Application of deep eutectic solvents and ionic liquids
in the extraction of catechins from tea

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SUPPLEMENTARY MATERIALS

Summary

This supporting information file includes additional results and information as described in the text of the main article including:

Figure S1. UHPLC-UV chromatogram of a standard solution containing the analyzed catechins and IS.

Figure S2. Effect of water content in DESs on extraction efficiency of catechins.

Figure S3. The experimental data vs predicted data for extraction efficiency of the conversion of (A) EGC, (B) EGCG, (C) GCG, (D) ECG.

Figure S4. Pareto chart showing the values of effects from variables using the extraction efficiency of (A) EGC, (B) EGCG, (C) GCG, (D) ECG.

Figure S5. Representative TIC-MRM chromatogram obtained for an extract of leaves of green tea after DES-SLE procedure using UHPLC-MS/MS method.

Table S1. List of tested deep eutectic solvents (DES).

Table S2. Polarity, viscosity and density of tested deep eutectic solvents (DESs).

Table S3. Rotatable central composite design setting in the original and coded form of the independent variables (X_1, X_2 and X_3).

Table S4. MRM transitions and mass spectrometer parameters.

Table S5. Analysis of variance (ANOVA) for fit of extraction efficiency from central composite design.

Table S6. Summary of calibration curve and linearity range of catechins (n=6).

Table S7. Summary of accuracy, precision and recovery of catechins (n=6).

