

Supplementary Materials

Green Waste from Cucumber (*Cucumis sativus* L.) Cultivation Is a Source of Bioactive Flavonoids with Hypolipidemic Potential

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Table S1. Basic characteristics of six flavonoid-rich cucumber cultivars.

Cultivar (supplier code ^a)	Breeding Code ^b	Direction of Use	Ripening Period	Type of Pollination	Crop Yield, kg/m ²	Growing Conditions	Seasonal Green Biomass (Waste) Yield ^c , kg/m ²
Altai (A)	7806183	Pickling	Early (25–30 days)	Bee-pollinated	3–4	Protected ground, commodity production	10–15
Konkurent (B)	7605854	Pickling	Early (25–30 days)	Bee-pollinated	3–5	Protected ground, commodity production	8–10
Masha (C)	9905332	Salad, canning	Early (25–30 days)	Parthenocarpic	10–11	Horticultural, commodity production	18–25
Parizhskii Kornishon (D)	9609847	Salad, canning	Early (30–32 days)	Parthenocarpic	1–3	Horticultural, commodity production	5–8
Zasolochnii (E)	9900764	Pickling	Middle (42–44 days)	Bee-pollinated	2–3	Protected ground, commodity production	11–17
Zozula (F)	7306415	Salad	Middle (46–48 days)	Parthenocarpic	15–25	Protected ground, commodity production	18–22

^a A—Federal Scientific Center of Vegetable Growing (Vniisok, Russia); B—Vavilov’s All-Russian Institute of Plant Genetic Resources (Saint Petersburg, Russia); C—Monsanto Holland BV (Amstelveen, Netherlands); D—Agrofirma Poisk, Ltd. (Vereya, Russia); E—Vavilov’s Volgograd Experimental Station (Krasnoslobodsk, Russia); F—Edelstein’s Vegetable Experimental Station (Moscow, Russia). ^b Breeding code is an original number assigned to the cultivars by State Commission for Testing and Protection of Breeding Achievements (Gossortkomissia). ^c Dry weight.

Table S2. Total flavonoid yield from cucumber leaves and stems (cv. Masha) in various extraction condition, mg/g DPW^a ± S.D.

Material	Extraction Parameter					
	Solvent Type					
	Water	Methanol	Ethanol	Propanol	Isopropanol	Acetonitrile
Leaves	26.93 ± 0.53	48.63 ± 0.97	49.63 ± 0.99	32.18 ± 0.64	25.83 ± 0.51	9.14 ± 0.18
Stems	0.93 ± 0.02	1.59 ± 0.03	1.83 ± 0.03	1.63 ± 0.03	0.69 ± 0.01	0.11 ± 0.00
	Ethanol Concentration, % of water					
	10	30	50	70	90	
Leaves	29.63 ± 0.59	32.84 ± 0.65	50.75 ± 1.02	51.89 ± 1.02	50.61 ± 1.00	
Stems	1.14 ± 0.02	1.85 ± 0.03	2.03 ± 0.04	2.14 ± 0.04	1.95 ± 0.04	
	Extraction Type ^b					
	USE	MWAE	BWBE	RTE		
Leaves	51.09 ± 1.02	48.12 ± 0.96	45.14 ± 0.90	27.69 ± 0.55		
Stems	2.10 ± 0.04	1.89 ± 0.03	1.53 ± 0.03	0.93 ± 0.02		
	Extraction Duration, min					
	10	20	30	40	50	60
Leaves	21.14 ± 0.42	46.14 ± 0.92	52.11 ± 1.04	52.22 ± 1.04	52.79 ± 1.05	52.83 ± 1.05
Stems	1.20 ± 0.02	1.79 ± 0.03	2.17 ± 0.04	2.18 ± 0.04	2.18 ± 0.04	2.19 ± 0.04

^aDPW—dry plant weight. ^bUSE – ultrasound extraction (50°C), MWAE – microwave-assisted extraction (20°C), BWBE – boiled water bath extraction (95°C), RTE – room temperature extraction (20°C).

