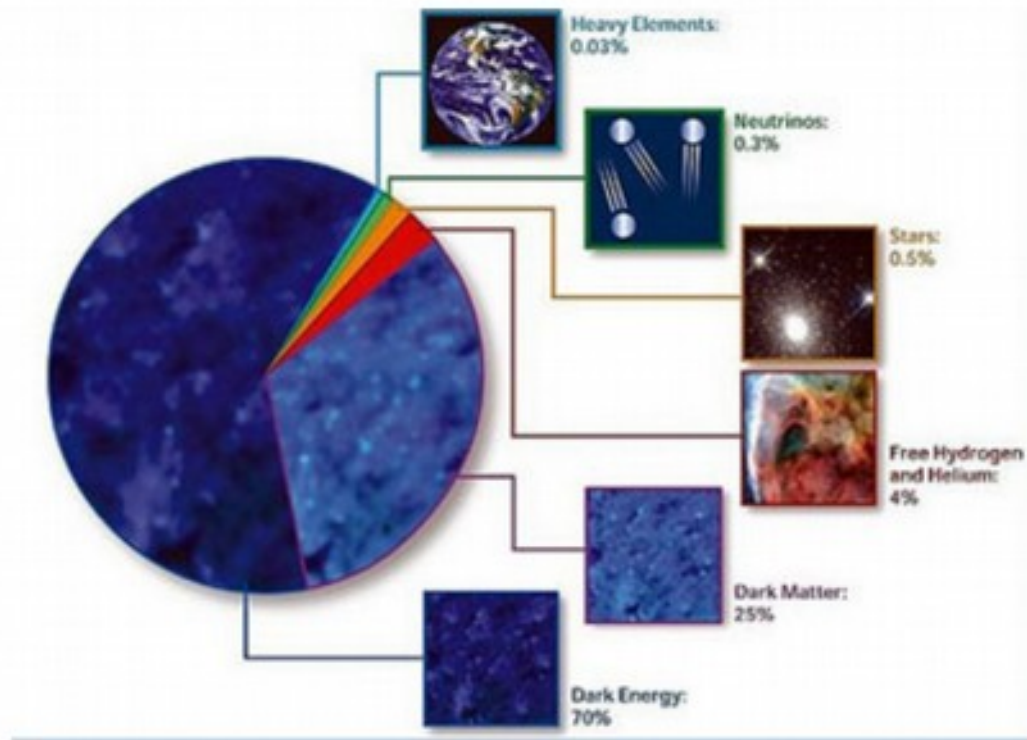


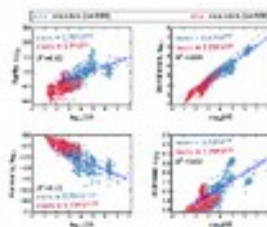
# **Çevresel mikrobiyoloji**



# karanlık madde

elektromanyetik dalgalar ile etkileşime girmeyen, varlığı yalnız diğer maddeler üzerindeki kütle çekimsel etkisi ile belirlenebilen maddeler.

~10<sup>4</sup> kültürü yapılanlar



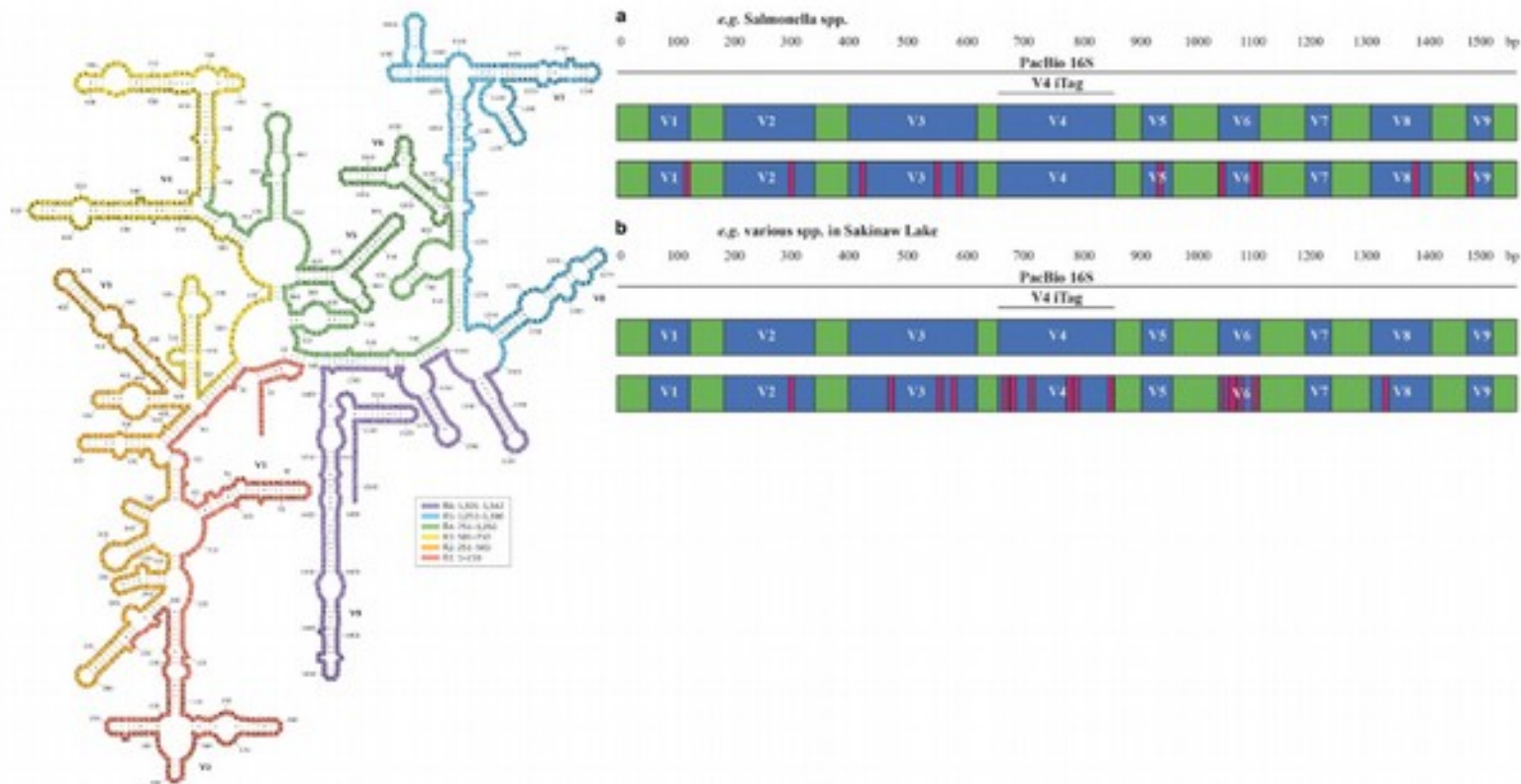
10<sup>5</sup> sekansı bulunan

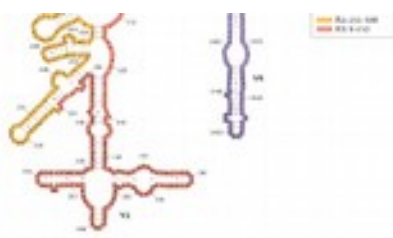
10<sup>7</sup> Earth Microbiome Project  
katalogunda...

10<sup>12</sup> mikrobiyal  
karanlık  
madde

# metagenomik sekanslama stratejileri

## 1- ampikon sekanslama (16S rDNA)

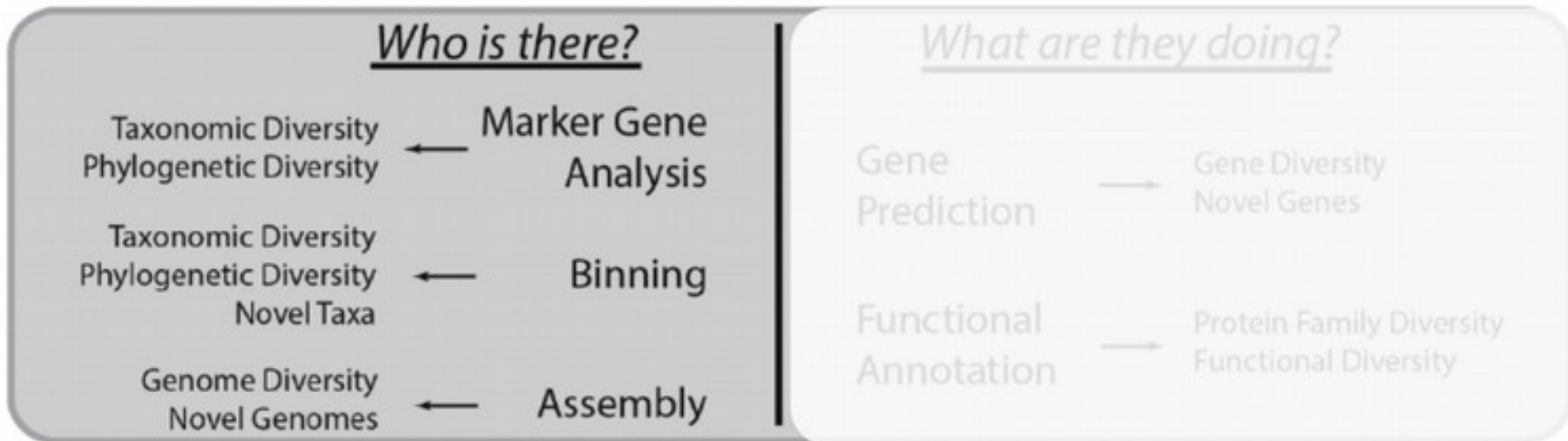




sekanslama hataları  
birleştirme (assebly) sırasında kimera oluşumu  
türler arasında 16 s rDNA lokusu aktarımı  
toksonlar --?--> biyolojik fonksiyon

