for the broad-band SW albedos. However, the magnitude of the changes would be smaller because of absorption at selected wavelengths and diminished scattering efficiencies at longer wavelengths.

The variations of daily mean albedo with LW flux during August 1991 compared with those for the 5-year period between 1985 and 1989 (Fig. 9, B and C) show some evidence of the trend that is expected from the direct effect. The albedo increase for both the clearest ocean ($M_{LW} \approx 295$ W m^{-2}) and land areas ($M_{LW} \approx 305$ W m^{-2}) was 0.022 (Fig. 9C). After a sharp drop to 0.006 at $M_{LW} \approx 280$ W m^{-2} and a recovery to 0.020 at $M_{LW} \approx 260$ W m^{-2}, the differences exhibit a generally downward trend to reflect relatively cloudy scenes ($M_{LW} \approx 220$ W m^{-2} and $\alpha \approx 0.290$). The albedo differences tend to increase, however, with decreasing LW fluxes for $M_{LW} < 220$ W m^{-2}. The greatest albedo differences occurred for deep convective clouds ($M_{LW} \approx 175$ W m^{-2}). The reversed trend in albedo differences at the lower LW fluxes is inconsistent with the direct effect. This reversal suggests that the aerosol layer caused an indirect effect on the albedo by altering the microphysical characteristics of the deeper convective clouds. Such changes may explain why the largest decreases in net radiation occurred over the tropical convective storm regions (Fig. 7).

After an eruption, volcanic debris in the stratosphere filters into the lower stratosphere and troposphere, primarily in the vicinity of tall convective storms and in tropopause folds. Significantly enhanced tropospheric aerosol loading due to Mount Pinatubo was observed by lidars as far north as 40°N during August 1991 (*22*). Thus, significant amounts of volcanic aerosols were available for incorporation into the clouds, especially at the upper levels. Sulfate aerosols function as efficient cloud condensation nuclei, and volcanic ash can act as an ice nucleus at temperatures below −16°C (*23*). Greater concentrations of cloud and ice nuclei tend to increase the number and reduce the effective radius of the hydrometeors in the cloud (*21*). If a cloud has a constant liquid or ice water content, its albedo increases as the effective particle radius decreases (*21*, *24*). This indirect effect could explain the increased albedos for the LW fluxes that correspond to deep convective clouds without changing the LW flux, because optically thick (high-albedo) clouds are opaque to LW radiation. Although the indirect effects are most noticeable for deep clouds (that is, $M_{LW} < 220$ W m^{-2}), they may also occur for other cloud types. For example, diffusion of volcanic aerosols across the tropopause may alter the optical properties of high, thin cirrus clouds or enhance the generation of clouds (*25*).

Volcanic radiative impacts are more complex than the model that depicts direct forcing by a single aerosol layer that is distributed uniformly over the background. Indirect effects are not now included in most climate models. These results provide additional impetus to include indirect aerosol effects such as variable cloud particle size in climate prediction models.

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Molecular Matchmakers

Aziz Sancar and John E. Hearst

Molecular matchmakers are a class of proteins that use the energy released from the hydrolysis of adenosine triphosphate to cause a conformational change in one or both components of a DNA binding protein pair to promote formation of a metastable DNA-protein complex. After matchmaking the matchmaker dissociates from the complex, permitting the matched protein to engage in other protein-protein interactions to bring about the effector function. Matchmaking is most commonly used under circumstances that require targeted, high-avidity DNA binding without relying solely on sequence specificity. Molecular matchmaking is an extensively used mechanism in repair, replication, and transcription and most likely in recombination and transposition reactions, too.

In the 1960s and 1970s models were proposed for protein folding and for binding of proteins to DNA. According to these models, proteins folded by following a kinetic pathway dictated by their primary structures (*1*); similarly, it was proposed that DNA binding proteins bound to specific sequences on DNA by presenting a set of hydrogen bond donors and acceptors complementary to those in the major or minor groove of the recognition sequences (*2*). The research of the past 25 years has generally supported these models (*3*). However, in recent years evidence has accumulat-

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ed that in many cases both the folding of proteins and the binding of proteins to DNA are aided and promoted by other proteins expressly made for these purposes. The group of proteins involved in preventing other proteins from making nonproductive associations or from folding improperly have been called chaperones (*4*). In fact, chaperones are an entire class of proteins that carry out unique functions along the protein folding pathways (*5*). Chaperones have been the subject of intensive studies and a great deal is known now about these proteins.

