Available bioassays to measure disruption of thyroid hormone transport via transthyretin binding

In the literature, several *in vitro* assays have been reported for the determination of the potency of environmental pollutants to bind to TTR. The most common *in vitro* assay is the radioligand binding assay (RLBA), which is a competitive binding assay using T4 or T3 (native and labeled) in a mixture with the competitor (compounds or extracts) to bind to TTR. The specific parameters of the assay differ slightly between laboratories, such as incubation temperature and time, buffer solution and packing material of the column for separating bound from free (labeled) TH.

Another approach is the non-radioligand binding assays (BA) using covalently bound ligands to different material. Three different BA approaches are represented here:

1. The use of polyclonal TTR antibodies covalently bound to a sepharose resin that is mixed with plasma pretreated with interfering compounds, i.e. the thyroid hormone disrupting compounds (THDCs). The TTR-THDC complex is washed and finally the THDC is eluted with high pH solution and injected onto an HPLC column for analysis and quantification [1]. The antibody capture of the complex can underestimate the THDC binding stoichiometry, owing to the weak
association between TTR and THDC and hence the THDC can dissociate during the wash step.

2. The weak association between transport protein and ligand is a property used in the second assay, the surface plasmon resonance (SPR) assay. In the SPR assay the thyroid hormone (TH) is covalently bound to a gold-layered chip, and an incubation mixture of the transport protein and competitors (THDCs) are injected in a flow cell passing over the bound thyroid hormone layer [2]. The association TTR-THDC complex is not stable and there will be a competition in the flow cell where available TTR will bind to the bound TH and affect the surface of the gold-layered chip. The change of the surface layer composition is detected in real time with SPR technique.

3. The third non-radioligand binding assay is the FLU-TTR, based on a protein-binding fluorescence probe (8-anilino-1-naphthalenesulfonic acid ammonium salt – ANSA or fluorescein-thyroxine), which becomes highly fluorescent after its binding to protein [3,4]. If an analyte binds to the protein at the same site as the probe, it will displace the probe from the protein and reduce the fluorescence intensity.

The radioligand and non-radioligand assays give comparable results (Table S1) but it is still difficult to compare studies using different parameters in their test setup, such as the use of T₄ or T₃, and different sources of TTR. For instance, the use of TTR from human, bird, fish and amphibians has been reported, both in purified and recombinant forms which influences the structure of the ligand binding site and the affinity.

One of the most commonly used bioassay is the well-established radioligand TTR binding assay based on the method described by Somack and co-workers [5], but with several minor modifications between laboratories [6-8]. The assay successfully detected TTR-binding activity of several pure compounds [6,7,9] and was also implemented for sediment [10,11] and biota extracts [12]. To test the applicability of the TTR-binding assay in EDA studies we present here the results from a sediment sample extract with a high TTR-binding potency.
**Table S1:** Compounds that have been tested in the *in vitro* TTR-binding assays reported in the literature are summarized. The backbone structure of the compound groups are presented together with the number of aromatic rings (ar). The compound name, the assay type, species and source of TTR (recombinant [rTTR], purified [pTTR] or non-specified) are given. The TTR binding potency is given as the concentration at 50% inhibition (IC\(_{50}\)) for either T\(_3\) or T\(_4\) and the corresponding relative effect potency (REP). All study references are given in the last column.

| Backbone | Compound       | Assay | TTR          | T\(_4\) IC\(_{50}\) (nM) | T\(_4\) REP | T\(_3\) IC\(_{50}\) (nM) | T\(_3\) REP | Ref |
|----------|----------------|-------|--------------|--------------------------|-------------|--------------------------|-------------|-----|
| Triiodothyronine (T\(_3\)) | BA | Human TTR | 280 | | | | | [3] |
| | FLU-TTR | Human TTR | 1472 | 0.16 | | | | [4] |
| | RLBA | Gull rTTR | 6 | 1 | | | | [8] |
| | RLBA | Chicken pTTR | 22.5 | 1 | | | | [13] |
| | RLBA | Rana pTTR | 1.43 | 1 | | | | [13] |
| | RLBA | Chicken pTTR | 21 | 1 | | | | [14] |
| | RLBA | Rana pTTR | 0.5 | 1 | | | | [14] |
| | RLBA | SeabreamrTTR | 24 | 0.9 | | | | [15] |
| | RLBA | Rana rTTR | 9 | | | | | [16] |
| | RLBA | Xenopus TTR | 400-550 | | | | | [16] |
| Diphenyl ether, 2 ar | Thyroxine (T\(_4\)) | RLBA | Human pTTR | 60-80 | 1 | | TS | |
| | | SPR | Human pTTR | 1 | | | | [2] |
| | | SPR | Human rTTR | 1 | 0.06 | | | [2] |
| | | BA | Human TTR | 30 | | | | [3] |
| | | FLU-TTR | Human TTR | 260 | 1 | | | [4] |
| | | RLBA | Human pTTR | 55 | 1 | | | [6,17] |
| | | RLBA | Human rTTR | 88, 138 | 1 | | | [7] |
| | | RLBA | Human TTR | 60 | | | | [9] |
| | | RLBA | Human TTR | 80.7 | 1 | | | [18] |
| | | RLBA | Human rTTR | 57 | 1 | | | [19] |
| Backbone Compound | Assay | TTR | T<sub>4</sub> IC<sub>50</sub> (nM) | T<sub>4</sub> REP | T<sub>3</sub> IC<sub>50</sub> (nM) | T<sub>3</sub> REP | Ref |
|-------------------|-------|-----|-------------------------------|-------------|-------------------------|-------------|-----|
| RLBA Human rTTR   | 81    | 1   |                               |             |                         |             | [20]|
| RLBA Human rTTR   | 79    |     |                               |             |                         |             | [21]|
| RLBA Human TTR    | 79    |     |                               |             |                         |             | [22]|
| RLBA Human rTTR   | 75    |     |                               |             |                         |             | [23]|
| RLBA Human TTR    | 400   | 1   |                               |             |                         |             | [24]|
| RLBA Human TTR    | 62    |     |                               |             |                         |             | [25]|
| ANSA Human TTR    | 260   | 1   |                               |             |                         |             | [26]|
| RLBA Gull rTTR    | 16    | 1   |                               |             |                         |             | [8]  |
| RLBA Seabream rTTR| 21    | 1   |                               |             |                         |             | [15]|
| RLBA Rana rTTR    | 500-700 | |                       |             |                         |             | [16]|
| RLBA Xenopus TTR  | >10000 |   |                               |             |                         |             | [16]|
| RLBA Human TTR    | 0<sup>c</sup> |   |                               |             |                         |             | [24]|
| Acetochlor Rana TTR | 79<sup>b</sup> |   |                               |             |                         |             | [14]|
| RLBA Salmon TTR   | 87.3<sup>b</sup> |   |                               |             |                         |             | [14]|
| RLBA Human TTR    | n.d.  | n.d.|                               |             |                         |             | [25]|
| Alachlor Rana TTR | 99.7<sup>b</sup> |   |                               |             |                         |             | [14]|
| 1,2-Dichlorobenzene RLBA Human TTR | 0<sup>c</sup> |   |                               |             |                         |             | [24]|
| 2,4-Dichlorophenoxyacetic acid RLBA Rana TTR | 99.7<sup>b</sup> |   |                               |             |                         |             | [14]|
| RLBA Salmon TTR   | 91<sup>b</sup> |   |                               |             |                         |             | [14]|
| 2,4,6-Tribromaniline RLBA Human rTTR | n.d. | n.d. |                               |             |                         |             | [20]|
| 2,3,5,6-Tetra brom-p-xylene RLBA Human rTTR | n.d. | n.d. |                               |             |                         |             | [20]|
| 2,3,4,5,6-Pentabromtoluene RLBA Human rTTR | n.d. | n.d. |                               |             |                         |             | [20]|
| Hexabrombenzene RLBA Human rTTR | n.d. | n.d. |                               |             |                         |             | [20]|
| Ioxynil RLBA Fish rTTR | 39 | 0.5 |                               |             |                         |             | [15]|
| RLBA Rana pTTR    | 100   | 0.005 |                               |             |                         |             | [14]|