Table S8. Content of catechins determined in infusion of tea.
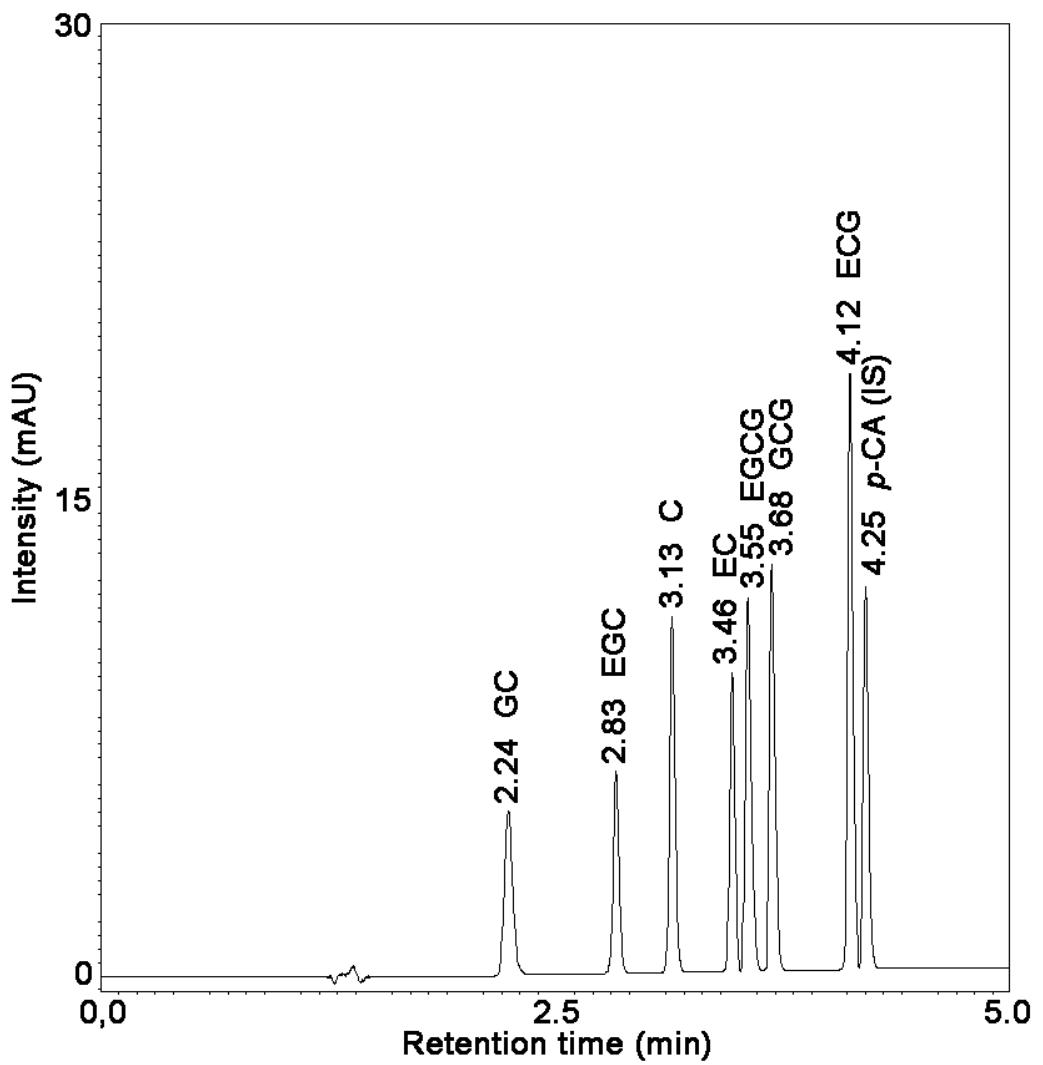
Figure S1. UHPLC-UV chromatogram of a standard solution containing the analyzed catechins (concentration corresponding GC, EGC, C, EC 5 µg/g and EGCG, GCG and ECG 10 µg/g) and IS (concentration corresponding 10 µg/g).