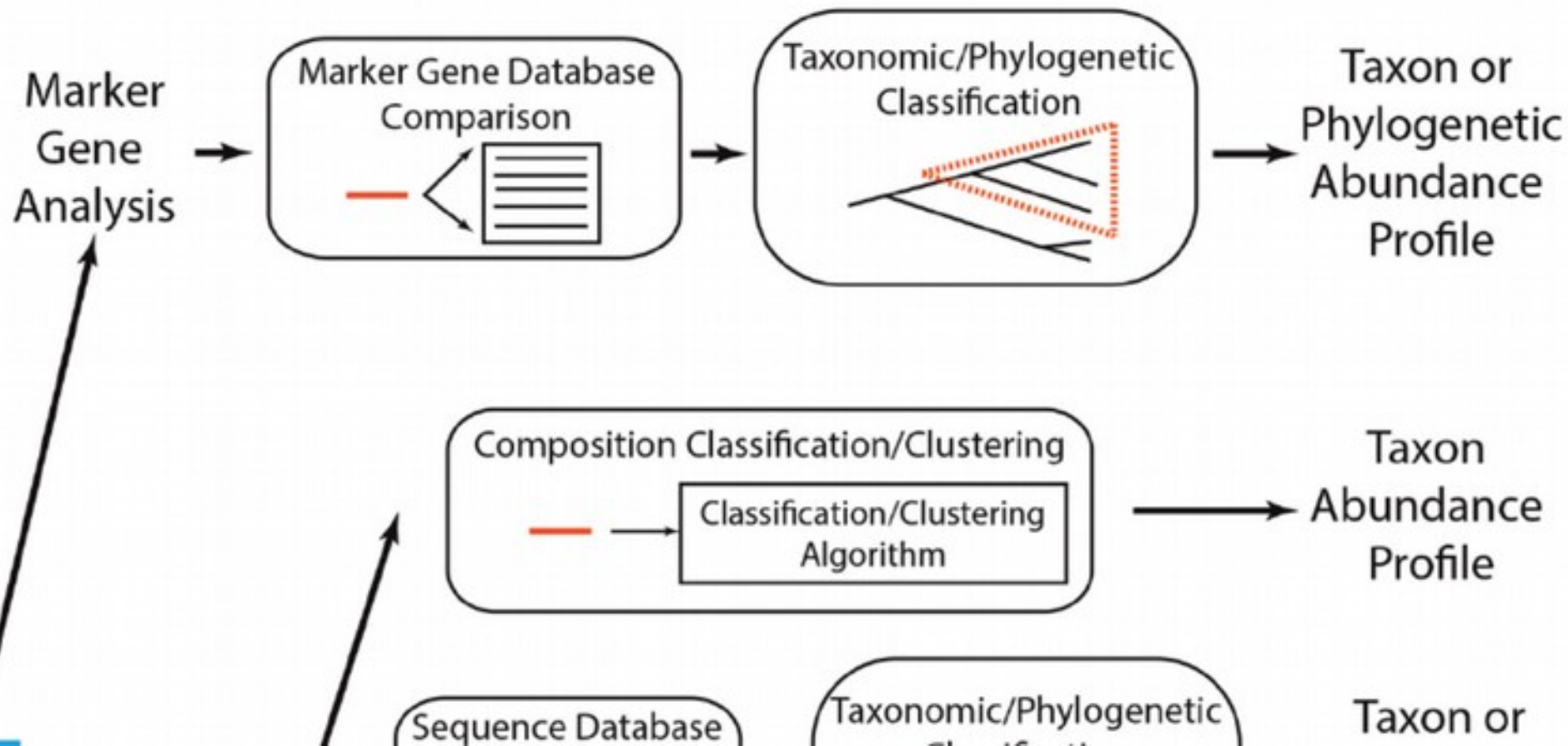
### ***1b- marker gen sekanslama***

faklı lokusların çeşitliliği çözümlene gücü farklı  
yeni / fazla iraksanmış türlerde hangi gen?



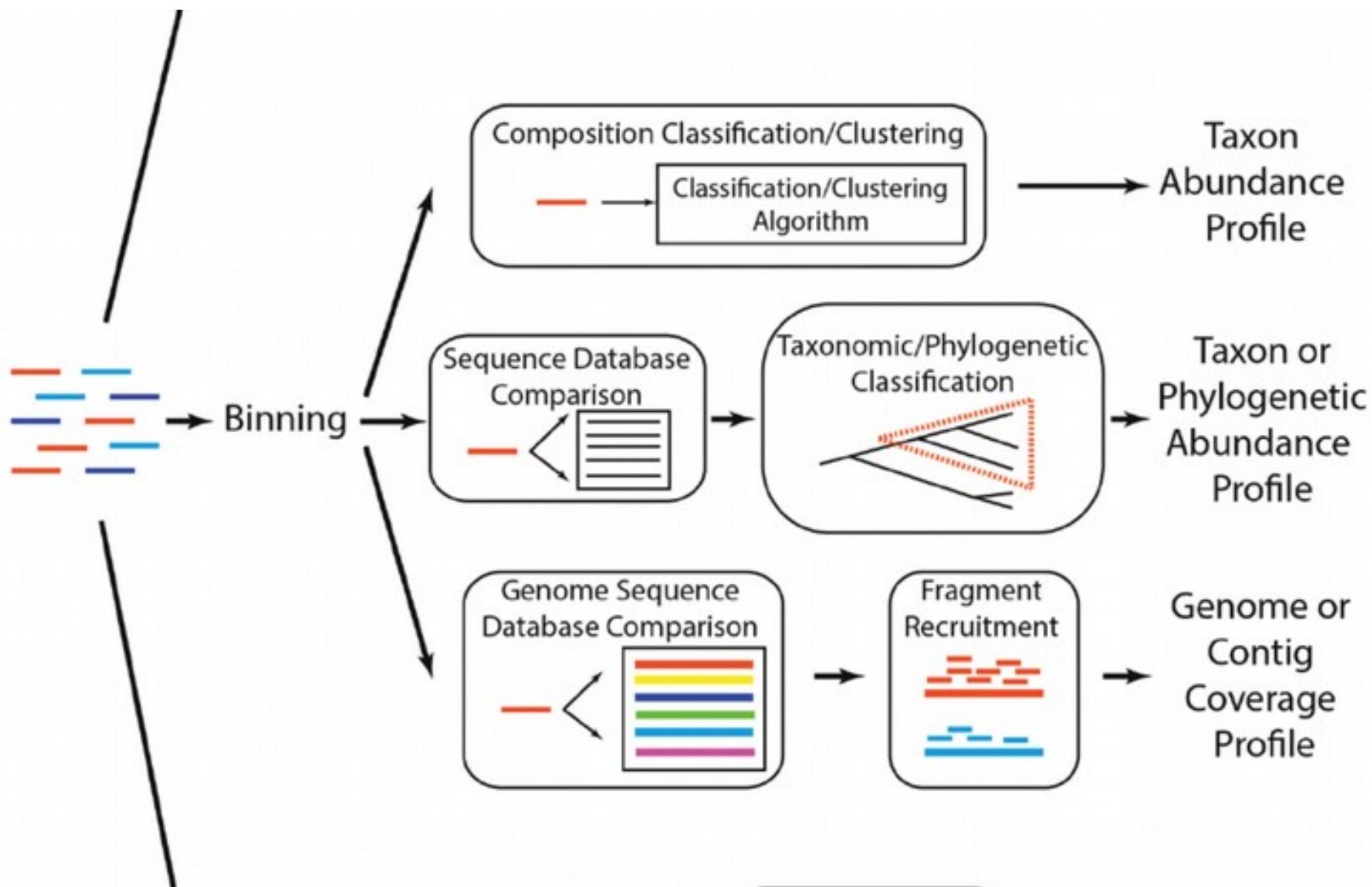
Sharpton, T.J., 2014. An introduction to the analysis of shotgun metagenomic data. Front Plant Sci 5. doi:10.3389/fpls.2014.00209

## 2- tüm metagenom sekanslama



rDNA, özgün marker genler  
küçük veritabanı  
çok hızlı...





## binning - verini akümüle edilmesi

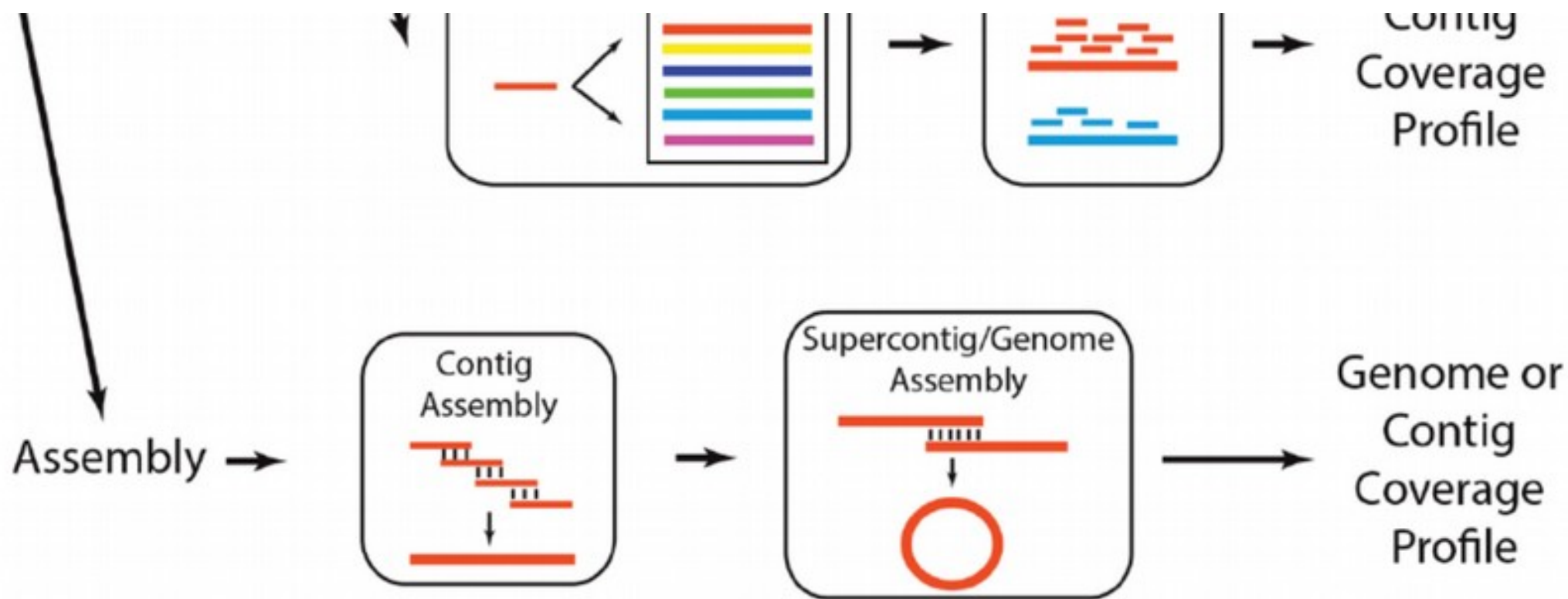
- %GC, tetramer frekansı gibi bir kriter ile gruplanması

