Supplementary material for the article:

Stanković, I. M.; Blagojević Filipović, J. P.; Zarić, S. D. Carbohydrate – Protein Aromatic Ring Interactions beyond CH/π Interactions: A Protein Data Bank Survey and Quantum Chemical Calculations. *Int. J. Biol. Macromol.* **2020**, *157*, 1–9.
https://doi.org/10.1016/j.ijbiomac.2020.03.251

Supplemental information

CARBOHYDRATE – PROTEIN AROMATIC RING INTERACTIONS BEYOND CH/π INTERACTIONS: A PROTEIN DATA BANK SURVEY AND QUANTUM CHEMICAL CALCULATIONS

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Figure S1. The example of a and b carbohydrate faces, β-D-glucose.
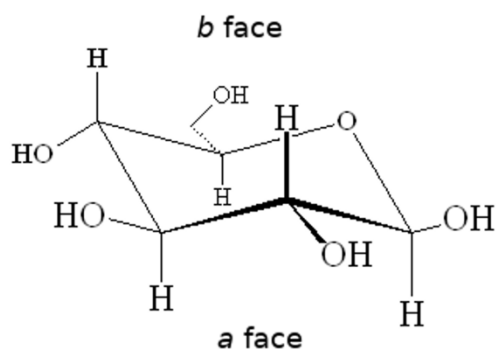
The contacts with CH/O interactions, where aromatic amino acids are hydrogen donors are prone to larger dihedral angle and offset values (Figure S2). There is no particular tendency for contacts with CH/O interactions, where carbohydrates are hydrogen donors (Figure S2). Figures S3-S5 show that smaller dihedral angle and offset values are more common for contacts with CH/N, OH/π and classical hydrogen bonds, respectively.
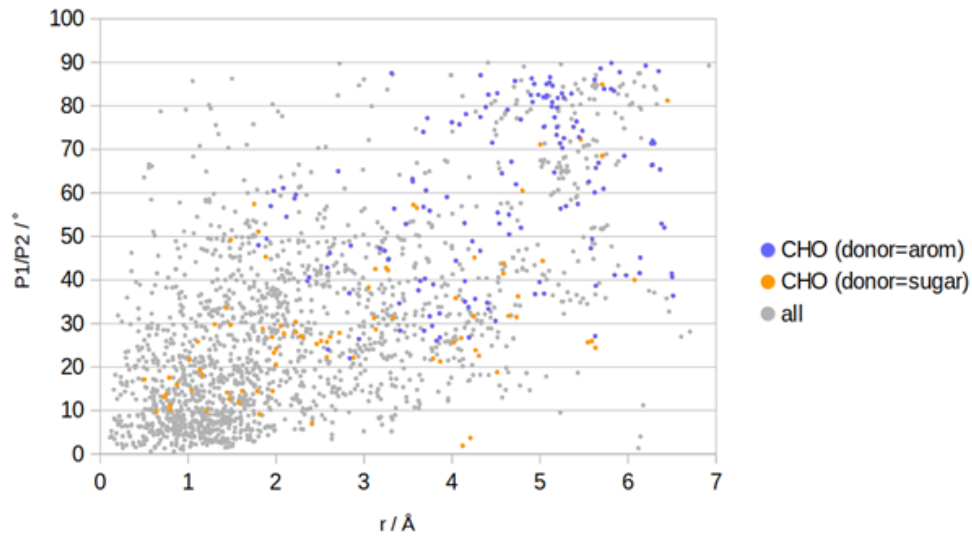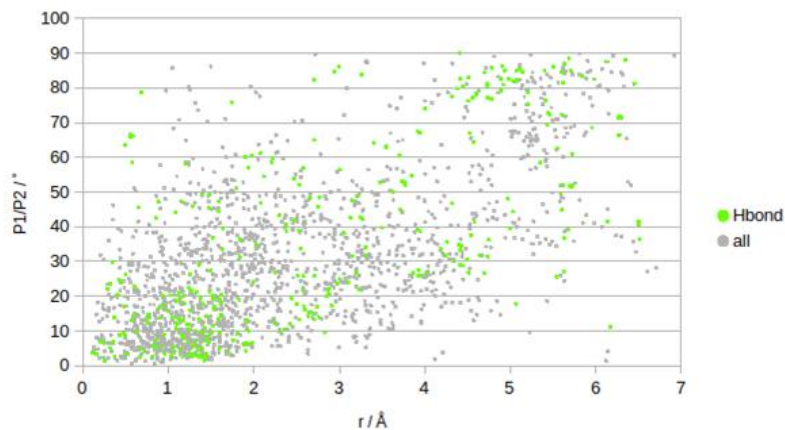
Figure S2. Depencences of the angles between rings on offset values, for all contacts and contacts with CH/O interactions, when hydrogen donor is the aromatic amino acid or sugar molecule
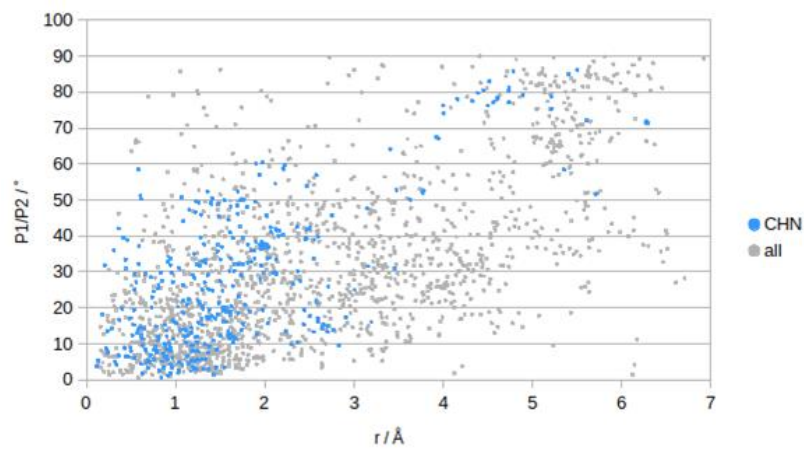
Figure S3. Depencences of the angles between rings on offset values, for all contacts and contacts with CH/N interactions
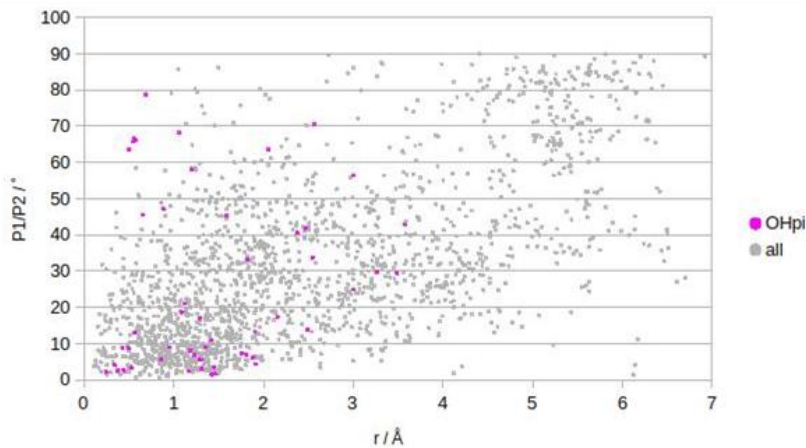
Figure S4. Depencences of the angles between rings on offset values, for all contacts and contacts with OH/π interactions