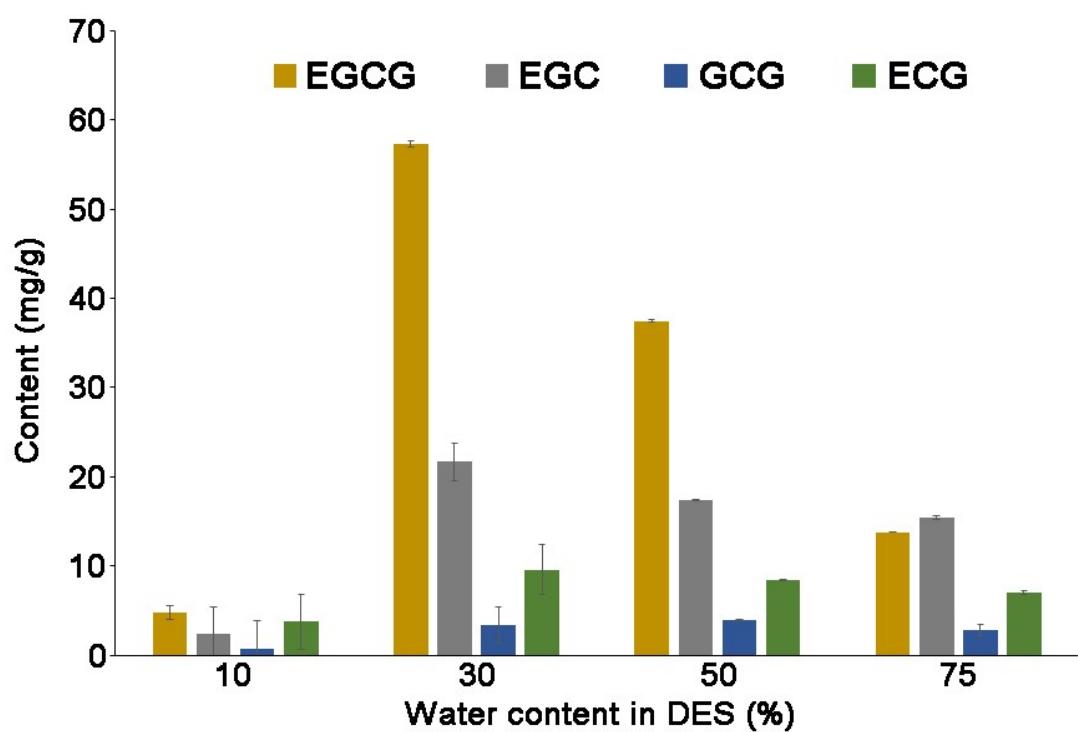
Figure S2. Effect of water content in DESs on extraction efficiency of catechins.
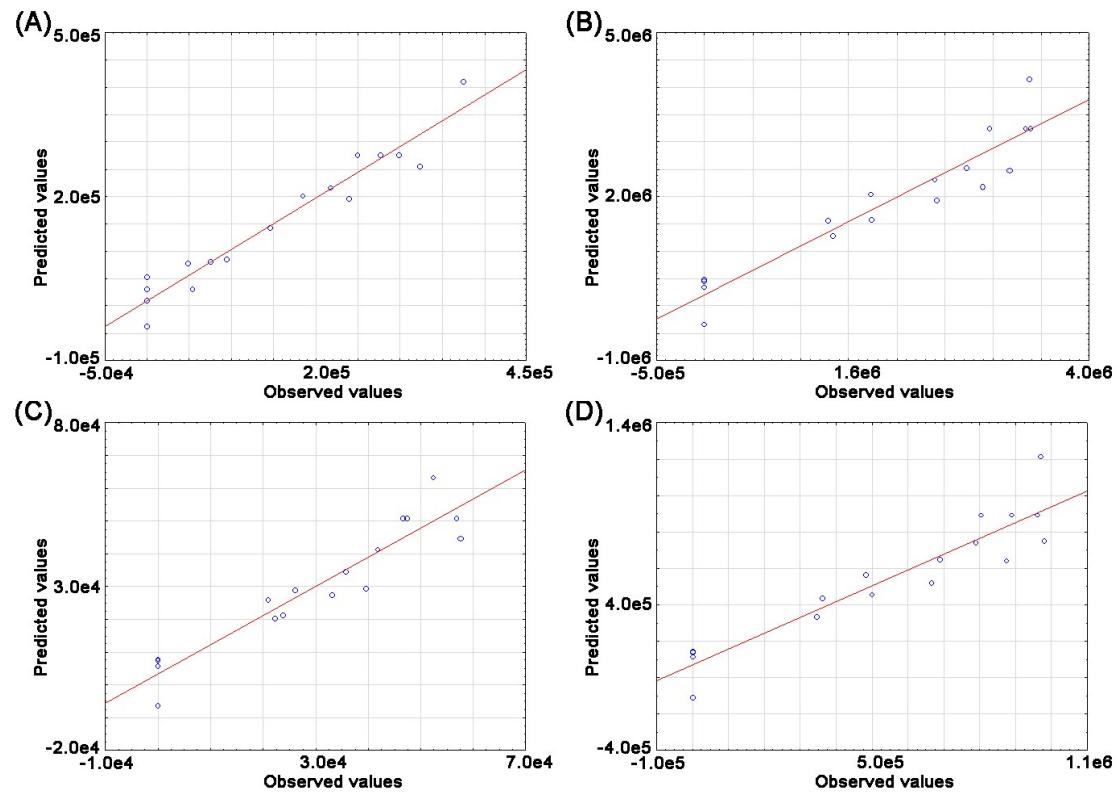
Figure S3. The experimental data vs predicted data for extraction efficiency of the conversion of (A) EGC, (B) EGCG, (C) GCG, (D) ECG.