ya da,

- elde edilen her bir sekansın bir OTU'ye atanması

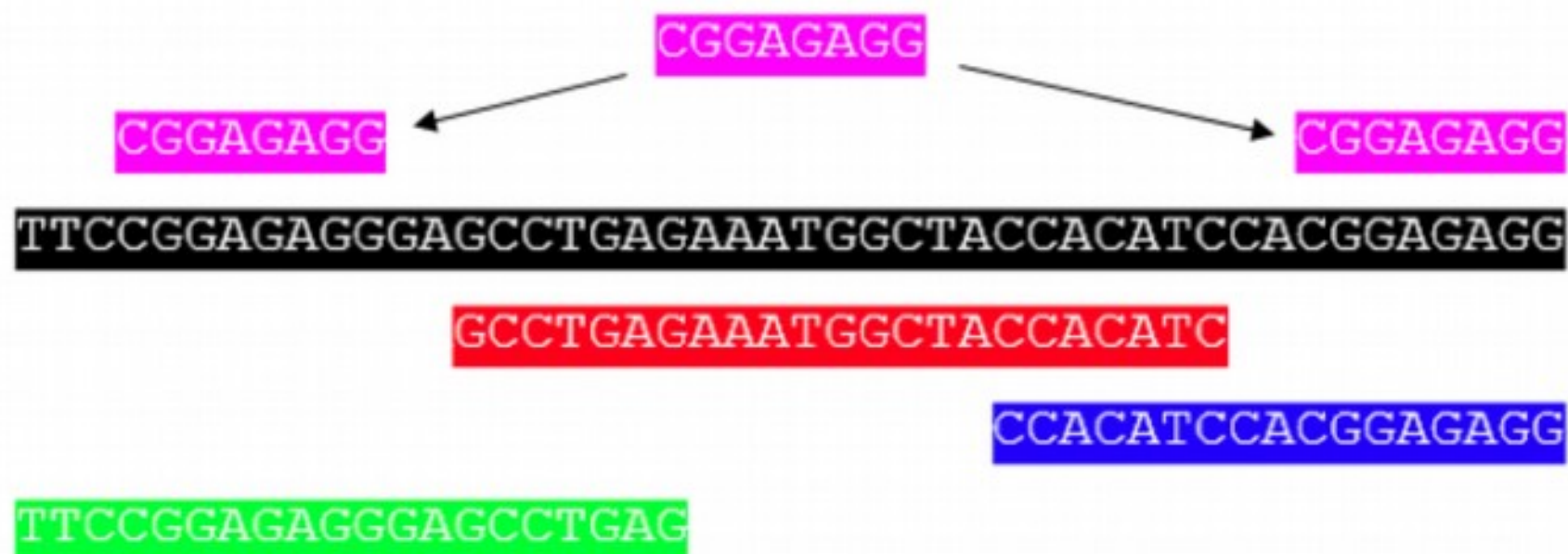
ya da,

- fragmentlerin toparlanması

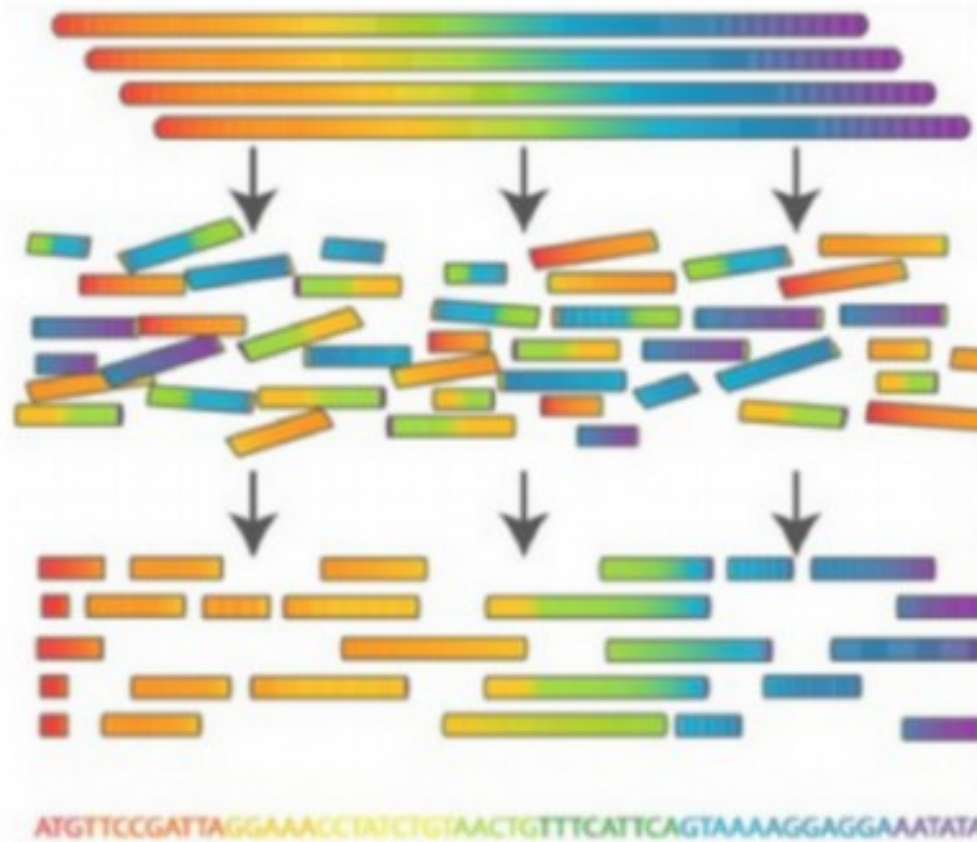


Sharpton, T.J., 2014. An introduction to the analysis of shotgun metagenomic data. *Front Plant Sci* 5. doi:10.3389/fpls.2014.00209

## assembly - birleştirme



aynı genom ait (?) kolineer okumaların devamlı tek bir dizi oluşturacak şekilde birleştirilmesi -> KONTİGler

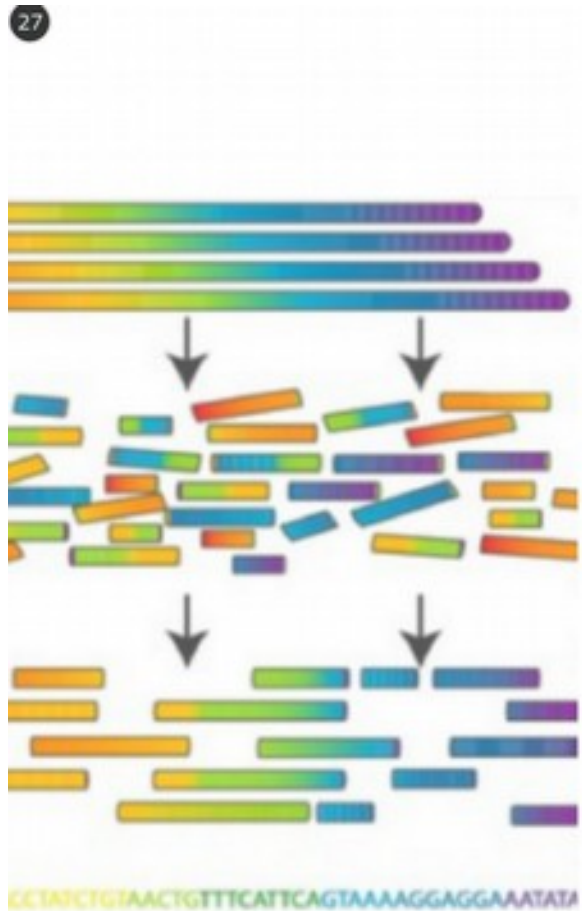


Tüm parçalar tek bir herşeyi yerli yerine c



Birçok bireyin parçala

Tüm genler / genomlar başarıyla k  
kimera gen / genomlar da

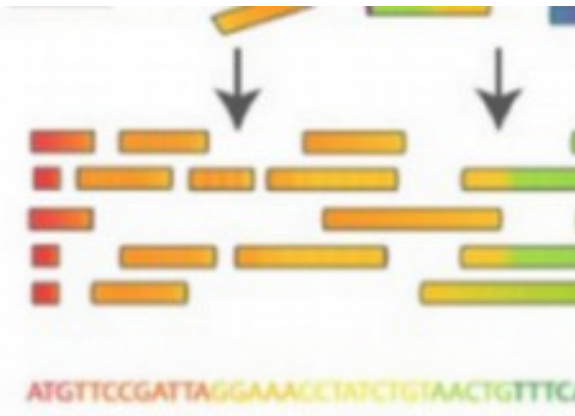


Tüm parçalar tek bir genomdan geliyor olsaydı, herşeyi yerli yerine oturtma çok kolay olacaktı.