Figure S5. Depencences of the angles between rings on offset values, for all contacts and contacts with classical hydrogen bonds

Higher P_1/P_2 angles

The sugar-aromatic contacts involving CH/π interactions tend to be parallel (Figure 2A, 3 and 4), as the consequence of the sugar CH group nearly perpendicular to the sugar plane. However, in some of the contacts there are higher P_1/P_2 angles between rings (Figure 2A) that are the consequence of distorted high-energy sugar rings, or deoxy sugars with missing OH groups. The example is shown in Figure S6.
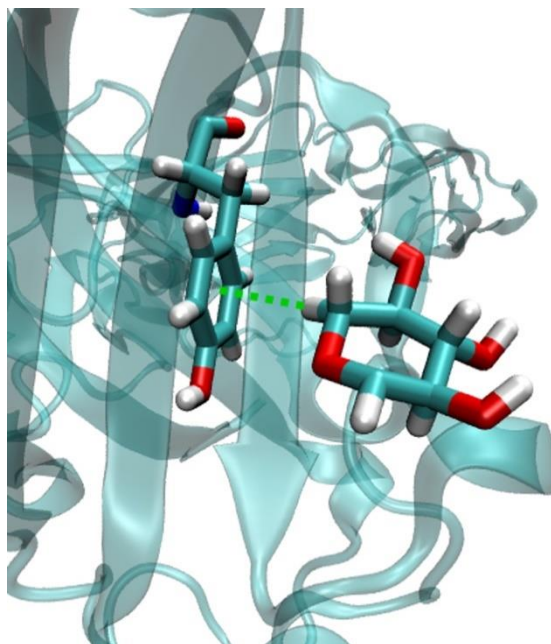
Figure S6: An example of the deoxy sugar forming non-parallel contact to the aromatic ring. Green dotted line represents CH/π interaction.

