

Site-specific Recombination: Points to consider

1. How does site-specific recombination differ from homologous recombination?
2. Are site-specific recombination systems found in prokaryotes or eukaryotes or both? Do you know some examples?
3. What is meant by conservative site-specific recombination?
4. What are the two well characterized families of site-specific recombinases?
5. What is the chemistry of strand breakage during conservative site-specific recombination?
6. What is the chemistry of strand joining during conservative site-specific recombination?
7. What is the difference between breaking a phosphodiester bond by transesterification and by hydrolysis? What is the mode of strand cleavage mediated by the conservative site-specific recombinases?
8. What is the cleavage nucleophile during Integrase/Tyrosine family site-specific recombination? What are the two DNA ends formed as a result of strand cutting?
9. Do the enzymes of this family make single or double stranded cuts?
10. What is the nucleophile for the strand joining reaction?
11. Is a Holliday junction an intermediate in the Integrase/Tyrosine family recombination reaction?
12. What is the nucleophile for the Resolvase/Invertase family site-specific recombination? What DNA ends are formed as a result of strand cutting?
13. What is the nucleophile for strand joining during the Invertase/Resolvase reaction?
14. Do the Invertase/Resolvase enzymes make single strand or double strand cuts?
15. Is a Holliday intermediate formed during the Resolvase/Invertase reaction?
16. Do the invertases and resolvases follow the same chemical mechanism or different chemical mechanisms during recombination?
17. Can you recall how site-specific recombination is used to bring about different biological consequences?
18. Can site-specific recombination be used to turn on or turn off a particular gene (think of flagellin phase variation in *Salmonella*)?

19. How does site-specific recombination help a bacterial pathogen to escape the host immune system, at least temporarily?
20. Is site-specific recombination useful for altering host-specificity of bacterial viruses? In what way is this going to be useful for the virus?
21. How is site-specific recombination employed to ensure the faithful segregation of replicated bacterial chromosomes?
22. Does the yeast plasmid use site-specific recombination as a mechanism for copy number control (or copy number amplification)?
23. How can site-specific recombination be applied as a tool in biotechnology?