

SUPPLEMENTARY MATERIAL

Table 1. Interactions of buried waters (networked as well as isolated) with secondary-structural elements in the protein.

Secondary structures involved in the water-mediated interactions	Number of waters involved	
	Networked buried waters (12)	Isolated buried waters (8)
α -helix - β -strand	-	1
β -strand - β -strand	2	1
$\beta\alpha$ loop - α -helix	1	1
$\beta\alpha$ loop - β -strand	6*	2
$\beta\alpha$ loop - $\beta\alpha$ loop	1	1
$\alpha\beta$ loop - α -helix	1	1
$\alpha\beta$ loop - β -strand	1	-
$\beta\alpha$ loop, α -helix and β -strand	-	1

* It may be seen that more networked buried waters are involved in bridging $\beta\alpha$ loops and β -strands.

Table 2. Buried water interactions in RTUX and CTUX**(a). Isolated buried waters**

Water residue number (as in CTUX)	Protein atoms	
	Main Chain	Side Chain
Wat501	Ile235 O (β_7)	Tyr17 OH (β_1) Thr265 OG1 (β_8)
Wat525	Pro45 O (β_2) Asn47 O ($\beta_2\alpha_2$) Met49 N ($\beta_2\alpha_2$) His83 O (β_3)	-
Wat504	Met49 O ($\beta_2\alpha_2$) Thr84 O (β_3)	His111 NE2 (α_3)
Wat538	Met116 O (α_3)	Trp126 NE1 (β_4) Asp164 OD2 (α_4)
Wat509	Ala132 O ($\beta_4\alpha_4$) Arg140 O ($\beta_4\alpha_4$)	Tyr152 OH (α_4)
Wat530	Asp164 O (α_4) Ala167 O ($\alpha_4\beta_5$)	Arg161 NH1 (α_4)
Wat506	Ala132 N ($\beta_4\alpha_4$) Asn172 O (β_5)	Asp173 OD2 (β_5)
Wat502	Gly268 O ($\beta_8\alpha_8$) Val269 O ($\beta_8\alpha_8$) Leu283 N ($\beta_8\alpha_8$)	-

The location in the secondary-structural element of the given residue is indicated in parentheses. Note that isolated buried waters are involved in more interactions (17) with main-chain atoms than with side-chain atoms (8). Also, among the interactions with the main-chain atoms, carbonyl oxygen is involved in 14 interactions and amide nitrogen in only three interactions.

(b). Buried waters which are networked with other waters

Water residue number (as in CTUX)	Protein atoms		Heteroatoms
	Main chain	Side chain	Water
Wat511	Thr236 O (β 7)	Thr44 OG1 (β 2) Asp81 NH1 (β 3)	Wat533
Wat533	-	Asn127 OD2 (β 4) Glu237 OE2 (β 7)	Wat511
Wat518	Trp51 N (β ₂ α ₂) Thr84 O (β 3)	-	Wat545
Wat545	Trp87 O (β ₃ α ₃) Ser89 O (β ₃ α ₃)	-	Wat518
Wat529	-	Thr142 OG1 (β ₄ α ₄) Tyr152 OH (α 4)	Wat514
Wat514	His88 N (β ₃ α ₃) Asn130 O (β 4) Glu131 O (β 4)	His88 ND1 (β ₃ α ₃)	Wat529
Wat515	-	Asp177 OD1 (β ₅ α ₅) Ser206 OG (β 6) Thr208 OG1 (β ₆ α ₆)	Wat535
Wat535	Leu176 O (β ₅ α ₅)	-	Wat515
Wat519	-	Tyr17 OH (β 1) Arg81 NE (β 3) Tyr170 OH (β 5)	Wat563
Wat503	Leu176 N (β ₅ α ₅) Thr208 N (β ₆ α ₆)	Asp177 OD1 (β ₅ α ₅) Ser206 OG (β 6)	Wat505
Wat558	Leu225 O (α 6) Thr230 N (α ₆ β ₇) Ala226 O (α 6)	Thr230 OG1 (α ₆ β ₇)	Wat579
Wat524	Trp267 O (β ₈ α ₈)	Asp239 OD2 (β 7) Ser274 OG (β ₈ α ₈)	Wat561

The location in the secondary-structural element of the given residue is indicated in parentheses. The first eight waters in the table define four water dimers. Note that networked buried waters are involved in more interactions (18) with side-chain atoms than with the main-chain atoms (15). Also among the interactions with the main-chain atoms, carbonyl oxygen is involved in ten interactions and amide nitrogen in five interactions.