In contrast, proteins that act catalytically to promote and aid the binding of other proteins to DNA have not been generally recognized as a unique entity, although examples of such proteins have existed in the literature for many years. Recently, during our studies on DNA repair, we discovered that the UvrA protein of *Escherichia coli* participates in nucleotide excision repair by recognizing DNA damage and delivering UvrB to the damage site (*6*). We named this activity molecular matchmaking (*7*) and noted the presence of many proteins of similar function. We defined the molecular matchmaker as a protein that, in an adenosine triphosphate (ATP)–dependent reaction, brings two compatible yet solitary macromolecules together, promotes their association, and then leaves the complex.

For a protein to be classified as a molecular matchmaker it must satisfy the following conditions: (i) In the absence of the matchmaker the affinity of the matched molecules to one another should be so low as to be physiologically insignificant; (ii) the molecular matchmaker should promote formation of a stable complex between the matched components; (iii) the matchmaker (or one of the matched proteins) should be an adenosine triphosphatase (ATPase) or an ATP binding protein; (iv) the matchmaker should form a complex with the matched components, causing conformational change but no covalent modification in one or both molecules that leads to their association; and (v) after matchmaking the mediator must dissociate from the complex it helped form to permit the complex to carry out its specific function. In the next sections we review examples of molecular matchmakers from the various fields of molecular biology. Although most of the examples satisfy all five criteria, some have not been characterized in sufficient detail for us to determine whether they meet all the requirements. However, the modes of action of proteins in the latter group are sufficiently close to those of bona fide matchmakers to justify their inclusion in this general category.

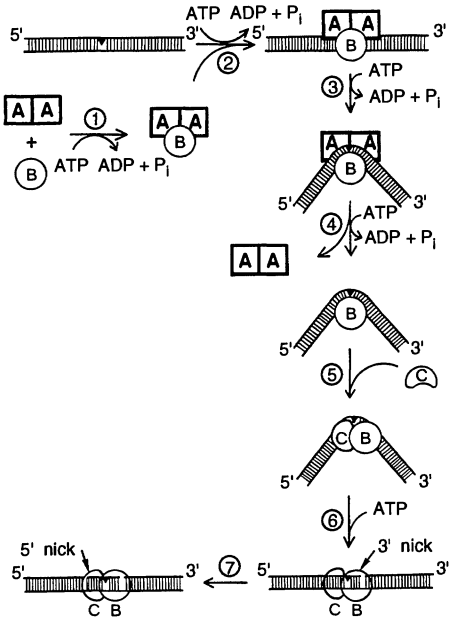

Fig. 1. Mechanism of action of (A)BC excinuclease. UvrA (in bold) is the molecular matchmaker that delivers UvrB to the damage site and primes DNA for double incisions by UvrB and UvrC. [Adapted from (*11*)]

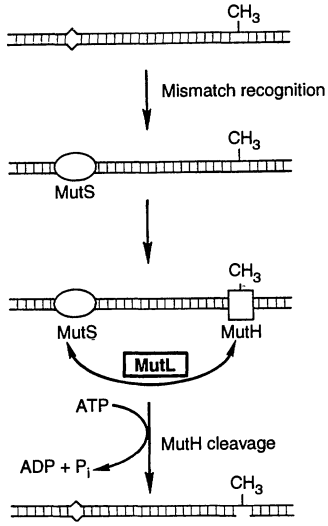

Fig. 2. Model for methyl-directed mismatch repair. MutL (in bold) is the potential matchmaker for MutH endonuclease. [Adapted from (*13*)]

DNA Repair

Molecular processes that eliminate damaged bases from DNA or correct mismatches in DNA are called DNA repair. There are many repair enzymes of narrow substrate specificity and two repair systems of wide substrate range, nucleotide excision repair, and methyl- or nick-directed mismatch repair. The nucleotide excision repair enzyme removes all damaged bases regardless of the chemical nature of the lesion, and methyl-directed mismatch repair corrects all eight possible mismatches. These repair systems attain high-specificity DNA-protein interactions that are not dictated by DNA sequence and thus are uniquely qualified for molecular matchmaking. The following is a brief description of nucleotide excision and methyl-directed mismatch repair in *E. coli*, with emphasis on the matchmaking step in these multistep systems.