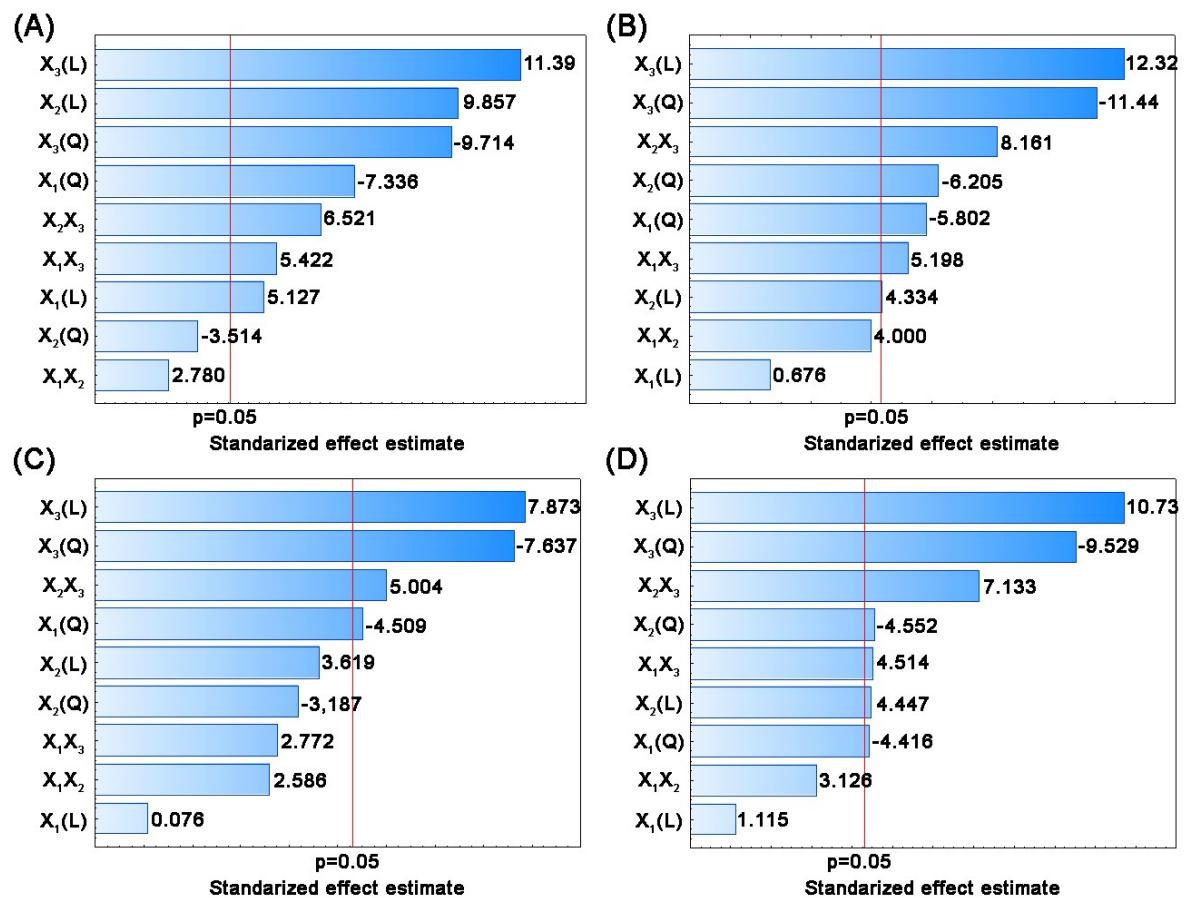
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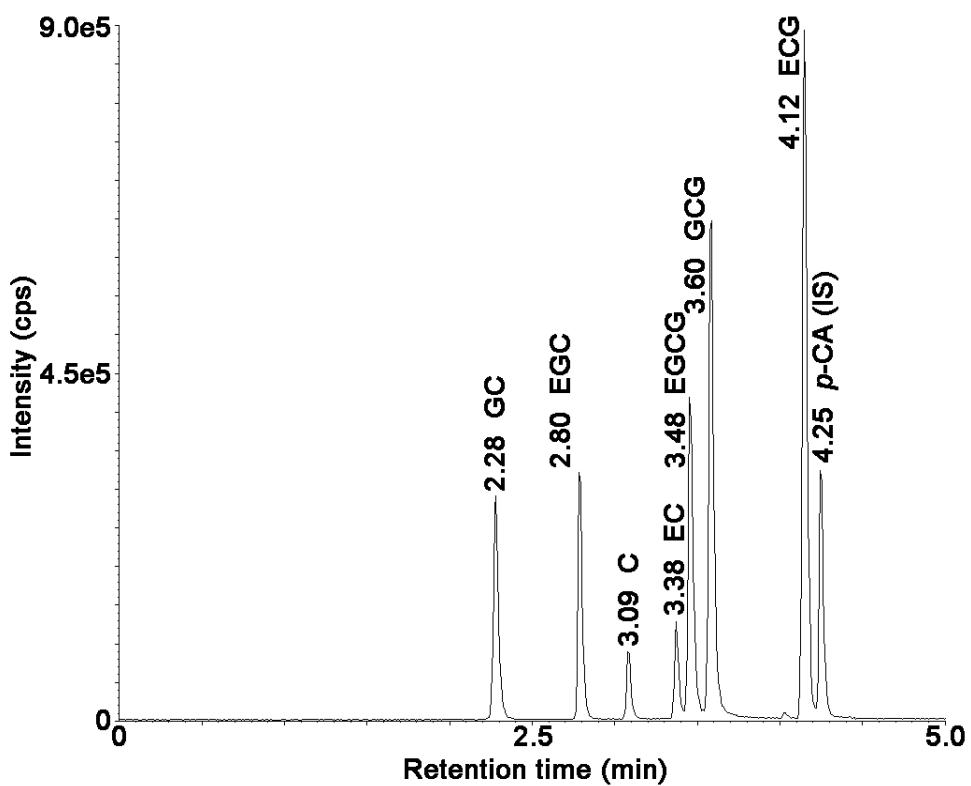
Figure S5. Representative TIC-MRM chromatogram obtained for an extract of leaves of green tea after DES-SLE procedure using UHPLC-MS/MS method.

