

Analysis of MBD-Seq data of Korean CLL patients:

As an example of KOBIC Research Support Service

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CLL (Chronic Lymphocytic Leukemia)

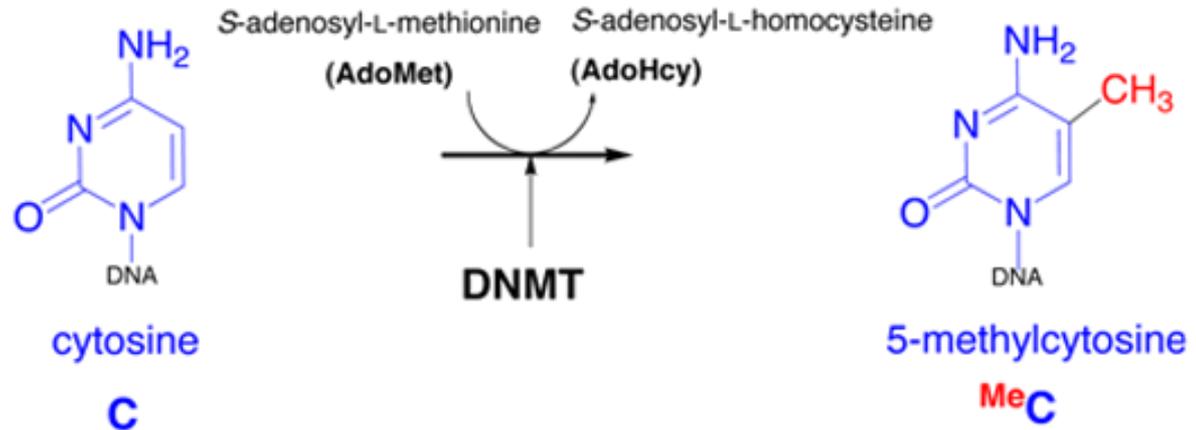


- **CLL is a cancer of the blood;** the bone marrow makes too many lymphocytes \Rightarrow the buildup of premature B-cell lymphocytes, which do not function well \Rightarrow crowd out healthy blood cells
- **Ethnic differences in CLL**

	Incidence rate (per 100,000 person-years)	Median age of initial diagnosis
Western countries	High (3.83)	Late (70)
Korea	Low (0.04)	Early (61)

DNA methylation

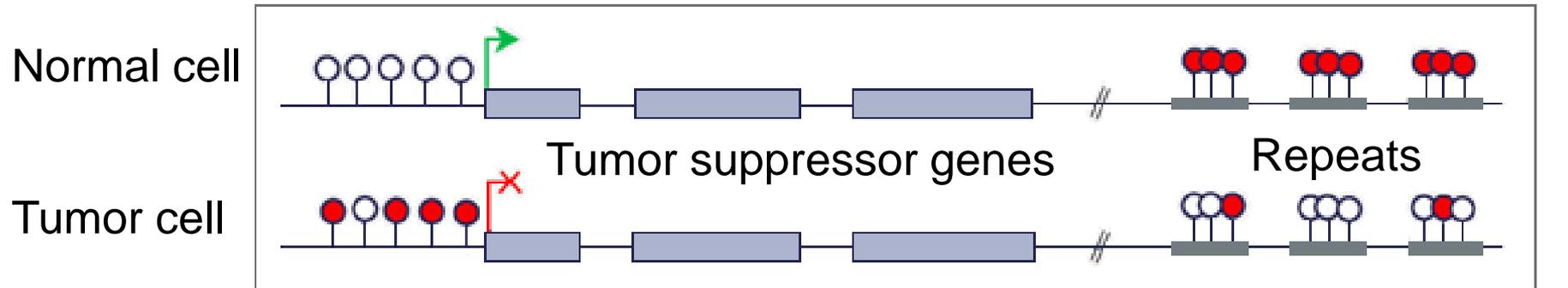
- DNA methylation refers to the addition of a **methyl group** to the cytosine ring.



- DNA methylation controls **gene transcription** by changing **chromatin structure** and, in turn, influences **the affinity and accessibility of TFBSs**.
- **In normal cells**, DNA methylation plays a useful role in **embryonic development**, **X-chromosome inactivation**, **genomic imprinting**, **maintaining genome integrity** (by repressing transcription at repeated sequences), and **tissue-specific gene expression**.