Nucleotide excision repair. The (A)BC excinuclease repairs virtually all DNA damages (but not mismatches) by excising a dodecamer carrying the modified nucleotide (*8*). The nuclease activity results from sequential and partly overlapping actions of three subunits, UvrA, UvrB, and UvrC (Fig. 1). UvrA, which is an overt ATPase (*8*) and a damage recognition protein, associates with UvrB, which is a cryptic ATPase with no measurable affinity for DNA, to form an A_2B_1 complex. This complex binds DNA nonspecifically and tracks along the double helix at least partly fueled by the UvrB ATPase activity, which now becomes overt (*9*); on encountering a structural abnormality, the complex binds to the damage site. The specificity provided by the A_2B_1 complex is modest in that the complex binds to a damaged site with about 10^4-fold higher affinity as compared with undamaged DNA. However, at the second stage of the recognition process, the molecular matchmaking activity of UvrA provides higher specificity. After formation of the A_2B_1-DNA ternary complex, the DNA undergoes a drastic change, being locally denatured and kinked by 130°. This is accompanied by a possible change in UvrB conformation, with the result that UvrB becomes a DNA binding protein. Both kinking and local unwinding of DNA as well as the conformational change in UvrB require ATP hydrolysis by UvrA, UvrB, or both subunits (*10, 11*). UvrA dissociates, leaving behind an essentially irreversible UvrB-DNA complex (half-life of about 2 hours at a psoralen-thymine adduct) in which the DNA is now primed for the dual incisions that result when it encounters the third subunit of this enzyme system, UvrC (*12*). In this system, UvrA is the matchmaker and satisfies all five criteria stated above.

Mismatch repair. In *E. coli*, methyl-directed mismatch repair corrects mismatches resulting from replication errors, recombination, or base deamination. The correct strand is identified by the methylation state of adenine in the GATC sequence (*13*). The MutH, MutL, and MutS proteins carry out the reaction with the help of replication proteins such as DNA polymerase (Pol) III and helicase II. In isolation, MutH has very weak incision activity on the unmethylated strand 5′ to the GATC sequence; MutS is a

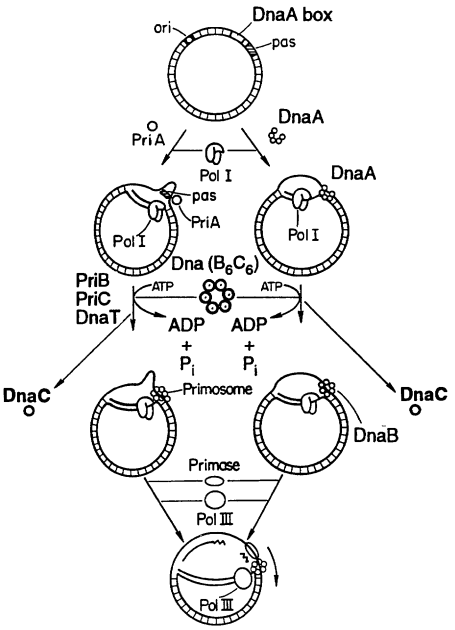

Fig. 3. The "ABC primosome" (oriC-type) (right) and the "ϕX-type" primosome (left). DnaC (in bold) is the molecular matchmaker for DnaB in both types of primosomes that synthesize primers at the origin, and for Okazaki fragments, respectively. [Adapted from (*15*)]