Table S1. List of tested deep eutectic solvents (DES).

| Number	DES			Mole ratio
	Component 1	**Component 2**	**Component 3**	
DES-1	L-lactic acid	Girard's reagent	–	2:1
DES-2	citric acid	Girard's reagent	–	2:1
DES-3	malic acid	Girard's reagent	–	2:1
DES-4	malic acid	Girard's reagent	–	1:1
DES-5	malic acid	Girard's reagent	–	1:2
DES-6	malic acid	Girard's reagent	L-lactic acid	1:1:1
DES-7	malic acid	Girard's reagent	choline chloride	1:1:1
DES-8	malic acid	Girard's reagent	citric acid	1:1:1

Table S2. Polarity, viscosity and density of tested deep eutectic solvents (DESs).

Solvent	Polarity (kcal/mol)	Viscosity (27ºC) mPa·s	Density (27ºC) g/cm³
DES-1	49.15	13.63	1.169
DES-2	48.08	70.64	1.306
DES-3	47.35	29.10	1.267
DES-4	47.46	30.90	1.241
DES-5	49.42	33.71	1.220
DES-6	48.02	20.11	1.221
DES-7	49.13	17.61	1.182
DES-8	47.21	40.63	1.288

Table S3. Rotatable central composite design setting in the original and coded form of the independent variables (X_1, X_2 and X_3).

Experiment	(X_1) Time (min)	(X_2) Temperature (°C)	(X_3) Solid/liquid ratio (mg/mL)
1	6.5	27.5	1:7
2	20	15	1:4
3	20	15	1:10
4	20	40	1:4
5	20	40	1:10
6	40	6.5	1:7
7	40	27.5	1:2
8	40 (C)	27.5	1:7
9	40 (C)	27.5	1:7
10	40 (C)	27.5	1:7
11	40	27.5	1:12
12	40	48.4	1:7
13	60	15	1:4
14	60	15	1:10
15	60	40	1:4
16	60	40	1:10
17	73.5	27.5	1:7

Table S4. MRM transitions and mass spectrometer parameters.

Compound	$Q1^a$ (m/z)	$Q2^b$ (m/z)	DP^c (V)	CE^e (V)	CXP^f (V)
GC	304.8	124.8	-90	-30	-7
EGC	304.8	124.8	-95	-30	-7
C	288.9	108.6	-70	-36	-3
EC	288.9	108.6	-70	-34	-3
EGCG	456.9	168.9	-75	-26	-3
GCG	456.9	168.8	-95	-27	-7
ECG	441.0	168.9	-75	-26	-3
p-CA (IS)	162.9	118.6	-50	-18	-5

[a] Q1, precursor ion
[b] Q2, fragment ion
[c] DP, declustering potential
[d] EP, entrance potential
[e] CE, collision energy
[f] CXP, cell exit potential

Table S5. Analysis of variance (ANOVA) for fit of extraction efficiency from central composite design.