Table S3. Regression equation, correlation coefficient (r^2), standard deviation (S_{yx}), limit of detection (LOD), limit of quantification (LOQ), intra- and inter-day precision, repeatability, stability and recovery linear range for isovitexin in spectrophotometric assay.

Regression equation	r^2	S_{yx}	LOD ($\mu\text{g/mL}$)	LOQ ($\mu\text{g/mL}$)	Linear range ($\mu\text{g/mL}$)
$y = 0.057 \cdot x - 0.001$	0.9999	$8.59 \cdot 10^{-3}$	0.46	1.40	2.0–500.0
Precision intra-day (RSD%); $n = 5$	Precision inter-day (RSD%); $n = 4$	Repeatability (RSD%); $n = 7$	Stability (RSD%); $n = 7$	Recovery (%); $n = 5$	
1.12	1.50	1.45	1.40	99.32	

Table S4. Mass-spectrometric (ESI-MS) data of compounds 1–38 found in flavonoid extracts of cucumber waste green biomass.

No.	Compound	ESI-MS data, <i>m/z</i> (Intensity, %)
1	Cucumerin C	MS: 715 [M+H] ⁺ (35), 553 [(M+H)-162] ⁺ (100) MS ² [553]: 463 [(M+H)-162-90] ⁺ (70), 433 [(M+H)-162-120] ⁺ (100)
2	Cucumerin D	MS: 715 [M+H] ⁺ (30), 553 [(M+H)-162] ⁺ (100) MS ² [553]: 463 [(M+H)-162-90] ⁺ (65), 433 [(M+H)-162-120] ⁺ (100)
3	Cucumerin A/B <i>O</i> -pentoside	MS: 685 [M+H] ⁺ (28), 553 [(M+H)-132] ⁺ (100) MS ² [553]: 463 [(M+H)-132-90] ⁺ (69), 433 [(M+H)-132-120] ⁺ (100)
4	Apigenin-C-hexoside-tri- <i>O</i> -hexoside	MS: 919 [M+H] ⁺ (14), 757 [(M+H)-162] ⁺ (3), 595 [(M+H)-162×2] ⁺ (12), 433 [(M+H)-162×3] ⁺ (100) MS ² [433]: 343 [(M+H)-162×3-90] ⁺ (70), 313 [(M+H)-162×3-120] ⁺ (100)
5	Isovitexin-7,4'- <i>O</i> -glucoside	MS: 757 [M+H] ⁺ (10), 595 [(M+H)-162] ⁺ (7), 433 [(M+H)-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-162×2-90] ⁺ (71), 313 [(M+H)-162×2-120] ⁺ (100)
6	Cucumerin A/B <i>O</i> -desoxyhexoside	MS: 699 [M+H] ⁺ (25), 553 [(M+H)-146] ⁺ (100) MS ² [553]: 463 [(M+H)-146-90] ⁺ (70), 433 [(M+H)-146-120] ⁺ (100)
7	Isovitexin-4',2''- <i>O</i> -glucoside	MS: 757 [M+H] ⁺ (14), 595 [(M+H)-162] ⁺ (1), 433 [(M+H)-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-162×2-90] ⁺ (68), 313 [(M+H)-162×2-120] ⁺ (100)
8	Cucumerin A/B <i>O</i> -desoxyhexoside	MS: 699 [M+H] ⁺ (23), 553 [(M+H)-146] ⁺ (100) MS ² [553]: 463 [(M+H)-146-90] ⁺ (72), 433 [(M+H)-146-120] ⁺ (100)
9	Apigenin-C-hexoside-di- <i>O</i> -hexoside- <i>O</i> - <i>p</i> -coumarate	MS: 903 [M+H] ⁺ (31), 757 [(M+H)-146] ⁺ (5), 595 [(M+H)-146-162] ⁺ (11), 433 [(M+H)-146-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162×2-90] ⁺ (68), 313 [(M+H)-146-162×2-120] ⁺ (100)
