

Figure I. MS analysis of native PAF (**A**) and DTT/iodacetamide-treated PAF (**B**), respectively. Inset in (B): Coomassie-stained SDS-PAGE of DTT/iodacetamide-treated (reduced) PAF. Lane 1: Marker (Mark 12^{TM} MW Standard, Invitrogen), lane 2: reduced PAF (1 µg), lane 3: native PAF, control (1 µg). The arrow indicates MW 6 kDa.

Supplements

Figure II



Figure II. Fluorescent micrographs of an *A. nidulans* germling. (**A**) The localization of PAF was determined by indirect immunofluorescence staining using a rabbit anti-PAF serum and a FITC-conjugated goat anti-rabbit IgG. (**B**) The nuclei were visualized by DAPI staining. (**C**) Merge picture of (A) and (B). (**D**) Phase contrast of images (A-C).



Figure III. The assigned 700 MHz ¹H-¹⁵N HSQC NMR spectrum of PAF. Two signals (G21 and K15) are folded in ¹⁵N dimension.



Figure IV. Correspondence of the cysteines and disulfide bond assignments in AFP (top) and PAF (bottom). Patterns **abcabc** (7-36, 14-43, 28-54) and **abbacc** (7-36, 14-28, 43-54) in PAF have the related topologies in AFP as **abcdabcd** (7-33, 14-40, 26-49, 28-51) and **abbcadcd** (7-33, 14-26, 28-49, 40-51).



Figure V. Calculated S² values in the course of the MUMO dynamics simulations for the different disulfide pairings. On the x-axis the progress of calculations is monitored by the number of MD steps. On average, the mean value agrees well with the S² = 0.81 ± 0.05 measured experimentally by ¹⁵N relaxation NMR.



Figure VI. Overlayed mean structures of CA chains of PAF (blue) and AFP (yellow) with no disulfide bond constraints.

Table I. Selected NMR data of PAF as measured at 304K in H₂O/D₂O solution and described in the experimental part. T₁ and T₂ are the ¹⁵N auto-relaxation times, NOEDs are the ¹⁵N-{¹H} NOE difference values, η_{zzN} , η_{xyN} and η_{xyH} are the longitudinal (zz) and transversal (xy) ¹⁵N or ¹H CSA/DD cross-correlated relaxation rates, ³J_{HN,HA} stands for three-bond homonuclear spin-spin couplings determined from high-resolution HSQC spectra. S² order parameters were calculated from T₁, T₂ and NOEDs using the model-free method of Lipari and Szabo, supposed a constant –160 ppm ¹⁵N CSA value.

Residue	T ₁ (s)	NOED	T ₂ (s)	$\eta_{zzN}(s^{\text{-}1})$	$\eta_{xyN}(s^{-1})$	$\eta_{xyH}(s^{\text{-}1})$	³ J _{HNHA} (Hz)	S^2
2	0.4426	-0.5123	0.2464	1.4377	2.035	1.09	9.07	0.70481
3	0.3942	-0.4254	0.2269	1.4145	2.6738	1.69	10	0.78875
4	0.3967	-0.3925	0.2185	1.4484	2.7427	1.88	9.95	0.80836
5	0.3782	-0.3354	0.2186	1.389	2.9566	1.95	8.75	0.81927
6	0.3625	-0.3958	0.2149	1.6369	3.1508	1.5	9.06	0.84888
7	0.3599	-0.3953	0.2159	1.9151	2.9765	1.75	9.84	0.84961
8	0.3641	-0.3366	0.2182	1.6338	3.1082	1.43	10.12	0.8423
9	0.364	-0.4423	0.2084	1.8126	2.8262	1.31	4.7	0.85386
10	0.3821	-0.4489	0.2114	1.4128	2.4134	1.35	-	0.82871
11	0.4121	-0.4721	0.2264	1.3385	3.0202	0.42	7.21	0.7683
12	0.3713	-0.4026	0.1933	1.7049	3.0208	1.12	7.71	0.89169
13	0.3641	-0.3948	0.2097	1.7044	3.0075	1.06	10.2	0.85831
14	0.3684	-0.4036	0.217	1.7572	2.9338	1.87	9.38	0.83666
15	0.3576	-0.3588	0.2121	1.7687	3.1694	1.54	9.67	0.86627
16	0.3535	-0.4118	0.2147	1.8645	2.9911	1.25	8.74	0.85688

