FEBS Letters 586 (2012) 3324–3329

FEBS Letters

journal homepage: www.FEBSLetters.org

The link between restriction endonuclease fidelity and oligomeric state: A study with Bse634I

Mindaugas Zaremba [1], Giedrius Sasnauskas [1], Virginijus Siksnys *

Institute of Biotechnology, Vilnius University, Graiciuno 8, LT-02241 Vilnius, Lithuania

ARTICLE INFO

Article history:
Received 5 April 2012
Revised 2 July 2012
Accepted 6 July 2012
Available online 22 July 2012

Edited by Miguel De la Rosa

Keywords:
Protein–DNA interaction
Specificity
DNA cleavage
Allostery

ABSTRACT

Type II restriction endonucleases (REases) exist in multiple oligomeric forms. The tetrameric REases have two DNA binding interfaces and must synapse two recognition sites to achieve cleavage. It was hypothesised that binding of two recognition sites by tetrameric enzymes contributes to their fidelity. Here, we experimentally determined the fidelity for Bse634I REase in different oligomeric states. Surprisingly, we find that tetramerisation does not increase REase fidelity in comparison to the dimeric variant. Instead, an inherent ability to act concertedly at two sites provides tetrameric REase with a safety-catch to prevent host DNA cleavage if a single unmodified site becomes available.

1. Introduction

The Type II REases recognise short, usually palindromic sequences of 4-8 bp and cleave DNA within or near their recognition sites [1]. From a mechanistic point of view, the high specificity of REases is achieved first by making numerous specific contacts with a DNA target sequence and then by securing tight coupling between recognition and catalysis to prevent cleavage at non-cognate DNA sites [1]. Typically, palindromic DNA sites are recognised and cleaved by homodimeric Type IIP REases that share the dyad symmetry with the target sequence and use two active sites to introduce a double-stranded break in DNA. These cross-talking interactions between individual enzyme subunits interacting with separate DNA half-sites ensure that DNA cleavage is triggered only when all base-specific contacts have occurred [1]. This mechanism allows for discrimination between the specific recognition site and one differing by 1 bp by a factor of 10^6 or more [2].

Though homodimeric arrangement seems to be optimal for palindromic DNA sites, many Type IIP REases (e.g., SfiI, NgoMIV, Bse634I and Cfr10I) are homotetramers composed of two primary dimers and are, therefore, capable of simultaneous binding of two recognition sites [3,4]. Moreover, some enzymes like SgrAI or Ecl18kI that are dimers in solution form transient tetramers upon DNA binding [5,6]. The catalytic activity of tetrameric (or Type IIF) REases is regulated by allosteric interactions at the tetramerisation interface. If a single copy of the recognition site is present, the dimer not bound to DNA inhibits the activity of the DNA-bound dimer. In contrast, concomitant DNA binding by both dimers results in rapid and concerted cleavage at both recognition sites [4,5,7–9].

The relationship between the oligomeric structure and REase function is not yet clear. It has been shown in the cases of Bse634I and SfiI that the tetrameric state contributes to the stabilisation of the functional dimeric unit [10,11]. This finding is not surprising because both Bse634I and SfiI exhibit optimal activity at elevated temperatures (50–60 °C). Despite of the fact that some REases indeed achieve stabilisation of the functional dimer via tetramerisation, the latter strategy is not unique. For example, PspGI REase functions as a dimer and retains its catalytic activity up to 90 °C [12].

Alternatively, it has been proposed that tetramerisation contributes to the fidelity of the archetypal tetrameric REase SfiI [11,13]. Binding studies revealed that SfiI binds to a single cognate or non-cognate site; however, the catalytically competent synaptic complex is formed only by bridging two copies of cognate DNA [13]. Previous studies have shown that even if wt SfiI forms a syn-

Abbreviations: REase, restriction endonuclease; wt, wild-type; SC, supercoiled; OC, open-circular; FLL, full-length linear; FI, fidelity index; BSA, bovine serum albumin; DTT, dithiothreitol; Tris, tris(hydroxymethyl)aminomethane; EDTA, ethylenediaminetetraacetic acid

* Corresponding author. Fax: +370 5 2602116.
E-mail address: siksnys@ibt.lt (V. Siksnys).
[1] Joint first authors.