Aberrant DNA methylation in cancer

- A simplified view



↑Hyper-methylation in TSG promoter
⇒ TSG silencing

↓Hypo-methylation in repeats
⇒ genomic instability

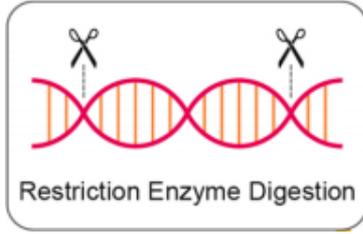
- The association between DNA methylation and gene expression is much more **ambiguous** and **controversial**

- The role of DNA methylation in **non-promoter regions** (inter-genic, intra-genic)
- The role of DNA methylation in **noncoding RNA** (miRNA, lncRNA) **promoters**
- The role of DNA methylation in **pseudogenes**
- ...

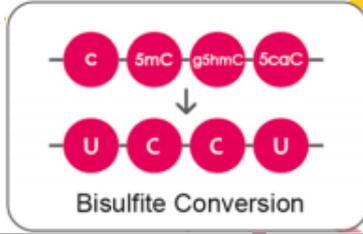
Platforms for genome-wide DNA methylation profiling

Pre-treatment of genomic DNA

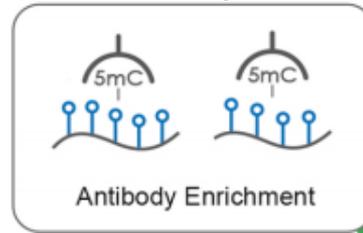
Restriction enzyme digestion:
with GC-rich recognition seq



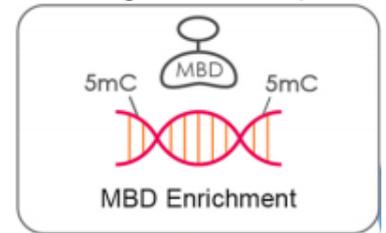
Bisulfite conversion:
 $C \Rightarrow U$ but $5\text{-mC} \Rightarrow C$



MeDIP: immunoprecipitation
with antibody for 5-mC



MBD: capture with Methyl-
CpG-Binding Domain protein



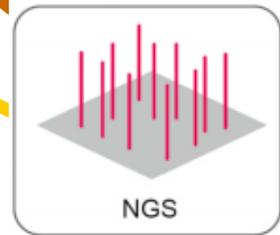
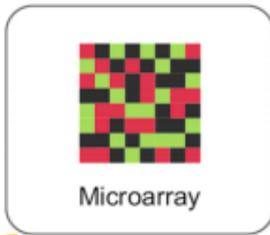
Reduced Representation
Bisulfite Sequencing

MeDIP-Seq

MBD-Seq

Whole Genome
Bisulfite Sequencing

Infinium Human
Methylation chip



Interrogation

Three routes for genome data analysis

- **In-house**
 - Best, but works for **large labs only**
- **Collaboration** with bioinformatics experts
 - Finding a **reliable collaborator** is not easy
 - There is a **luck factor** (collaboration with a prominent bioinformatics group, but the actual work done by a novice graduate student)
 - There is a potential **credit dispute**
 - **(Extreme) view from biology:** You are just support personnel
 - **(Extreme) view from bioinformatics:** I am the one who magically pulled out science from your junky data
- **Outsourcing** to an NGS service company
 - **Customer's perspective:** OK to pay for sequencing. Resistant to pay extra for data analysis. Even with extra payment, not sure about full support until publishable results come out.
 - **Company's perspective:** Going beyond an automated analysis takes time and money. In-depth analysis may not be much of a profitable business model.



We have KOBIC, a friendly neighborhood of Korean biologists

KOBIC Research Support Service

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TOP



목적

KOBIC이 보유한 생명정보 분석 기술의 무상 제공을 통한 국내 BT 연구 활성화.

지원 범위

- 주로 대용량 유전체 데이터, 단백질 구조 등의 bioinformatics 분석 지원을 함.
- 또한 데이터의 NCBI 등록 지원, 생명정보 프로그램 설치 자문 등의 서비스도 제공함.