10	Isovitexin-7- <i>O</i> -(6''- <i>O</i> -glucosyl)-glucoside	MS: 757 [M+H] ⁺ (9), 595 [(M+H)-162] ⁺ (4), 433 [(M+H)-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-162×2-90] ⁺ (69), 313 [(M+H)-162×2-120] ⁺ (100)
11	Isovitexin-4'- <i>O</i> -glucoside-2''- <i>O</i> -(6'''- <i>O</i> - <i>p</i> -coumaroyl)-glucoside	MS: 903 [M+H] ⁺ (12), 757 [(M+H)-146] ⁺ (5), 595 [(M+H)-146-162] ⁺ (2), 433 [(M+H)-146-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162×2-90] ⁺ (69), 313 [(M+H)-146-162×2-120] ⁺ (100)
12	Apigenin-C-hexoside-di- <i>O</i> -hexoside- <i>O</i> -ferulate	MS: 933 [M+H] ⁺ (18), 757 [(M+H)-176] ⁺ (7), 595 [(M+H)-176-162] ⁺ (5), 433 [(M+H)-176-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-176-162×2-90] ⁺ (70), 313 [(M+H)-176-162×2-120] ⁺ (100)
13	Isovitexin-4'- <i>O</i> -glucoside-2''- <i>O</i> -(6'''- <i>O</i> -feruloyl)-glucoside	MS: 933 [M+H] ⁺ (10), 757 [(M+H)-176] ⁺ (2), 595 [(M+H)-176-162] ⁺ (1), 433 [(M+H)-176-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-176-162×2-90] ⁺ (69), 313 [(M+H)-176-162×2-120] ⁺ (100)
14	Apigenin-C-hexoside-di- <i>O</i> -hexoside- <i>O</i> - <i>p</i> -coumarate	MS: 903 [M+H] ⁺ (11), 757 [(M+H)-146] ⁺ (10), 595 [(M+H)-146-162] ⁺ (8), 433 [(M+H)-146-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162×2-90] ⁺ (69), 313 [(M+H)-146-162×2-120] ⁺ (100)
15	Isovitexin-2''- <i>O</i> -glucoside	MS: 595 [M+H] ⁺ (15), 433 [(M+H)-162] ⁺ (100) MS ² [433]: 343 [(M+H)-162-90] ⁺ (70), 313 [(M+H)-162-120] ⁺ (100)
16	Isovitexin-4'- <i>O</i> -glucoside	MS: 595 [M+H] ⁺ (8), 433 [(M+H)-162] ⁺ (100) MS ² [433]: 343 [(M+H)-162-90] ⁺ (71), 313 [(M+H)-162-120] ⁺ (100)
17	Apigenin-C-hexoside-di- <i>O</i> -hexoside- <i>O</i> -ferulate	MS: 933 [M+H] ⁺ (14), 757 [(M+H)-176] ⁺ (11), 595 [(M+H)-176-162] ⁺ (2), 433 [(M+H)-176-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-176-162×2-90] ⁺ (71), 313 [(M+H)-176-162×2-120] ⁺ (100)
18	Apigenin-C-hexoside-di- <i>O</i> -hexoside- <i>O</i> - <i>p</i> -coumarate	MS: 903 [M+H] ⁺ (16), 757 [(M+H)-146] ⁺ (5), 595 [(M+H)-146-162] ⁺ (4), 433 [(M+H)-146-162×2] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162×2-90] ⁺ (69), 313 [(M+H)-146-162×2-120] ⁺ (100)
19	Apigenin-7- <i>O</i> -(6''- <i>O</i> -feruloyl)-glucoside (saponarin-6''- <i>O</i> -ferulate)	MS: 609 [M+H] ⁺ (25), 433 [(M+H)-176] ⁺ (39), 271 [(M+H)-176-162] ⁺ (100) MS ² [271]: 153 [(M+H)-176-162-118] ⁺ (100), 145 [(M+H)-176-162-126] ⁺ (29), 121 [(M+H)-176-162-150] ⁺ (16), 119 [(M+H)-176-162-152] ⁺ (30)