0014-5793/$36.00
http://dx.doi.org/10.1016/j.febslet.2012.07.009

aptic complex binding either two non-cognate or one non-cognate and one cognate site, cleavage at the alternative site is restrained unless the cross-talking interactions between the two primary dimers are disrupted, for example, by the Y68F mutation [11]. The linkage between tetramerisation and the fidelities of other homotetrameric REases remains to be established.

Type II REase Bse634I, which is specific for the DNA sequence 5′-RCCGGY-3′ (where R is a purine and Y is a pyrimidine) [4], provides a unique experimental platform to study the relationship between enzyme tetramerisation and DNA cleavage specificity. Currently, four biochemically characterised Bse634I variants exist (Table 1) that differ in their oligomeric structures and allosteric communication pathways: (i) the wt tetramer that displays two types of intersubunit communication signals: a 'stopper' inhibits cleavage at a single site, and a 'sync' promotes concerted cleavage upon binding of two recognition sites [10]; (ii) the dimeric W228A mutant that is similar to an orthodox homodimeric REase [7]; (iii) the tetrameric N262A mutant that lacks the 'stopper' signal and therefore cuts a single-site DNA efficiently [10]; and (iv) the tetrameric V263A mutant that retains the 'stopper' but lacks the 'sync' signal and therefore cleaves DNA slowly even if the enzyme binds two DNA copies [10]. To test whether binding of two recognition sites by tetrameric enzymes contributes to their fidelity, here we experimentally determined the fidelity index and non-cognate DNA cleavage rates for Bse634I REase in different oligomeric states.

2. Materials and methods

2.1. Oligoduplexes

All oligonucleotides used in this study were synthesised and HPLC purified by Metabion. 5'-ends of oligonucleotides were phosphorylated using T4 DNA polynucleotide kinase (Thermo Scientific). Oligoduplexes were assembled by slow annealing of complementary strands from 95 °C to room temperature.

2.2. Proteins

Wt Bse634I and its mutant variants were expressed and purified as described [10].

2.3. DNA cleavage experiments

Phage λ DNA (1 μg/50 μl) was incubated with varying protein amounts (0.1 nM to 5 μM in terms of monomer) in Reaction Buffer (10 mM Tris-HCl (pH 8.6), 100 mM KCl, 10 mM $MgCl_2$ and 0.1 mg/ml BSA) for 4 h at 25 °C. The final glycerol concentration (2% (v/v)) was adjusted in all reactions by addition of protein storage buffer (10 mM potassium phosphate (pH 7.4), 100 mM KCl, 2 mM DTT, 0.1 mM EDTA and 50 % (v/v) glycerol). Reactions were quenched with phenol/chloroform, samples were mixed with loading dye and analysed by agarose gel electrophoresis. pUC0 plasmid (2.3 nM) was incubated with 5 μM enzyme (in terms of monomer) at 25 °C in Reaction Buffer containing 2% (v/v) glycerol. The reactions either lacked oligonucleotides or contained 1 μM 'cognate', 'double product' and non-phosphorylated 'double product' oligoduplexes or 2 μM 'product' oligoduplex. Reactions were quenched at timed intervals with phenol/chloroform, and cleavage products were analysed and quantified as described [7,10].

3. Results

First, we examined the specificity of the four characterised Bse634I variants using the conventional phage λ assay. Phage λ DNA, which contains 61 Bse634I recognition sites, was incubated with various enzyme concentrations (0.1 to 5000 nM in terms of monomer) for 4 h at 25 °C, and reaction products were analysed by agarose gel electrophoresis. Under these conditions, wt Bse634I and the N262A mutant provided a specific fragmentation pattern at ~20 nM concentrations (Fig. 1). The dimeric W228A mutant and the tetrameric V263A mutant required 25-fold higher (500 nM) protein concentrations to achieve specific phage λ DNA fragmentation. Further increases in protein concentration resulted in star activity of wt Bse634I and the N262A mutant; however, the dimeric W228A mutant and the tetrameric V263A mutant displayed no star activity, even at the highest enzyme concentration tested (Fig. 1). To quantify star activity, we determined the fidelity index (FI), defined as the ratio of the maximum enzyme concentration showing no star activity to the minimum enzyme concentration needed for complete digestion at the cognate recognition sites [14]. The calculated FIs for wt Bse634I and the N262A mutant under experimental conditions tested were 100 and 5, respectively (Fig. 1). Thus, disruption of the 'stopper' signal in the tetrameric N262A mutant results in a 20-fold decrease in enzyme fidelity. The FI values for the W228A and V263A mutants could not be determined due to a lack of star activity even at the highest affordable enzyme concentration (5000 nM in terms of monomer) (Fig. 1). Nevertheless, the estimated lower limit of FI for the W228A mutant ($\geqslant$10, Fig. 1) indicates that complete disruption of the tetramerisation interface in this mutant is less deleterious to enzyme fidelity than removal of the 'stopper' signal in the N262A tetramer.