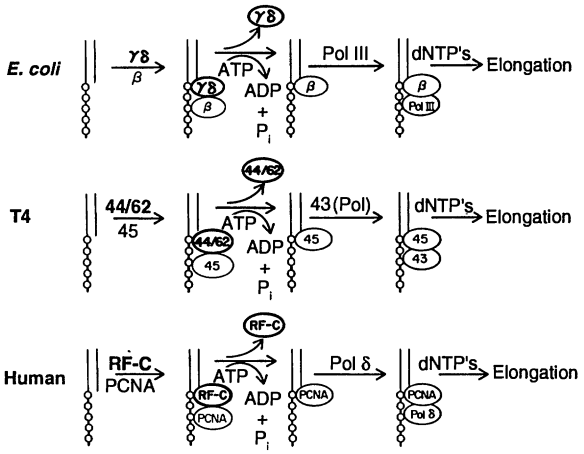

Fig. 4. Molecular matchmakers and DNA polymerase clamps in *E. coli*, T4 phage, and humans. In *E. coli*, the "γ complex" (γδ in bold) is the matchmaker that clamps β onto DNA to enable Pol III to act processively. In T4, the gp44-gp62 complex (in bold) is the molecular matchmaker that delivers the gp45 to the primer-template to permit gp43 to synthesize DNA processively. In humans, RF-C (in bold) is the matchmaker that facilitates Pol δ or Pol ϵ–PCNA complex to form and carry out processive DNA synthesis. [Adapted from (*25*)]

marginal ATPase and binds to mismatches; and MutL has no detectable activity. However, in combination, MutL interacts with MutS-mismatch complex in an ATP-dependent manner (*14*) and activates the MutH endonuclease in a mismatch- and MutS-dependent manner. A plausible mechanism for the initiation of mismatch repair would be as follows (Fig. 2) (*14*): MutL is the matchmaker; it binds to MutS-mismatch complex and activates the latent ATPase of MutS, which (alone or together with MutL) enables MutH to incise the unmethylated strand of the GATC sequence. MutL satisfies all the criteria of a matchmaker with the possible exception of condition (v), because it is not known whether it dissociates from the MutS-MutH-DNA complex before MutH carries out its effector function.

DNA Replication

Replication of a DNA duplex involves two basic steps: initiation of synthesis at a unique site (or sites) in the chromosome called origin of replication and at intervals in the lagging strand, and elongation of the DNA chains. Both processes require high avidity DNA-protein interactions that are not (or are only partly) sequence-specific. Molecular matchmakers help form the DNA-protein complexes (primosomes) that initiate synthesis and those that carry out chain elongation in a highly processive manner (DNA polymerase clamps).

Primosomes. In *E. coli*, the mobile protein complex containing DnaB and DnaG (primase) and perhaps some accessory proteins is called a primosome, and the multiprotein complex that helps form a stable DnaB-DNA complex is called a preprimosome. Depending on the nature of the DNA-targeting protein, two types of primosomes have been described (Fig. 3) (*15*). In both cases DnaC is the matchmaker that delivers DnaB to the primer synthesis site.

1) The ABC-primosome is also called the "oriC-type primosome." DnaA(ATP) binds specifically to the *oriC* sequence (DnaA box) at the *E. coli* origin of replication. The prepriming proteins DnaB and DnaC form a B_6C_6 complex in which the ATPase activity of DnaB helicase is completely inhibited by DnaC. This complex binds to oriC-DnaA to form the transient ABC preprimosome (*16*). DnaC must come off the complex for initiation to occur. The B_6C_6 complex is stabilized by the binding of ATP, and the delivery of DnaB to, and dissociation of DnaC from, the preprimosome require ATP hydrolysis. After dissociation of DnaC from the ABC primosome, DnaB moves away bidirectionally from the DnaA-OriC complex using its helicase activity. Primase (DnaG) recognizes the DnaB-DNA complex and synthesizes the RNA primers. In this system the sole role of DnaC is to deliver DnaB onto DNA in an ATP hydrolysis–dependent reaction, and DnaC is absent from the primer-synthesizing complex and thus satisfies all five criteria of a molecular matchmaker. The initiation of λ phage DNA replication makes use of a similar strategy except that λP substitutes for DnaC and λO substitutes for DnaA (ATP) (*17*). Similarly, in yeast an origin recognition complex made up of six subunits binds to replication origins in an ATP-dependent manner (*18*), suggesting that OriC-type primosomes may initiate DNA replication in eukaryotes.

2) The ϕX-type primosome catalyzes the initiation of Okazaki fragments. The primosome assembly site (pas) on single-stranded DNA binding protein (SSB)–covered DNA is recognized by the PriA, PriB, and PriC proteins that assemble at the site. The B_6C_6 complex of DnaB and DnaC aided by DnaT recognizes the pas-PriABC complex (*15*) and delivers DnaB to the complex (Fig. 3). The preprimosome contains some or all five of the preprimosome proteins PriA, PriB, PriC, DnaT, and DnaB. Of these proteins, PriA is a 3′ → 5′ helicase, whereas DnaB is a 5′ → 3′ helicase. Thus, the complex can move in either direction. Considering that the direction of tracking of DnaB is the appropriate one for discontinuous DNA synthesis, it is plausible that the ϕX-type primosome may also contain only DnaB to target primase (DnaG) to initiate Okazaki fragments. Thus, the function of the other preprimosome proteins may consist of aiding DnaC in its matchmaking function.