Birçok bireyin parçaları birbirine uyumlu olabilir

Tüm genler / genomlar başarıyla bireştirilebileceği gibi kimera gen / genomlar da oluşturulabilir.



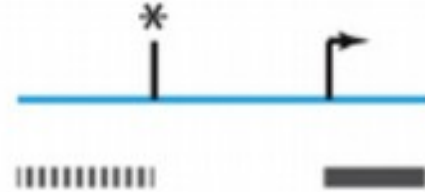
Birleřtirme nce yapılırsa  
daha bařarılı binning yapılabilir.

Binning nce yapılırsa  
daha bařarılı birleřtirme  
elde edilebilir...

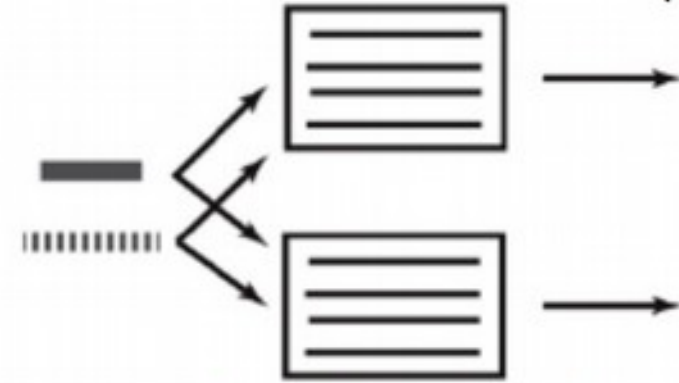
# Biyolojik fonksiyon çıkarımı



metagenom



gen tahmini



fonksiyonel açıklama  
(annotation)

- gen fragmentlerinin yakalanması
- protein ailesi sınıflandırması

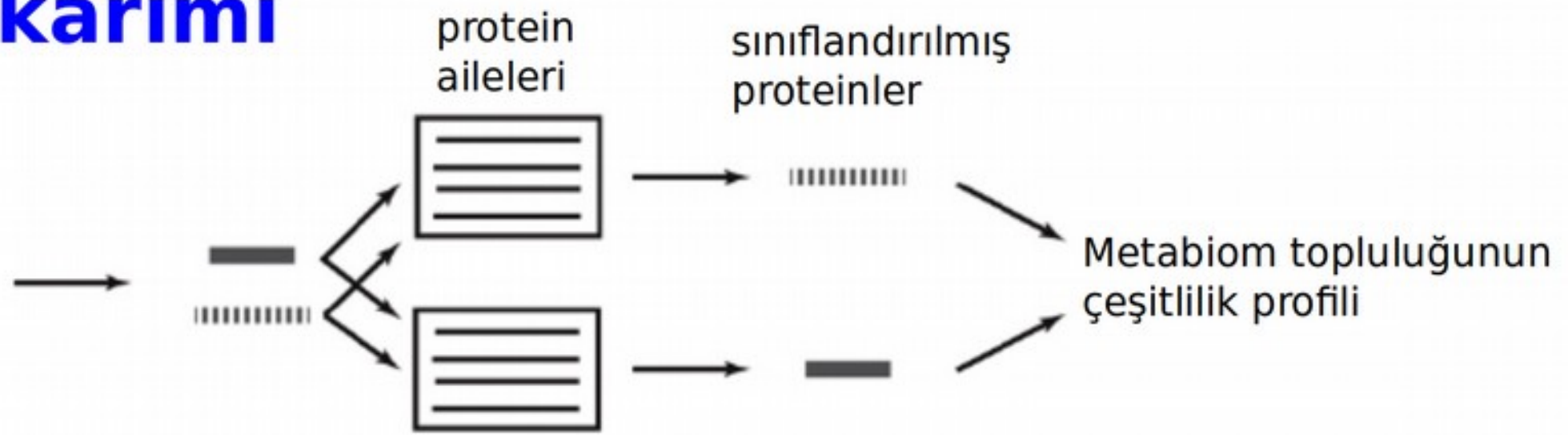
homoloji:  
nükleotid sekansı << protein sekansı

- de novo gen bulma algoritmaları





# Çıkarımı



fonksiyonel açıklama (annotation)

ı yakalanması  
ndırması

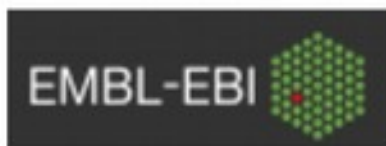
< protein sekansı

a algoritmaları



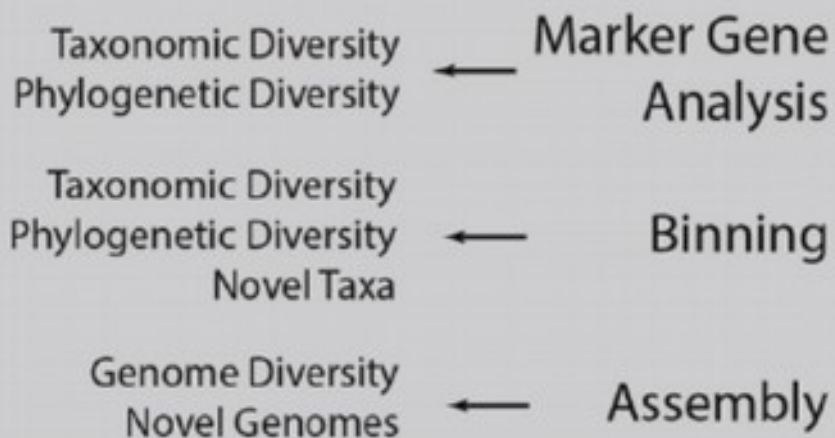
VS

g g a g a c t g t a g a c a g c t a a t g c t a t a  
 g a a c g c c c t a g c c a c g a g c c c t a t c

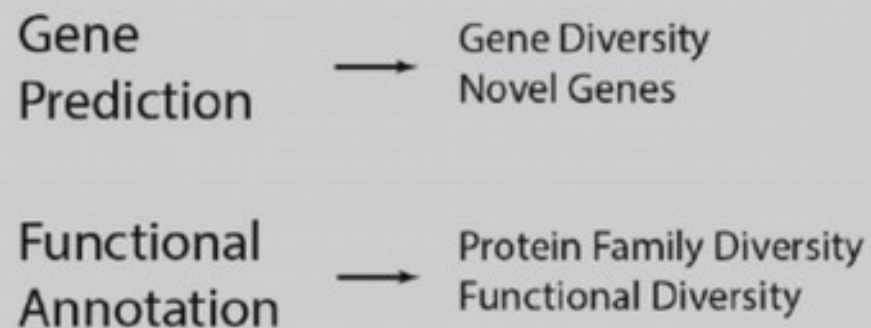


Sistem biyolojisi veritabanları  
- verinin zenginleştirilmesi

### Who is there?



### What are they doing?

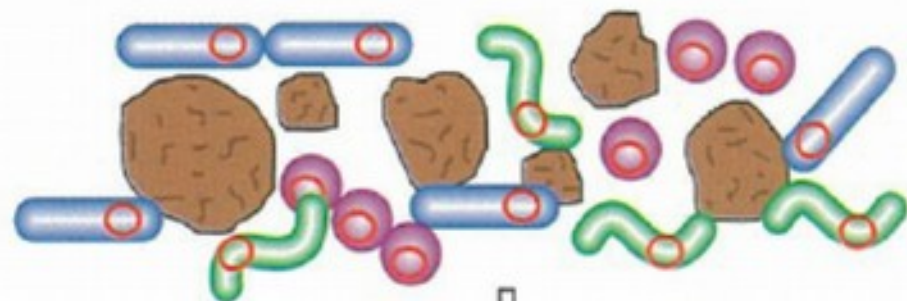


Mevcut veritabanları ile...

Kültürü yapılabilen mikroorganizmalarda bile:

- putative protein nnnnnn
- unknown protein xxxxxx

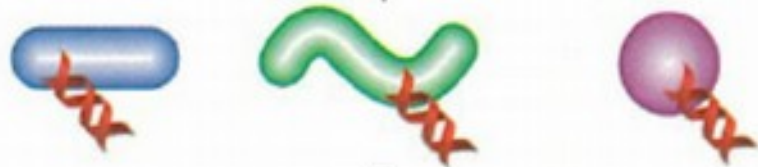
Handelsman, J., Rondon, M.R., Brady, S.F., Clardy, J., Goodman, R.M., 1998.