<sup>a</sup> Benzene, 1 ar

<sup>b</sup> Acetochlor

<sup>c</sup> Benzene, 1 ar
| Backbone | Compound                  | Assay   | TTR                | $T_4$ IC$_{50}$ (nM) | $T_4$ REP | $T_3$ IC$_{50}$ (nM) | $T_3$ REP | Ref |
|----------|--------------------------|---------|--------------------|-----------------------|-----------|-----------------------|-----------|-----|
|          |                          | RLBA    | Chicken pTTR       | 15                    | 1.4       |                       |           | [14]|
|          |                          | RLBA    | Rana TTR           | 25.8$^b$              |           |                       |           | [14]|
|          |                          | RLBA    | Salmon TTR         | 9.2$^b$               |           |                       |           | [14]|
| Phenol   |                          | RLBA    | Human TTR          | 10$^c$                |           |                       |           | [24]|
| Octylphenol |                      | RLBA    | Human pTTR         | n.d.                  | n.d.      | TS                    |           |     |
|          |                          | RLBA    | Rana TTR           | 84.2$^b$              |           |                       |           | [14]|
|          |                          | RLBA    | Salmon TTR         | 85.5$^b$              |           |                       |           | [14]|
| Nonylphenol |                       | RLBA    | Seabream rTTR      | 17040                 | 0.001     |                       |           | [15]|
|          | technical mix           | RLBA    | Human pTTR         | 10000$^d$             | 0.008     | TS                    |           |     |
|          | linear                   | RLBA    | Human TTR          | n.d.$^f$              | n.d.      |                       |           | [23]|
|          | branched                 | RLBA    | Human TTR          | 23434$^e$             | 0.003     |                       |           | [23]|
|          |                          | RLBA    | Chicken pTTR       | 1330                  | 0.02      |                       |           | [13]|
|          |                          | RLBA    | Rana pTTR          | 2730                  | 0.0005    |                       |           | [13]|
|          |                          | RLBA    | Rana TTR           | 60.1$^b$              |           |                       |           | [14]|
|          |                          | RLBA    | Salmon TTR         | 44.3$^b$              |           |                       |           | [14]|
| 2.4-Dinitrophenol |                  | RLBA    | Rana TTR          | 96.4$^b$              |           |                       |           | [14]|
|          |                          | RLBA    | Salmon TTR         | 91.3$^b$              |           |                       |           | [14]|
| 4-Phenoxyphenol |                      | RLBA    | Human rTTR        | n.d.                  | n.d.      |                       |           | [20]|
|          | 4-(Phenylmethyl)phenol  | RLBA    | Human pTTR         | >20000                | <<1       |                       |           | TS  |
|          | 4-(1,1,3,3-tetramethylbutyl)phenol | RLBA  | Human pTTR       | n.d.                  | n.d.      |                       |           | TS  |
|          | 2-methyl-4-(1,1,3,3-tetramethylbutyl)phenol | RLBA | Human pTTR   | n.d.                  | n.d.      |                       |           | TS  |
|          | 4-tert-Pentyphenol      | RLBA    | Human pTTR         | n.d.                  | n.d.      |                       |           | TS  |
| 2-Chloro-4-nonylphenol |                  | RLBA    | Chicken pTTR     | 165                   | 0.14      |                       |           | [13]|
|          |                          | RLBA    | Rana pTTR          | 597                   | 0.002     |                       |           | [13]|

$^a$ Denotes hydrophobicity increase.
$^b$ Denotes hydrophobicity decrease.
$^c$ Denotes hydrophobicity comparable.
$^d$ Denotes polarity increase.
$^e$ Denotes polarity decrease.
$^f$ Denotes polarity comparable.

