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Supporting information for article:

Cloning, expression, crystallization and structure determination of the putative polysaccharide deacetylase Ba0331

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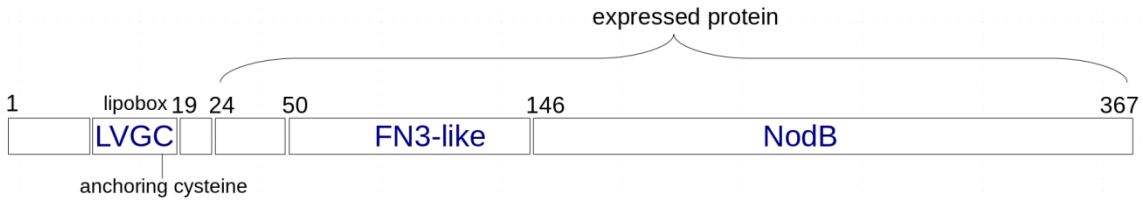

Figure S1 Linear representation of the Ba0331 sequence showing key features of the protein. On position 19 is located the membrane anchoring cysteine residue, while the targeting lipobox preceding Cys19 is shown explicitly. The portion of the protein that was expressed using recombinant technology is shown in curly bracket. The crystal structure revealed electron density for the range 50-367, covering the Fn3-*like* and the NodB domains.