The distribution of the number of CH/π interactions

The distribution of the number of CH/π interactions per sugar-aromatic pair demonstrates tendency towards only one or two CH/π bonds per sugar-aromatic pair (Figure S7). It means that one or two CH/π bonds per sugar/aromatic pair are sufficient for holding them together. PHE and TYR form only one or two, rarely three CH/π interactions, while TRP can form one, two, three or four because of its two aromatic rings. We plotted R(r) dependences for each one of these groups: with one, two, three or four CH/π interactions (Figure S7). For the higher number of CH/π interactions, the tendency is towards less scattered R values and lower r values, which proves that the CH/π interactions are responsible for tighter bonding between carbohydrate and aromatic moieties, even though one such bond is sufficient for holding the carbohydrate in place.
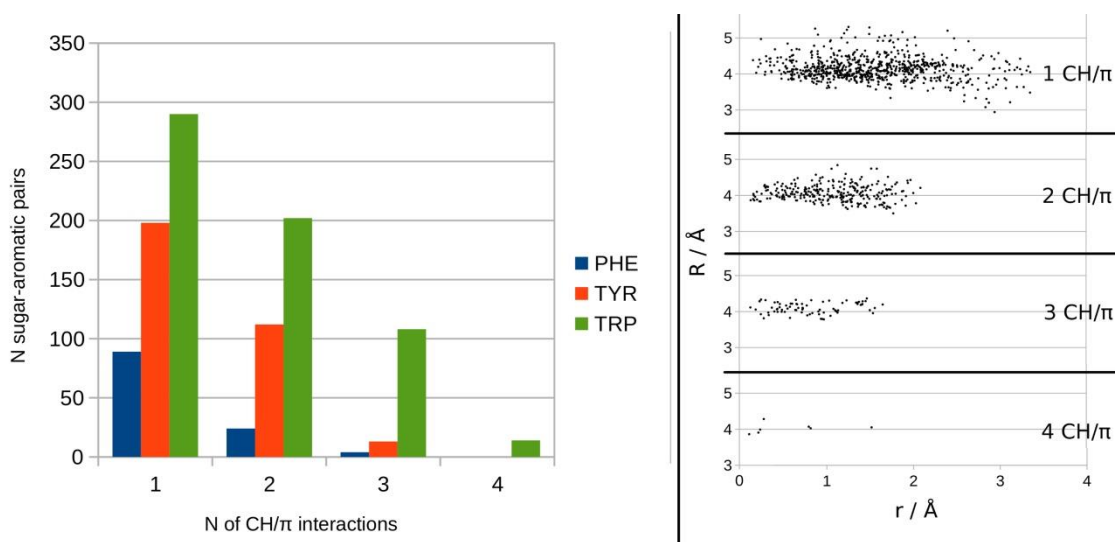
Figure S7: The distribution of the number of CH/π interactions per sugar-aromatic pair. The R(r) diagrams related to each number of CH/π interactions are shown on the right side.