Ref: Reference number for the study.
| Compound                        | Assay | TTR                  | T₄ IC₅₀ (nM) | T₄ REP | T₃ IC₅₀ (nM) | T₃ REP | Ref |
|--------------------------------|-------|----------------------|-------------|--------|-------------|--------|-----|
| 2,6-Dichloro-4-nonylphenol     | RLBA  | Chicken pTTR         | 7.5         | 3.0    | [13]        |        |     |
|                                | RLBA  | Rana pTTR            | 48          | 0.03   | [13]        |        |     |
| 2-Chlorophenol                 | RLBA  | Human TTR            | 1 000 000   | 0.0004 | [24]        |        |     |
| 3-Chlorophenol                 | RLBA  | Human TTR            | 1 400 000   | 0.0003 | [24]        |        |     |
| 2.6-Dichlorophenol             | RLBA  | Human TTR            | 22 000      | 0.02   | [24]        |        |     |
| 2.3-Dichlorophenol             | RLBA  | Human TTR            | 130 000     | 0.003  | [24]        |        |     |
| 2.4.5-Trichlorophenol          | RLBA  | Human TTR            | 2700        | 0.15   | [24]        |        |     |
| 2.4.6-Trichlorophenol          | RLBA  | Human TTR            | 1200        | 0.33   | [24]        |        |     |
| Pentachlorophenol              | RLBA  | Seabream rTTR        | 2480        | 0.008  | [15]        |        |     |
|                                | SPR   | Human rTTR           |             | 0.53   | [2]         |        |     |
|                                | SPR   | Human pTTR           |             | 0.64   | [2]         |        |     |
|                                | RLBA  | Human pTTR           |             | 230    | 1.74        | [24]   |     |
|                                | RLBA  | Rana pTTR            |             | 45     | 0.01        | [14]   |     |
|                                | RLBA  | Chicken pTTR         |             | 6      | 3.5         | [14]   |     |
|                                | RLBA  | Rana TTR             |             | 15.4ᵇ  | [14]        |        |     |
|                                | RLBA  | Salmon TTR           |             | 15.1ᵇ  | [14]        |        |     |
| 4-OH-Heptachlorostyrene        | RLBA  | Human TTR            | 72          | 1.1    | [22]        |        |     |
| 2,4-Dibromphenol               | RLBA  | Human rTTR           | 1400        | 0.06   | [20]        |        |     |
| 2,4,6-Tribromphenol            | RLBA  | Human pTTR           | 4.8         | 10     | [6]         |        |     |
|                                | RLBA  | Seabream rTTR        | 30          | 0.7    | [15]        |        |     |
|                                | RLBA  | Human rTTR           | 67          | 1.2    | [20]        |        |     |
|                                | RLBA  | Human rTTR           | 26          | 3      | [21]        |        |     |
| Pentabromphenol                | RLBA  | Human rTTR           | 12          | 7.14   | [20]        |        |     |
|                                | SPR   | Human rTTR           |             | 0.64   | [2]         |        |     |
|                                | SPR   | Human pTTR           |             | 0.4    | [2]         |        |     |
| Piperine                       | RLBA  | Human pTTR           | n.d.        | n.d.   | TS          |        |     |
| Backbone | Compound       | Assay  | TTR          | $T_{4}IC_{50}$ (nM) | $T_{4}$ REP | $T_{3}IC_{50}$ (nM) | $T_{3}$ REP | Ref |
|----------|----------------|--------|--------------|---------------------|------------|---------------------|------------|-----|
| Musks, 1 ar | Isosafrole     | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Celestolide    | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Galaxolide     | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Phantolide     | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Tonalide       | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Traseolide     | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Musk ambrette  | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Musk ketone    | RLBA   | Human pTTR   | n.d.                | n.d.       |                     |            | TS  |
|          | Biphenyl       | RLBA   | Seabream rTTR| n.d.                | n.d.       |                     |            | [15]|
| Biphenyl, 2 ar | CB 5          | RLBA   | Human TTR    | 87$^c$             |            |                     |            | [24]|
|          | CB 14          | SPR    | Human rTTR   | <0.001              |            |                     |            | [2] |
|          |               | SPR    | Human pTTR   | <0.001              |            |                     |            | [2] |
|          |               | RLBA   | Human TTR    | 11$^c$             |            |                     |            | [24]|
|          | CB 19          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 28          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 38          | BA     | Human pTTR   | 0.18$^a$            |            |                     |            | [27]|
|          | CB 47          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 51          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 52          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 53          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 74          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 77          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [25]|
|          | CB 80          | BA     | Human pTTR   | 0.62$^a$            |            |                     |            | [27]|
|          |               | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 95          | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 100         | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 101         | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
|          | CB 104         | RLBA   | Human TTR    | n.d.                | n.d.       |                     |            | [19]|
| Backbone | Compound | Assay | TTR | $T_4$ IC$_{50}$ (nM) | $T_4$ REP | $T_3$ IC$_{50}$ (nM) | $T_3$ REP | Ref |
|----------|----------|-------|-----|---------------------|----------|---------------------|----------|-----|
| CB 105   | RLBA     | Human rTTR | > 1000 | <<1 | [7] |
| CB 110   | BA       | Human pTTR | n.d.$^a$ | | | [27] |
| CB 111   | BA       | Human pTTR | 0.06$^a$ | | | [27] |
| CB 118   | RLBA     | Human TTR | n.d. | n.d. | [19] |
| CB 122   | RLBA     | Human TTR | 790 | 0.083 | [19] |
| CB 125   | RLBA     | Human TTR | 3700 | 0.014 | [19] |
| CB 126   | RLBA     | Human TTR | n.d. | n.d. | [25] |
| CB 127   | BA       | Human pTTR | 1.5$^a$ | | | [27] |
| CB 128   | RLBA     | Human TTR | n.d. | n.d. | [19] |
| CB 136   | RLBA     | Human TTR | n.d. | n.d. | [19] |
| CB 138   | BA       | Human pTTR | n.d.$^a$ | | | [27] |
|         | RLBA     | Human TTR | >15 000 | <0.008 | [19] |
| CB 153   | RLBA     | Human TTR | >15 000 | <0.008 | [19] |
| CB 162   | BA       | Human pTTR | 0.19$^a$ | | | [27] |
| CB 168   | RLBA     | Human TTR | 1300 | 0.041 | [19] |
| CB 169   | BA       | Human pTTR | 0.05$^a$ | | | [27] |
| CB 170   | RLBA     | Human TTR | n.d. | n.d. | [19] |
| CB 180   | RLBA     | Human TTR | n.d. | n.d. | [19] |
| CB 187   | RLBA     | Gull rTTR | 91 | 0.17 | 791 | 0.012 | [8] |
| CB 190   | RLBA     | Human TTR | n.d. | n.d. | [19] |
| 2'-OH CB 105 | RLBA | Human rTTR | 950 | 0.09 | [7] |
| 3'-OH CB 138 | RLBA | Human pTTR | 3.3 | | | [21] |
| 3'-OH CB 180 | BA     | Human pTTR | 1.67$^a$ | | | [27] |
|         | RLBA     | Human TTR | 1.9 | | | [21] |
|         | RLBA     | Human TTR | 17.8 | 3.4 | | [28] |
| Backbone | Compound       | Assay | TTR     | T4 IC50 (nM) | T4 REP | T3 IC50 (nM) | T3 REP | Ref |
|----------|----------------|-------|---------|--------------|--------|--------------|--------|-----|
| 3'-OH CB 182 | RLBA Human TTR | 19    | 3.2     |              |        |              |        | [28]|
| 3'-OH CB 30 | RLBA Human TTR | n.d.  | n.d.    |              |        |              |        | [25]|
| 3-OH CB 203 | RLBA Human TTR | 44.1  | 1.59    |              |        |              |        | [23]|
| 4-OH Biphenyl | RLBA Human TTR | 14c   |         |              |        |              |        | [24]|
| 4-OH CB 14  | SPR Human rTTR | 4.36  |         |              |        |              |        | [2] |
|           | SPR Human pTTR | 3.04  |         |              |        |              |        | [2] |
|           | RLBA Human TTR | 16    | 3.88    |              |        |              |        | [25]|
| 4'-OH CB 35 | RLBA Human rTTR| 10.5  | 8.4     |              |        |              |        | [7]  |
| 4'-OH CB 50 | BA Human pTTR  | 0.81a |         |              |        |              |        | [27]|
| 4-OH CB 56  | RLBA Human rTTR| 8.8   | 10.2    |              |        |              |        | [7]  |
| 4'-OH CB 61 | BA Human pTTR  | 0.83a |         |              |        |              |        | [27]|
|           | RLBA Human TTR | 89    | 0.7     |              |        |              |        | [25]|
| 4'-OH CB 69 | BA Human pTTR  | 0.96a |         |              |        |              |        | [27]|
|           | RLBA Human TTR | 33    | 1.88    |              |        |              |        | [25]|
| 4'-OH CB 79 | BA Human pTTR  | 1.86a |         |              |        |              |        | [27]|
|           | ANSA Human TTR | 510   | 0.52    |              |        |              |        | [26]|
| 4'-OH CB 82 | RLBA Human rTTR| 17.5  | 5       |              |        |              |        | [7]  |