연구지원 신청시 주의사항

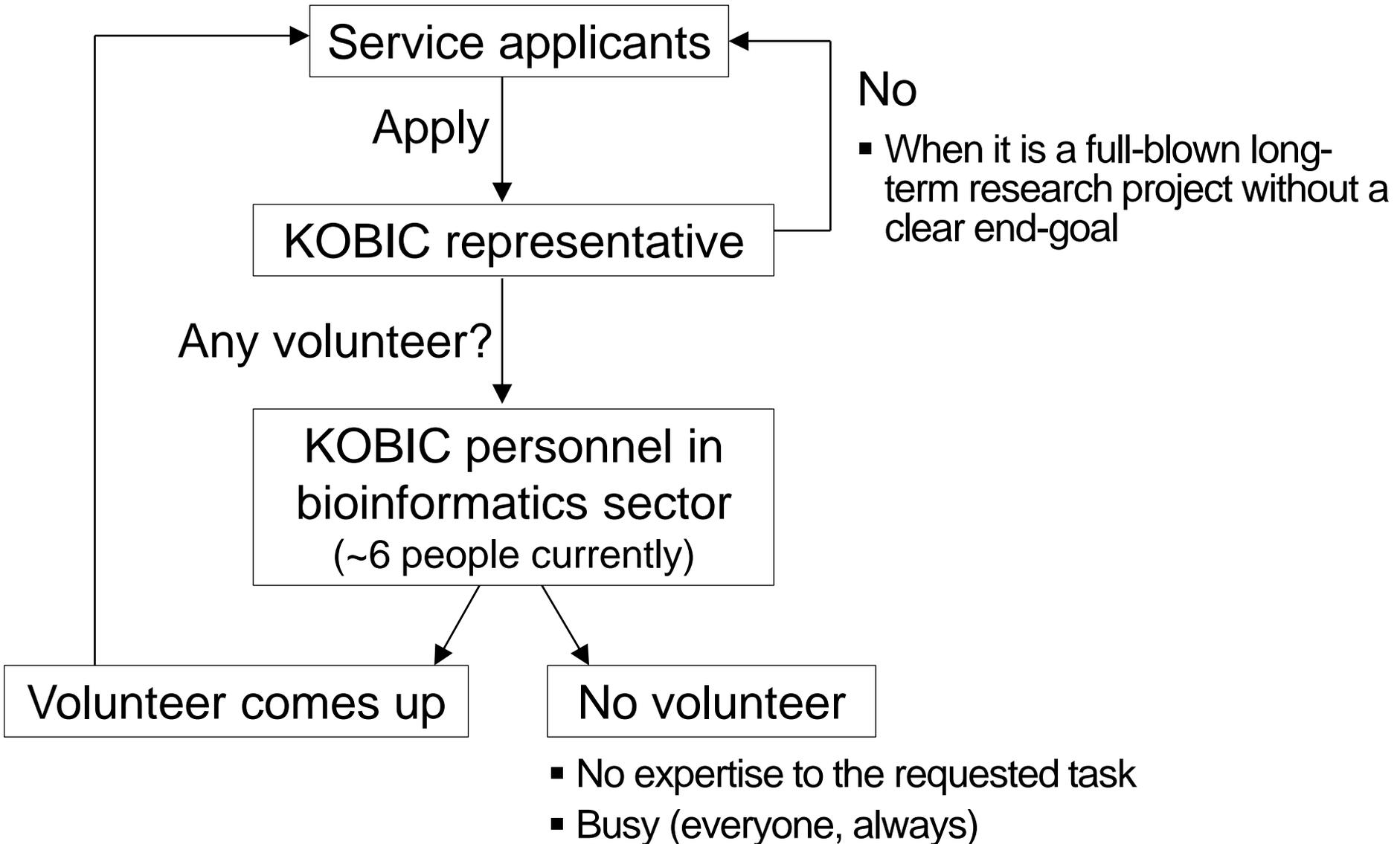
- 신청서 작성 시, 지원을 필요로 하는 내용을 명확하게 기술 요망
- NCBI 서열 등록 지원 신청 시, 생명정보 연구성과물 등록 시스템 에도 등록 요망

다음에 해당하는 경우에는 연구지원이 불가할 수도 있음을 알려드리며 미리 양해를 구합니다.

- 해당 분야에 충분한 전문성을 보유한 담당자가 없는 경우
- 이미 수행 중인 연구 지원 등 업무의 과다로 인하여, 센터 내 인원 활용 여력이 없는 경우
- 정기적이고 전주기적인 연구 지원을(데이터 분석-생물학적 해석-논문 작성까지) 수행 가능한 담당자가 없는 경우