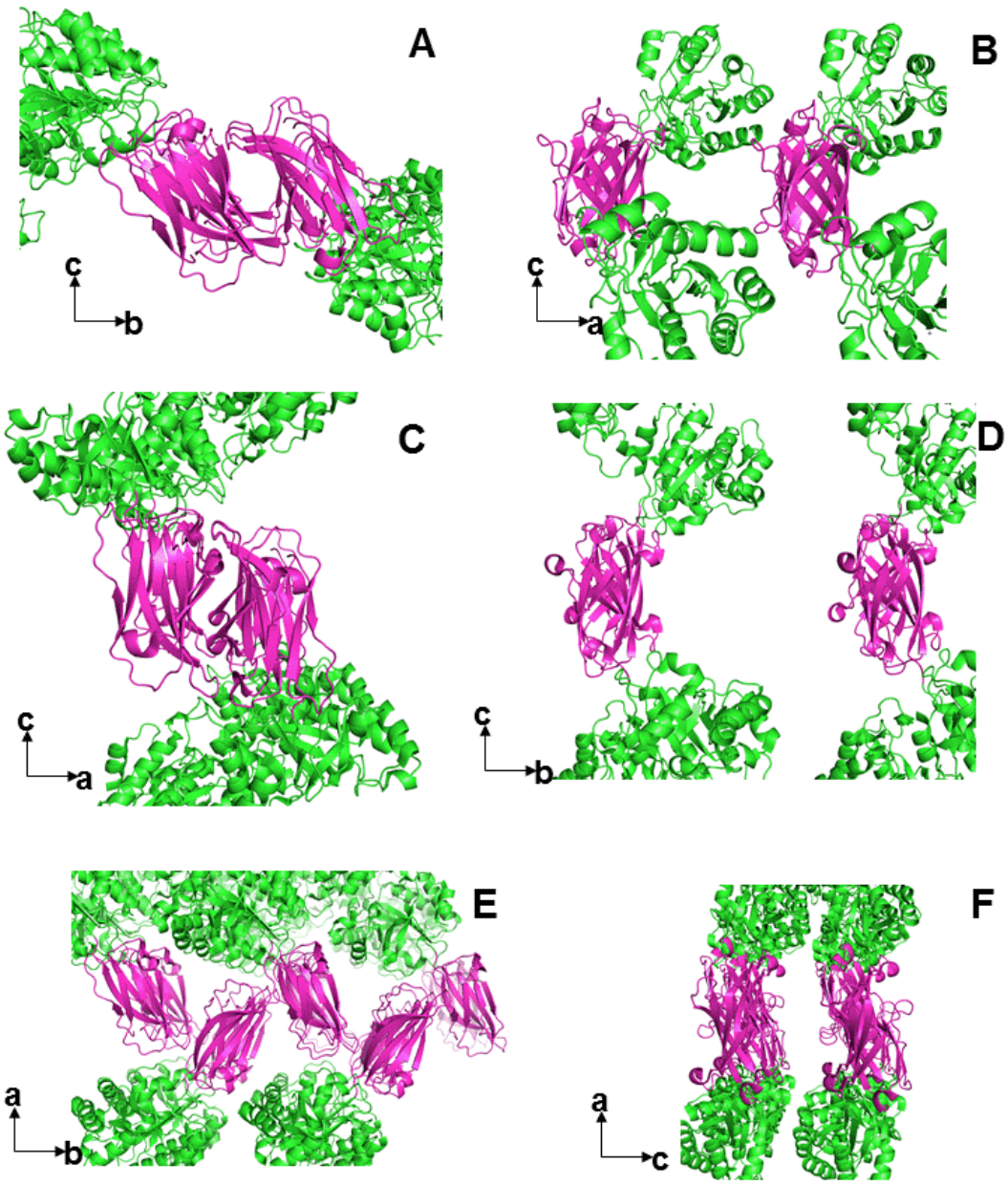

Figure S2 Crystal packing interactions of the Fn3-*like* domain in Ba0331 (**A** and **B**), in Ba0330 (**C** and **D**) and in Bc0361 (**E** and **F**).The three proteins have been crystallized in different space groups and their two domains are involved in distinct crystal packing contacts. Ba0331 was crystallized in P21 having two molecules in the asymmetric unit and the Fn3-*like* domain interacts with the corresponding domain of an antiparallel symmetry related molecule along axis **b**, forming an interface of 462.4 Å^2 as calculated by the PISA server (Krissinel and Henrick, 2007) (Fig. S2 a and b). Similarly for Ba0330, which however was crystallized in C2, the Fn3-*like* domain interacts with a symmetry related Fn3-*like* domain along axis **a** placed again in an antiparallel mode (Fig. S2 c and d). The interface area between the adjacent Fn3-*like* domains in Ba0330 structure was calculated 564.8Å^2. The crystal packing contacts in Bc0361,

which was crystallized in P21, differ substantially. Each Fn3-*like* domain interacts in an angled mode with two symmetry related Fn3-*like* domains placed on either side along the axis **b** and also with two NodB domains, forming interactions along the axis **a** (Fig.S2 e and f). As a result, the interface area with each of the Fn3-*like* adjacent molecules was calculated at 310.9 Å^2, and with the two NodB domains at 362.1 Å^2 in total. From the crystal packing analysis we conclude that the counterbalancing forces impose an additional strain to the Fn3-*like* domain resulting in rotational movements among the three proteins. Collectively, the initially thought structural rigidity of the PDA family's Fn3-*like* containing proteins seems to break in convenience of a relative interdomain flexibility.

Table S1 DynDom motion analysis

Ratio, ratio of interdomain to intradomain displacement; Rmsd, all atom rmsd on this domain; D1, fixed domain; D2, moving domain; RA, rotation angle; TR, translation; a_CM, angle between screw axis and line joining centres of mass; d_CM, distance between screw axis and line joining centres of mass;, distance closure. The Ba0330 and Bc0361 structures were compared to Ba0331 deducing the rigid-body movement of the Fn3 domain (considered as the moving domain; D2) relative to the NodB domain (fixed domain; D1). Default parameters were used for the analysis. Parametric equation of the screw axis for Ba0331- Ba0330 pair is (x,y,z) = (0.340, -0.043, 1.036) + t(-0.051, 0.0085, -0.025) while for the Ba0331-Bc0361 is (x,y,z) = (0.366, -0.048, 0.953) + t(-0.032, 0.010, -0.037) in the reference system of Ba0331 and expressed as fractional components of ***a***, ***b***, ***c*** unit cell parameters.

Ba0331-Ba0330								
	Ratio	**rmsd (Å)**		**RA (º)**	**TR (Å)**	**a_CM (º)**	**d_CM (Å)**	**CL (%)**
	1.022	D1	D2	11.03	-0.185	61.47	1.944	77.8
		0.998	1.223					
Ba0331-Bc0361								
	1.462	D1	D2	15.9	-0.346	73.63	4.148	92.055
		0.936	1.386					

Table S2 Main chain torsion angles in linker residues.

The phi (ϕ), psi (ψ) torsion angles for Ba0331, Ba0330, Bc0361 and $\Delta\phi$, $\Delta\psi$ of the superimposed residues for the pairs Ba0331-Ba0330 and Ba0331-Bc0361. The analysis shows that twisting for both pairs, as indicated by the $\Delta\varphi$ and $\Delta\psi$ values (shown in bold), is mainly due to φ and ψ changes of Thr144-Phe145 and Gln146-Gln147 (the number of residues are given for the Ba0331).

Ba0331						
	Thr144	Phe145	Gln146	Gln147	His148	Val149
ϕ(°)	–121.4	-112.3	-58.2	-110.9	-74.6	-129.2
ψ(°)	132.4	110.3	141.1	145.5	140.0	114.3
Ba0330						
	Thr139	Phe140	Glu141	Gln142	Lys143	Val144
ϕ(°)	–120.8	-96.50	-51.20	-91.60	-72.40	-132.50
ψ(°)	119.3	104.80	129.40	143.70	147.50	102.00
Bc0361						
	Thr139	Phe140	Glu141	Gln142	Lys143	Val144
ϕ(°)	–120.8	-96.50	-51.20	-91.60	-72.40	-132.50
ψ(°)	119.3	104.80	129.40	143.70	147.50	102.00
Ba0331 - Ba0330						
$\Delta\phi$(°)	0.6	**15.8**	7.0	**19.3**	2.2	-3.3
$\Delta\psi$(°)	**-13.1**	-5.5	**-11.7**	-1.8	7.5	-12.3
Ba0331 - Bc0361						
$\Delta\phi$(°)	-4.6	**22.7**	4.4	**8.0**	1.8	-2.7
$\Delta\psi$(°)	**-20.2**	-0.8	**-8.2**	6.9	7.5	-8.2