ELSEVIER

Biophysical Chemistry 75 (1998) 105–113

Biophysical Chemistry

Analysis of hydrophobic and charged patches and influence of medium- and long-range interactions in molecular chaperones

T.S. Kumarevel[a], M. Michael Gromiha[b], M.N. Ponnuswamy[a,*]

[a] *Department of Crystallography and Biophysics, University of Madras, Guindy Campus, Chennai, 600 025 Tamil Nadu, India*

[b] *Tsukuba Life Science Center, The Institute of Physical and Chemical Research (RIKEN), 3-1-1 Koyadai, Tsukuba, Ibaraki 305, Japan*

Received 27 May 1998; received in revised form 17 August 1998; accepted 17 August 1998

Abstract

The amino acid composition of the aromatic residues Phe, Tyr and Trp are much less significant in chaperones and the residues Cys, Glu, His, Met and Pro vary significantly in chaperones compared to normal globular proteins. In the present work, we have analysed the hydrophobic and charged patches in molecular chaperones which provide more insight for a better understanding of chaperone folding. Also, we have investigated the role of medium- and long-range contacts in chaperones and the preference of amino acid residues influenced by these interactions. Furthermore, the role of hydrophobic and helix-forming residues and disulfide bonding in these interactions have been discussed.

Keywords: Molecular chaperones; Amino acid composition; Hydrophobic and charged patches; Medium- and long-range interactions

1. Introduction

Anfinsen and co-workers [1,2] suggested that proteins contain the information necessary for folding within their amino acid sequences. However, the discovery of molecular chaperones has dramatically changed the concept of cellular protein folding. Rather than folding spontaneously, many newly synthesized polypeptide chains seem to acquire their native conformation in a reaction mediated by these versatile helper proteins. Molecular chaperones, of different classes, are now known to participate in a large variety of cellular functions. They assist in de novo protein folding, stabilize proteins under stress conditions and maintain polypeptide chains in a loosely-folded state component for translocation across organellar membranes [3–8].

It has been reported that the hydrophobicity might be an important factor for the recognition of chaperones [9–13]. The structural and functio-

* Corresponding author.

0301-4622/98/$ - see front matter
PII S0301-4622(98)00198-7

nal studies of GroEL support this concept [14–16]. The structure-based mutational analysis of GroEL indeed suggests the presence of a complementary hydrophobic binding surface that lines the cavity of the cylinder [15]. In contrast to Hsp70, GroEL does not seem to recognize short peptide segments in extended conformations [17]. A number of studies have been examined towards the contributions of hydrophobicity, using a spectrum of molecules [18–20]. Recently, the three-dimensional structures of chaperones Hsc70 [21], PapD [22], GroEL [13], GroES [23] and a complex of GroEL–GroES [24] were solved by X-ray crystallography.

Several investigations have been made to analyse the residue contacts and stability of globular proteins from their three-dimensional structures. Based on this, the free energy contacts between pairs of amino acid residues [25], the preferred environment and co-operative behaviour of amino acid residues [26], interaction of each kind of side chains with specified atom type and other side chains [27], residue–residue preference/association potentials [28–30], effective inter-residue contact energies [31], side-chain clusters [32] and side-chain structures on the residue–residue associations [33] and conformational stability [34] of globular proteins were reported.

In this article, we present the results of amino acid composition, hydrophobic and charged patches and medium- and long-range interactions for chaperone proteins. Since a protein molecule is composed of 20 different amino acid residues in a specific sequence and composition, the environment will be different for different types of residues. The three-dimensional atomic coordinates available for the four chaperone proteins have enabled us to carry out this objective and compare them with the monomeric globular proteins.

2. Materials and methods

2.1. Computation of amino acid composition in proteins

The amino acid composition and percentage of residues in a given protein is defined as follows:

The number of amino acids of type i in a given protein is calculated by

$$X_i = \sum_{j=1}^{N} X_{ij} \quad (1)$$

where $X_{ij} = 1$ if amino acid type at sequence position j is i; and $X_{ij} = 0$ otherwise.

