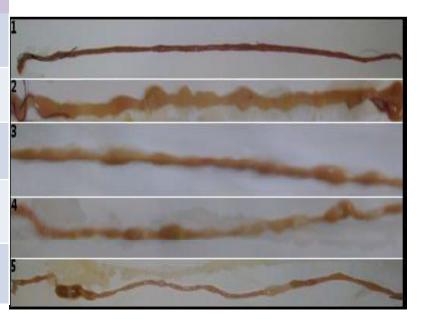
# Compilation of the Result Study Zhang and Dalin 2003

RIL V. cholerae

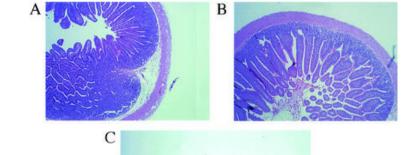


Faisal 2013 MILL V. cholerae

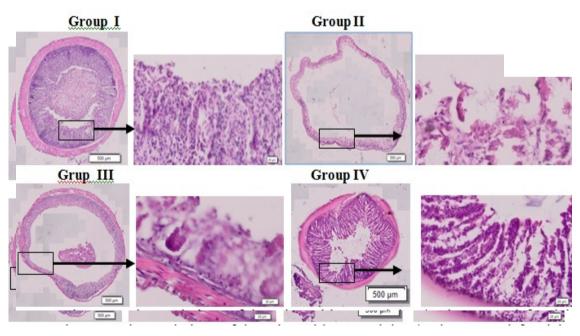
Group of Mice	No
Without immunized not injected by <i>V. cholerae</i> (control negative)	1
Without immunized injected by <i>V. cholerae</i> ( control positive)	2
Immunized with Sumbawa fermented mare's milk injected by <i>V. cholerae</i>	3
Immunized with Sumbawa fermented mare's milk i+ CT B injected by <i>V. cholerae</i>	4
Immunized with Sumbawa fermented mare's milk i+ CT B + mol adhesive pili injected by <i>V. cholerae</i>	5



## Zhang and Dalin 2003 RIL *V. cholerae*



# Setyorini MILL *S. dysenteriae* 2013



#### Conclusion

- Moving solution prevention from intestine to lumen was found in the second, third, and forth groups
- The MLIL test can be used for studying bacterial diarrhea in animal model.
  - Can Mice Legated Ilea Loop (MLIL) Model Replace Rabbit Ilea Loop (RIL) for studyng bacterial Diarrhae???

(Ethical Clearance in Animal Model ???)

# THANK YOU