The star activity of Bse634I and its mutant variants imply that cleavage occurs at alternative sites that differ from the canonical recognition sites. To monitor reactions at the non-cognate sites, we employed the pUC0 plasmid that is free of cognate Bse634I sites but contains ~50 DNA sequences differing by 1 bp from the canonical Bse634I site, RCCGGY [7]. Cleavage reactions were performed using the highest possible enzyme concentration (5000 nM monomer). Although these reaction conditions are not physiological, high protein concentrations, however, are absolutely necessary to monitor DNA cleavage at non-cognate sites. Furthermore, at large excess of protein with respect to DNA (1000:1), the tetrameric variants of Bse634I will presumably bind to a single non-cognate site, and no synaptic complexes between two non-cognate target sites will be formed [7]. Therefore, pUC0 plasmid

Table 1
Proteins, oligomeric structures and catalytic features.

Protein	Oligomeric state	DNA cleavage		Allosteric communication signals between the primary dimers
		1 site	2 sites	
wt Bse634I	Tetramer	Slow	Fast	'sync'[a] and 'stopper'[b]
W228A	Dimer	Fast	Fast	–
N262A	Tetramer	Fast	Fast	'sync'[b]
V263A	Tetramer	Slow	Slow	'stopper'[a]

[a] A 'sync' signal promotes concerted and fast DNA cleavage upon binding of two recognition sites by a tetramer (analogous to positive cooperativity).
[b] A 'stopper' signal inhibits DNA cleavage when a tetramer is bound to a single recognition site.

