Understanding Xylose Isomerase from *Burkholderia cenocepacia*: insights into structure and functionality for ethanol production

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Additional Figures
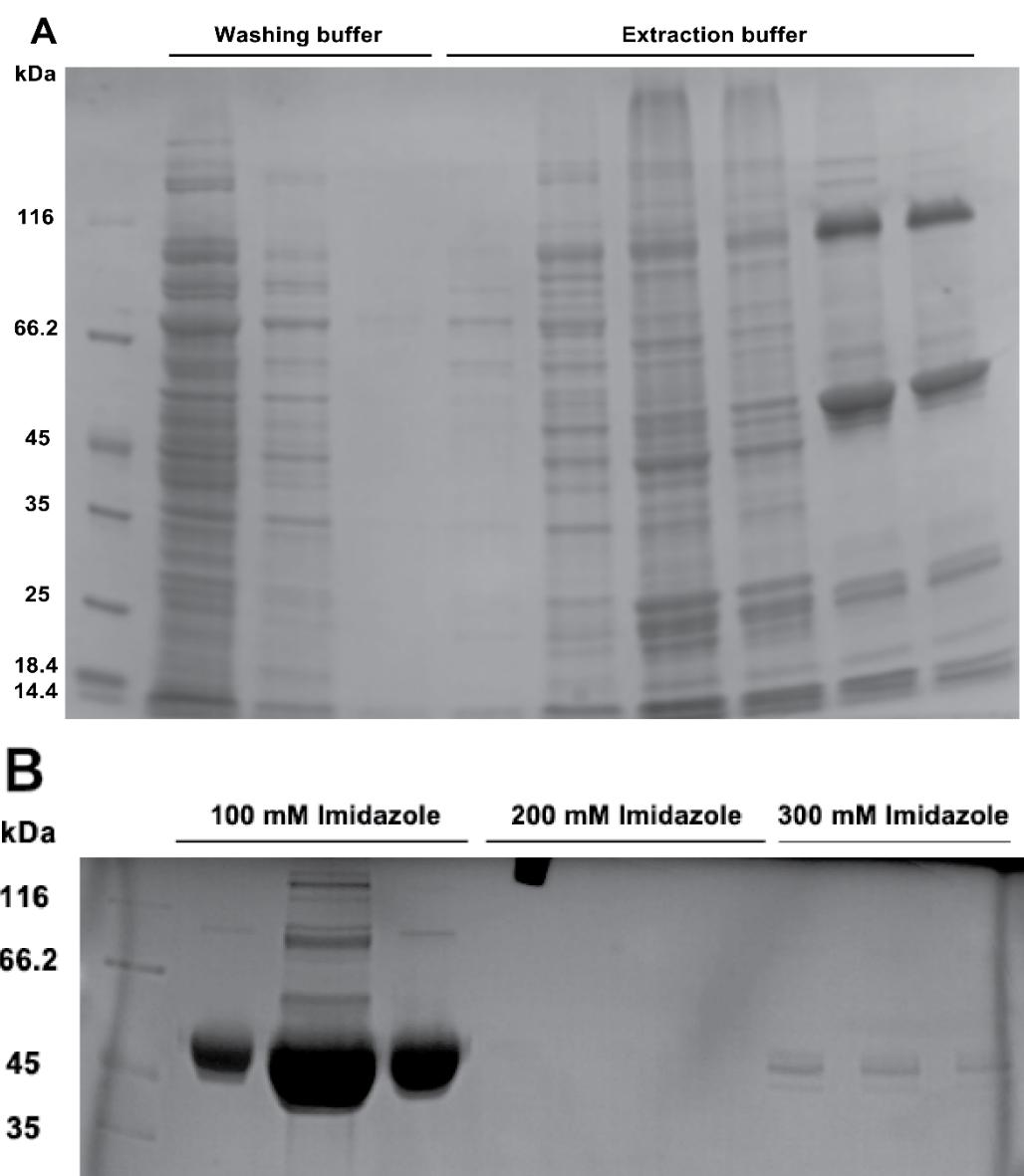
Figure S1. Protein extraction from *B. cenocepacia.* Only soluble proteins loaded onto the gel.