## The sandwich aromatic amino acid/carbohydrate arrangements

The survey pointed to rare sandwich arrangements (two aromatics per sugar monomer, Figure S8) for the CH/π interacting pairs: from 1054 contacts with CH/π only 49 of them are positioned in aromatic sandwich.
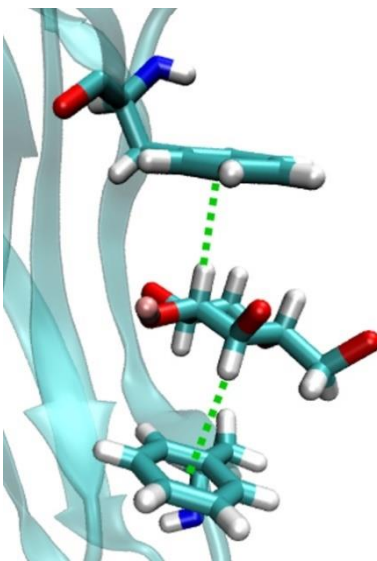
Figure S8: Example of sandwich interaction: two aromatic rings interacting simultaneously with one sugar ring. PDB id: 5DPN.
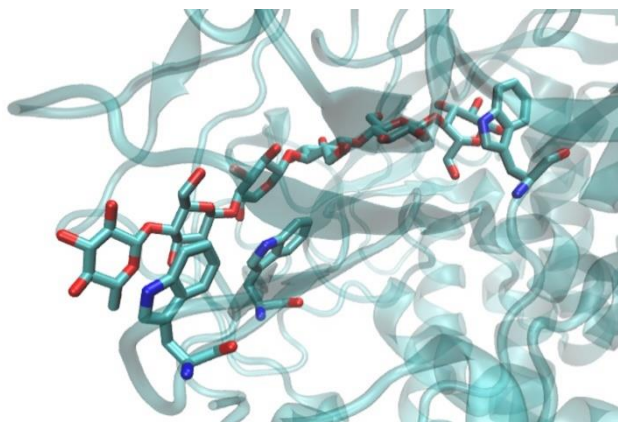
The graphs in Figure S9 represent the projections of monosaccharide centers onto aromatic ring plane of the six-membered rings of PHE, TYR and TRP. Data for contacts with CH/π interactions (Figure S9) show no specificity in offset nor φ angle (Figure 2B), except for TRP. The clustering in the fourth quadrant for TRP is due to interactions with five-membered TRP ring (Figure 2B, Figure S9A). The distribution of the φ angle in contacts without CH/π interactions (Figure S9B) shows that there is clustering in the second and third quadrant for all three aromatic rings, probably due to steric hindrances caused by the protein backbone.
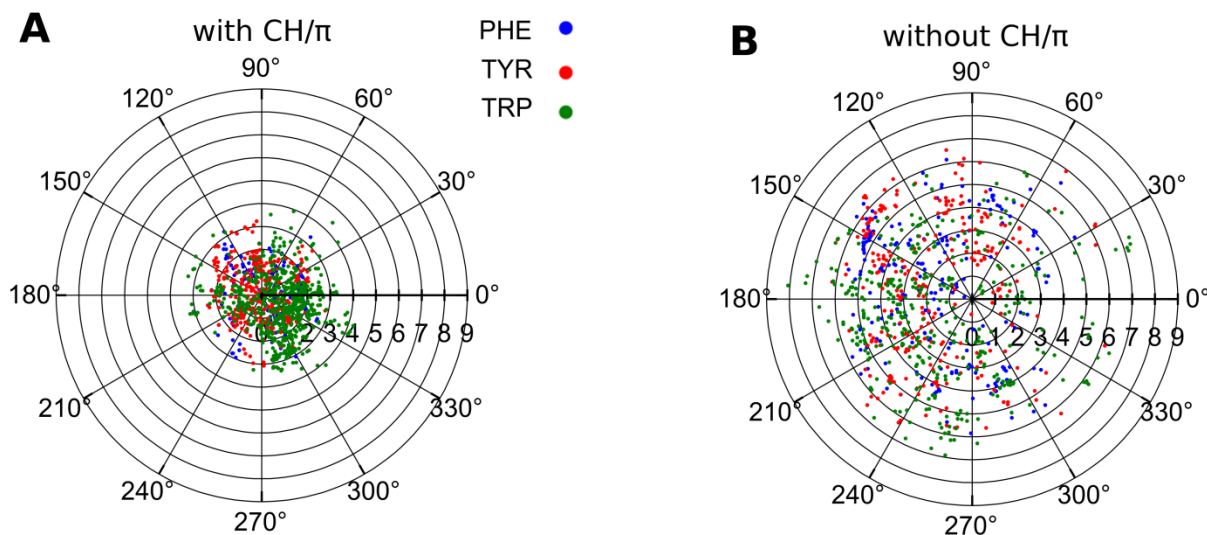
Figure S9. The polar graph offset - φ angle (Figure 2B) for the contacts involving CH/π interactions (A) and without CH/π interactions (B). Different aromatic amino acids are represented separately.

Table S1. Pattern of the average number of aromatic rings per carbohydrate substrate (both monomers and oligomers) and per monomer, with and without CH/π interactions.

Number of monomers per oligomer unit	N of substrates	Any contact		With CH/π		Without CH/π	
		N of aromatics per substrate	N of aromatics per monomer	N of aromatics per substrate	N of aromatics per monomer	N of aromatics per substrate	N of aromatics per monomer
1	799	0.68	0.68	0.32	0.32	0.36	0.36
2	243	1.45	0.72	0.99	0.50	0.46	0.22
3	156	1.40	0.47	0.85	0.28	0.55	0.19
4	95	1.92	0.48	1.33	0.33	0.59	0.15
5	54	2.28	0.46	1.39	0.28	0.89	0.18
6	49	3.14	0.52	1.82	0.30	1.32	0.22
7	26	2.78	0.40	1.62	0.23	1.16	0.17
8	20	2.95	0.39	1.40	0.18	1.55	0.21

Figure S10. An example of a six units long carbohydrate substrate interacting with three aromatic rings in the protein cleft. PDB id: 1L2A.