Percentage of amino acid residues,

$$P_i = X_i * 100/N \quad (2)$$

where N is the total number of amino acid residues in a protein.

The amino acid composition of a protein is expressed as (P_A, P_D, P_C,..., P_Y), where A, D, C,..., Y stands for the corresponding amino acid single letter code.

2.2. Hydrophobic and charged patches

The hydrophobic and charged patches from protein sequences have been determined by a simple procedure described as follows: to construct a continuous window of desired length (= 4,6,8,12), the number of hydrophobic (Ala, Ile, Leu, Val, Met, Phe and Trp) and charged (Asp, Glu, His, Lys, Arg) residues present are counted. If the number of residues are equal to or below the desired length (> 50% of the considered window size) then the above segment (window) is considered as one hydrophobic/charged patch. In this way the hydrophobic and charged patches are counted from N- to C-terminal.

2.3. Short-, medium- and long-range interactions in molecular chaperones

We followed the method of Gromiha and Selvaraj [35] to compute the medium- and long-range interactions in chaperones. The composition of surrounding residues of each amino acid residue within the sphere of radius 8 Å are computed using Eq. (3) and the contribution from short-range (± 2 residues along the sequence), medium-range (± 4 residues along the sequence)

and long-range ($> \pm 4$ residues) contacts are evaluated.

$$N_j = \Sigma n_{i,j} \quad (3)$$

where $n_{i,j}$ is the number of surrounding residues of type i around the jth residue of the protein.

2.3.1. Database

The crystallographic data for four chaperone protein molecules and amino acid sequences of 92 chaperones and 74 monomeric globular proteins form the source for our study. The three-dimensional structures have been taken from the recent release of the Protein Data Bank of Brookhaven National Laboratory [36,37]. The sequence information for a set of 92 chaperones and 74 globular proteins are obtained from the SWISS-PROT [38] database.

2.4. Chaperone proteins

1GRL, GroEL; 3HSC, Heat shock cognate; 3DPA, PapD. The coordinates of GroES were kindly provided by Dr John F. Hunt, Howard Hughes Medical Institute Laboratories, USA.

2.4.1. The chaperone sequences

CH60–ACYPS, CH60–AGRTU, CH60–AMOPS, CH60–ARATH, CH60–BACSU, CH60–BRUAB, CH60–CHLPN, CH60–CHLTR, CH60–CHRVI, CH60–CLOAB, CH60–CLOPE, CH60–COXBU, CH60–CYACA, CH60–ECOLI, CH60–HAEDU, CH60–LEGMI, CH60–LEGPN, CH60–MAIZE, CH60–MYCLE, CH60–MYCTU, CH60–PESAE, CH60–RICTS, CH60–SYNP7, CH60–SYNY3, CH60–THEP3, CH61–STRAL, CH62–STRAL, CH63–HELVI, CH60–BACST, CH60–BARBA, CH60–BRANA, CH60–LACLA, CH60–LEPIN, CH60–PLAFG, CH60–RHILV, CH60–STAAU, CH61–CUCMA, CH61–MYCLE, CH61–RHIME, CH61–STRCO, CH61–SYNY3, CH62–BRAJA, CH62–CUCMA, CH62–RHIME, CH63–BRAJA, CH63–RHIME, CH61–ECOLI, HS7A–YEAST, HSCB–ECOLI, HS82–BASCU, HSP7–YEAST, HSLU–BACSU, HSCA–ECOLI, HS9B–RAT, HS82–TOBAC, HS82–MAIZE, HS82–ASPFU, HS80–LYCES, DNAK–STRCO, DNAK–BRUOV, TCPZ–YEAST, TCPZ–MOUSE, TCPZ–HUMAN, TCPH–MOUSE, TCPG–YEAST, TCPG–MOUSE, TCPE–YEAST, TCPE–MOUSE, TCPE–AVESA, TCPD–YEAST, TF55–SULSH, TCPB–MOUSE, TCP1–YEAST, TCP1–RAT, TCP1–HUMAN, TCP1–DROME, TCP1–OCCRIGR, TCP1–ARATH, TCPD–MOUSE, TCPB–YEAST, TCPB–MOUSE, P60–RAT, P60–MOUSE, P60–HUMAN, P60–GRIGR, RUBB–BRANA, RUBB–ARATH, RUBA–RICCO, RUBA–WHEAT, RUBB–BRANA, RUBA–ARATH, RUB2–BRANA.