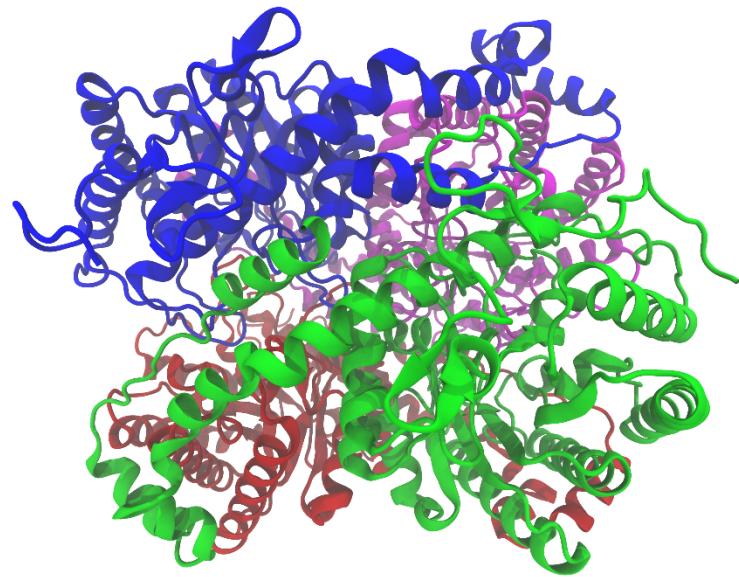
Figure S2. XylA predicted structure for *C. phytofermentans*.
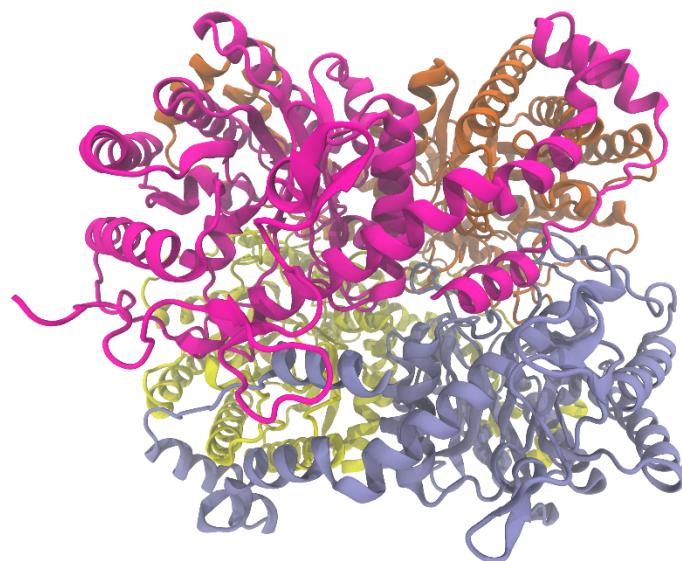
Figure S3. XylA predicted structure for *Piromyces sp.* E2.

Ramachandran Plot
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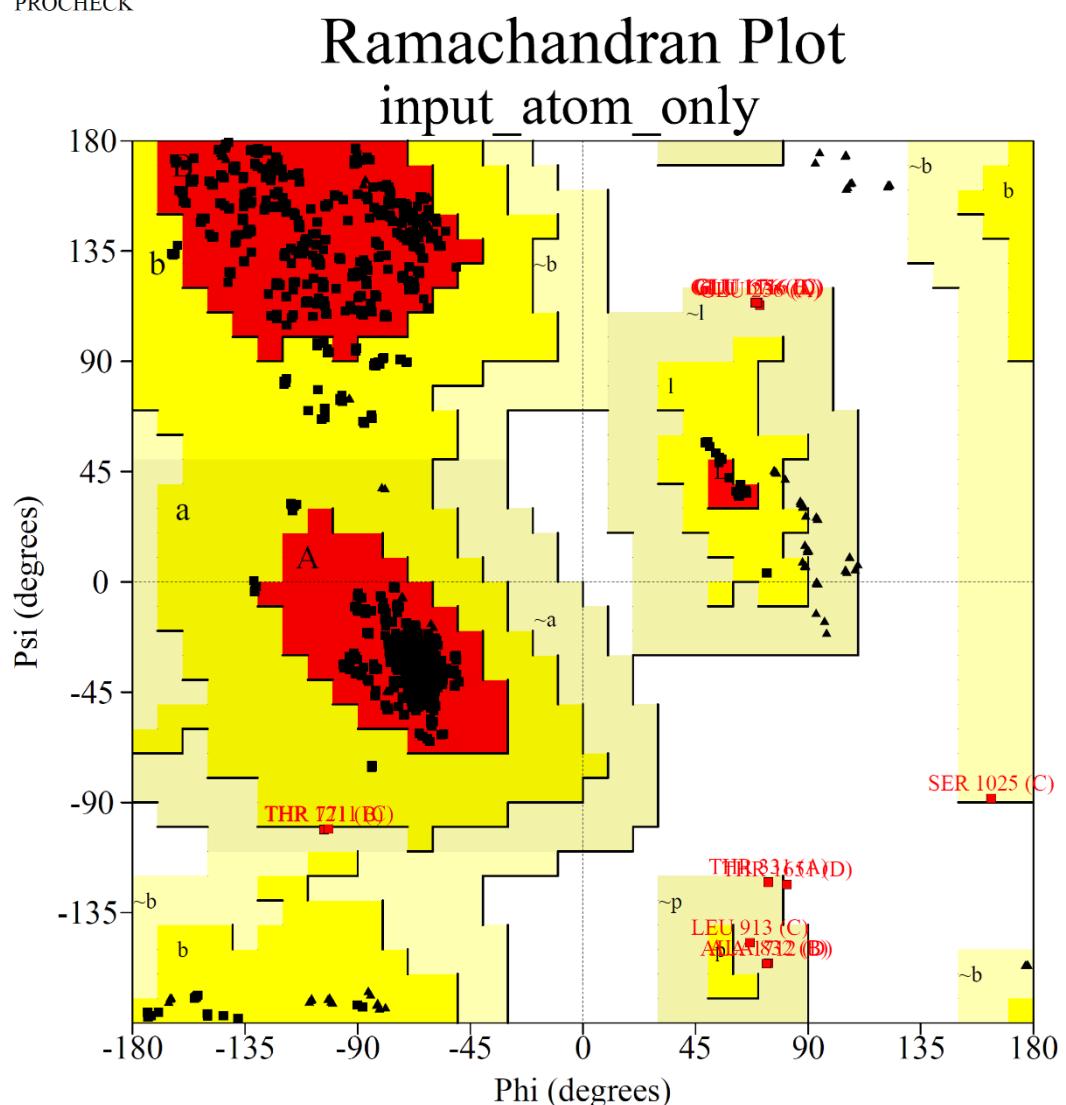