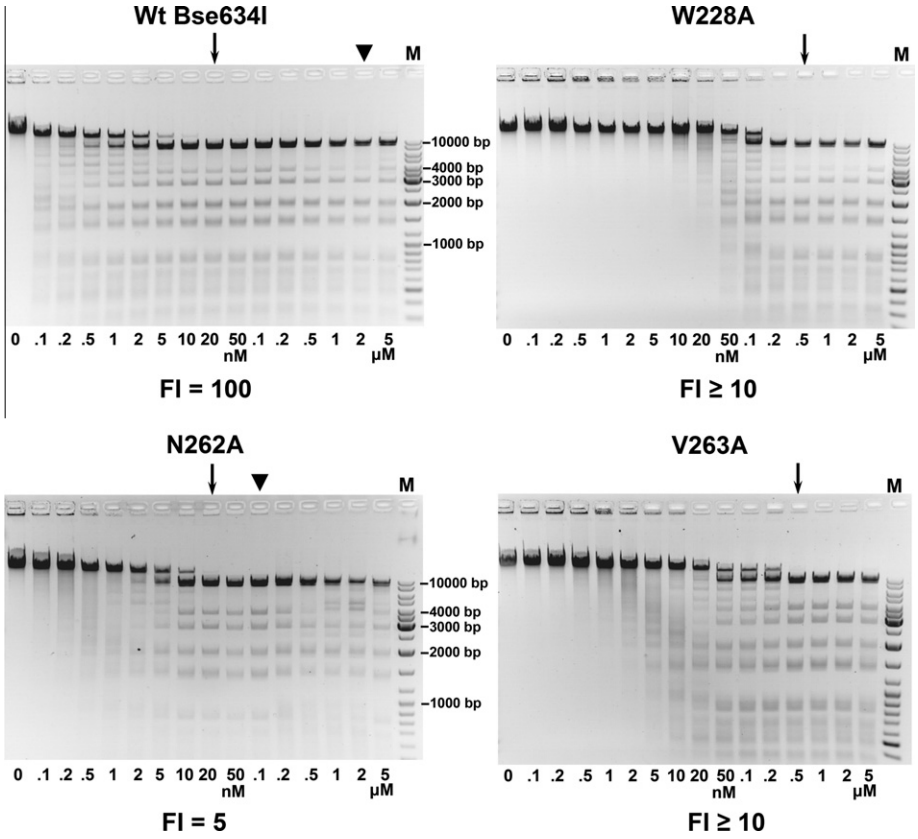

Fig. 1. Phage λ DNA cleavage by wt Bse634I and the Bse634I mutants W228A, N262A and V263A. Reactions were performed as described in Section 2. Final concentrations of proteins are below each lane. The fidelity index (FI) is defined as the ratio of the maximal enzyme concentration showing no star activity (indicated by a downward-facing triangle) to the minimal enzyme concentration needed for complete digestion at the cognate recognition sites (indicated by an arrow). Gel lanes "M" contain DNA length marker (300–10000 bp). The ~4 kb fragment is formed due to annealing of the terminal 1.9 and 1.8 kbp λ DNA fragments via the sticky *cos* ends.

cleavage under these reaction conditions should report the activity of Bse634I and its variants acting on individual copies of non-cognate DNA. Because wt Bse634I is able to form various mixed synaptic complexes (e.g., cognate–non-cognate, cognate–product DNA) [7], we also studied pUC0 cleavage reactions in the presence of 1000 nM of cognate oligoduplex ('cognate' DNA), 2000 nM oligoduplex with one sticky end mimicking the reaction product ('product' DNA), or 1000 nM oligoduplex with two sticky ends ('double product' DNA, Fig. 2a, Supplementary Table S1). The latter oligoduplex resembles DNA fragments that may be formed during Bse634I reactions on DNA containing multiple cognate sites (e.g., phage λ DNA). In addition to the phosphorylated 'double product' oligoduplex, we also employed its non-phosphorylated variant because a similar oligoduplex activated the star activity of the SgrAI REase [15]. To quantify the reactions, we determined the apparent first order rate constant for the conversion of the supercoiled plasmid substrate into nicked products (Fig. 2b and Supplementary Table S2).

3.1. Wt Bse634I tetramer

Under the experimental conditions tested, the wt enzyme slowly nicked pUC0 DNA (Fig. 2b). Non-cognate oligoduplex lacking a Bse634I recognition site had no effect on pUC0 cleavage (data not shown). However, addition of the 'cognate' or 'product' oligoduplexes increased the pUC0 cleavage rate ~2-fold. Because under these conditions, wt Bse634I rapidly converts the 'cognate' oligoduplex into 'product' DNA (data not shown), the observed activation in both cases must be due to formation of the mixed synaptic complexes involving a non-cognate site in one DNA binding cleft and the 'product' DNA in the second cleft. Surprisingly, more pronounced acceleration of plasmid cleavage was observed in the presence of the 'double product', especially the non-phosphorylated 'double product' oligoduplexes, despite the fact that concentrations of the product-mimicking ends were the same as in the experiments with 'product' DNA (Fig. 2b and Supplementary Table S2). Prolonged pUC0 incubation with the 'double product' oligoduplexes yielded a small amount of linearised plasmid, indicative of double-stranded DNA cleavage at non-cognate sites (Fig. 3). We propose that each primary dimer of Bse634I may simultaneously bind two product termini, thereby reconstituting the cleaved recognition sequence. On 'double product' oligoduplexes (but not on 'product' oligonucleotides), this may result in formation of large 3-D protein–DNA networks (Supplementary Fig. S1). If the pUC0 plasmid is captured within such an aggregate, it is cleaved more rapidly due to increased local protein concentration. In turn, stronger activation observed with the non-phosphorylated 'double product', in comparison to the phosphorylated 'double product' DNA, can be attributed to tighter REase binding to the product termini lacking the negatively charged 5′-terminal phosphates. It remains to be determined if the star activity of the related SgrAI REase observed in the presence of the 'double product' oligoduplex [15] is also mediated by protein–DNA aggregation.

3.2. W228A dimer

The W228A mutant nicked the pUC0 plasmid at the same rate as the wt enzyme (Fig. 2b, Supplementary Table S2). In contrast to wt Bse634I, none of the oligoduplexes stimulated cleavage of non-cognate DNA.