2.4.2. Monomeric globular proteins

CYC–ORYSA, ACYM–HORSE, C51–PSEAE, C562–ECOLI, CYB5–BOVIN, PA2–BOVIN, C551–PSEAE, CRAM–CRAAB, AAT–ECOLI, ACTN–ACTCH, ACON–PIG, RS6–THETH, ATXA–STRGL, CAT3–ECOLI, CISY–PIG, KAD1–PIG, A1AT–HUMAN, ADHE–HORSE, CPXA–PSEPU, CCPR–YEAST, CBPA–BOVIN, DGAL–ECOLI, HBA–HUMAN, HBB–HUMAN, EBA1–FLAME, FENR–SPIOL, CYPH–HUMAN, FLAV–GLOBE, DGAL–ECOLI, FER–SPIOL, CRG–BOVIN, CRG4–BOVIN, FER1–AZOVI, GOX–SPIOL, IAA–STRTE, GSHR–ECOLI, CALM–HUMAN, MDH–ECOLI, LAMB–ECOLI, NXL1–BUNMV, HEMM–THEZO, FABA–MOUSE, FABB–MOUSE, LGB2–LUPLU, MIIB–HUMAN, BBP–PIEBR, TRPR–ECOLI, RNP–BOVIN, ADA–ECOLI, RUBR–DESDE, PMGY–YEAST, ASO–CVCPM, PAPA–CARPA, PYR–YEAST, THTR–BOVIN, ICIC–HIRME, CTRA–BOVIN, RNBR–BACAM, SODC–YEAST, AMYA–ASPOR, LYCV–BPT4, KMLC–RABIT, LIVK–ECOLI, PHHY–PSEFL, AZUP–ALCFA, SYY–BACST, NUG–STAAU, THER–BACTH, NXL1–NAJKA, RPC1–BP434, UBIQ–HUMAN, UTER–RABBIT, PLAS–POPNI, SCX3–GENSC.

Table 1
Amino acid composition (%) in molecular chaperones and globular proteins

Residue	Chaperone	Globular	Ratio	Difference
Ala (A)	10.80 (2.26)	8.86 (2.93)	1.22	1.94
Asp (D)	6.42 (1.16)	6.14 (1.63)	1.05	0.28
Cys (C)	0.70 (0.64)	2.10 (1.02)	0.33	−1.40
Glu (E)	8.34 (1.98)	5.90 (2.03)	1.41	2.44
Phe (F)	1.94 (1.08)	3.76 (2.45)	0.52	1.82
Gly (G)	8.44 (2.04)	8.24 (2.33)	1.02	0.20
His (H)	0.94 (0.67)	2.14 (1.06)	0.44	−1.20
Ile (I)	7.08 (1.44)	5.56 (1.61)	1.27	1.52
Lys (K)	8.01 (1.54)	6.66 (2.93)	1.20	1.35
Leu (L)	8.71 (1.33)	8.01 (2.28)	1.09	0.70
Met (M)	2.76 (0.98)	1.95 (1.04)	1.42	0.81
Asn (N)	3.84 (1.01)	4.44 (1.52)	0.86	−0.6
Pro (P)	2.85 (0.87)	4.50 (1.41)	0.63	−1.65
Gln (Q)	3.14 (1.25)	3.58 (1.47)	0.88	−0.44
Arg (R)	4.56 (1.07)	3.67 (2.01)	1.24	0.81
Ser (S)	5.06 (1.27)	6.72 (1.97)	0.75	−1.66
Thr (T)	5.72 (0.94)	5.63 (1.62)	1.02	0.09
Val (V)	8.97 (1.52)	7.25 (1.79)	1.24	1.72
Trp (W)	0.20 (0.23)	1.31 (0.80)	0.15	−1.11
Tyr (Y)	1.51 (0.77)	3.55 (1.95)	0.42	−2.04

3. Results and discussion

3.1. Amino acid composition of chaperones

The composition of amino acid residues derived from a set of 92 molecular chaperones are presented in Table 1.