17	0.3938	-0.4044	0.232	1.4638	2.8614	1.43	8.68	0.78257
18	0.3816	-0.4731	0.2341	1.7433	3.0109	1.32	7.8	0.78015
19	0.4011	-0.4358	0.2326	1.3775	2.4198	0.85	-	0.77052
20	0.4091	-0.401	0.2404	1.6331	2.9479	0.9	8.42	0.75491
21	0.3841	-0.4522	0.229	1.5451	2.7981	1.78	6.64, 6.82	0.79015
22	0.38	-0.4691	0.229	1.8362	3.0296	1.2	8.77	0.79133
23	0.3847	-0.4259	0.225	1.6506	2.9587	1.57	4.5	0.80109
24	0.3846	-0.3959	0.2327	1.4488	2.4143	1.68	9.83	0.79165
25	0.3826	-0.373	0.2278	1.5915	2.737	1.79	9.8	0.8059
26	0.3687	-0.3595	0.2197	1.6848	2.8625	1.55	8.35	0.83802
27	0.3766	-0.3322	0.2239	1.6877	2.6263	1.43	-	0.81287
28	0.3747	-0.4245	0.2137	1.5646	2.7289	1.49	7.3	0.83401
29								
30	0.4008	-0.4336	0.2191	1.4711	2.528	1.49	6.79	0.79781
31	0.4841	-0.5484	0.2734	1.286	2.5083	0.3	5.77	0.63479
32	0.3939	-0.4235	0.2345	1.3577	2.4671	1.24	-	0.77526
33	0.3964	-0.4955	0.2268	1.3204	2.4363	1.03	7.25	0.77676
34	0.3863	-0.4639	0.2208	1.5974	2.7235	1.25	10.14	0.80215
35	0.3986	-0.4679	0.2231	1.5984	2.9248	0.73	6.8	0.78623
36	0.3714	-0.3986	0.2167	1.7788	2.8113	1.47	7.3	0.83505
37	0.3733	-0.4163	0.1799	1.815	3.01	1.87	8.54	0.9306
38	0.3671	-0.363	0.2041	1.7056	3.311	0.7	6.87	0.87311
39	0.379	-0.3298	0.2034	1.6326	2.9753	2	-	0.83348
40	0.3714	-0.3714	0.226	1.5981	2.758	1.59	6.66	0.82133
41	0.363	-0.4228	0.2116	1.8635	3.0201	0.87	6.43	0.85085
42	0.3747	-0.4245	0.2137	1.5646	2.7289	1.49	4.5	0.83401

43	0.3702	-0.3494	0.2322	1.6068	3.1634	1.07	7.44	0.8155
44	0.3718	-0.362	0.2125	1.7292	2.9213	1.78	9.86	0.84926
45	0.3662	-0.399	0.2176	1.7044	2.7489	1.64	9.98	0.83871
46	0.3847	-0.4259	0.225	1.6506	2.9587	1.57	9.6	0.80109
47	0.3866	-0.4297	0.2296	1.4118	2.6698	1.43	6.85	0.79006
48	0.3892	-0.4116	0.2306	1.772	3.0067	0.9	6.74	0.78835
49	0.3982	-0.4656	0.2322	1.1909	2.9927	0.88	9.32	0.76936
50	0.4053	-0.4066	0.2336	1.6562	3.0094	0.95	9.95	0.76927
51	0.4075	-0.4064	0.2316	1.6056	2.9401	0.89	7.87	0.77099
52	0.4004	-0.3924	0.2301	1.616	2.5234	1.43	10.1	0.78188
53	0.3713	-0.3982	0.2018	1.5731	2.8527	1.47	9.78	0.86944
54	0.3959	-0.4741	0.2221	1.2832	2.5605	1.67	9.6	0.78978
55	0.3858	-0.4263	0.22	1.6479	2.6611	1.05	8.3	0.81