20	Isovitexin-2''-O-(6'''-O- <i>p</i> -coumaroyl)-glucoside	MS: 741 [M+H] ⁺ (20), 595 [(M+H)-146] ⁺ (17), 433 [(M+H)-146-162] ⁺ (100) MS ² [433]: 343 [(M+H)-162-90] ⁺ (70), 313 [(M+H)-162-120] ⁺ (100)
21	Isovitexin-2''-O-glucoside-6''-O- <i>p</i> -coumarate	MS: 741 [M+H] ⁺ (10), 595 [(M+H)-146] ⁺ (2), 433 [(M+H)-146-162] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162-90] ⁺ (67), 313 [(M+H)-146-162-120] ⁺ (100)
22	Isovitexin-2''-O-glucoside-6''-O-ferulate	MS: 771 [M+H] ⁺ (9), 595 [(M+H)-176] ⁺ (1), 433 [(M+H)-176-162] ⁺ (100) MS ² [433]: 343 [(M+H)-176-162-90] ⁺ (68), 313 [(M+H)-176-162-120] ⁺ (100)
23	Apigenin-C-hexoside-O-hexoside-O- <i>p</i> -coumarate	MS: 741 [M+H] ⁺ (12), 595 [(M+H)-146] ⁺ (3), 433 [(M+H)-146-162] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162-90] ⁺ (68), 313 [(M+H)-146-162-120] ⁺ (100)
24	Isovitexin-2''-O-(6'''-O-feruloyl)-glucoside	MS: 771 [M+H] ⁺ (14), 595 [(M+H)-176] ⁺ (8), 433 [(M+H)-176-162] ⁺ (100) MS ² [433]: 343 [(M+H)-176-162-90] ⁺ (69), 313 [(M+H)-176-162-120] ⁺ (100)
25	Isovitexin-2''-O- <i>p</i> -coumarate	MS: 579 [M+H] ⁺ (16), 433 [(M+H)-146] ⁺ (100) MS ² [433]: 343 [(M+H)-146-90] ⁺ (72), 313 [(M+H)-146-120] ⁺ (100)
26	Cucumerin C/D O-hexoside	MS: 877 [M+H] ⁺ (41), 715 [(M+H)-162] ⁺ (30), 553 [(M+H)-162×2] ⁺ (100) MS ² [553]: 463 [(M+H)-162×2-90] ⁺ (67), 433 [(M+H)-162×2-120] ⁺ (100)
27	Cucumerin C/D O-hexoside	MS: 877 [M+H] ⁺ (45), 715 [(M+H)-162] ⁺ (25), 553 [(M+H)-162×2] ⁺ (100) MS ² [553]: 463 [(M+H)-162×2-90] ⁺ (69), 433 [(M+H)-162×2-120] ⁺ (100)
28	Isovitexin-7-O-glucoside	MS: 595 [M+H] ⁺ (10), 433 [(M+H)-162] ⁺ (100) MS ² [433]: 343 [(M+H)-162-90] ⁺ (70), 313 [(M+H)-162-120] ⁺ (100)
29	Apigenin-C-hexoside-O-hexoside-O- <i>p</i> -coumarate	MS: 741 [M+H] ⁺ (12), 595 [(M+H)-146] ⁺ (1), 433 [(M+H)-146-162] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162-90] ⁺ (68), 313 [(M+H)-146-162-120] ⁺ (100)
30	Apigenin-C-hexoside-O-hexoside-O- <i>p</i> -coumarate	MS: 741 [M+H] ⁺ (16), 595 [(M+H)-146] ⁺ (4), 433 [(M+H)-146-162] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162-90] ⁺ (68), 313 [(M+H)-146-162-120] ⁺ (100)
31	Apigenin-C-hexoside-O-hexoside-O- <i>p</i> -coumarate	MS: 741 [M+H] ⁺ (11), 595 [(M+H)-146] ⁺ (4), 433 [(M+H)-146-162] ⁺ (100) MS ² [433]: 343 [(M+H)-146-162-90] ⁺ (67), 313 [(M+H)-146-162-120] ⁺ (100)
32	Isovitexin-2''-O-rhamnoside	MS: 579 [M+H] ⁺ (18), 433 [(M+H)-146] ⁺ (100) MS ² [433]: 343 [(M+H)-146-90] ⁺ (70), 313 [(M+H)-146-120] ⁺ (100)
33	Isovitexin	MS: 433 [M+H] ⁺ (100), 343 [(M+H)-90] ⁺ (35), 313 [(M+H)-120] ⁺ (31)
34	Isovitexin-2''-O-(6'''-O-feruloyl)-glucoside-6''-O-ferulate	MS: 947 [M+H] ⁺ (25), 771 [(M+H)-176] ⁺ (10), 595 [(M+H)-176×2] ⁺ (3), 433 [(M+H)-176×2-162] ⁺ (100) MS ² [433]: 343 [(M+H)-176×2-162-90] ⁺ (69), 313 [(M+H)-176×2-162-120] ⁺ (100)
35	Isovitexin-6''-O- <i>p</i> -coumarate	MS: 579 [M+H] ⁺ (17), 433 [(M+H)-146] ⁺ (100) MS ² [433]: 343 [(M+H)-146-90] ⁺ (70), 313 [(M+H)-146-120] ⁺ (100)
36	Apigenin-C-hexoside-di-O- <i>p</i> -coumarate	MS: 725 [M+H] ⁺ (8), 579 [(M+H)-146] ⁺ (2), 433 [(M+H)-146×2] ⁺ (100) MS ² [433]: 343 [(M+H)-146×2-90] ⁺ (65), 313 [(M+H)-146×2-120] ⁺ (100)
37	Apigenin-C-hexoside-O- <i>p</i> -coumarate-O-ferulate	MS: 755 [M+H] ⁺ (12), 609 [(M+H)-146] ⁺ (1), 579 [(M+H)-176] ⁺ (1), 433 [(M+H)-146-176] ⁺ (100) MS ² [433]: 343 [(M+H)-146-176-90] ⁺ (68), 313 [(M+H)-146-176-120] ⁺ (100)
38	Apigenin-C-hexoside-di-O-ferulate	MS: 785 [M+H] ⁺ (18), 609 [(M+H)-176] ⁺ (7), 433 [(M+H)-176×2] ⁺ (100) MS ² [433]: 343 [(M+H)-176×2-90] ⁺ (69), 313 [(M+H)-176×2-120] ⁺ (100)