Ala is found to be the most abundant residue in chaperones followed by Val, Gly, Glu, Lys and Leu, whereas Trp, Cys and His are the least abundant.

The composition of 74 monomeric globular proteins are given for comparison. Ala and Gly are almost equally abundant followed by Leu, Val, Ser, Lys and Thr. The least abundant residues are Trp, Met, Cys and His in decreasing order. We computed the standard deviation in the composition of all the amino acid residues and these are included in Table 1. We found a minimum deviation for most of the residues and the compositions for both chaperones and globular proteins are statistically significant. We also found a lesser deviation of 0.01–0.05 for the amino acid composition of the 20 residues when one protein was omitted at a time, indicating the significance of the dataset. The average amino acid composition of chaperone and globular proteins can be visualized in Fig. 1. The study reveals that the amino acid composition of the aromatic residues Phe, Trp and Tyr are less in chaperones when compared to globular proteins.

The percentage ratio between the chaperone

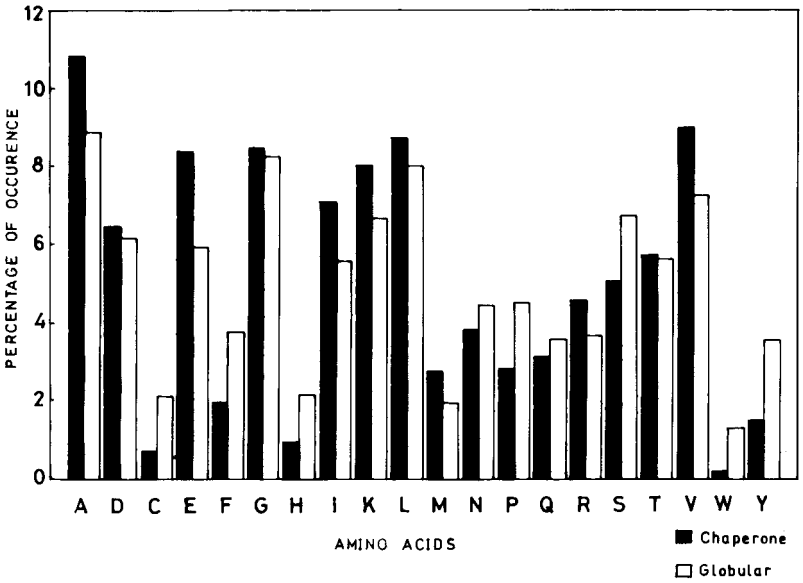

Fig. 1. Percentage amino acid compositions in chaperones and monomeric globular proteins.

and globular protein compositions are also included in Table 1. The ratio stands out higher for the residues Met, Glu, Ile, Arg and Val and lower for Trp, Cys, Tyr, His, Phe and Pro.

We have analysed the amino acid composition of chaperone and globular proteins based on their size and folding type. Most of the chaperones are larger in size and fall under the α/β type of proteins. We observed a similar kind of behaviour between chaperone and globular proteins as that of the complete dataset.

3.1.1. Comparison

The residues Cys, Phe, His, Pro, Ser, Trp and Tyr seem to be present in lower percentages in chaperones compared to globular proteins. Interestingly, Ala, Val, Glu, Lys, Leu, Arg and Met are occurring at higher levels in chaperones than in monomeric globular proteins.

Based on the functional groups, the amino acid composition of acidic (Asp, Glu), basic (His, Lys, Arg), hydrophobic (Ala, Leu, Val, Ile, Met, Phe, Trp) and polar uncharged (Asn, Cys, Gln, Gly, Ser, Thr, Tyr) groups are analysed for chaperones and globular proteins and given below.