연구지원 문의 전화 : 042-879-8544 메일 : swhwang@kribb.re.kr

Process



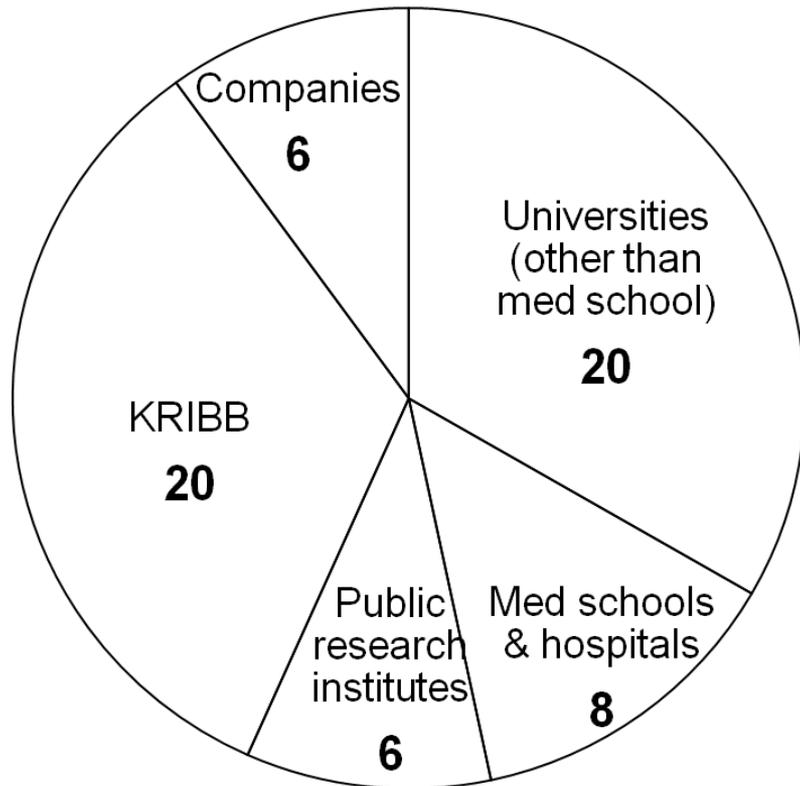
Methods for the MBD-Seq data analysis

Task	Software
Alignment	Bowtie2
Differential methylation analysis	MEDIPS
Annotation of differentially methylated windows	ChIPseeker
GO/Pathway analysis	clusterProfiler
Network analysis	Cytoscape with BisoGenet and ReactomeFIViz apps (for retrieval of protein-protein and protein-DNA interaction data)
Gene-Disease relation analysis	DisGeNET

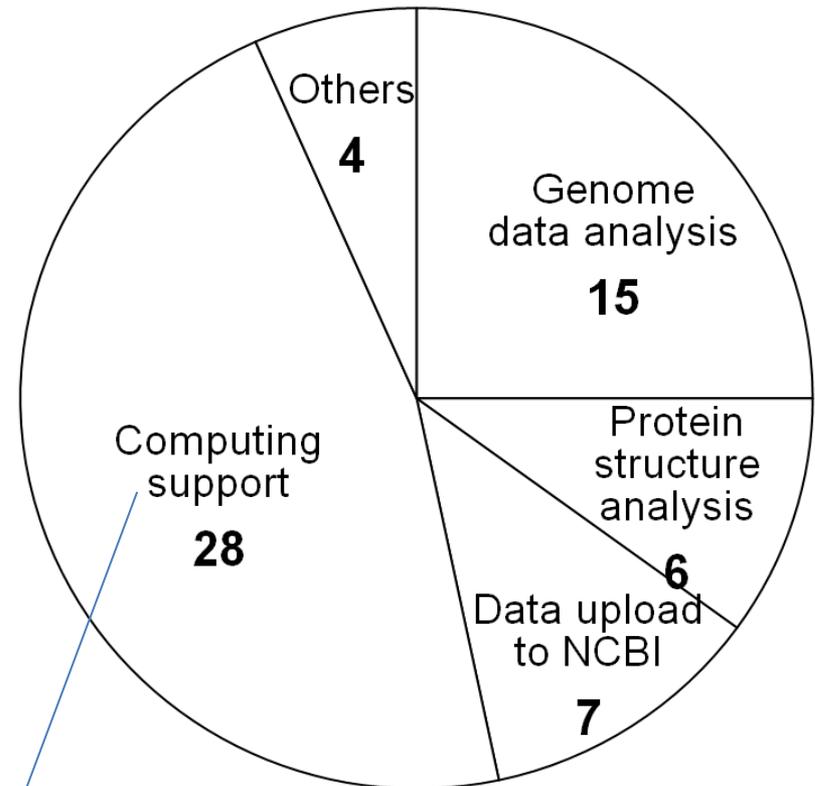
Service requests come in all varieties

Services offered in 2017 (total 60)

By type of applicants



By service topics



Mostly about providing computer servers
(web server, DB server, servers for bioinformatics short courses)

Conclusion and some thoughts

- Need for a **change of perception** for bioinformatics support
 - Perception as just a support ↔ yet sometimes the responsibility to get something useful out of data is put on the shoulders of bioinformaticians
 - Perception that bioinformatics does not cost a lot, or at least customer's emotional resistance (**inkjet printer – ink cartridge scam**)



Cheap



Expensive



- Need for **appropriate price tag** for bioinformatics support
 - Very hard to quote a price: **all projects are different**
- Should KOBIC start to commercialize the service?
 - **Currently:** Free of charge. We don't owe them anything. We may work at our own convenience.
 - **Commercialization:** An age-old internal discussion in KOBIC