DNA polymerase clamps. DNA polymerases whose main function is DNA replication (rather than repair) are highly processive (*15*). The processivity is not an intrinsic property of the polymerizing subunit but is conferred by a special class of proteins called polymerase clamps or sliding clamps (*19*) that are applied to DNA by molecular matchmakers (Fig. 4).

1) The T4 DNA polymerase (gp43), which is not highly processive under physiologically relevant ionic strength (*20*), becomes so in the presence of gp44, gp45, and gp62 proteins. The gp44 and gp62 proteins form a very tight complex (4:1 stoichiometry) that has ATPase activity, whereas gp45 is a trimer with no ATPase function (*21*). Although the precise mechanism is still not clear, recent studies (*21*) suggest that the gp44-gp62 ATPase-helicase loads gp45 at the template-primer site to produce a sliding clamp for gp43 polymerase and permit it to act in a processive manner (Fig. 4). It has not been shown unambiguously that gp44-gp62 is absent in the elongation complex [condition (v)]; however, it is

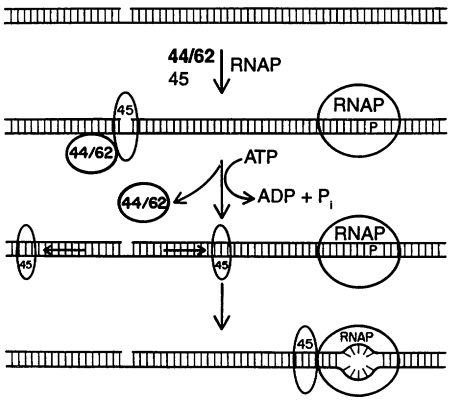

Fig. 5. The T4 phage mobile transcriptional enhancer. The gp44-gp62 complex (in bold) is the matchmaker that helps clamp gp45 on DNA at a nick. The clamp tracks along DNA, encounters an RNAP-promoter complex, and activates transcription [Adapted from (*30*)]

known that at very high concentrations of gp45, the gp44-gp62 complex is not needed for binding of gp45 to DNA polymerase (*22*). Thus, the sole function of gp44-gp62 is to promote this interaction at physiological concentration, and therefore gp44-gp62 meets all the criteria of a molecular matchmaker.

2) The β sliding clamp of *E. coli* Pol III is the best characterized and the most elaborate of all the sliding clamps. The β clamp is a dimer of 40 kD; it is loaded onto primer-template by the γ complex, which includes five proteins (γ, δ, δ′, χ, and ψ). Of these, γ has the ATPase sequence motifs, and the γ complex (γδδ′χψ) loads β2 onto the primer template in a reaction that requires ATP hydrolysis (*15, 23*). β2 itself is not an ATPase but enhances the γ complex ATPase in the presence of primed template and thus couples ATP hydrolysis to clamping of β onto DNA. Once loaded, the β clamp, which is in the form of a ring around the duplex, can diffuse on DNA in a bidirectional manner and does not require ATP hydrolysis (*23*). The β clamp interacts with the two core components of Pol III, the polymerizing subunit α and the proofreading subunit ε, to form a complex that synthesizes DNA with high processivity and fidelity (Fig. 4). In terms of molecular matchmaking, the γ complex satisfies all the criteria of a matchmaker, including induction of a conformational change in the β protein.

3) Proliferating cell nuclear antigen (PCNA) has been found in all eukaryotes (*15*). This protein increases the processivity of Pol δ (*15*) and Pol ε (*24*). Two proteins, RF-A (eukaryotic SSB) and RF-C (a DNA-dependent ATPase composed of five polypeptides), promote the interaction of PCNA with these polymerases. RF-C mediates the binding of PCNA to primer-template, and eventually to polymerase, in an ATP-dependent manner (Fig. 4), and PCNA stimulates the DNA-dependent ATPase activity of RF-C three- to fourfold (*25, 26*). However, ATP binding without hydrolysis appears to be sufficient for RF-C–mediated binding of PCNA to DNA. In terms of molecular matchmaking, RF-C satisfies all of our criteria except condition (v) [it has not been shown that RF-C dissociates from the PCNA-(polδ)-primer-template complex once DNA synthesis starts]. Recent work has revealed significant sequence homology between the T4 gp44 matchmaker and the large subunit of RF-C (*23*), providing further evidence that RF-C performs a gp44-gp62–like function.