Functional groups	Chaperones	Globular
Acidic	14.76	12.04
Basic	13.51	12.47
Hydrophobic	40.46	36.70
Polar uncharged	28.41	34.26
	97.14	95.47

The results show that the charged and hydrophobic residues are more abundant in chaperones than in globular proteins to an extent of 3% and 4%, respectively, whereas the polar uncharged residues are less by approx. 6%. The increased contribution may result in the formation of the hydrophobic and charged patches in chaperones. The average percentage of functional residues present is 97.14% and 95.47% for chaperones and globular proteins, respectively, an increase of 1.67% is noted. The clustered hydrophobic and charged patches may assist the nascent proteins to attain their proper folding rather than misfolding/aggregation. To understand this aspect we analyse here the cluster of those residues.

3.2. Hydrophobic and charged patches in molecular chaperones

The hydrophobic and charged patches of the molecular chaperones are calculated as stated in Section 2 and the results are listed in Table 2. The results show that the charged and hydrophobic patches in molecular chaperones are higher than the monomeric proteins.

The observed average length of the monomeric globular protein is 200.7 and for the chaperone is 537.02. To scale these hydrophobic and charged patch quantities, we divided the hydrophobic and charged patches by a factor of 2.68 (537.02/200.7) and the values are tabulated in Table 2.

3.2.1. The four (4) residue window

The considered four residues are in the hydrophobic and charged set, the observed values 3.99, 1.86 and 3.50, 0.43, respectively, are for chaperones and globular proteins. A difference of 0.49 and 1.43 is noted for hydrophobic and charged patches in chaperones compared to the globular proteins. When three residues are equal out of the considered four residues, the hydrophobic values are 37.71, 33.70 and the charged patches are 14.30, 9.14, respectively, for chaperones and globular proteins.

3.2.2. The six (6) residue window

The considered six residues are observed in the set of hydrophobic ones, the values are 0.63 and 0.31, whereas for all charged, the values are 0.42 and 0.13 for chaperones and globular proteins, respectively. If only five residues are equal out of six, then the difference is 1.50 for hydrophobic and 2.03 for charged patches in chaperones and globular proteins. Suppose only four residues are equal, then 4.55 and 5.79 are the difference in values noted for hydrophobic and charged patches in chaperones, respectively. It is interesting to note that in all the cases, the values against hydrophobic and charged patches are more in chaperones compared to globular proteins and this may provide data to understand the functions of chaperones. Furthermore, from the analysis on

Table 2
Hydrophobic and charged patches calculated from chaperones and globular proteins

Window length	Chaperone		Globular		% Difference	
	Hydro	Charged	Hydro	Charged	Hydro	Charged
Four-residue						
4	3.99	1.86	3.50	0.43	0.12	0.77
3	37.71	14.30	33.70	9.14	0.11	0.36
Six-residue						
6	0.63	0.42	0.31	0.13	0.51	0.69
5	6.96	2.58	5.46	0.55	0.22	0.79
4	37.54	12.78	32.99	6.99	0.12	0.45
Eight-residue						
8	0.01	0.19	0.00	0.00	1.00	1.00
7	0.65	0.86	0.61	0.00	0.06	1.00
6	8.22	3.07	6.19	0.54	0.25	0.82
5	36.48	10.67	30.82	4.68	0.16	0.56
Twelve-residue						
12	0.00	0.07	0.00	0.00	0.00	1.00
11	0.00	0.26	0.00	0.00	0.00	1.00
10	0.13	0.52	0.15	0.00	−0.15	1.00
9	1.39	1.18	1.04	0.01	0.25	0.99
8	8.11	2.79	7.19	0.31	0.11	0.89
7	31.49	8.12	25.84	2.40	0.18	0.70

the occurrence of higher number of patches in chaperones and globular proteins, and the difference between hydrophobic and charged patches are also given in Table 2. From Table 2, we suggest that (i) a subsegment of six residues in a segment of eight-residue window length and (ii) a subsegment of eight residues in a segment of 12-residue window size may be proper to identify the hydrophobic and charged patches. The work on the analysis of the binding site properties to reveal the functional implications of the proposed hydrophobic and charged patches are in progress.