Figure S4. Ramachandran plot of *B. cenocepacia* XI theoretical model.

Ramachandran Plot
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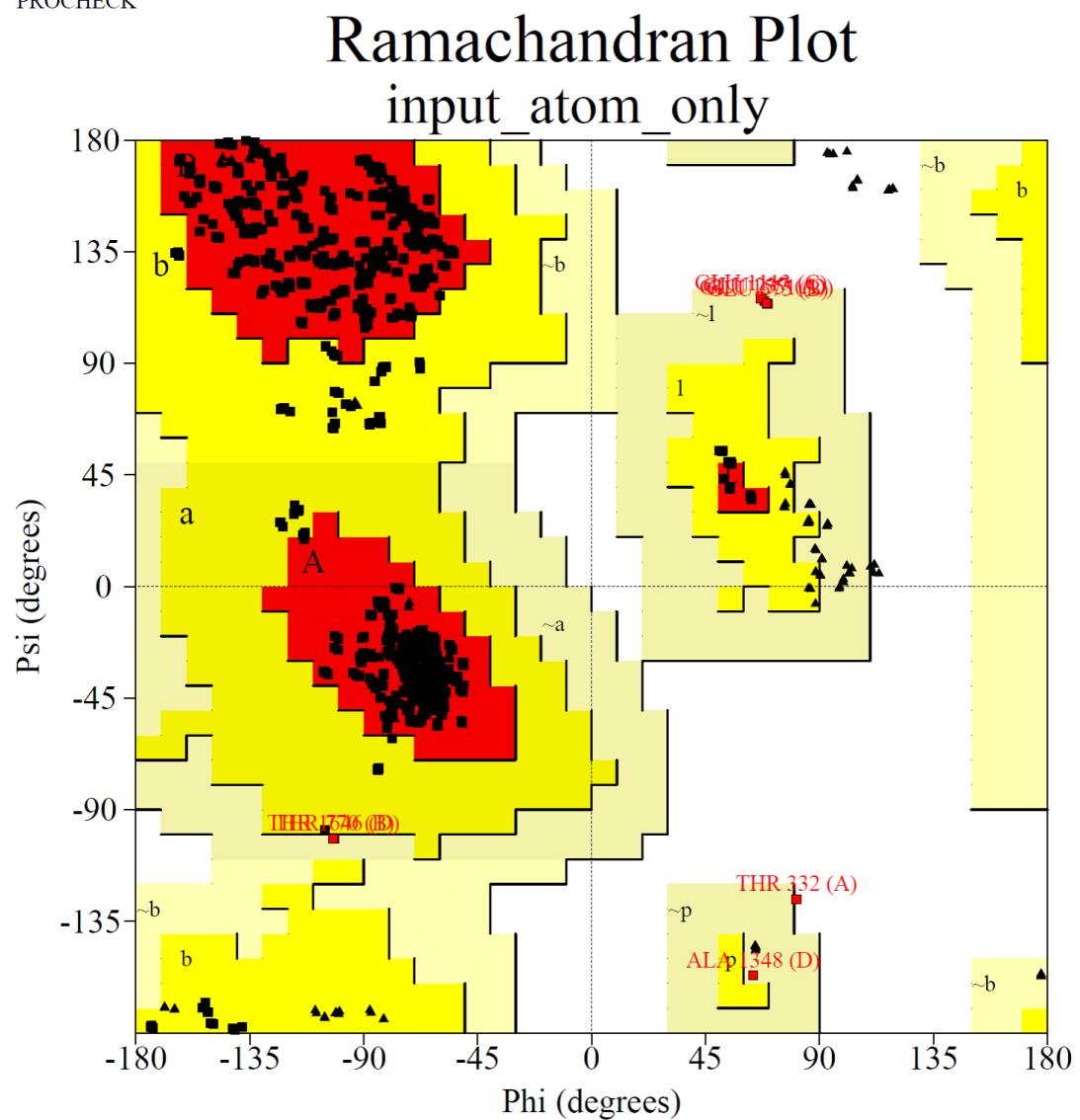
Figure S5. Ramachandran plot of *C. phytofermentans* XI theoretical model.

Ramachandran Plot