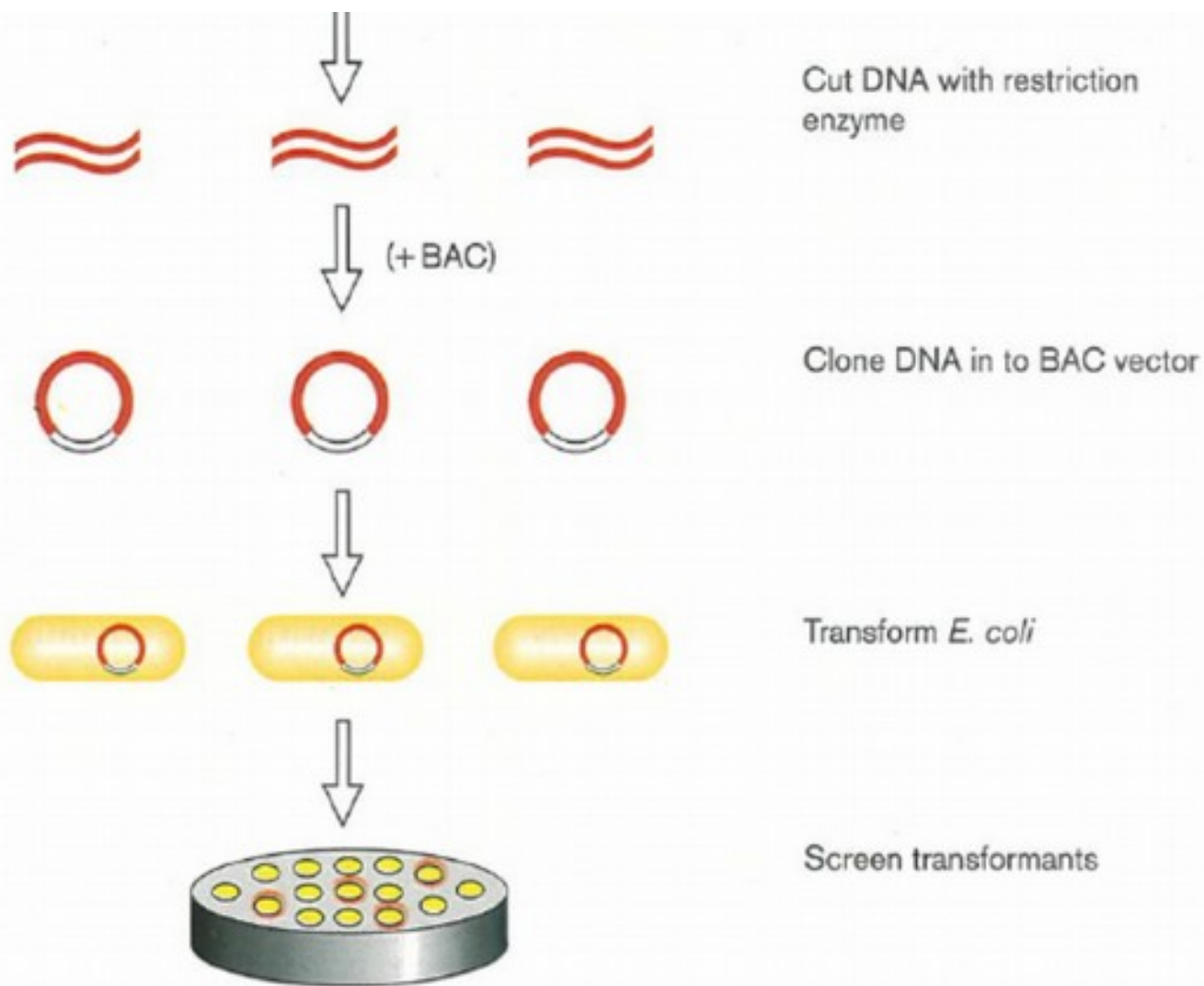
Soil sample



Separate bacteria

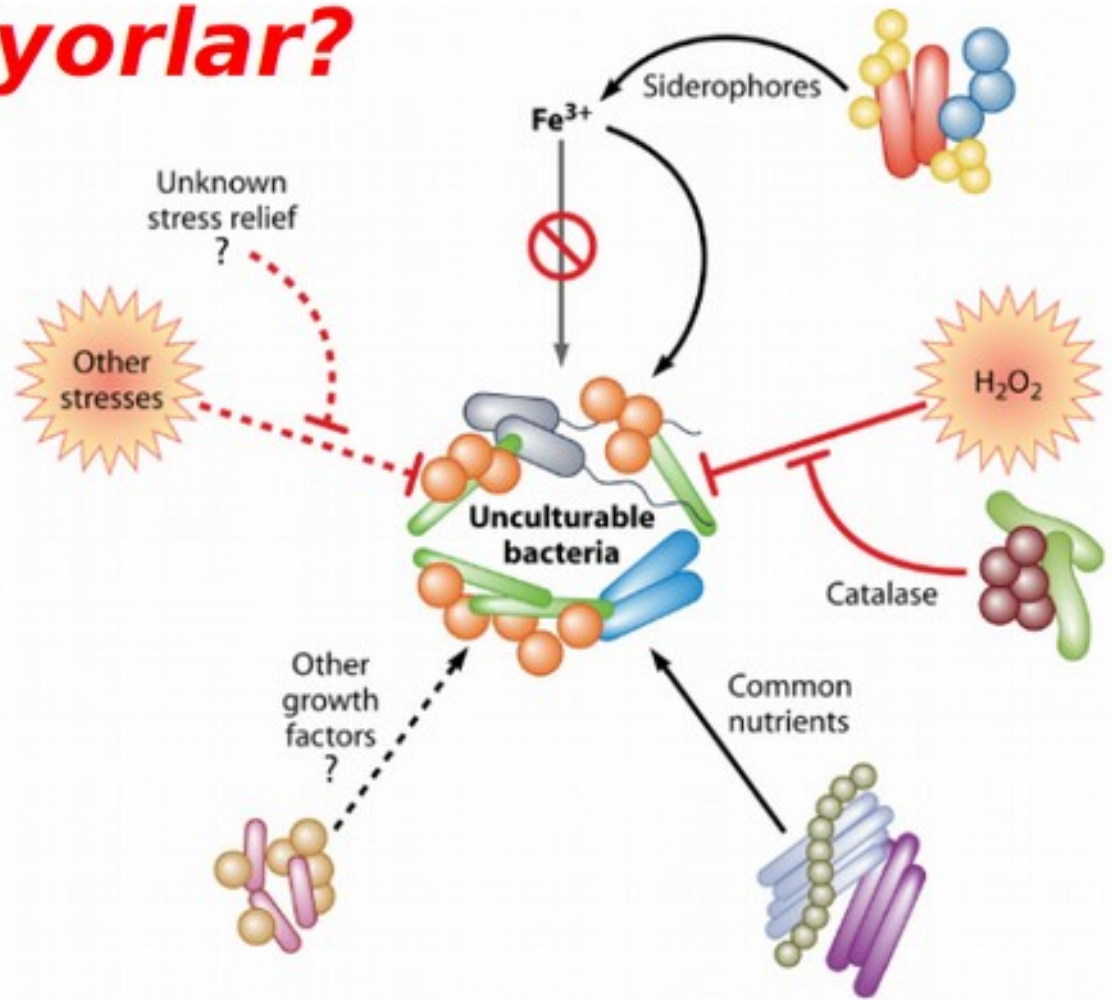
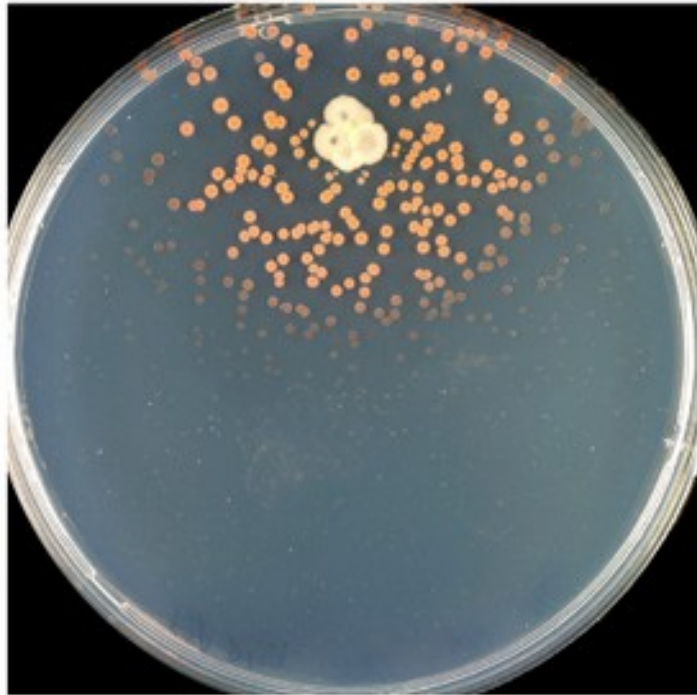