The previous studies [8–16,24] support our results and the recent complex structure of GroEL–GroES explains the importance of hydrophobic and charged residues for the chaperonin function [24]. The surface of the expanded *cis* cavity now presents mostly charged residues. Hydrophobic residues that bound the non-native polypeptide in the cavity of the *trans* ring are now used to stabilize interfaces that supports the GroES complex and have been replaced on the cavity walls by mostly charged residues. This switch in the chemical character of the cavity lining triggers dissociation of the non-native polypeptide from the wall of the cavity. The released polypeptide is now free to re-initiate folding as an isolated molecule in a much-enlarged cavity that has a hydrophilic lining conducive to burial of the substrate polypeptide's hydrophobic residues as it initiates folding into a native structure.

3.3. Short-, medium- and long-range interactions in chaperones

The total and the average contacts per residue for short-, medium- and long-range interactions for all the four chaperones are given in Table 3. For all the 11 788 residues, 4676 short-range, 2178 medium-range and 4934 long-range interactions have been found. On average, within the sphere of 8 Å radius, 4, 2 and 4 residues contribute towards the short-, medium- and long-range interactions, respectively. It is interesting to note that the medium-range contacts is much less, 0.7 and 0.8, respectively, for PapD and GroES due to the absence of alpha helices. These results are similar to those obtained with all beta class proteins [35,40]. Furthermore, the average medium-range interactions obtained for HSC and GroEL

Table 3
Medium- and long-range contacts in chaperones

Name	Nres	Short		Medium		Long	
		Average	Total	Average	Total	Average	Total
HSC	382	3.984	1522	2.016	770	4.162	1590
GroEL	518	3.988	2066	2.336	1210	4.139	2144
PapD	201	3.881	780	0.677	136	4.239	852
GroES	80	3.850	308	0.775	62	4.350	348
Globular proteins[a]							
All α	1728	3.951	6826	2.802	4834	2.361	4076
All β	2353	3.950	9294	0.921	2168	5.201	12210
All $\alpha+\beta$	2067	3.951	8166	1.925	3978	3.722	7694
All α/β	4729	3.970	18776	1.879	8886	4.242	20066

[a] Data from [35,40].

are similar to those of (α/β) type proteins. Recent analysis on the influence of long-range interactions in a set of 36 $(\alpha/\beta)_8$ barrel proteins [39,41] showed the presence of 4.27 contacts per residue which is comparable to all molecular chaperones. Also, we note that all the four chaperone proteins have similar long-range contacts although two of them are all beta type.

3.3.1. Preference of amino acid residues influenced by medium- and long-range interactions

The average medium- and long-range contacts per residue and the ratio between these contacts are presented in Table 4. A perusal of the table reveals that for all the residues long-range interactions are higher than the medium-range interactions. However, apart from the residue Cys which is involved in disulfide bonding, the hydrophobic residues Tyr, Val, Ile and Phe have relatively high long-range contacts (more than three times that of medium-range contact). It is to be noted that the helix breaking residue Pro possess high long-range contact. We note that the residues Ala, Met, Gln, Arg and Ser have higher while Tyr, Trp, Cys, Phe and His have lower medium-range contacts in chaperones than in globular proteins. It is noteworthy that all the aromatic residues and the disulfide bond forming residues have lower medium-range contacts in chaperones. The residues Pro, Ala, Cys, Val, Leu and Asp possess higher long-range interactions whereas His, Trp, Met and Arg have lower long-range contacts in chaperones compared to globular proteins.

In long-range contacts, the residue Cys has the highest contribution (6.14) followed by the hydrophobic residues Val, Ile, Tyr, Leu and Phe. The lowest value is observed for His (1.5). Other residues having lowest long-range contacts are Glu (2.67) and Lys (2.91).