Table S5. Hypolipidemic and antioxidant activity of plant total flavonoid extracts.

Source	Model	Hypolipidemic Manifestation	Antioxidant Manifestation
<i>Actinidia kolomikta</i> leaves [63]	Mice hyperlipidemia; lard-based high fat diet; 3 weeks	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, GPx↑, CAT↑, serum MDA↓
<i>Chrysanthemum morifolium</i> flowers [53]	Rat hyperlipidemia; lard-based high fat diet; 6 weeks	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, GPx↑, CAT↑, serum MDA↓
<i>Citrus aurantium</i> pericarp [64]	Golden hamster hyperlipidemia; 2% cholesterol high fat diet; 6 weeks	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, serum MDA↓
<i>Crataegus pinnatifida</i> leaves [55]	Rat hyperlipidemia; 1.2% cholesterol high fat diet; 6 weeks	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, GPx↑, CAT↑
<i>Cucumis sativus</i> leaves (present study)	Golden hamster hyperlipidemia; 1% cholesterol high fat diet; 6 months	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, GPx↑, CAT↑, serum MDA↓
<i>Polygonum capitatum</i> herb [65]	Rat hyperlipidemia; high fat diet; 6 weeks	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, GPx↑, CAT↑, serum MDA↓
<i>Polygonum perfoliatum</i> tubers [66]	Rat hyperlipidemia; 3% cholesterol high fat diet; 6 weeks	Serum TC↓, TG↓, LDLC↓, HDLC↑	Liver SOD↑, GPx↑, CAT↑, serum MDA↓
<i>Solanum melongena</i> fruits [48]	Rat hyperlipidemia; 2% cholesterol high fat diet; 45 days	Serum TC↓, TG↓	-
<i>Trichosanthes kirilowii</i> seeds [67]	Mice hyperlipidemia; lard-based high fat diet; 6 weeks	Serum TC↓, TG↓, LDLC↓	Liver SOD↑, GPx↑, serum MDA↓