Isolate large DNA fragments



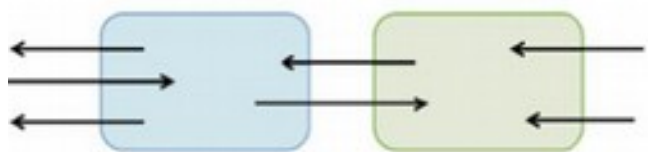
Chemistry & Biology

# Neden üremiyorlar?

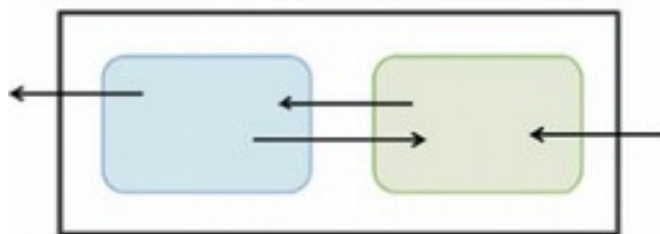


# doğal etkileşimler

in nature

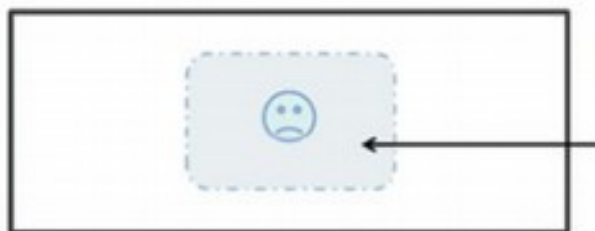


in co-culture

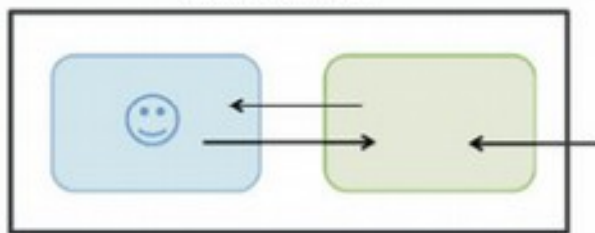


# (mono)kültürü yapılamayanlar

monoculture



co-culture

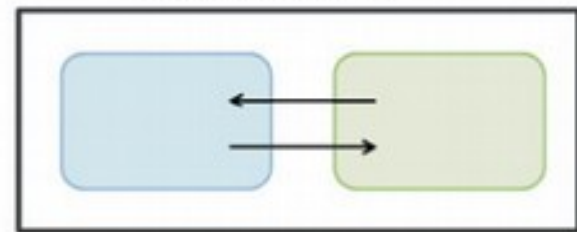


# sentetik...

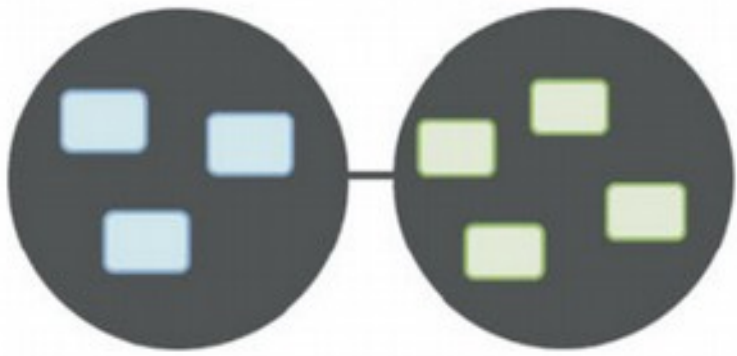
in nature



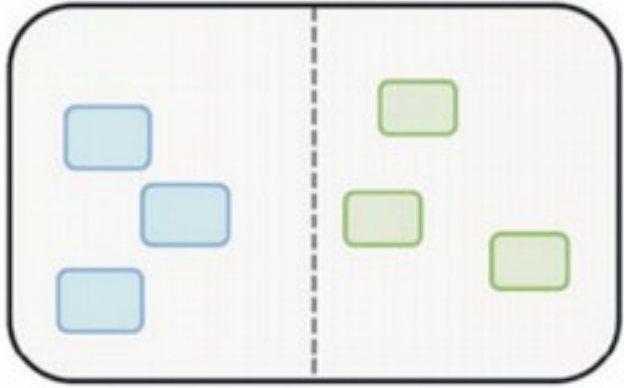
in co-culture



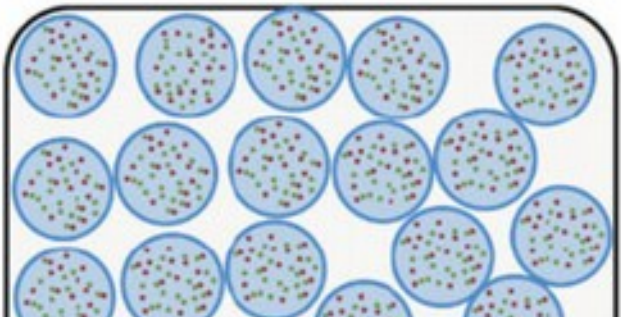




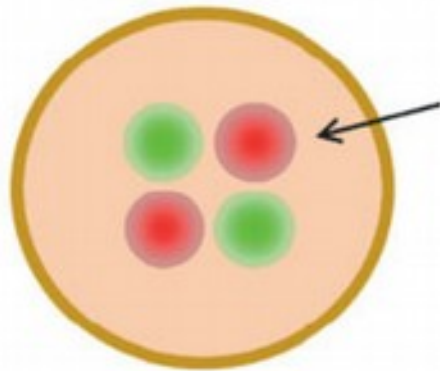
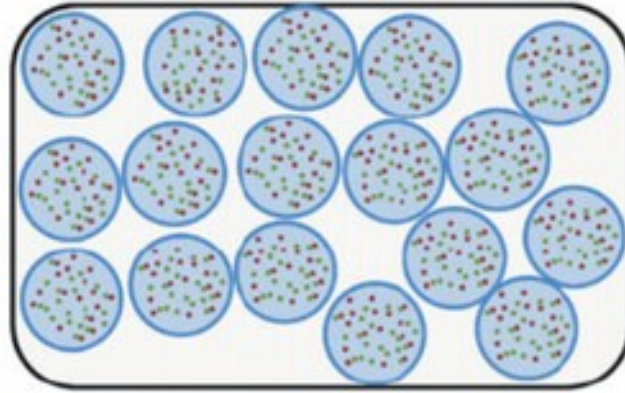
mikrofluidik kanallar