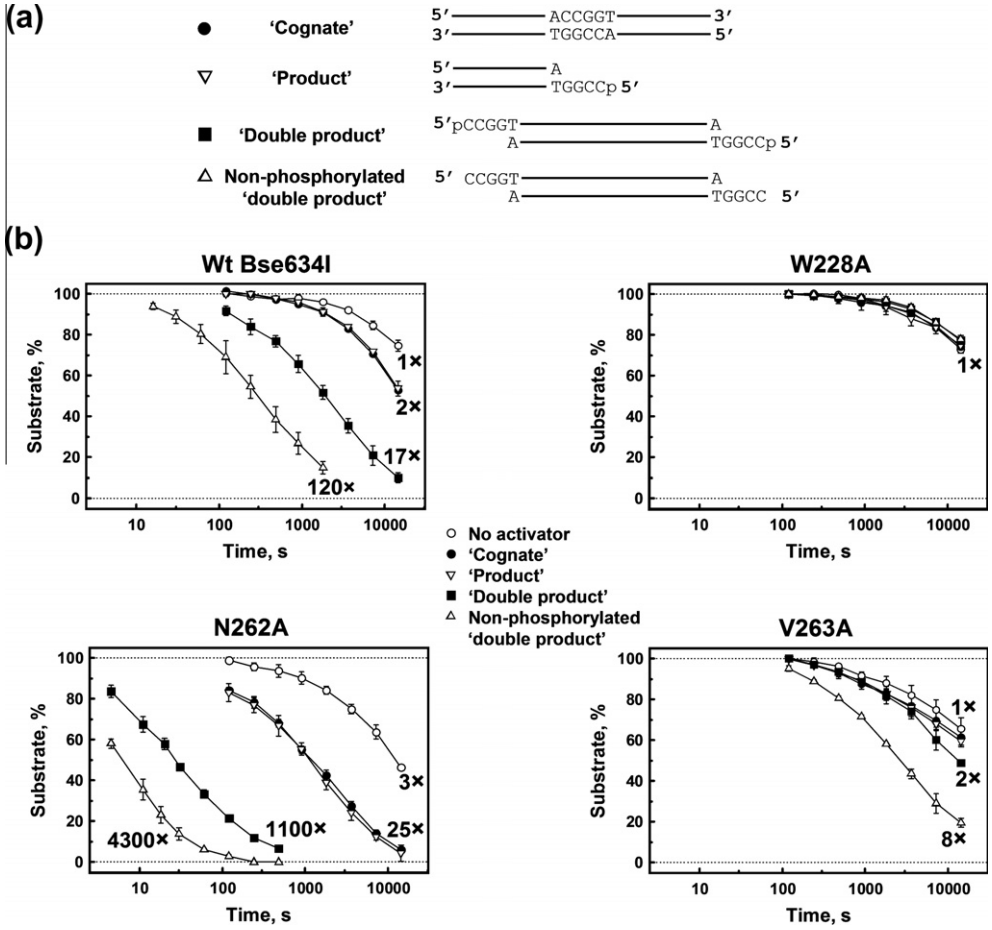

Fig. 2. Bse634I reactions on pUC0 plasmid DNA lacking cognate recognition sites. (a) Schematic representation of oligoduplexes used in this study. The Bse634I recognition sequence and the sticky ends mimicking the Bse634I cleavage product are shown in text format. Oligoduplex sequences are presented in Supplementary Table S1. (b) Cleavage of the supercoiled pUC0 DNA by wt Bse634I, the homodimeric W228A mutant, and the tetrameric N262A and V263A mutants. The reactions either lacked oligonucleotides (open circles) or contained 'cognate', 'double product' and non-phosphorylated 'double product' oligoduplexes (filled circles, filled squares or upward-facing triangles, respectively) or 'product' oligoduplex (downward-facing triangles). For clarity, only depletion of the supercoiled plasmid form is shown. All data points are presented as the mean values from 3 independent experiments ± 1 SD. Numbers on graphs indicate the cleavage rates normalised to the wt Bse634I reaction in the absence of any oligoduplex. Rate constants are provided in Supplementary Table S2.