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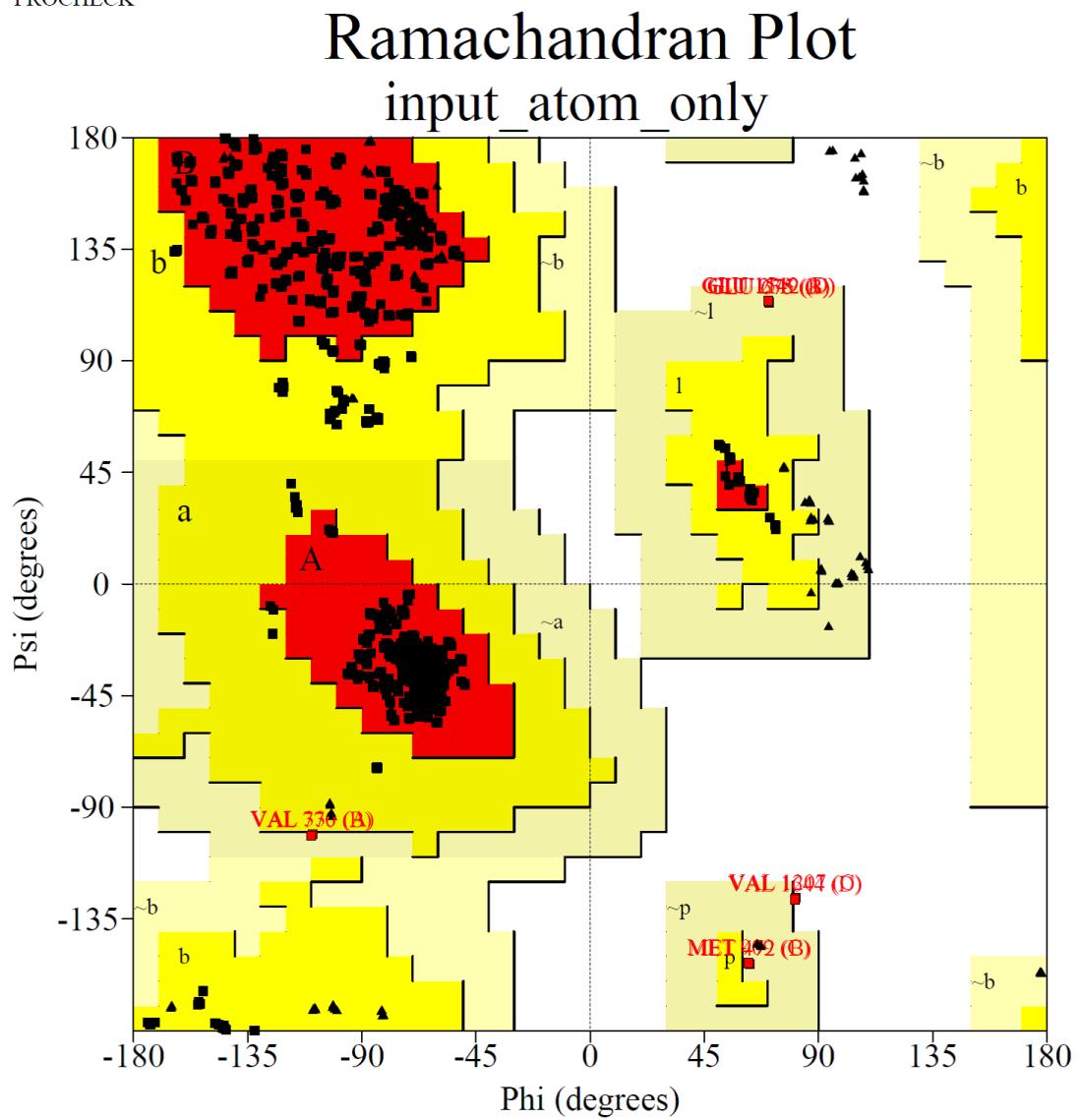
Figure S6. Ramachandran plot of *Piromyces sp.* XI theoretical model.
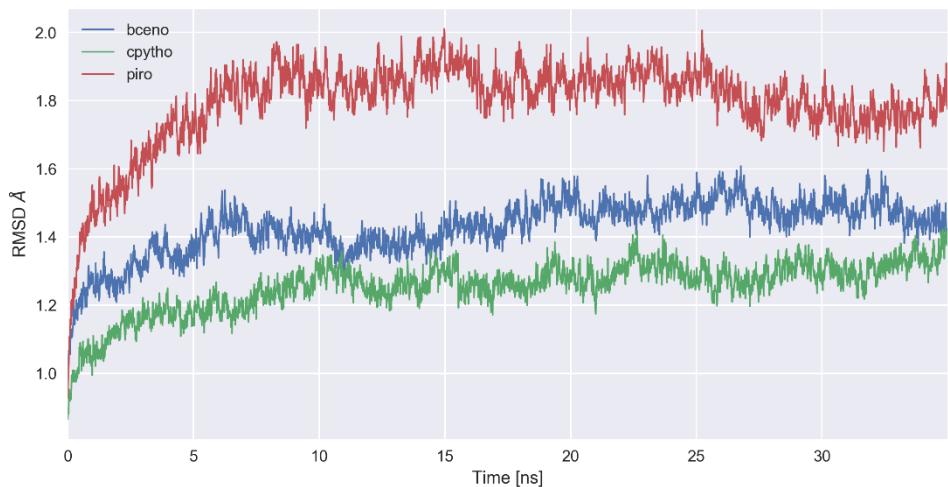
Figure S7. Global RMSD during dynamics. Blue: *B. cenocepacia*. Green: *C. phytofermentans*. Red: *Piromyces sp.*
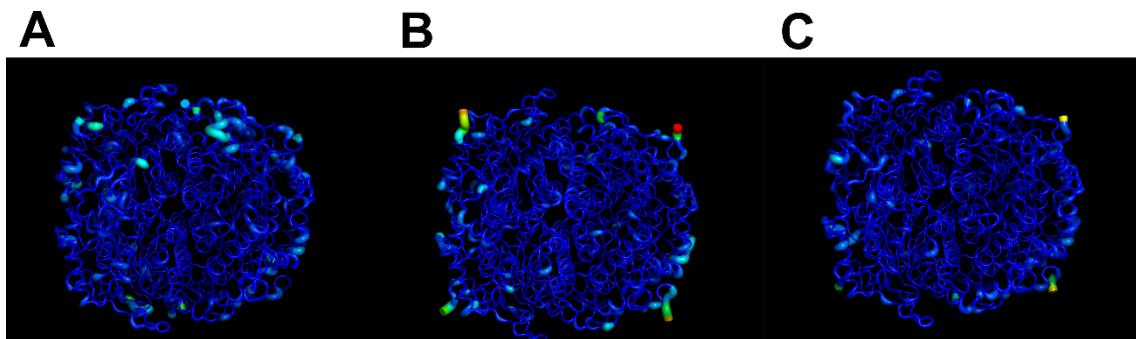