membranlar

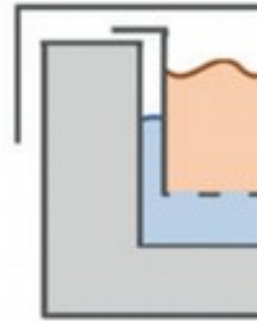


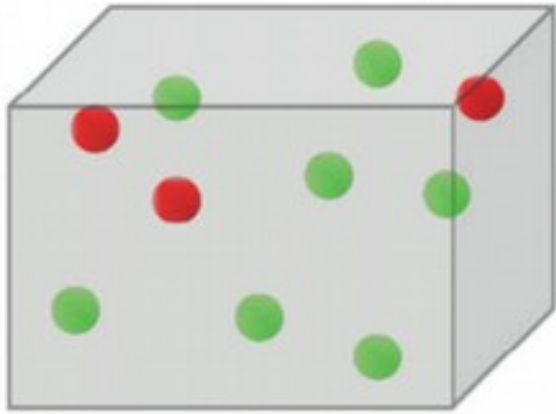
damlacıklar



damlacıklar

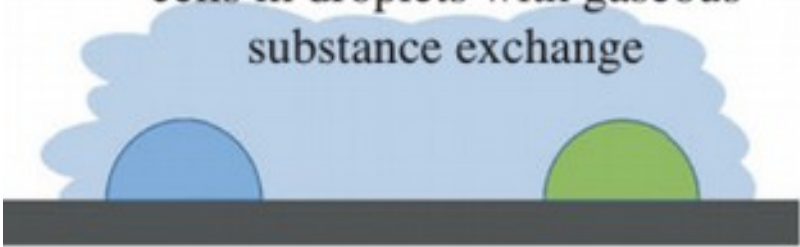
petri plağı





Jel içine gömülü hücreler

cells in droplets with gaseous  
substance exchange



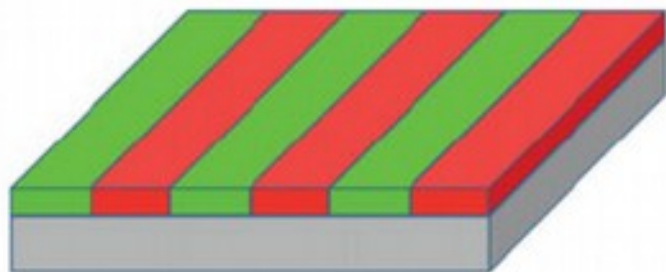
gaz deęiřimi



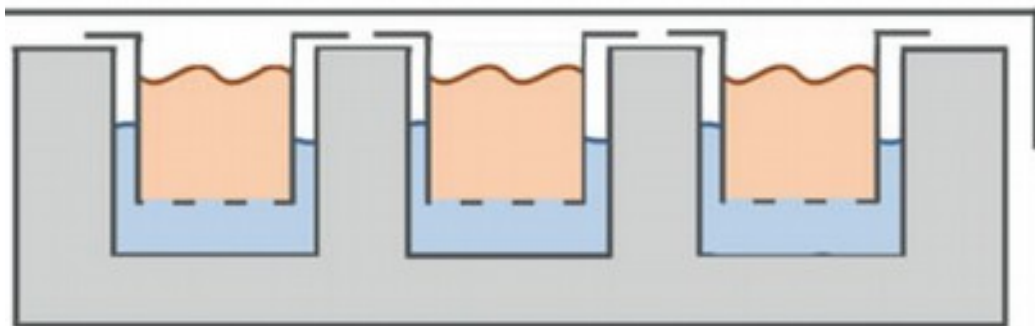
yan yana jel dilimleri



yan yana jel dilimleri

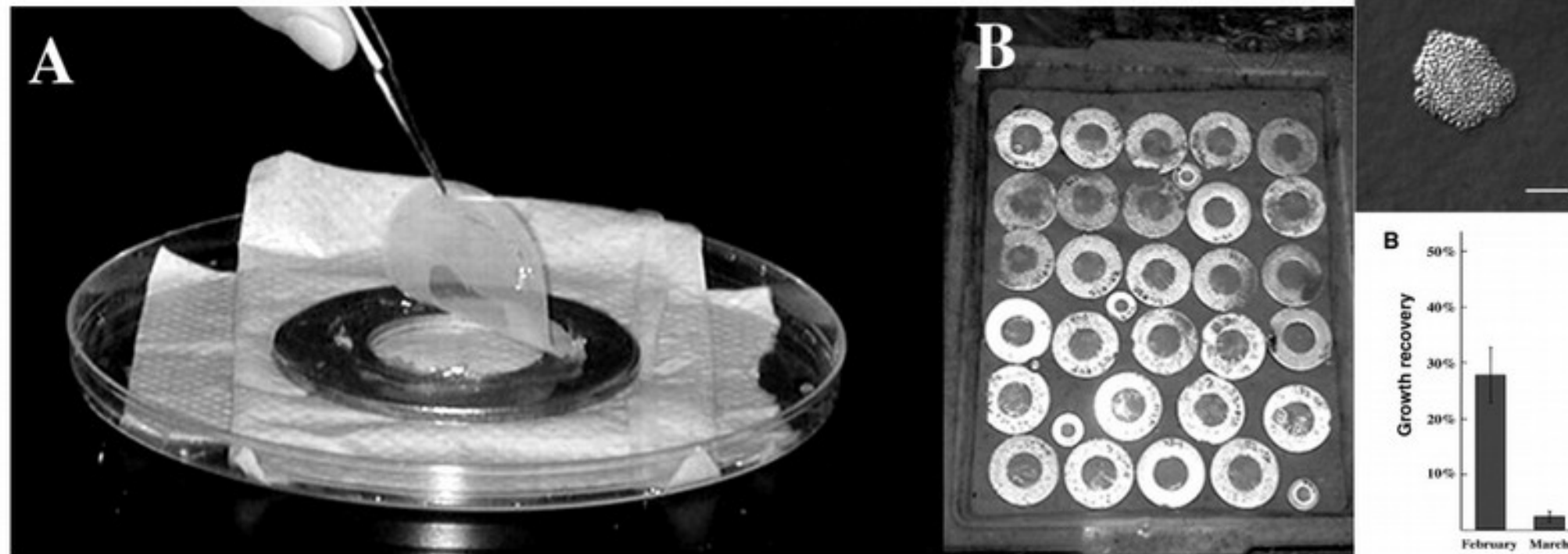


yan yana jel dilimleri

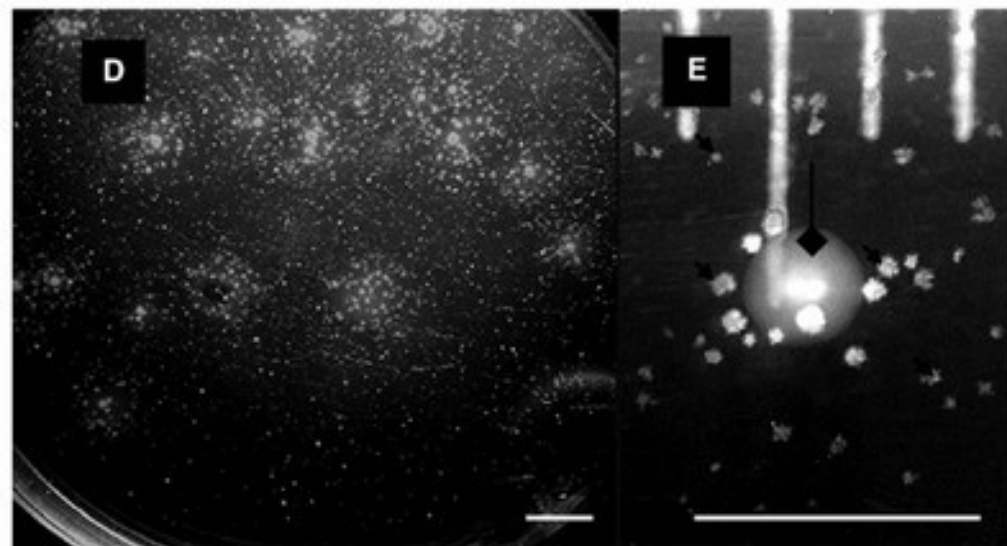
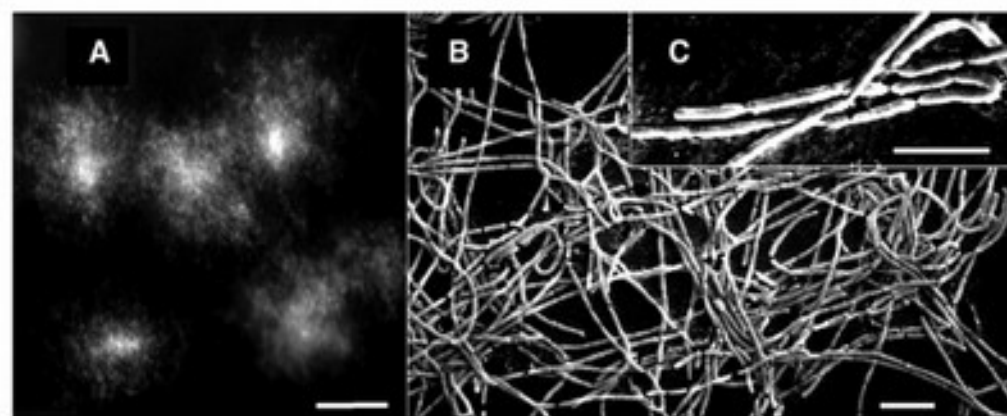
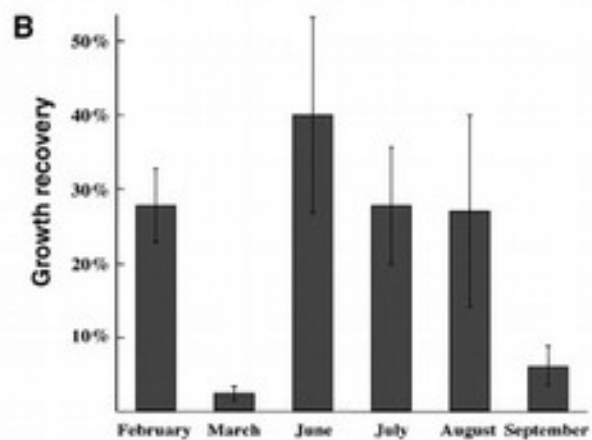
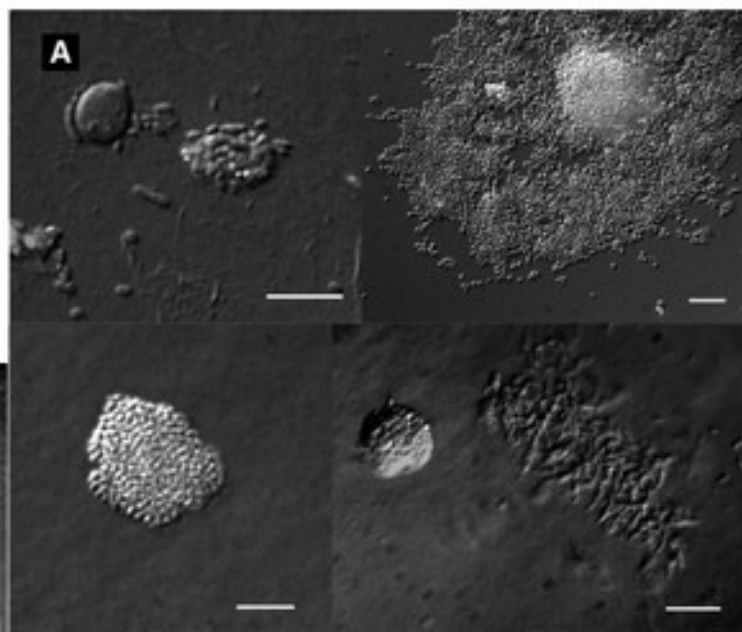


kuyucuklar

# 0.03 $\mu\text{m}$ por çaplı polikarbonat membranlar arasında deniz suyu sedimenti örneklerinin inkübasyonu

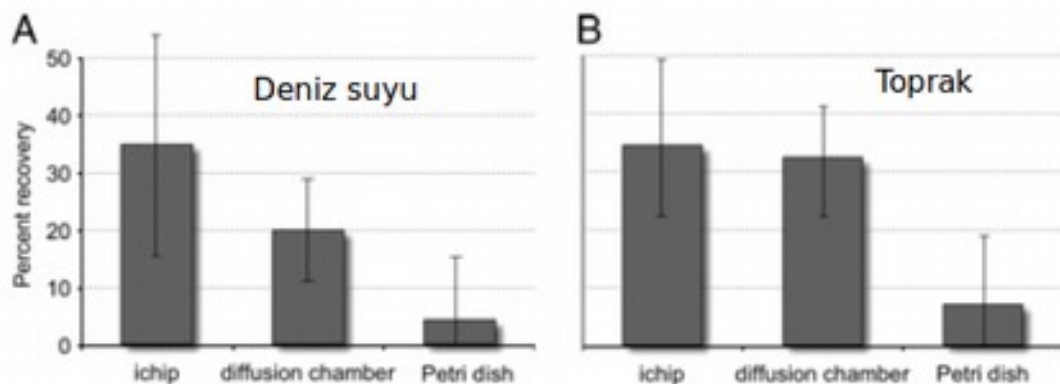
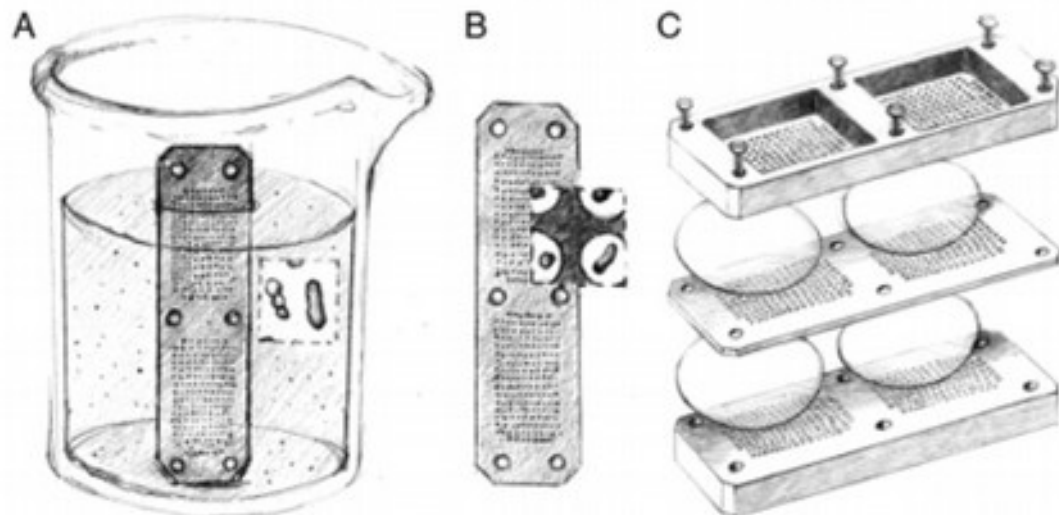