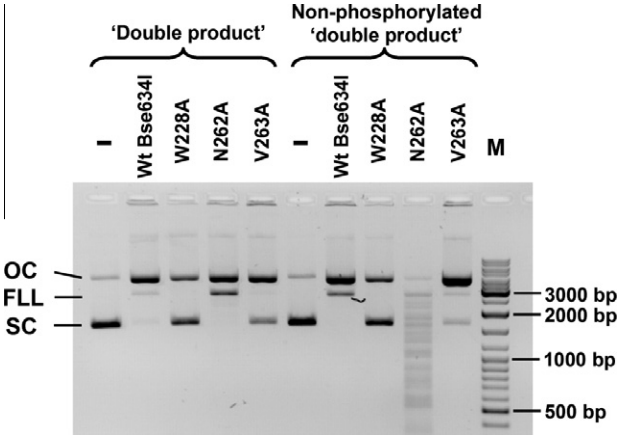

Fig. 3. Analysis of the pUC0 DNA cleavage products formed in the presence of 'double product' and non-phosphorylated 'double product' oligoduplexes. Plasmid DNA (2.3 nM) was incubated with 5 μM (in terms of monomer) wt Bse634I and its mutants in the presence of 1 μM oligoduplex for 4 h at 25 °C in. The electrophoretic mobilities of the supercoiled (SC), open-circular (OC) and full-length linear (FLL) forms of the DNA are marked on the left; lane "M" contains the 400–10000 bp DNA ladder (UAB Fermentas).

3.3. N262A tetramer

In the absence of oligoduplexes, the tetrameric N262A mutant cleaved the pUC0 plasmid ~3-fold faster than the wt enzyme (Fig. 2b, Supplementary Table S2). Addition of either the substrate or product oligoduplex resulted in a more pronounced stimulation of DNA cleavage at non-cognate sites compared to the wt enzyme. For example, in the presence of the non-phosphorylated 'double product', the N262A mutant cleaved the pUC0 plasmid ~4300-fold faster than wt Bse634I (Fig. 2b). Prolonged incubation under these reaction conditions resulted in multiple linear DNA fragments, indicative of double-stranded DNA cleavage at multiple non-cognate sites (Fig. 3). The non-cognate oligoduplex had no effect on the plasmid cleavage rate (data not shown).

3.4. V263A tetramer

The V263A mutant cleaved the pUC0 plasmid at the same rate as the wt enzyme. However, contrary to the wt enzyme, the 'cognate' and 'product' oligoduplexes did not activate DNA cleavage at all, while 'double product' oligoduplexes stimulated cleavage to a much lesser extent (Fig. 2b, Supplementary Table S2). The maximum ~8-fold acceleration was observed in the presence of the non-phosphorylated 'double product' DNA.

4. Discussion

Cleavage data presented here show that the W228A mutant cleaves the pUC0 plasmid at the same rate as wt Bse634I and the V263A mutant but ~3-fold slower than the N262A mutant (Fig. 2b and Supplementary Table S2). Thus, the disruption of all communication pathways in the dimeric mutant W228A does not impair enzyme fidelity in comparison to the wt tetramer (both 'stopper' and 'sync' pathways present), the V263A mutant ('sync' impaired), or the N262A mutant ('stopper' impaired). In other words, tetramerisation does not per se increase DNA cleavage fidelity of the Bse634I primary dimer. Furthermore, the star activity of all tetrameric variants (wt, N262A and V263A) is stimulated, at least to some extent, by the 'cognate' or '(double) product' oligoduplexes (Fig. 2b, Supplementary Table S2). Thus, under certain reaction conditions (presence of 'product' DNA), tetramerisation may even promote Bse634I promiscuity.

The activation of non-cognate DNA cleavage by tetrameric Bse634I variants must be due to formation of mixed synaptic complexes with non-cognate DNA bound by one primary dimer and cleavage products bound by the second dimer. Moreover, this activation implies that allosteric communication signals are transmitted via the dimer–dimer interface. In the absence of allosteric regulation, product binding by one of the dimers should not accelerate cleavage of non-cognate DNA bound by the other dimer. The promiscuity of the tetrameric variants (N262A > wt Bse634I > V263A) in the presence of 'product' DNA implies that the 'stopper' and the 'sync' allosteric communication signals controlling cognate DNA cleavage by the wt tetramer are also operational during DNA cleavage at non-cognate sites. The 'sync' signal, responsible for rapid and concerted reactions of wt enzyme on two cognate DNA sites [10], also contributes to the acceleration of non-cognate DNA cleavage in the mixed synaptic complexes. Indeed, disruption of the 'sync' signal in the V263A mutant results in low non-cognate activity. In contrast, the 'stopper' signal that normally prevents hydrolysis at a single recognition target site by wt Bse634I also prevents activation of non-cognate DNA cleavage by product DNA. The counterbalance of the 'stopper' and 'sync' pathways results in the wt enzyme's moderate product activation (Fig. 2b). Conversely, the disruption of the 'stopper' pathway in the N262A mutant boosts the non-cognate activity in the presence of product ends (Fig. 2b). Intriguingly, a similar effect (rapid hydrolysis of single-site DNA and a significant decrease in fidelity) has been reported for the Y68F mutant of the tetrameric SfiI enzyme [11], suggesting that the Y68F substitution impaired a SfiI communication signal similar to the 'stopper' in Bse634I.