Figure S8. 3D RMS fluctuation. A) *B. cenocepacia.* B) *C. phytofermentans*. C) *Piromyces sp.* Structures are in a tube representation where thickness and color represents flexibility. Hot colors represent a higher fluctuation during dynamics simulation.
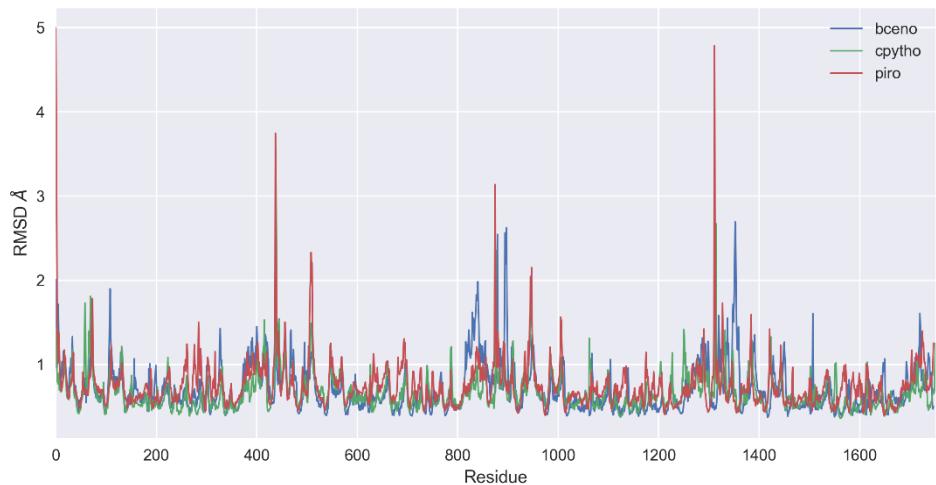