| 4-OH CB 93  | BA Human pTTR  | 0.7a  |         |              |        |              |        | [27]|
| 4'-OH CB 106| BA Human pTTR  | 1.23a |         |              |        |              |        | [27]|
|           | RLBA Human TTR | 141   | 0.44    |              |        |              |        | [25]|
| 4-OH CB 107 | BA Human pTTR  | 1.48a |         |              |        |              |        | [27]|
|           | RLBA Human TTR | 24.4  | 3.3     |              |        |              |        | [18]|
|           | RLBA Human TTR | 22.6  | 3.5     |              |        |              |        | [21]|
| 4'-OH CB 108| BA Human pTTR  | 1.63a |         |              |        |              |        | [27]|
|           | RLBA Human rTTR| 15    | 5.9     |              |        |              |        | [7]  |
| 4'-OH CB 109| BA Human pTTR  | 0.84a |         |              |        |              |        | [27]|
| 4'-OH CB 112| BA Human pTTR  | 1.12a |         |              |        |              |        | [27]|
| Backbone | Compound | Assay | TTR | $T_4\text{IC}_{50}$ (nM) | $T_4\text{REP}$ (nM) | $T_3\text{IC}_{50}$ (nM) | $T_3\text{REP}$ | Ref |
|---------|---------|-------|-----|-----------------|-----------------|-----------------|----------------|-----|
| 4'-OH CB 118 | RLBA | Human TTR | 19.3 | 4.1 | [21] |
| 4'-OH CB 121 | RLBA | Human TTR | 40 | 1.55 | [25] |
| 4'-OH CB 127 | RLBA | Human rTTR | 10.3 | 8.5 | [7] |
| 4'-OH CB 130 | BA | Human pTTR | 1.02a | | [27] |
| | RLBA | Human TTR | 21.4 | 3.7 | [21] |
| 4'-OH CB 172 | BA | Human pTTR | 1.4a | | [27] |
| | RLBA | Human pTTR | 13 | 4.6 | [28] |
| | RLBA | Human TTR | 3.8 | | [21] |
| 4-OH-CB 187 | RLBA | Gull rTTR | 14 | 1.19 | 2 | 3.15 | [8] |
| | RLBA | Human TTR | 19.8 | 4 | | [21] |
| 4'-OH CB 201 | RLBA | Human TTR | 21 | 3.35 | [23] |
| 4,4'-OH CB 80 | RLBA | Human rTTR | 16.5 | 5.4 | [7] |
| | BA | Human pTTR | 1.36a | | [27] |
| | RLBA | Human TTR | 11 | 5.6 | [25] |
| 4,4'-OH CB 111 | RLBA | Human rTTR | 6.5 | 13.6 | [7] |
| 4,4'-OH CB 202 | RLBA | Human TTR | 16.2 | 4.34 | [23] |
| 5-OH CB 77 | RLBA | Human rTTR | 25 | 3.5 | [7] |
| 5'-OH CB 105 | RLBA | Human rTTR | 19 | 4.6 | [7] |
| 5-OH CB 183 | RLBA | Human TTR | 19.6 | 3.1 | [28] |
| 4-MeO CB 187 | RLBA | Gull rTTR | 981 | 0.016 | 55 | 0.108 | [8] |
| | | | | | | | |
| Bisphenol A | RLBA | Human rTTR | n.d. | n.d. | [20] |
| | RLBA | Seabream rTTR | n.d. | n.d. | [15] |
| | SPR | Human rTTR | <0.001 | | [2] |
| | SPR | Human pTTR | <0.001 | | [2] |
| | RLBA | Chicken pTTR | 9070 | 0.002 | [13] |
| | RLBA | Rana pTTR | 1530 | 0.0009 | [13] |
| | RLBA | Rana rTTR | 500-700 | | [16] |
| | RLBA | Xenopus TTR | >10 000 | | [16] |
| Backbone | Compound                      | Assay | TTR            | \(T_4\) IC\(_{50}\) (nM) | \(T_4\) REP | \(T_3\) IC\(_{50}\) (nM) | \(T_3\) REP | Ref |
|----------|-------------------------------|-------|----------------|--------------------------|------------|--------------------------|------------|-----|
|          |                               | RLBA  | Rana pTTR      | 1500                     | 0.0003     |                          |            | [14]|
|          |                               | RLBA  | Chicken pTTR   | 6000                     | 0.004      |                          |            | [14]|
|          |                               | RLBA  | Rana TTR       | 19.3\(^b\)              |            |                          |            | [14]|
|          |                               | RLBA  | Salmon TTR     | 74\(^b\)                |            |                          |            | [14]|
|          |                               | RLBA  | Human pTTR     | n.d.                     | n.d.       | TS                       |            |     |
|          | 3-Chlorobisphenol A           | RLBA  | Chicken pTTR   | 367                      | 0.06       |                          |            | [13]|
|          |                               | RLBA  | Rana pTTR      | 173                      | 0.008      |                          |            | [13]|
|          | 3,3',Dichlorobisphenol A      | RLBA  | Chicken pTTR   | 86.3                     | 0.26       |                          |            | [13]|
|          |                               | RLBA  | Rana pTTR      | 18.3                     | 0.078      |                          |            | [13]|
|          | 3,5-Dichlorobisphenol A       | RLBA  | Chicken pTTR   | 19                       | 1.18       |                          |            | [13]|
|          |                               | RLBA  | Rana pTTR      | 44.7                     | 0.032      |                          |            | [13]|
|          | 3,3',5-Trichlorobisphenol A   | RLBA  | Chicken pTTR   | 33.7                     | 0.67       |                          |            | [13]|
|          |                               | RLBA  | Rana pTTR      | 10.8                     | 0.13       |                          |            | [13]|
|          | 3,3',5,5'-Tetrachlorobisphenol A | RLBA  | Chicken pTTR  | 22.7                     | 0.99       |                          |            | [13]|
|          |                               | RLBA  | Rana pTTR      | 18.7                     | 0.076      |                          |            | [13]|
|          |                               | RLBA  | Human rTTR     | 107                      | 0.76       |                          |            | [20]|
|          |                               | SPR   | Human rTTR     | 0.75                     |            |                          |            | [2] |
|          | Bisphenol A bis (2,3-dihydroxypropyl) ether | RLBA  | Human rTTR     | n.d.                     | n.d.       |                          |            | [20]|
|          | Bisphenol A bis (3-chloro-2-hydroxypropyl) ether | RLBA  | Human rTTR     | n.d.                     | n.d.       |                          |            | [20]|
|          | Bisphenol A diglycidyl ether  | RLBA  | Human rTTR     | n.d.                     | n.d.       |                          |            | [20]|
|          | Bisphenol A diglycidyl ether, brominated | RLBA  | Human rTTR     | n.d.                     | n.d.       |                          |            | [20]|
| Compound                        | Assay | TTR          | T\textsubscript{4} IC\textsubscript{50} (nM) | T\textsubscript{4} REP | T\textsubscript{3} IC\textsubscript{50} (nM) | T\textsubscript{3} REP | Ref |
|--------------------------------|-------|--------------|---------------------------------------------|------------------------|---------------------------------------------|------------------------|-----|
| Monobromobisphenol A           | RLBA  | Human rTTR   | n.d.                                        | n.d.                   |                                             |                        | [20]|
| Dibromo bisphenol A            | RLBA  | Human rTTR   | n.d.                                        | n.d.                   |                                             |                        | [20]|
| Tribromobisphenol A            | RLBA  | Human rTTR   | 140                                         | 0.58                   |                                             |                        | [20]|
| Tetrabromo bisphenol A (TBBPA) | RLBA  | Human pTTR   | 31                                          | 1.6                    |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| 2.1                                         | 10                     |                                             |                        | [15]|
|                                | RLBA  | Human rTTR   | 7.7                                         | 10.6                   |                                             |                        | [20]|
|                                | SPR   | Human rTTR   |                                             | 1.5                    |                                             |                        | [2] |
|                                | SPR   | Human pTTR   |                                             | 1.06                   |                                             |                        | [2] |
| TBBPA-DBPE                     | RLBA  | Human pTTR   | 5200                                        | 0.0086                 |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| n.d.                                        | n.d.                   |                                             |                        | [15]|
| Pentamix PBDE                  | RLBA  | Seabream rTTR| 8.3                                         | 2.5                    |                                             |                        | [15]|
| Octamix PBDE                   | RLBA  | Human pTTR   | n.d.                                        | n.d.                   |                                             |                        | [6] |
| BDE 19                         | RLBA  | Human pTTR   | n.d.                                        | n.d.                   |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| 44                                          | 0.5                    |                                             |                        | [15]|
| BDE 28                         | RLBA  | Human pTTR   | n.d.                                        | n.d.                   |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| 14.9                                       | 1.4                    |                                             |                        | [15]|