Table 4
Medium- and long-range contacts for all the 20 amino acid residues

Residue	Chaperones			Globular proteins[a]		
	Medium	Long	Ratio	Medium	Long	Ratio
Ala	2.58	4.60	1.79	2.11	3.92	1.86
Asp	1.62	3.16	1.95	1.80	2.85	1.58
Cys	1.43	6.14	4.30	1.88	5.55	2.95
Glu	1.97	2.67	1.36	2.09	2.72	1.30
Phe	1.53	4.88	3.18	1.98	4.53	2.29
Gly	1.67	4.56	2.73	1.53	4.31	2.82
His	1.62	1.50	0.92	1.98	3.77	1.90
Ile	1.65	5.66	3.44	1.77	5.58	3.15
Lys	1.84	2.91	1.58	1.96	2.79	1.42
Leu	1.97	4.98	2.53	2.19	4.59	2.10
Met	2.59	3.63	1.40	2.27	4.14	1.82
Asn	1.78	3.26	1.83	1.84	3.64	1.98
Pro	1.24	4.63	3.73	1.32	3.57	2.70
Gln	2.34	2.97	1.27	2.03	3.06	1.51
Arg	2.13	3.32	1.55	1.94	3.78	1.95
Ser	1.37	3.60	2.63	1.57	3.75	2.39
Thr	1.75	4.08	2.33	1.57	4.09	2.61
Val	1.69	5.87	3.48	1.63	5.43	3.33
Trp	1.33	3.00	2.25	1.90	4.83	2.54
Tyr	1.15	5.07	4.42	1.67	4.93	2.95

[a] Data from [35].

The residues having minimum long-range interactions are charged residues. This indicates that the probability of ion pairs in the sequence level in this range ($> \pm 4$ residues) is less and the ion pairs are formed between neighbouring residues. Another important aspect is that the charged residues are mainly exposed to the solvent and therefore do not have as many partners as those core residues. A similar trend is observed for globular proteins also [35].

In medium-range, the residue Met has the highest contribution (2.59) among all the 20 amino acid residues. Also the residues Ala, Gln and Arg have more than two contacts per residue. It is interesting to note that Met and Ala are very strong helix formers and the residue Gln is also a helix former [42,43].

4. Conclusions

The study reveals the presence of many hydrophobic and charged patches in molecular chaperone proteins. The amino acid composition of all the aromatic residues are low in chaperones. Residues Cys, Val, Ile, Tyr, Leu and Phe have higher long-range interactions due to the formation of disulfide bridges and hydrophobic clusters. The helix forming residues Met, Ala and Gln are preferred for medium-range interactions.

Acknowledgements

TSK thanks the Council of Scientific and Industrial Research, Government of India for the award of Senior Research Fellowship.