Tetrameric IIF REases interacting with two DNA sites belong to distinct nuclease families. For example, Bse634I, SgrAI and SfiI are PD-(D/E)XK family REases, while Cfr42I belongs to the unrelated GIY–YIG nuclease family [8]. Furthermore, the tetramerisation mechanisms of the related Bse634I and SgrAI enzymes are not conserved [4,16], suggesting that tetrameric REases have evolved independently multiple times. Despite their structural differences, SgrAI, SfiI and Bse634I cut non-cognate DNA in the presence of product termini [11,15], raising a question regarding the evolutionary advantage of the tetrameric REase that cuts non-cognate DNA in the presence of DNA cleavage products over a standard homodimeric variant.

It is likely that the inherent ability of the tetrameric REase to act concertedly at two recognition sites provides a safety-catch that prevents host DNA cleavage if a single unmodified site becomes available [17]. The tetrameric REase will effectively cleave multiple cognate sites present in the foreign DNA but will refrain from cutting the host genome if a single unmethylated site is accidentally formed. However, this host DNA protection mechanism misfires if cleavage at a single Bse634I site occurs. In this case, the Bse634I 'sync' pathway, which is usually responsible for acceleration of cognate DNA cleavage when bound to two cognate sites, stimulates cleavage of the non-cognate DNA in the mixed synaptic complex with product DNA. It is important to note that the star activity of wt Bse634I is amplified in the in vitro reactions (high protein and 'double-product' concentration). In the cell, star activity may be much less pronounced due to the lower protein and product termini concentration, and it would be restrained to generating DNA nicks that will be repaired by DNA ligase. Even after bacteriophage DNA cleavage into multiple fragments, it is likely that the resultant reaction products will not activate wt Bse634I to that the extent where double-strand breaks will be introduced at non-cognate sites of the host genome. In any case, the key requirement to prevent cellular DNA cleavage at accidentally formed unmethylated sites presumably outweighs the potential harm due to the partial loss of fidelity in the presence of reaction products.

A requirement for synapsis of at least two cognate sites for efficient DNA hydrolysis is common among REases, and this prerequisite is achieved by different molecular mechanisms. For example, Type I, Type III and Type IV REases are large multisubunit complexes that utilise NTP hydrolysis and one-dimensional movement along DNA to achieve communication between the two target sites [18]. The Type IIE enzymes (e.g., NaeI and EcoRII) contain two different DNA binding sites that both must be filled with cognate DNA to trigger cleavage at one of the sites [19,20]. Furthermore, the archetypal Type IIS REase FokI forms an active complex upon dimerisation of two DNA-bound monomers [21]. It remains to be determined whether the requirement for multiple recognition sites also provides a built-in safety-catch for Type IIE and IIS REases, thereby protecting host DNA.

Acknowledgement

This work was supported by the Research Council of Lithuania (Grant MIP-102/2010 to M.Z.).

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.febslet.2012.07.009.