Figure S9. RMSF variation during dynamics simulation. Blue: *B. cenocepacia.* Green: *C. phytofermentans*. Red: *Piromyces sp*
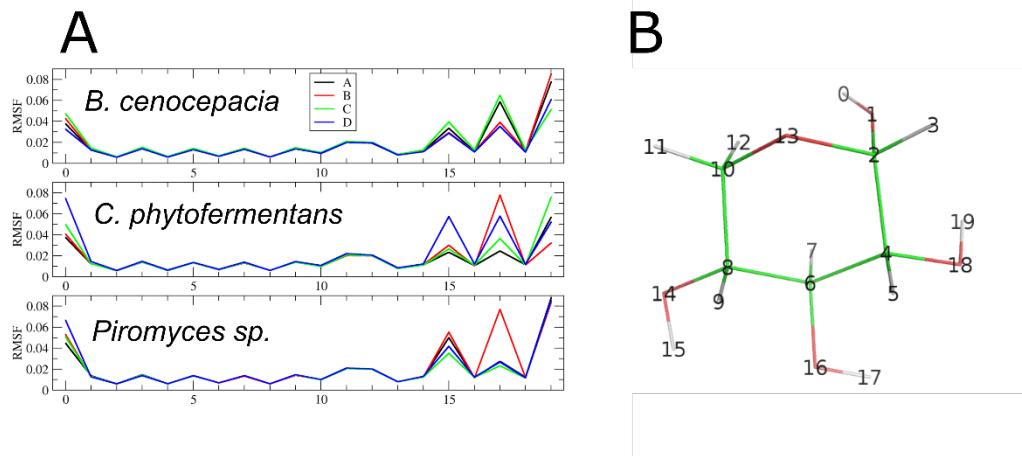
Figure S10. D-xylose RMSF inside the active site during dynamics simulation. A) RMSF for each xylose atom. B) D-xylose structure with each atom numbered.