Transcription

Transcription is the synthesis by an RNA polymerase (RNAP) of an RNA copy of one strand of the DNA duplex. As in replication, transcription involves two basic steps, initiation and elongation. However, there are two fundamental differences between replication and transcription. First, replication cannot start without a primer, whereas transcription does; and second, replication results in the synthesis of a duplex, whereas transcription involves the synthesis of a single strand. As a consequence, special DNA-protein interactions requiring molecular matchmaking occur almost exclusively at the initiation stage. Matchmaking is a particularly suitable mechanism for the physical punctuation of DNA, that is, marking the site on DNA where an effector macromolecule exerts its specification before a transcriptional start by RNAP. We find the following model of molecular matchmaking in transcription particularly attractive. A matchmaker produces a conformational change in the promoter region (melting and kinking) as well as in RNAP to induce new conformations in both components that are optimal for strong binding and initiation of transcription. In fact, such conformational changes do occur in all transcription systems that have been studied. However, these changes occur by multiple pathways, only some of which make use of molecular matchmakers. In the next section we analyze the various transcription systems in terms of molecular matchmaking and point out the differences that separate matchmaking from closely related modes of macromolecular interactions.

Escherichia coli *RNA polymerase.* The holoenzyme, with the subunit composition of $\alpha_2\beta\beta'\sigma$, is guided by the σ ($\sigma^{70}$) subunit to the promoter. After transcriptional initiation, which involves conversion of B-DNA to open complex by local melting of the duplex (*27*), the σ subunit dissociates, and elongation is carried out by the core RNAP ($\alpha_2\beta\beta'$). The σ subunit satisfies all the criteria of a molecular matchmaker except that it is not an ATPase [condition (iii)]. In contrast to RNAP (σ^{70}), RNAP (σ^{54}) requires ATP hydrolysis for initiation. This form of RNAP transcribes positively regulated genes and is guided to the promoter region by interaction between σ^{54} and an activator bound upstream of the promoter (*28*). This interaction activates the latent ATPase of the upstream activator, and the energy of ATP hydrolysis is used in the formation of an open complex. Thus, σ^{54} satisfies all five criteria of a matchmaker.

T4 RNA polymerase. This is *E. coli* RNAP associated with T4 phage–encoded gp55 (σ^{55}) and gp33 proteins that direct RNAP to T4 late promoters. The T4 "mobile enhancer" of late promoters constitutes an unusual example of molecular matchmaking. In fact, this matchmaker is identical to the T4 replicational matchmaker discussed above. The gp44-gp62-gp45 complex (4:1:3 stoichiometry) (*29*) binds to a nick in an ATP hydrolysis–dependent manner; some components, most likely gp44-gp62, leave the complex (*21*), and the gp45 clamp, very much like the β clamp of *E. coli* DNA Pol III, tracks along the duplex, encounters the closed promoter complex of gp33-σ^{55} RNAP, and converts it to an open complex (Fig. 5) (*30*). Here, gp44-gp62 satisfies the criteria of a molecular matchmaker except that it has not been shown by physical methods to leave DNA after loading gp45 [condition (v)], although indirect evidence suggests that it does (*21*).

Eukaryotic RNA polymerases. Eukaryotic transcription is carried out by three multisubunit-RNAPs targeted to their cognate promoters by sets of overlapping basal transcription initiation factors that regulate the basal level of transcription. This level can be modulated by regulatory factors that bind upstream of promoters (*31*). RNAP I transcribes ribosomal RNA genes; RNAP II synthesizes heterogeneous nuclear RNA; and RNAP III transcribes the transfer RNA, 5SRNA, and small nuclear RNA genes. It is not known with certainty whether any of these RNAPs make use of a matchmaker for initiation. However, evidence exists that both RNAP II and RNAP III make use of strategies very similar to matchmaking reactions. In the case of RNAP II, of the five basal initiation factors, transcription factor IIE (TFIIE) is a DNA-dependent ATPase, and the conversion of the preinitiation to the initiation complex requires ATP hydrolysis (*31, 32*). Thus, TFIIE meets all the criteria of a matchmaker except that it has not been shown unambiguously to leave the complex [condition (v)] on conversion of the preinitiation complex to its active form (*30*).