References

[1] Pingoud, A., Fuxreiter, M., Pingoud, V. and Wende, W. (2005) Type II restriction endonucleases: structure and mechanism. Cell. Mol. Life Sci. 62, 685–707.
[2] Engler, L.E., Welch, K.K. and Jen-Jacobson, L. (1997) Specific binding by EcoRV endonuclease to its DNA recognition site GATATC. J. Mol. Biol. 269, 82–101.
[3] Vanamee, E.S., Viadiu, H., Kucera, R., Dorner, L., Picone, S., Schildkraut, I. and Aggarwal, A.K. (2005) A view of consecutive binding events from structures of tetrameric endonuclease SfiI bound to DNA. Embo J. 24, 4198–4208.
[4] Siksnys, V., Grazulis, S. and Huber, R. (2004) Structure and function of the tetrameric restriction enzymes in: Restriction Endonucleases (Pingoud, A., Ed.), pp. 237–259, Springer-Verlag, Berlin.
[5] Daniels, L.E., Wood, K.M., Scott, D.J. and Halford, S.E. (2003) Subunit assembly for DNA cleavage by restriction endonuclease SgrAI. J. Mol. Biol. 327, 579–591.
[6] Zaremba, M. et al. (2010) DNA synapsis through transient tetramerization triggers cleavage by Ecl18kI restriction enzyme. Nucleic Acids Res. 38, 7142–7154.
[7] Zaremba, M., Sasnauskas, G., Urbanke, C. and Siksnys, V. (2005) Conversion of the tetrameric restriction endonuclease Bse634I into a dimer: oligomeric structure-stability-function correlations. J. Mol. Biol. 348, 459–478.
[8] Gasiunas, G., Sasnauskas, G., Tamulaitis, G., Urbanke, C., Razaniene, D. and Siksnys, V. (2008) Tetrameric restriction enzymes: expansion to the GIY-YIG nuclease family. Nucleic Acids Res. 36, 938–949.
[9] Wentzell, L.M., Nobbs, T.J. and Halford, S.E. (1995) The SfiI restriction endonuclease makes a four-strand DNA break at two copies of its recognition sequence. J. Mol. Biol. 248, 581–595.
[10] Zaremba, M., Sasnauskas, G., Urbanke, C. and Siksnys, V. (2006) Allosteric communication network in the tetrameric restriction endonuclease Bse634I. J. Mol. Biol. 363, 800–812.

[11] Bellamy, S.R., Mina, P., Retter, S.E. and Halford, S.E. (2008) Fidelity of DNA sequence recognition by the SfiI restriction endonuclease is determined by communications between its two DNA-binding sites. J. Mol. Biol. 384, 557–563.
[12] Morgan, R., Xiao, J. and Xu, S. (1998) Characterization of an extremely thermostable restriction enzyme, PspGI, from a Pyrococcus strain and cloning of the PspGI restriction-modification system in Escherichia coli. Appl. Environ. Microbiol. 64, 3669–3673.
[13] Embleton, M.L., Williams, S.A., Watson, M.A. and Halford, S.E. (1999) Specificity from the synapsis of DNA elements by the SfiI endonuclease. J. Mol. Biol. 289, 785–797.
[14] Wei, H., Therrien, C., Blanchard, A., Guan, S. and Zhu, Z. (2008) The Fidelity Index provides a systematic quantitation of star activity of DNA restriction endonucleases. Nucleic Acids Res. 36, e50.
[15] Bitinaite, J. and Schildkraut, I. (2002) Self-generated DNA termini relax the specificity of SgrAI restriction endonuclease. Proc. Natl. Acad. Sci. USA 99, 1164–1169.
[16] Park, C.K., Joshi, H.K., Agrawal, A., Ghare, M.I., Little, E.J., Dunten, P.W., Bitinaite, J. and Horton, N.C. (2010) Domain swapping in allosteric modulation of DNA specificity. PLoS Biol. 8, e1000554.
[17] Bickle, T.A. and Kruger, D.H. (1993) Biology of DNA restriction. Microbiol. Rev. 57, 434–450.
[18] Dryden, D.T.F., Murray, N.E. and Rao, D.N. (2001) Nucleoside triphosphate-dependent restriction enzymes. Nucleic Acids Res. 29, 3728–3741.
[19] Huai, Q., Colandene, J.D., Topal, M.D. and Ke, H. (2001) Structure of NaeI-DNA complex reveals dual-mode DNA recognition and complete dimer rearrangement. Nat. Struct. Biol. 8, 665–669.
[20] Golovenko, D., Manakova, E., Tamulaitiene, G., Grazulis, S. and Siksnys, V. (2009) Structural mechanisms for the 5'-CCWGG sequence recognition by the N- and C-terminal domains of EcoRII. Nucleic Acids Res. 37, 6613–6624.
[21] Catto, L.E., Bellamy, S.R., Retter, S.E. and Halford, S.E. (2008) Dynamics and consequences of DNA looping by the FokI restriction endonuclease. Nucleic Acids Res. 36, 2073–2081.