| BDE 38                         | RLBA  | Human pTTR   | >25 000                                     | n.d.                   |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| 65                                          | 0.3                    |                                             |                        | [15]|
| BDE 39                         | RLBA  | Human pTTR   | n.d.                                        | n.d.                   |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| 9.5                                         | 2.2                    |                                             |                        | [15]|
| BDE 47                         | RLBA  | Human pTTR   | >25 000                                     | n.d.                   |                                             |                        | [6] |
|                                | RLBA  | Human rTTR   | 36 000                                      | 0.0025                 |                                             |                        | [17]|
|                                | RLBA  | Seabream rTTR| 5.3                                         | 4                      |                                             |                        | [15]|
|                                | RLBA  | Gull rTTR    | 89                                          | 0.176                  | 529                                         | 0.011                  | [8] |
| BDE 49                         | RLBA  | Human pTTR   | >25 000                                     | n.d.                   |                                             |                        | [6] |
|                                | RLBA  | Seabream rTTR| 0.5                                         | 42                     |                                             |                        | [15]|
| Compound | Assay | TTR | T₄ IC₅₀ (nM) | T₄ REP | T₃ IC₅₀ (nM) | T₃ REP | Ref |
|----------|-------|-----|-------------|--------|-------------|--------|-----|
| BDE 79   | RLBA  | Human pTTR | n.d. | n.d. | [6] |
| BDE 99   | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | 6.7 | 3.1  | [15] |
| BDE 100  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | 56 | 0.4  | [15] |
| BDE 127  | RLBA  | Human pTTR | 22 000 | 0.0025 | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 153  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 155  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 169  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 181  | RLBA  | Human pTTR | 7000 | 0.01 | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 183  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 185  | RLBA  | Human pTTR | 7400 | 0.01 | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 190  | RLBA  | Human pTTR | 9000 | 0.008 | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 206  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| BDE 209  | RLBA  | Human pTTR | n.d. | n.d. | [6] |
|          | RLBA  | Seabream rTTR | n.d. | n.d. | [15] |
| 2'-OH BDE 3 | BA  | Human TTR | 200 | 0.15 | 1.4 | [3] |
|          | FLU-TTR | Human TTR | 4132 | 0.06 | [4] |
| 2'-OH BDE 7 | BA  | Human TTR | 100 | 0.3  | 2.8 | [3] |
| Backbone | Compound     | Assay       | TTR           | $T_4$ IC$_{50}$ (nM) | $T_4$ REP | $T_3$ IC$_{50}$ (nM) | $T_3$ REP | Ref |
|----------|--------------|-------------|---------------|----------------------|-----------|----------------------|-----------|-----|
| 2'-OH BDE 28 | FLU-TTR Human TTR | 864 | 0.3 | [4] |
| 2'-OH BDE 28 | BA Human TTR | 10 | 3.0 | 28 | [3] |
| 2'-OH BDE 28 | RLBA Human rTTR | n.d. | n.d. | [20] |
| 2'-OH BDE 66 | FLU-TTR Human TTR | 900 | 0.29 | [4] |
| 2'-OH BDE 68 | RLBA Human TTR | 170 | 0.65 | [17] |
| 2'-OH BDE 68 | ANSA Human TTR | 230 | 0.93 | [26] |
| 2'-OH BDE 68 | BA Human TTR | 8 | 3.75 | 35 | [3] |
| 3'-OH BDE 7 | BA Human TTR | 50 | 0.6 | 5.6 | [3] |
| 3'-OH BDE 7 | FLU-TTR Human TTR | 375 | 0.69 | [4] |
| 3'-OH BDE 28 | BA Human TTR | 30 | 1.0 | 9.3 | [3] |
| 3'-OH BDE 28 | FLU-TTR Human TTR | 360 | 0.72 | [4] |
| 3-OH BDE 47 | RLBA Human TTR | 17 | 4.0 | [17] |
| 3-OH BDE 47 | BA Human TTR | 7 | 4.3 | 40 | [3] |
| 3-OH BDE 47 | FLU-TTR Human TTR | 110 | 2.36 | [4] |
| 3-OH BDE 100 | FLU-TTR Human TTR | 219 | 1.19 | [4] |
| 3-OH BDE 154 | BA Human TTR | 10 | 3.0 | 28 | [3] |
| 3-OH BDE 154 | FLU-TTR Human TTR | 200 | 1.3 | [4] |
| 4'-OH BDE 17 | BA Human TTR | 20 | 1.5 | 14 | [3] |
| 4-OH BDE 42 | RLBA Human TTR | 19 | 3.5 | [17] |
| 4-OH BDE 42 | BA Human TTR | 9 | 3.3 | 31 | [3] |
| 4-OH BDE 42 | ANSA Human TTR | 290 | 0.93 | [26] |
| 4'-OH BDE 49 | RLBA Human TTR | 19 | 3.5 | [17] |
| 4'-OH BDE 49 | RLBA Gull rTTR | 7.7 | 2.05 | 4.89 | 1.12 | [8] |
| 4'-OH BDE 49 | BA Human TTR | 10 | 3.0 | 28 | [3] |
| 4-OH BDE 188 | FLU-TTR Human TTR | 190 | 1.37 | [4] |
| 5-OH BDE 47 | RLBA Human TTR | 25 | 3.0 | [17] |
| 5-OH BDE 47 | BA Human TTR | 4 | 7.5 | 70 | [3] |
| 5-OH BDE 47 | FLU-TTR Human TTR | 218 | 1.19 | [4] |
| Backbone | Compound                   | Assay   | TTR       | $T_4$ IC$_{50}$ (nM) | $T_4$ REP | $T_3$ IC$_{50}$ (nM) | $T_3$ REP | Ref     |
|-----------|----------------------------|---------|-----------|----------------------|----------|----------------------|----------|---------|
| 6-OH BDE 47 | RLBA Human rTTR           | 180     | 0.26      |                      |          |                      |          | [6]     |
|           | RLBA Human TTR            | 150     | 0.39      |                      |          |                      |          | [17]    |
|           | RLBA Seabream rTTR        | 700     | 0.03      |                      |          |                      |          | [15]    |
|           | RLBA Gull rTTR            | 11.9    | 1.32      | 69                   | 0.07     |                      |          | [8]     |
|           | BA Human TTR              | 15      | 2.0       | 19                   |          |                      |          | [3]     |
|           | ANSA Human TTR            | 630     | 0.42      |                      |          |                      |          | [26]    |
|           | FLU-TTR Human TTR         | 323     | 0.81      |                      |          |                      |          | [4]     |
| 6-OH BDE 85 | BA Human TTR              | 7       | 4.3       | 40                   |          |                      |          | [3]     |
| 6-MeO BDE 47 | RLBA Gull rTTR           | 54.3    | 0.29      | 234                  | 0.025    |                      |          | [8]     |
| Triclosan  | RLBA Human pTTR           | 4179    | 0.015     |                      | TS       |                      |          |         |
| 2-Bromo-4-(2,4,6-tribromo-phenoxy) phenol | RLBA Human rTTR | 66 | 1.22 |                      |          |                      |          | [20]    |
| 2,6-Dibromo-4-(2,4,6-tribromo-phenoxy) phenol | RLBA Human rTTR | 57 | 1.42 |                      |          |                      |          | [20]    |
| 4-(2,4,6-Tribromo-phenoxy) phenol | RLBA Human rTTR | 199 | 0.41 |                      |          |                      |          | [20]    |
| o,p'-DDT   | RLBA Human TTR            | n.d.    | n.d.      |                      |          |                      |          | [25]    |
| o,p'-DDD   | RLBA Human TTR            | n.d.    | n.d.      |                      |          |                      |          | [25]    |
| p,p'-DDE   | RLBA Human TTR            | n.d.    | n.d.      |                      |          |                      |          | [25]    |
| p,p'-DDT   | RLBA Human TTR            | n.d.    | n.d.      |                      |          |                      |          | [25]    |
| Dicofol (Keltane) | RLBA Chicken pTTR | 5000 | 0.004 |                      |          |                      |          | [14]    |
|           | RLBA Rana TTR             | 196.3$^b$ |          |                      |          |                      |          | [14]    |
|           | RLBA Salmon TTR           | 96.7$^b$ |          |                      |          |                      |          | [14]    |
| Hexachlorophene | RLBA Human TTR | 600 | 0.67 |                      |          |                      |          | [24]    |
| Diethylstilbestrol | RLBA Seabream rTTR | 4850 | 0.004 |                      |          |                      |          | [15]    |
|           | RLBA Rana rTTR            | 9       |           |                      |          |                      |          | [16]    |
|           | RLBA Xenopus TTR          | 400-550 |          |                      |          |                      |          | [16]    |
| Backbone | Compound | Assay | TTR   | $T_4$ IC$_{50}$ (nM) | $T_4$ REP | $T_3$ IC$_{50}$ (nM) | $T_3$ REP | Ref |
|----------|----------|-------|-------|----------------------|-----------|----------------------|-----------|-----|
|          |          | RLBA  | Rana pTTR | 0.3                  | 1.7       |                      |           | [14]|
|          |          | RLBA  | Chicken pTTR | 0.4                  | 53        |                      |           | [14]|
|          |          | RLBA  | Rana TTR  | 4.1$^b$              |           |                      |           | [14]|
|          |          | RLBA  | Salmon TTR | 7.4$^b$              |           |                      |           | [14]|
| Dioxin, 2 ar | 2-OH-1,37,8-CDD | RLBA  | Human rTTR | 31.6                 | 4.37      |                      |           | [7] |
|          | 7-OH-2,3,8-CDD | RLBA  | Human rTTR | 136                  | 1         |                      |           | [7] |