ar  
in





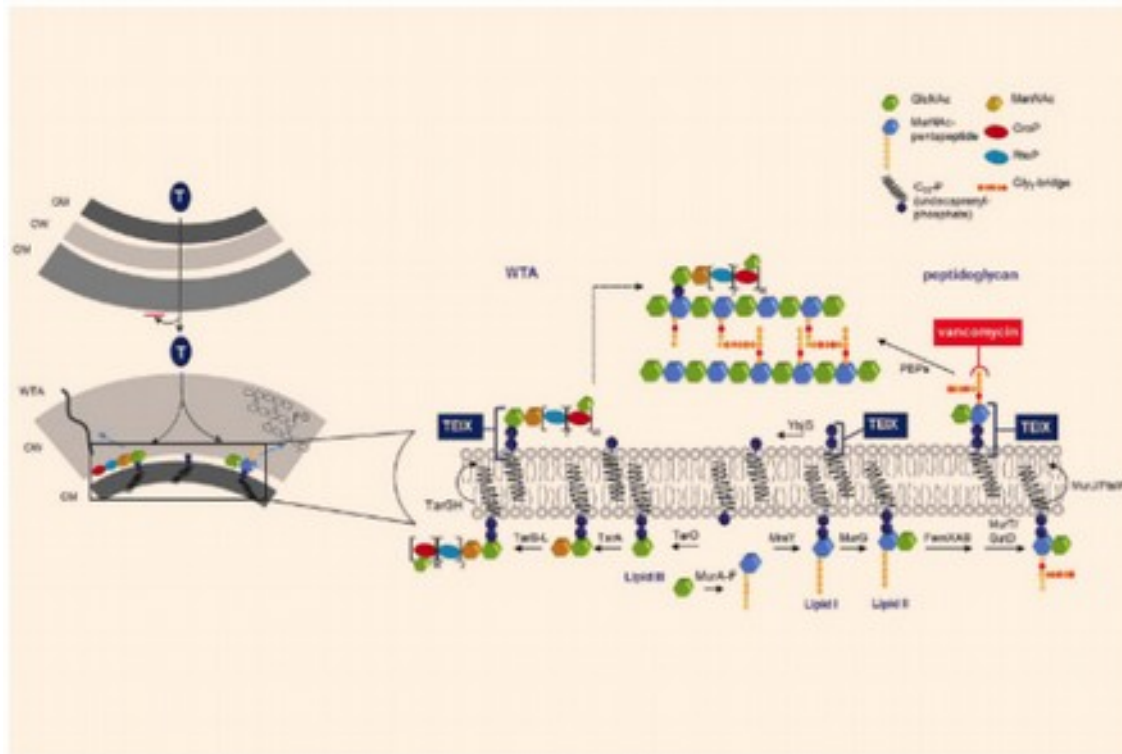
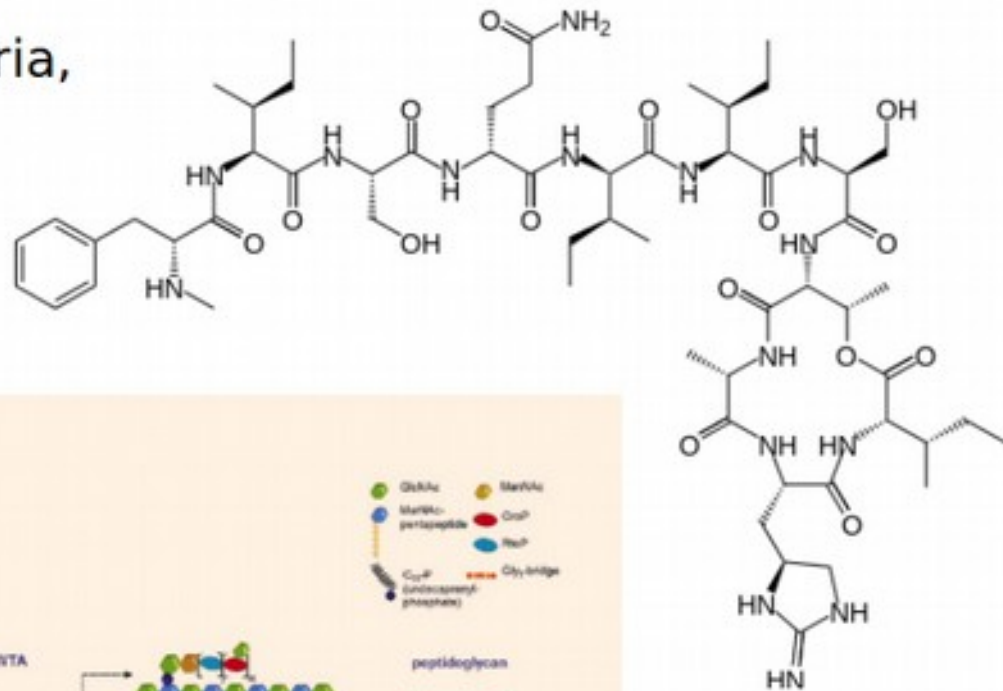
Ledford, H., 2015. Promising antibiotic discovered in microbial "dark matter."  
Nature. doi:10.1038/nature.2015.16675



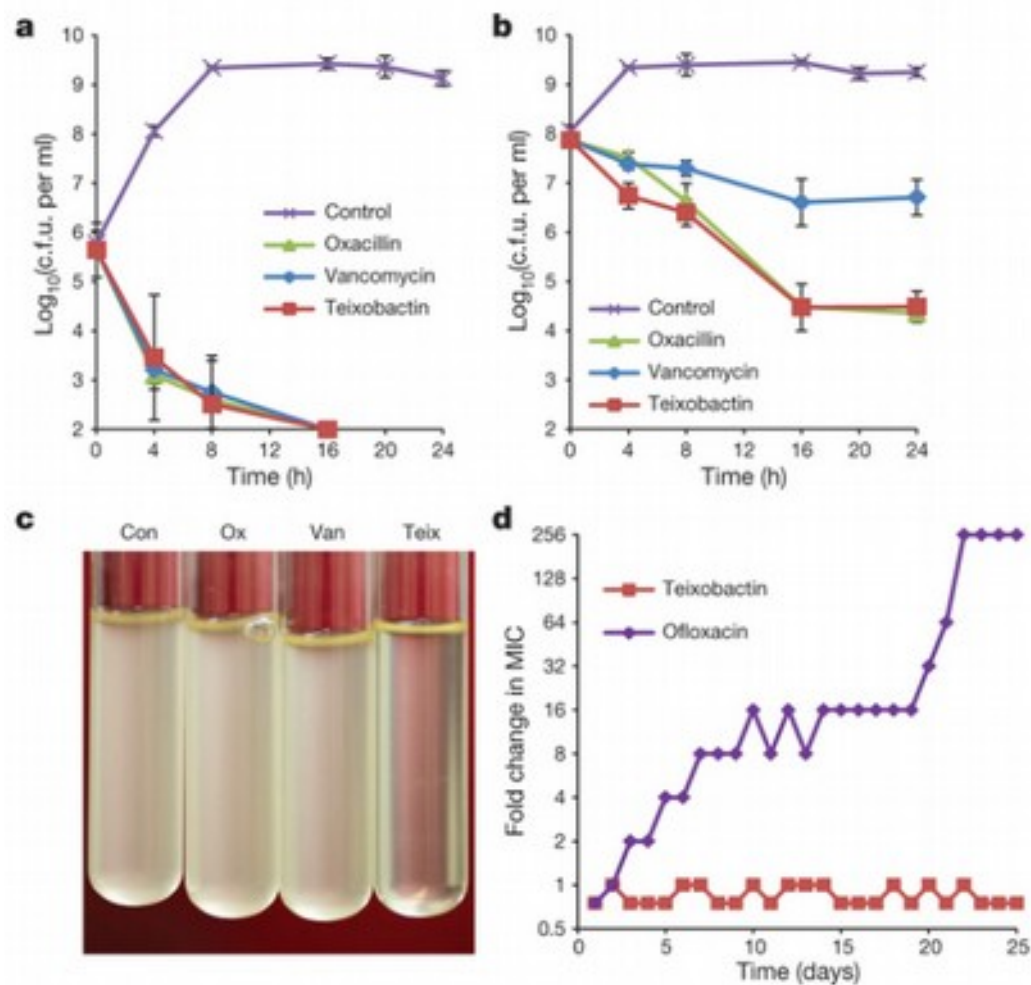
Nichols, D., Cahoon, N., Trakhtenberg, E.M., Pham, L., Mehta, A., Belanger, A., Kanigan, T., Lewis, K., Epstein, S.S., 2010. Use of Ichip for High-Throughput In Situ Cultivation of "Uncultivable" Microbial Species. *Appl. Environ. Microbiol.* 76, 2445-2450. doi:10.1128/AEM.01754-09



beta-proteobacteria,  
*Eleftheria terrae*



lipid II - peptidoglikan  
lipid III - lipoteikoik asit



Ling, L.L., Schneider, T., Peoples, A.J., Spoering, A.L., Engels, I., Conlon, B.P., Mueller, A., Schäberle, T.F., Hughes, D.E., Epstein, S., Jones, M., Lazarides, L., Steadman, V.A., Cohen, D.R., Felix, C.R., Fetterman, K.A., Millett, W.P., Nitti, A.G., Zullo, A.M., Chen, C., Lewis, K., 2015. A new antibiotic kills pathogens without detectable resistance. *Nature* 517, 455-459. doi:10.1038/nature14098