Source of variation	Sum of square	Df[a]	Mean square	*F*-value[b]	*P*-value
EGC					
X_1	$2.93 \cdot 10^{10}$	1	$2.93 \cdot 10^{10}$	$4.86 \cdot 10^{1}$	$1.99 \cdot 10^{-2}$
X_1^2	$2.59 \cdot 10^{10}$	1	$2.59 \cdot 10^{10}$	$4.30 \cdot 10^{1}$	$2.24 \cdot 10^{-2}$
X_2	$1.10 \cdot 10^{11}$	1	$1.10 \cdot 10^{11}$	$1.83 \cdot 10^{2}$	$5.39 \cdot 10^{-3}$
X_2^2	$5.90 \cdot 10^{7}$	1	$5.90 \cdot 10^{7}$	$9.81 \cdot 10^{-2}$	$7.84 \cdot 10^{-1}$
X_3	$7.81 \cdot 10^{10}$	1	$7.80 \cdot 10^{10}$	$1.29 \cdot 10^{2}$	$7.62 \cdot 10^{-3}$
X_3^2	$7.80 \cdot 10^{10}$	1	$7.80 \cdot 10^{10}$	$1.29 \cdot 10^{2}$	$7.62 \cdot 10^{-3}$
$X_1 X_2$	$4.65 \cdot 10^{9}$	1	$4.65 \cdot 10^{9}$	7.73	$1.09 \cdot 10^{-1}$
$X_1 X_3$	$1.77 \cdot 10^{10}$	1	$1.77 \cdot 10^{10}$	$2.94 \cdot 10^{1}$	$3.24 \cdot 10^{-2}$
$X_2 X_3$	$2.56 \cdot 10^{10}$	1	$2.56 \cdot 10^{10}$	$4.25 \cdot 10^{1}$	$2.27 \cdot 10^{-2}$
Lack of fit	$1.09 \cdot 10^{11}$	5	$2.19 \cdot 10^{10}$	$3.64 \cdot 10^{1}$	$2.69 \cdot 10^{-2}$
Pure error	$1.20 \cdot 10^{9}$	2	$6.01 \cdot 10^{8}$		
Total SS	$4.71 \cdot 10^{11}$	16			
EGCG					
X_1	$2.56 \cdot 10^{10}$	1	$2.56 \cdot 10^{10}$	$4.57 \cdot 10^{-1}$	$5.69 \cdot 10^{-1}$
X_1^2	$1.88 \cdot 10^{12}$	1	$1.88 \cdot 10^{12}$	$3.37 \cdot 10^{1}$	$2.84 \cdot 10^{-2}$
X_2	$1.05 \cdot 10^{12}$	1	$1.05 \cdot 10^{12}$	$1.88 \cdot 10^{1}$	$4.93 \cdot 10^{-2}$
X_2^2	$2.16 \cdot 10^{12}$	1	$2.16 \cdot 10^{12}$	$3.85 \cdot 10^{1}$	$2.50 \cdot 10^{-2}$
X_3	$8.49 \cdot 10^{12}$	1	$8.49 \cdot 10^{12}$	$1.52 \cdot 10^{2}$	$6.53 \cdot 10^{-3}$
X_3^2	$7.32 \cdot 10^{12}$	1	$7.32 \cdot 10^{12}$	$1.31 \cdot 10^{2}$	$7.56 \cdot 10^{-3}$
$X_1 X_2$	$8.95 \cdot 10^{11}$	1	$8.95 \cdot 10^{11}$	$1.60 \cdot 10^{1}$	$5.72 \cdot 10^{-2}$
$X_1 X_3$	$1.51 \cdot 10^{12}$	1	$1.51 \cdot 10^{12}$	$2.70 \cdot 10^{1}$	$3.51 \cdot 10^{-2}$
$X_2 X_3$	$3.73 \cdot 10^{12}$	1	$3.73 \cdot 10^{12}$	$6.66 \cdot 10^{1}$	$1.47 \cdot 10^{-2}$
Lack of fit	$2.70 \cdot 10^{12}$	5	$5.41 \cdot 10^{11}$	9.67	$9.64 \cdot 10^{-2}$
Pure error	$1.12 \cdot 10^{11}$	2	$5.60 \cdot 10^{10}$		
Total SS	$2.66 \cdot 10^{13}$	16			
GCG					
X_1	$1.92 \cdot 10^{5}$	1	$1.92 \cdot 10^{5}$	$5.84 \cdot 10^{-3}$	$9.46 \cdot 10^{-1}$
X_1^2	$6.70 \cdot 10^{8}$	1	$6.70 \cdot 10^{8}$	$2.03 \cdot 10^{1}$	$4.58 \cdot 10^{-2}$
X_2	$4.32 \cdot 10^{8}$	1	$4.32 \cdot 10^{8}$	$1.31 \cdot 10^{1}$	$6.86 \cdot 10^{-2}$
X_2^2	$3.35 \cdot 10^{8}$	1	$3.35 \cdot 10^{8}$	$1.02 \cdot 10^{1}$	$8.59 \cdot 10^{-2}$
X_3	$2.04 \cdot 10^{9}$	1	$2.04 \cdot 10^{9}$	$6.20 \cdot 10^{1}$	$1.58 \cdot 10^{-2}$
X_3^2	$1.92 \cdot 10^{9}$	1	$1.92 \cdot 10^{9}$	$5.83 \cdot 10^{1}$	$1.67 \cdot 10^{-2}$
$X_1 X_2$	$2.20 \cdot 10^{8}$	1	$2.20 \cdot 10^{8}$	6.69	$1.23 \cdot 10^{-1}$
$X_1 X_3$	$2.53 \cdot 10^{8}$	1	$2.53 \cdot 10^{8}$	7.69	$1.09 \cdot 10^{-1}$
$X_2 X_3$	$8.25 \cdot 10^{8}$	1	$8.25 \cdot 10^{8}$	$2.50 \cdot 10^{1}$	$3.77 \cdot 10^{-2}$
Lack of fit	$6.57 \cdot 10^{8}$	5	$1.31 \cdot 10^{8}$	3.99	$2.12 \cdot 10^{-1}$
Pure error	$6.59 \cdot 10^{7}$	2	$3.29 \cdot 10^{7}$		
Total SS	$6.62 \cdot 10^{9}$	16			
ECG					
X_1	$7.61 \cdot 10^{9}$	1	$7.61 \cdot 10^{9}$	1.24	$3.81 \cdot 10^{-1}$
X_1^2	$1.19 \cdot 10^{11}$	1	$1.19 \cdot 10^{11}$	$1.95 \cdot 10^{1}$	$4.76 \cdot 10^{-2}$
X_2	$1.21 \cdot 10^{11}$	1	$1.21 \cdot 10^{11}$	$1.98 \cdot 10^{1}$	$4.70 \cdot 10^{-2}$