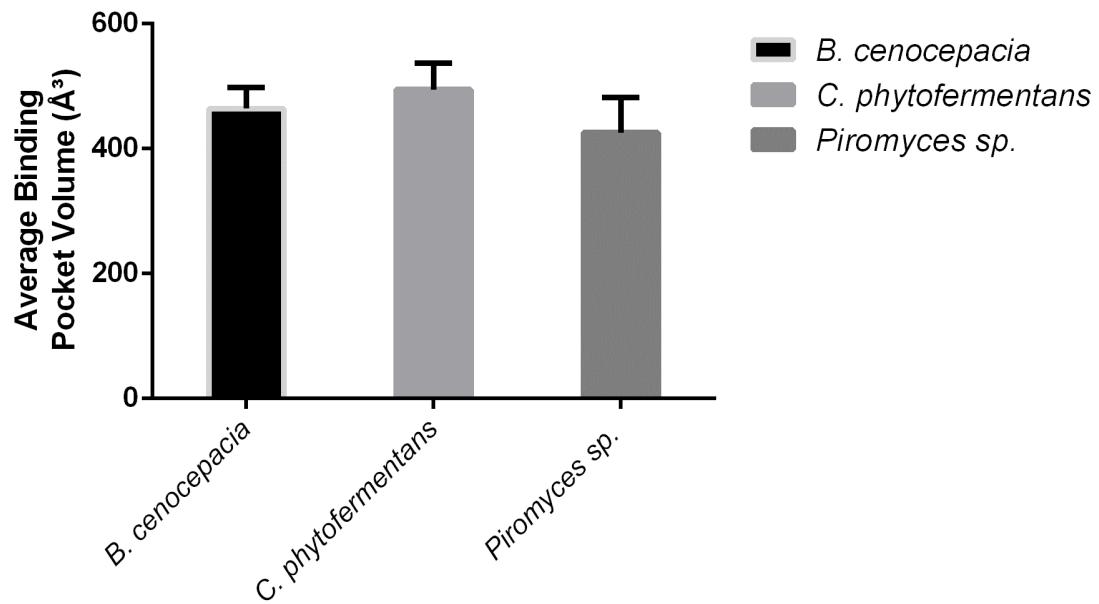
Figure S11. Binding site pocket volume. The volume of the protein binding site domain cavity was calculated from the overall average structure resulted from dynamics simulation. The column bars represent the standard error. Data shown are representative of an average of 4 domains from each protein.