References

[1] C.B. Anfinsen, E. Haber, M. Sela, F.H. White, Proc. Natl. Acad. Sci. USA 47 (1961) 1309.
[2] C.B. Anfinsen, Science 181 (1973) 223.
[3] F.U. Hartl, R. Hlodan, T. Langer, Trends. Biochem. Sci. 19 (1994) 20.
[4] M.J. Gething, J. Sambrook, Nature 355 (1992) 33.
[5] R.J. Ellis, Curr. Opin. Struct. Biol. 4 (1994) 117.
[6] F.U. Hartl, Nature 381 (1996) 571.
[7] J.P. Hendrick, F.U. Hartl, Annu. Rev. Biochem. 62 (1993) 349.
[8] W.A. Fenton, A.L. Horwich, Protein Sci. 6 (1997) 743.
[9] G.C. Flynn, J. Rohl, M.T. Flocco, J.E. Rothman, Nature 353 (1991) 726.
[10] G.C. Flynn, T.G. Chappel, J.E. Rothman, Science 245 (1989) 385.
[11] A. Gragerov, M.E. Gottesman, J. Molec. Biol. 241 (1994) 133.
[12] S. Blond-Eluindi, S.E. Cwirla, M.J. Dower, R.J. Lipshutz, S.R. Sprang, J.F. Sambrook, M.J.H. Gething, Cell 5 (1993) 717.
[13] G. Richarme, M. Kohiyama, J. Biol. Chem. 268 (1993) 24074.
[14] K. Braig, Z. Otwinoskz, R. Hegda, D. Boisvert, A. Joahimiak, A.L. Horwich, B. Sigler, Nature 371 (1994) 578.
[15] W.A. Fenton, Y. Kashi, K. Furtak, A.L. Horwich, Nature 371 (1994) 614.
[16] T.S. Kumarevel, M.M. Gromiha, M.N. Ponnuswamy, Biophys. Chem. 71 (1998) 99.
[17] S.J. Landry, R. Jordan, R. McMaaun, L.M. Gierasch, Nature 355 (1992) 455.
[18] G. Richarme, M. Kohiyama, J. Biol. Chem. 269 (1994) 7095.
[19] R. Hlodan, P. Tempst, F.U. Hartl, Nature Struct. Biol. 2 (1995) 587.
[20] S. Lin, F.P. Schwarz, E. Einstein, J. Biol. Chem. 270 (1997) 1011.
[21] K.M. Flaherty, C. Deluca-Flaherty, D.B. McKay, Nature 346 (1990) 623.
[22] A. Holmgren, C.I. Branden, Nature 342 (1989) 248.
[23] J. Hunt, A.J. Weaver, S.J. Landry, L. Gierash, J. Deisenhofer, Nature 379 (1996) 37.
[24] Z. Xu, A.L. Horwich, P.B. Sigler, Nature 388 (1997) 741.
[25] S. Tanaka, H.A. Scheraga, Macromolecules 9 (1976) 945.
[26] P. Manavalan, P.K. Ponnuswamy, Arch. Biochem. Biophys. 184 (1977) 476.
[27] P.K. Warme, R.S. Morgan, J. Molec. Biol. 118 (1978) 289.
[28] M.W. MacArthur, J.M. Thornton, J. Molec. Biol. 218 (1991) 397.
[29] M.Y.A. Karpeisky, V.A. Ilyin, J. Molec. Biol. 224 (1991) 629.
[30] S.V.L. Narayana, P. Argos, Int. J. Peptide Protein Res. 24 (1984) 25.
[31] S. Miyazawa, R.L. Jernigan, Macromolecules 18 (1985) 552.
[32] J.K. Herniga, P. Argos, J. Molec. Biol. 220 (1991) 151.
[33] R. Muthusamy, P.K. Ponnuswamy, J. Theor. Biol. 153 (1991) 25.
[34] P.K. Ponnuswamy, M.M. Gromiha, J. Theor. Biol. 166 (1994) 63.
[35] M.M. Gromiha, S. Selvaraj, J. Biol. Phys. 23 (1997) 151.
[36] F.C. Bernstein, T.F. Koetzle, J.B. Williams, E.F. Meyer, Jr., M.D. Brice, J.R. Rodgers, O. Kennard, T. Shimanouchi, M. Tasumi, J. Molec. Biol. 112 (1977) 535.
[37] E. Abola, F.C. Bernstein, S.H. Bryant, T.F. Koetzle, J. Weng, Protein data bank, in: F.H. Allen, G. Bergerhoff, R. Sievers (Eds.), Crystallographic Databases — Information Content, Software Systems, Scientific Applications, 1987, pp. 107.

[38] A. Bairoch, B. Boeckmann, The SWISS-PROT protein sequence data bank. Nucl. Acids. Res. 20 (1992) 2019.
[39] M.M. Gromiha, S. Selvaraj, J. Biol. Phys. 23 (1997) 209.
[40] M.M. Gromiha, S. Selvaraj, Protein Eng. 11 (1998) 249.
[41] S. Selvaraj, M.M. Gromiha, J. Protein Chem. 17 (1998) 697.
[42] G.D. Fasman, in: G.D. Fasman (Ed.), Prediction of Protein Structure and Principles of Protein Conformation, Plenum Press, New York, 1989, p. 193.
[43] M.M. Gromiha, P.K. Ponnuswamy, Int. J. Peptide Protein Res. 45 (1995) 225.