|          | 8-OH-2,3-CDD | RLBA  | Human rTTR | >1000                | <<1       |                      |           | [7] |
| Furan, 2 ar | 2-OH-7,8-CDF | RLBA  | Human rTTR | >1000                | <<1       |                      |           | [7] |
|          | 3-OH-2,6,7,8-CDF | RLBA  | Human rTTR | 30.2                 | 1         |                      |           | [7] |
|          | 8-OH-2,3,4-CDF | RLBA  | Human rTTR | >1000                | <<1       |                      |           | [7] |
|          | Diphenyl sulfoxide | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
|          | (2E)-1,4-Diphenyl-2- butene-1,4-dione | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
|          | Diclofenac | RLBA  | Human pTTR | 2200                 | 0.032     |                      |           | TS  |
|          | Triclocarban | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
|          | Perinaphthenone | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
| PAH, >2 ar | Aminopyrene | RLBA  | Rana TTR  | 69.4$^b$             |           |                      |           | [14]|
|          |          | RLBA  | Salmon TTR | 43.5$^b$             |           |                      |           | [14]|
|          | Benzo(a)pyrene | RLBA  | Rana TTR  | 96$^b$               |           |                      |           | [14]|
|          |          | RLBA  | Salmon TTR | 96.6$^b$             |           |                      |           | [14]|
|          | 7H-Benzo[de]anthracen-7-onea | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
| Organo phosphate | Tris(2-chloro-isopropyl) phosphate | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
|          | Tris(2-chloro-ethyl) phosphate | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
|          | Triphenyl phosphate | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
|          | Tris(2-ethylhexyl) phosphate | RLBA  | Human pTTR | n.d.                 | n.d.      |                      |           | TS  |
| Backbone | Compound                          | Assay | TTR              | $T_4IC_{50}$ (nM) | $T_4$ REP | $T_3IC_{50}$ (nM) | $T_3$ REP | Ref  |
|----------|----------------------------------|-------|------------------|-------------------|---------|------------------|---------|------|
| Phthalate| Cresyl diphenylphosphate         | RLBA  | Human pTTR       | n.d.              | n.d.    |                  |         | TS   |
|          | Butylbenzyl phthalate            | RLBA  | Rana TTR         | 75.4$^b$          |         |                  |         | [14] |
|          |                                  | RLBA  | Salmon TTR       | 78.9$^b$          |         |                  |         | [14] |
|          |                                  | RLBA  | Human pTTR       | n.d.              | n.d.    |                  |         | TS   |
| Phthalate| Di-2-ethylhexyl phthalate        | RLBA  | Rana TTR         | 97.6$^b$          |         |                  |         | [14] |
|          |                                  | RLBA  | Salmon TTR       | 82.8$^b$          |         |                  |         | [14] |
| Phthalate| Di-n-butyl phthalate             | RLBA  | Rana TTR         | 89.2$^b$          |         |                  |         | [14] |
|          |                                  | RLBA  | Salmon TTR       | 85.8$^b$          |         |                  |         | [14] |
|          |                                  | RLBA  | Human pTTR       | n.d.              | n.d.    |                  |         | TS   |
| Phthalate| Mirex                            | RLBA  | Rana TTR         | 30.1$^b$          |         |                  |         | [14] |
|          |                                  | RLBA  | Salmon TTR       | 92.4$^b$          |         |                  |         | [14] |
| Phthalate| alpha-HBCDD                       | RLBA  | Human rTTR       | 12 000            | 0.0027  |                  |         | [6]  |
|          |                                  | RLBA  | Seabream rTTR    | n.d.              | n.d.    |                  |         | [15] |
| Phthalate| beta-HBCDD                        | RLBA  | Human rTTR       | 25 000            | 0.0023  |                  |         | [6]  |
|          |                                  | RLBA  | Seabream rTTR    | n.d.              | n.d.    |                  |         | [15] |
| Phthalate| gamma-HBCDD                       | RLBA  | Human rTTR       | n.d.              |         |                  |         | [6]  |
|          |                                  | RLBA  | Seabream rTTR    | n.d.              | n.d.    |                  |         | [15] |
| Phthalate| HBCDD technical mixture           | RLBA  | Human rTTR       | n.d.              |         |                  |         | [6]  |
| Cyclo-alkane| Retinoic acid                  | RLBA  | Rana rTTR        | >50 000           |         |                  |         | [16] |
|          |                                  | RLBA  | Xenopus TTR      | >50 000           |         |                  |         | [16] |
| Cyclo-alkane| Retinol                     | RLBA  | Rana rTTR        | >50 000           |         |                  |         | [16] |
|          |                                  | RLBA  | Xenopus TTR      | >50 000           |         |                  |         | [16] |
| Alkanes  | Cholesterol                      | ANSA  | Human TTR        | > 30 000          | <0.01   |                  |         | [26] |
|          | Naphtenic acid                   | RLBA  | Human pTTR       | n.d.              | n.d.    |                  |         | TS   |
|          | Hexaethyleneglycol monododecyl ether| RLBA  | Human rTTR       | n.d.              | n.d.    |                  |         | TS   |
| Backbone | Compound | Assay | TTR | T₄ IC₅₀ (nM) | T₃ IC₅₀ (nM) | Ref |
|----------|----------|-------|-----|-------------|-------------|-----|
| Malathion | RLBA     | Rana pTTR | 27 000 | <<         |              | [14]|
|          | RLBA     | Chicken pTTR | 900 | 0.02 |              | [14]|
|          | RLBA     | Rana TTR | 87ᵇ |              |              | [14]|
|          | RLBA     | Salmon TTR | 91.7ᵇ |       |              | [14]|
| Methoprene | RLBA    | Human TTR | n.d. |       |              | [25]|
| Squalene   | RLBA    | Human pTTR | n.d. | n.d. |            | TS |
| Octanoic acid | RLBA | Human rTTR | n.d. | n.d. |              | [9] |
| Decanoic acid | RLBA | Human rTTR | n.d. | n.d. |              | [9] |
| Dodecanoic acid | ANSA | Human TTR | 11 000 | 0.02 |            | [26]|
| Myristic acid | ANSA | Human TTR | 6500 | 0.04 |            | [26]|
|          | RLBA | Human rTTR | n.d. | n.d. |            | [9] |
| Palmitic acid | ANSA | Human TTR | n.d. | 0.12ᵇ |            | [26]|
| Stearic acid | ANSA | Human TTR | > 300 000 | <0.001 |            | [26]|
| Arachidonic acid | ANSA | Human TTR | 2300 | 0.12 |            | [26]|
| Linoleic acid | ANSA | Human TTR | 6500 | 0.04 |            | [26]|
| Oleic acid | ANSA | Human TTR | 3300 | 0.08 |            | [26]|
| PFBA | RLBA | Human rTTR | n.d. | n.d. |            | [9] |
| PFPeA | RLBA | Human rTTR | 439 | 0.18 |            | [21]|
| PFHxA | RLBA | Human rTTR | 8220 | 0.007 |            | [9] |
| PFHpA | RLBA | Human rTTR | 1565 | 0.039 |            | [9] |
| PFOA | RLBA | Human rTTR | 949 | 0.064 |            | [9] |
| PFNA | RLBA | Human rTTR | 2737 | 0.022 |              | [9] |
| PFDCa | RLBA | Human rTTR | 8954 | 0.007 |            | [9] |
| PFUnA | RLBA | Human rTTR | 21560 | 0.003 |              | [9] |
| Backbone | Compound   | Assay | TTR            | $T_4$ IC$_{50}$ (nM) | $T_3$ IC$_{50}$ (nM) | $T_4$ REP | $T_3$ REP | Ref |
|-----------|------------|-------|----------------|----------------------|----------------------|-----------|-----------|-----|
|            | PFDoA      | RLBA  | Human rTTR     | 46894                | 0.001                | [9]       |
|            | PFTrDA     | RLBA  | Human rTTR     | 1234                 | 0.064                | [21]      |
|            | PFTedA     | RLBA  | Human rTTR     | 28996                | 0.002                | [9]       |
|            | DoFHpA     | RLBA  | Human rTTR     | 8637                 | 0.007                | [9]       |
|            | FHUEA      | RLBA  | Human rTTR     | 8848                 | 0.007                | [9]       |
|            | PFBS       | RLBA  | Human rTTR     | 19460                | 0.003                | [9]       |
|            | PFHxS      | RLBA  | Human rTTR     | 717                  | 0.085                | [9]       |
|            | PFOS       | RLBA  | Human rTTR     | 940                  | 0.065                | [9]       |
|            | L-PFDS     | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | L-PFOSi    | RLBA  | Human rTTR     | 1733                 | 0.035                | [9]       |
|            | FTSA (6:2) | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [21]      |
|            | FTOH (6:2) | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | FTOH (8:2) | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | N-MeFOSA   | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | N-EtFOSA   | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | FOSE       | RLBA  | Human rTTR     | 6124                 | 0.01                 | [9]       |
|            | N,N-Me3FOSA| RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | N-MeFOSA   | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |
|            | N-EtFOSA   | RLBA  | Human rTTR     | n.d.                 | n.d.                 | [9]       |