X_2^2	$1.27 \cdot 10^{11}$	1	$1.27 \cdot 10^{11}$	$2.07 \cdot 10^{1}$	$4.50 \cdot 10^{-2}$
X_3	$7.04 \cdot 10^{11}$	1	$7.04 \cdot 10^{11}$	$1.15 \cdot 10^{2}$	$8.57 \cdot 10^{-3}$
X_3^2	$5.55 \cdot 10^{11}$	1	$5.55 \cdot 10^{11}$	$9.08 \cdot 10^{1}$	$1.08 \cdot 10^{-2}$
$X_1 X_2$	$5.97 \cdot 10^{11}$	1	$5.97 \cdot 10^{10}$	9.77	$8.89 \cdot 10^{-2}$
$X_1 X_3$	$1.25 \cdot 10^{11}$	1	$1.25 \cdot 10^{11}$	$2.04 \cdot 10^{1}$	$4.57 \cdot 10^{-2}$
$X_2 X_3$	$3.11 \cdot 10^{11}$	1	$3.11 \cdot 10^{11}$	$5.09 \cdot 10^{1}$	$1.91 \cdot 10^{-2}$
Lack of fit	$2.69 \cdot 10^{11}$	5	$5.38 \cdot 10^{10}$	8.79	$1.05 \cdot 10^{-1}$
Pure error	$1.22 \cdot 10^{10}$	2	$6.11 \cdot 10^{9}$		
Total SS	$2.20 \cdot 10^{12}$	16			

Table S6. Summary of calibration curve and linearity range of catechins (n=6).

Analyte	logP	Retention time (min)	Regression equation	S_a	S_b	Linear range (µg/g)	Coefficient of determination r^2
GC	1.49	2.24	y=0.0062x + 0.0039	0.0001	0.0025	1 – 40	0.9988
EGC	1.49	2.83	y=0.0068x + 0.0025	0.0001	0.0028	1 – 40	0.9987
C	1.80	3.13	y=0.0111x + 0.0062	0.0002	0.0051	1 – 40	0.9983
EC	1.80	3.46	y=0.0168x – 0.0011	0.0003	0.0061	1 – 40	0.9990
EGCG	3.08	3.55	y=0.0283x + 0.0091	0.0005	0.0107	1 – 40	0.9989
GCG	3.08	3.68	y=0.0353x + 0.0068	0.0006	0.0128	1 – 40	0.9990
ECG	3.38	4.12	y=0.0434x+0.0053	0.0008	0.0171	1 – 40	0.9988

Table S7. Summary of accuracy, precision and recovery of catechins (n=6).