Similarly, Kassavetis *et al.* (*33*) have shown that of the three transcription factors in the RNAP III system, the potential matchmakers TFIIIA and TFIIIC are required only to load TFIIIB to the promoter region and that once bound, TFIIIB can catalyze many rounds of transcription by RNAP III in the absence of TFIIIA and TFIIIC. However, there is no evidence yet that ATP is involved in the reaction [condition (III)], nor is it known whether dissociation of TFIIIA and TFIIIC from the preinitiation complex [condition (v)] is essential for initiation of transcription.

In contrast to the uncertainties with eukaryotic cellular RNAP, the use of a molecular matchmaker by the vaccinia virus RNAP appears to be unambiguous. Transcription by this RNAP requires separate transcription factors for early and late genes. One of these transcription factors, vaccinia early transcription factor (VETF), has all the characteristics of a molecular matchmaker. VETF is a DNA-dependent ATPase, binds both upstream and downstream of transcription initiation sites, bends DNA, and targets RNAP to the promoter. ATP hydrolysis by VETF helps transcription initiation by RNAP and promotes rapid dissociation of the VETF itself from the promoter (*34*).

Recombination

General genetic recombination is the exchange of fragments between homologous duplexes. The process is initiated by a single- or double-stranded break followed by strand invasion and formation of a covalent, four-stranded Holliday intermediate, which is then resolved by an endonuclease to two duplexes (*35*). In *E. coli*, reciprocal strand transfer and formation of the Holliday intermediate is catalyzed by the RecA ATPase whose function is stimulated by the SSB. Although the precise mechanism of stimulation of RecA by the SSB is controversial, recent studies suggest that it aids in threading single-stranded DNA (ssDNA) into the ssDNA groove on the RecA filament by causing ATP-dependent conformational change in RecA; after the formation of the RecA-ssDNA filament, SSB is released from the complex (*36*). In this model, SSB satisfies all the criteria of a matchmaker.

In *E. coli*, resolution of the covalent recombination intermediates (Holliday) by the RuvA, RuvB, and RuvC proteins also seems to make use of a matchmaker mechanism (*37*): RuvA binds specifically to the Holliday junction; RuvB is a weak ATPase; and RuvC cleaves the junction. Although the details of RuvABC reaction mechanism are not known, RuvA bound to a Holliday junction stimulates RuvB ATPase and cleavage of the junction by RuvC. A plausible model is that RuvA acts as a molecular matchmaker and targets RuvB to the junction. The binding of RuvB to the junction permits it to carry out branch migration using the energy of ATP hydrolysis and facilitates the binding of RuvC to RuvB-DNA and thus the resolution of the Holliday intermediate by the RuvC endonuclease (*37*). RuvA appears to be a matchmaker, except that it is not yet known whether RuvA dissociates from the RuvB-DNA complex [condition (v)] before incision by RuvC.

Transposition

Transposition is the movement of DNA elements of discrete size from one DNA site to another. In conservative transposition the element moves from its original site to another on the same or a different DNA molecule, whereas in replicative transposition a copy of the transposon is inserted elsewhere (*38*). Transposition is widespread in prokaryotes and eukaryotes.

From the standpoint of molecular matchmaking, two transposition systems in *E. coli* are of interest. One of these is the bacteriophage Mu, which undergoes transposition by both replicative and conservative modes (*38*). Two Mu-encoded proteins, MuA and MuB, and the host protein HU are required for in vitro transposition. MuA is the nuclease and MuB is a DNA- and MuA-dependent ATPase. The second example is the Tn7 transposon. In Tn7, transposition occurs by a conservative mechanism and involves the Tn7-encoded TnsA, TnsB, TnsC, TnsD, and TnsE proteins. Of these proteins, TnsA might carry the nuclease active site; TnsB binds to the transposon ends; TnsC is a DNA-dependent ATPase; and TnsD binds to the target sequence (*39*). ATP hydrolysis is required for transposition. Although the detailed mechanisms of Mu and Tn7 transpositions have not yet been elucidated, it is conceivable that MuB and TnsC ATPases act as molecular matchmakers to initiate the series of ordered protein interactions that lead to transposition.