n.d. = not detected  
TS = This Study  
RLBA = Radio Ligand Binding Assay  
BA = Binding Assay  
SPR = Surface Plasmon Resonance based assay  
ANSA = ANSA displacement assay based on the salt 8-anilino-1-naphthalenesulfonic acid ammonium  
FLU-TTR = A fluorescein-thyroxine (F−T$_4$) conjugate assay  
$^a$ Antibody based binding assay with relative binding potencies in plasma (<0.2 is low binding affinity and >0.7 is high affinity).  
$^b$ Effects (%) of competitor (8 000nM) on $[^{125}]$T3 binding to chicken and salmon TTR. <90% is significant binding competition.  
$^c$ Competition (%) of competitor (100 000nM) on $[^{125}]$T4 binding to human. the higher percentage the more potent competitor.  
$^d$ Technical mixture CAS 25154-52-3  
$^e$ Branched CAS 84852-15-3  
$^f$ Linear CAS 104-40-5.  
$^g$ Calculated on IC20 value
### Table S2. Brief explanation of the chemical descriptors calculated using the Molecular Operating Environment software

| Descriptor | Description |
|------------|-------------|
| VDistEq   | If $m$ is the sum of the distance matrix entries then VDistEq is defined to be the sum of $\log_2 m - p_i \log_2 p_i / m$ where $p_i$ is the number of distance matrix entries equal to $i$. |
| VDistMa   | If $m$ is the sum of the distance matrix entries then VDistMa is defined to be the sum of $\log_2 m - D_{ij} \log_2 D_{ij} / m$ over all $i$ and $j$. |
| b_1rotR   | Fraction of rotatable single bonds: $b_1rotN$ divided by $b_{\text{heavy}}$. |
| Weight    | Molecular weight (including implicit hydrogens) with atomic weights taken from [CRC 1994]. |
| chi0      | Atomic connectivity index (order 0) from [Hall 1991] and [Hall 1977]. this is calculated as the sum of $1/\sqrt{d_i}$ over all heavy atoms $i$ with $d_i > 0$. |
| chi1      | Atomic connectivity index (order 1) from [Hall 1991] and [Hall 1977]. this is calculated as the sum of $1/\sqrt{d_id_j}$ over all bonds between heavy atoms $i$ and $j$ where $i < j$. |
| VAdjEq    | Vertex adjacency information (equality): $-(1-f) \log_2 (1-f) - f \log_2 f$ where $f = (n^2 - m) / n^2$, $n$ is the number of heavy atoms and $m$ is the number of heavy-heavy bonds. If $f$ is not in the open interval $(0,1)$, then 0 is returned. |
| VAdjMa    | Vertex adjacency information (magnitude): $1 + \log_2 m$ where $m$ is the number of heavy-heavy bonds. If $m$ is zero, then zero is returned. |
| balabanJ  | Balaban's connectivity topological index [Balaban 1982]. |
| PEOE_PC+  | Total positive partial charge: the sum of the positive $q_i$. $Q_{\text{PC+}}$ is identical to $PC+$ which has been retained for compatibility. |
| PEOE_PC-  | Total negative partial charge: the sum of the negative $q_i$. $Q_{\text{PC-}}$ is identical to $PC-$ which has been retained for compatibility. |
| PEOE_RPC+ | Relative positive partial charge: the largest positive $q_i$ divided by the sum of the positive $q_i$. $Q_{\text{RPC+}}$ is identical to $RPC+$ which has been retained for compatibility. |
| PEOE_RPC- | Relative negative partial charge: the smallest negative $q_i$ divided by the sum of the negative $q_i$. $Q_{\text{RPC-}}$ is identical to $RPC-$ which has been retained for compatibility. |
| PEOE_VSA_FHYD | Fractional hydrophobic Van der Waals surface area. this is the sum of the $v_i$ such that $\vert q_i \vert$ is less than or equal to 0.2 divided by the total surface area. the $v_i$ are calculated using a connection table approximation. |
| PEOE_VSA_FNEG | Fractional negative Van der Waals surface area. this is the sum of the $v_i$ such that $q_i$ is negative divided by the total surface area. the $v_i$ are calculated using a connection table approximation. |
| PEOE_VSA_FPNEG | Fractional negative polar Van der Waals surface area. this is the |
sum of the \( v_i \) such that \( q_i \) is less than -0.2 divided by the total surface area. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_FPOL** Fractional polar Van der Waals surface area. this is the sum of the \( v_i \) such that \( |q_i| \) is greater than 0.2 divided by the total surface area. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_FPOS** Fractional positive Van der Waals surface area. this is the sum of the \( v_i \) such that \( q_i \) is non-negative divided by the total surface area. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_FPPOS** Fractional positive polar Van der Waals surface area. this is the sum of the \( v_i \) such that \( q_i \) is greater than 0.2 divided by the total surface area. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_HYD** Total hydrophobic Van der Waals surface area. this is the sum of the \( v_i \) such that \( |q_i| \) is less than or equal to 0.2. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_NEG** Total negative Van der Waals surface area. this is the sum of the \( v_i \) such that \( q_i \) is negative. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_PNEG** Total negative polar Van der Waals surface area. this is the sum of the \( v_i \) such that \( q_i \) is less than -0.2. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_POL** Total polar Van der Waals surface area. this is the sum of the \( v_i \) such that \( |q_i| \) is greater than 0.2. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_POS** Total positive Van der Waals surface area. this is the sum of the \( v_i \) such that \( q_i \) is non-negative. the \( v_i \) are calculated using a connection table approximation.