CAT—liver catalase; GPx—liver glutathione peroxidase; HDLC—serum high-density lipoprotein-cholesterol; LDLC—serum low-density lipoprotein-cholesterol; MDA—serum malondialdehyde level; SOD—liver superoxide dismutase; TC—total cholesterol (TC); TG—serum total triglycerides; ↓—level decreased; ↑—level increased.

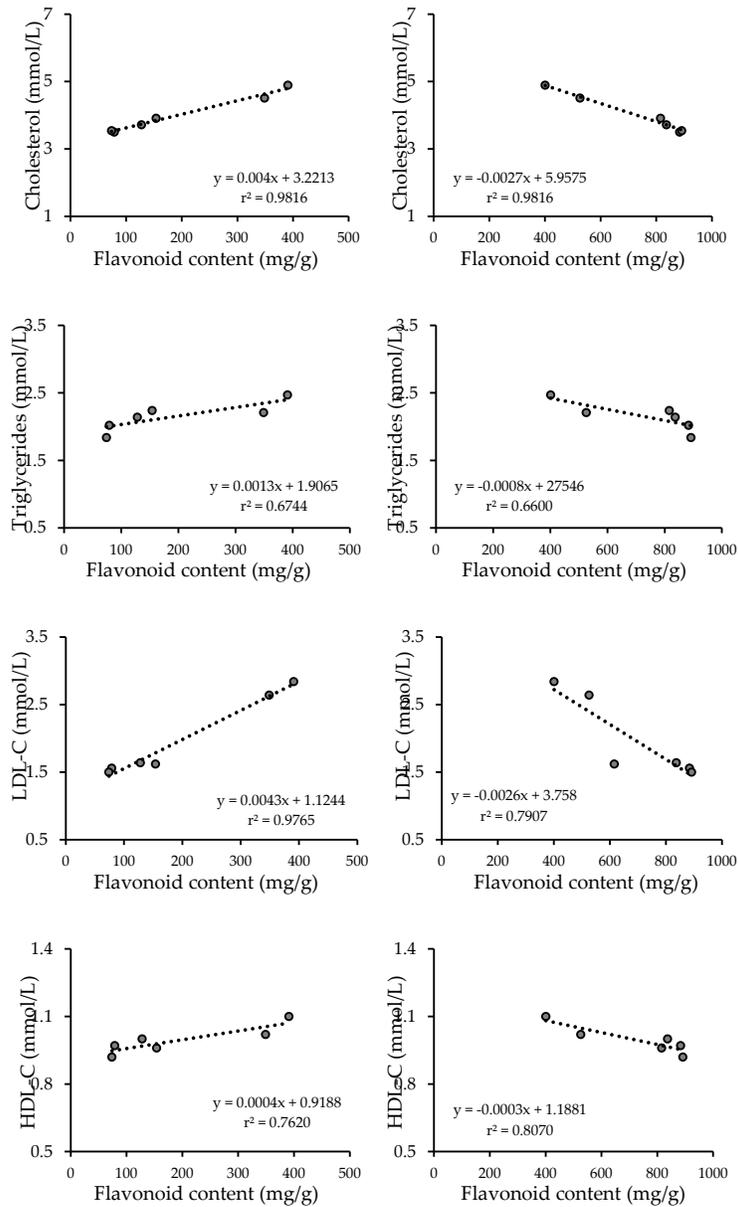


Figure S1. Correlation graphs between flavonoid content in flavonoid extracts of cucumber waste green biomass (mg/g) and values of serum total cholesterol, serum total triglycerides, serum low-density lipoprotein-cholesterol (LDL-C), and serum high-density lipoprotein-cholesterol (HDL-C).