CLUSTAL multiple sequence alignment by MUSCLE (3.8)

```
bcenocepacia_xyla        --MSYFEHIPAIRYEGPQSDNPLAYHHYDPDKRVLGKTLAEHLRIAVCYWHTFVWPGHDI   58
C_phytofermentans_xyla   -MKNYFPNVPEVKYEGPNSTNPFAFKYYDANKVVAGKTMKEHCRFALSWWHTLCAGGADP   59
piromyces_xyla           MAKEYFPQIQKIKFEGKDSKNPLAFHYYDAEKEVMGKKMKDWLRFAMAWWHTLCAEGADQ   60
                          .** ::  :.:** :* **:*:::**.:* * **.: :. *:*:..:***:   * *

bcenocepacia_xyla        FGQAAFRRPWQQPGDALERARMKADAAFEFFTKLGTPFYTFHDTDVAPEGDSLREYAANF   118
C_phytofermentans_xyla   FGVTTMDRTYGNITDPMELAKAKVDAGFELMTKLGIEFFCFHDADIAPEGDTFEESKKNL   119
piromyces_xyla           FGGGTKSFPWNEGTDAIEIAKQKVDAGFEIMQKLGIPYYCFHDVDLVSEGNSIEEYESNL   120
                         **  :    .: :  *.:* *. *.**.**:: ***   :: ***.*:..**::: *   *:

bcenocepacia_xyla        ARMVDYLGERQQASGVRLLWGTANLFSHPRFAAGAATNPNPDVFAWAATQVCHALDATHR   178
C_phytofermentans_xyla   FEIVDYIKEKMDQTGIKLLWGTANNFSHPRFMHGASTSCNADVFAYAAAKIKNALDATIK   179
piromyces_xyla           KAVVAYLKEKQKETGIKLLWSTANVFGHKRYMNGASTNPDFDVVARAIVQIKNAIDAGIE   180
                          :* *: *. . :*:.***.*** *.* *:   **:*. : **.* * .:: :*:**

bcenocepacia_xyla        LGGENYVLWGGREGYETLLNTDLKRERDQFARFLSMVVEHKHRIGFKGALLIEPKPQEPT   238
C_phytofermentans_xyla   LGGKGYVFWGGREGYETLLNTDLGLELDNMARLMKMAVEYGRANGFDGDFYIEPKPKEPT   239
piromyces_xyla           LGAENYVFWGGREGYMSLLNTDQKREKEHMATMLTMARDYARSKGFKGTFLIEPKPMEPT   240
                         **.:.**:******* .*****  * ::* ::.*. ::  . **.* : ***** ***

bcenocepacia_xyla        KHQYDYDVATVHGFLVQYGLQNEIRVNIEANHATLAGHSFHHEIANAFALGVFGSVDANR   298
C_phytofermentans_xyla   KHQYDFDTATVLAFLRKYGLEKDFKMNIEANHATLAGHTFEHELAMARVNGAFGSVDANQ   299
piromyces_xyla           KHQYDVDTETAIGFLKAHNLDKDFKVNIEVNHATLAGHTFEHELACAVDAGMLGSIDANR   300
                         ***** *. *. .**   :.*:::::.:***.*********.* **:* *   * :**:***.

bcenocepacia_xyla        GDPQNGWDTDQFPNSVEELTLAFYEILRHGGFTTGGMNFDAKVRRQSIDPEDLFYGHVGA   358
C_phytofermentans_xyla   GDPNLGWDTDQFPTDVHSATLAMLEVLKAGGFTNGGLNFDAKVRRGSFEFDDIAYGYIAG   359
piromyces_xyla           GDYQNGWDTDQFPIDQYELVQAWMEIIRGGGFVTGGTNFDAKTRRNSTDLEDIIIAHVSG   360
                         ** : ********  .  . . *  *::. ***..** *****.** * : :*:  .::..

bcenocepacia_xyla        IDVLALALERAAVLVENDRLDALRRQRYAQWDDAFGRKILAGGYTLESLAADALARGVDP   418
C_phytofermentans_xyla   MDTFALGLIKAAEIIDDGRIAKFVDDRYASYKTGIGKAIVDGTTSLEELEQYVLTHS-EP   419
piromyces_xyla           MDAMARALENAAKLLQESPYTKMKKERYASFDSGIGKDFEDGKLTLEQVYEYGKKNG-EP   420
                         :*.:* .* .** ::::.     :  :***.:. ..:*. :  *   :**.:     .. :*

bcenocepacia_xyla        QHASGAQERLENIVNQAIYGLR                                        440
C_phytofermentans_xyla   VMQSGRQEVLETIVNNILF--R                                        438
piromyces_xyla           KQTSGKQELYEAIV--AMY--Q                                        437
                          ** ** * **   ::  .
```

Similarity:
B. cenocepacia and *C. phytofermentans*: 52%
B. cenocepacia and *Piromyces sp*. E2: 50%
C. phytofermentans and *Piromyces sp*. E2: 54%

Figure S12. XylA protein sequence alignment. Arrows indicate substrate and metal binding sites.