**PEOE_VSA_PPOS** Total positive polar Van der Waals surface area. this is the sum of the \( v_i \) such that \( q_i \) is greater than 0.2. the \( v_i \) are calculated using a connection table approximation.

**Kier1** First kappa shape index: \((n-1)^2 / m^2 \) [Hall 1991].

**Kier2** Second kappa shape index: \((n-1)^2 / m^2 \) [Hall 1991].

**Kier3** Third kappa shape index: \((n-1) (n-3)^2 / p_5^2 \) for odd \( n \), and \((n-3) (n-2)^2 / p_5^2 \) for even \( n \) [Hall 1991].

**KierFlex** Kier molecular flexibility index: \((KierA1) (KierA2) / n \) [Hall 1991].

**logS** Log of the aqueous solubility this property is calculated from an atom contribution linear atom type model [Hou 2004] with \( r^2 = 0.90 \), \(~1,200\) molecules.

**apol** Sum of the atomic polarizabilities (including implicit hydrogens) with polarizabilities taken from [CRC 1994].

**bpol** Sum of the absolute value of the difference between atomic polarizabilities of all bonded atoms in the molecule (including implicit hydrogens) with polarizabilities taken from [CRC 1994].
**Mr**
Molecular refractivity (including implicit hydrogens). This property is calculated from an 11 descriptor linear model [MREF 1998] with $r^2 = 0.997$, RMSE = 0.168 on 1,947 small molecules.

**SMR**
Molecular refractivity (including implicit hydrogens). This property is an atomic contribution model [Crippen 1999] that assumes the correct protonation state (washed structures). The model was trained on ~7000 structures and results may vary from the mr descriptor.

**TPSA**
Polar surface area calculated using group contributions to approximate the polar surface area from connection table information only. The parameterization is that of Ertl et al. [Ertl 2000].

**Density**
Molecular mass density: Weight divided by vdw_vol.

**vdw_area**
Area of Van der Waals surface calculated using a connection table approximation.

**vdw_vol**
Van der Waals volume calculated using a connection table approximation.

**logP(o/w)**
Log of the octanol/water partition coefficient (including implicit hydrogens). This property is calculated from a linear atom type model [LOGP 1998] with $r^2 = 0.931$, RMSE=0.393 on 1,827 molecules.

**Diameter**
Largest value in the distance matrix [Petitjean 1992].

**Radius**
If $r_i$ is the largest matrix entry in row $i$ of the distance matrix $D$, then the radius is defined as the smallest of the $r_i$ [Petitjean 1992].

**Wiener Path**
Wiener path number: half the sum of all the distance matrix entries as defined in [Balaban 1979] and [Wiener 1947].

**Wiener Polarity**
Wiener polarity number: half the sum of all the distance matrix entries with a value of 3 as defined in [Balaban 1979].

**$a_{aro}$**
Number of aromatic atoms.

**$b_{1rotN}$**
Number of rotatable single bonds. Conjugated single bonds are not included (e.g., ester and peptide bonds).

**$b_{ar}$**
Number of aromatic bonds.

**$b_{double}$**
Number of double bonds. Aromatic bonds are not considered to be double bonds.

**$rings$**
Number of rings.

**Zagreb**
Zagreb index: the sum of $d_i^2$ over all heavy atoms $i$.

**$b_{double/b_count}$**
Number of double bonds. / Number of bonds (including implicit hydrogens). This is calculated as the sum of $(d_i/2 + h_i)$ over all non-trivial atoms $i$.

**$b_{ar/b_count}$**
Number of aromatic bonds / Number of bonds

**$b_{single/b_count}$**
Number of single bonds / Number of bonds

**$a_{aro/a_count}$**
Number of aromatic atoms / Number of atoms

**$a_{don/a_count}$**
Number of hydrogen bond donor atoms / Number of atoms

**$a_{acc/a_count}$**
Number of hydrogen bond acceptor atoms (not counting acidic atoms but counting atoms that are both hydrogen bond donors
and acceptors such as -OH). / Number of atoms

| a_hyd / a_count | Number of hydrophobic atoms. / Number of atoms |
|-----------------|-----------------------------------------------|
| rings/a_count   | the number of rings. / Number of atoms        |
Fig. S1. Loading plot with first (p[1]) versus second (p[2]) loading vector from the PCA including the 186 compounds with data from the human TTR assay. Variables are abbreviated according to Table S2.
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