Analyte	Concentration (μg/g)	Precision[a] (%RSD)		Accuracy[b] (%RE)		Recovery (%)
		Intra-day	Inter-day	Intra-day	Inter-day	
GC	2.5	6.6	8.0	8.7	9.8	97.2
	20	3.2	4.6	1.4	3.3	98.5
	35	6.0	6.4	-4.7	-7.7	95.3
EGC	2.5	9.4	11	4.6	6.3	84.7
	20	2.6	3.5	-5.8	-8.6	97.2
	35	2.3	2.7	-7.6	-9.0	109
C	2.5	8.8	10	7.2	8.6	96.8
	20	3.7	5.7	-6.6	-7.2	86.5
	35	1.1	2.6	-2.3	-4.1	87.7
EC	2.5	7.6	8.5	4.5	5.6	80.2
	20	1.2	2.4	-4.4	-5.0	94.3
	35	1.8	1.7	-9.3	-10	85.2
EGCG	2.5	4.8	5.7	7.2	9.7	77.6
	20	2.6	5.0	-7.5	-9.3	85.2
	35	2.1	1.8	-8.9	-9.1	87.4
GCG	2.5	4.8	5.1	1.9	3.9	69.4
	20	2.4	3.7	-4.8	-5.6	74.1
	35	1.8	2.6	-7.6	-8.4	73.8
ECG	2.5	3.5	5.4	2.4	3.0	67.5
	20	1.7	4.0	-4.6	-6.0	76.3
	35	1.3	1.4	-9.3	-10	77.5

[a] Expressed as relative error (RE (%)).
[b] Expressed as relative standard deviation (RSD (%)).

Table S8. Content of catechins determined in infusion of tea.

Sample		Concentration (mg/g of dry weight[a])						
		GC	EGC	C	EC	EGCG	GCG	ECG
Black tea	1	8.37±0.04[b]	4.25±0.02	1.15±0.10	0.85±0.06	1.54±0.05	0.37±0.01	3.29±0.07
	2	12.4±0.30	2.73±0.08	0.98±0.02	0.38±0.03	2.52±0.01	0.19±0.01	3.30±0.04
	3	6.89±0.69	2.41±0.05	0.86±0.07	0.33±0.03	1.77±0.18	0.28±0.01	1.56±0.07
	4	14.1±1.27	4.21±0.53	0.54±0.04	0.64±0.03	3.40±0.83	0.34±0.03	3.29±0.40
	5	20.3±0.50	1.88±0.02	0.55±0.07	0.52±0.02	2.64±0.10	0.24±0.01	3.20±0.16
	6	2.47±0.04	2.19±0.01	1.09±0.02	0.92±0.09	2.14±0.07	0.06±0.01	1.29±0.01
	7	4.02±0.07	2.88±0.06	0.86±0.15	0.97±0.06	0.93±0.06	0.04±0.01	1.41±0.02
	8	8.23±0.07	0.24±0.02	0.19±0.07	0.34±0.01	0.33±0.08	0.11±0.01	0.59±0.01
	9	5.27±0.03	0.24±0.01	0.20±0.04	0.33±0.08	0.32±0.08	0.10±0.02	0.45±0.01
Green tea	10	4.10±0.29	14.9±0.85	0.16±0.04	3.23±0.18	14.5±1.54	2.61±0.20	3.02±0.30
	11	8.22±0.28	17.3±0.30	1.91±0.15	3.17±0.25	26.7±1.27	3.81±0.11	5.60±0.21
	12	16.6±0.90	15.8±0.21	1.23±0.14	2.34±0.30	31.2±1.06	0.80±0.09	8.47±0.35
	13	3.17±0.25	14.8±0.02	2.45±0.23	3.31±0.14	16.9±0.25	1.35±0.06	4.44±0.04
	14	7.88±0.14	12.2±0.48	1.75±0.23	2.78±0.12	20.3±0.12	2.65±0.06	3.79±0.04
	15	6.98±0.06	11.4±0.42	1.78±0.20	2.16±0.16	31.5±0.09	1.10±0.10	9.20±0.21
	16	5.37±0.15	7.66±0.09	1.99±0.16	5.40±0.15	23.7±0.12	3.41±0.03	8.88±0.01
	17	6.29±0.22	9.48±0.07	0.57±0.01	3.16±0.24	18.8±0.44	1.39±0.10	4.34±0.01
	18	7.49±0.16	16.3±0.74	0.20±0.02	3.90±0.13	24.8±0.37	3.96±0.08	9.79±0.03
Fruit tea	19	ND[c]	8.00±0.09	2.07±0.07	0.14±0.01	0.10±0.01	ND	0.11±0.01
	20	ND	6.00±0.08	1.11±0.06	0.12±0.01	0.13±0.01	ND	0.09±0.01

[a] Each value is the mean (µg/g of dry weight used for prepare of infusion) of three replications
[b] SD relative standard deviation
[c] ND not