Discussion

Molecular matchmakers, by acting on one or both reactants, make surfaces not readily available on these reactants by thermal fluctuation rapidly available for interactions. The matchmaker uses the energy released from ATP hydrolysis to affect the conformational change. ATP is hydrolyzed either by the matchmaker (the γ complex that loads the β clamp) or the matched protein (DnaB) activated by the matchmaker (DnaC); in some cases both the matchmaker (UvrA) and the matched protein (UvrB) are ATPases.

When is molecular matchmaking the mechanism of choice for the cell? We suspect that matchmaking is most frequently used under circumstances where there is a need for high-avidity DNA binding but where specificity conferred by DNA base sequence must be avoided. Thus, the matchmaking event, in addition to causing a conformational change in the matched protein, often also creates a conformational change in the DNA backbone that would have a negligible probability of occurring by thermal excitation at 37°C; nevertheless, once formed, this metastable conformation can rapidly and specifically interact with the matched protein. Although the resulting complex may be thermodynamically less stable than the corresponding B-DNA duplex and unbound protein pair, it is kinetically trapped by the large activation energy barrier.

Kinetic considerations. In many of the matchmaking reactions, the kinetics is a sensitive function of the structural context in which the matchmaking event is used. An example is the kinetic variability observed in damage removal by (A)BC excinuclease, which responds with highly variable kinetics depending on the chemical nature of the DNA damage. For example, psoralen monoadducts and cisplatin diadducts are repaired with greater kinetic efficiency than pyrimidine dimers. There is a similar kinetic variability in the mismatch repair reaction of MutHLS. In this case the variability is associated with the type of mismatch. Furthermore, in both of these systems, which are nominally sequence-independent, the sequence context of the damage or the mismatch has a moderate effect on the repair rate.

The matchmaking mechanism can account for such kinetic differences at three steps: formation of the matched pair, dissociation of the matchmaker, and the stability of the matched DNA-protein complex. Although in most cases the identification of a unique site on DNA (for example, mismatch, damage, or a promoter) will be limited only by diffusion and therefore rapid, the ATP-requiring reaction that creates the unique match could be very slow despite the availability of energy from ATP hydrolysis. Furthermore, this reaction would be expected to be sensitive to the chemical structures at the DNA site of the match. The second slow step might be the dissociation of the matchmaker from the matched partners. This step also is affected by the structural context of DNA, and the overall matchmaking reaction retains some reversibility until the matchmaker dissociates. The second step is rate limiting in the case of (A)BC excinuclease (*7, 12*). Finally, the matched

complexes may have variable dissociation rates.

Conclusion. Matchmaking is a general model for the complex events regulating some of the functions of DNA. In view of the diversity of reactions catalyzed by molecular matchmaking, more than one pathway exists for matchmaking reactions. Furthermore, in many instances the demarcation between binding by a matchmaking event and binding in complexes in which the role of one of the components is to increase the local concentration of the matched molecules might not be so well defined. Finally, in this paper we have only considered matchmaking events in which one of the partners is DNA and the other is a protein. The value of extending the notion of molecular matchmaking to other pairs of interacting molecules, such as RNA-protein, RNA-DNA, and macromolecule–small ligand, remains to be explored. For example, the prokaryotic translation initiation factor IF2 and the elongation factor EF-Tu meet all the criteria of a molecular matchmaker except that they hydrolyze guanosine triphosphate (GTP) instead of ATP. We wish to state, however, that in applying the molecular matchmaking concept to multicomponent reaction systems, the five criteria we have outlined should be satisfied. Otherwise, the model loses its rigor (*40*).

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39. P. Gamas and N. L. Craig, *Nucleic Acids Res.* **20**, 2525 (1992).
40. D. Ringe [*Curr. Biol.* **2**, 545 (1992)] used the term molecular matchmaker to denote ligands such as cyclosporin or GTP that promote macromolecular association not by allosteric effect but by acting as "double-sided sticky tape" and being an actual part of the binding surface. Although we recognize the usefulness of new terminology for these types of ligands, we suggest that the term molecular matchmaker should be reserved for macromolecules that match two partners and then dissociate, as was originally proposed (*7*).

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