Different effects of feeding pregnant and lactating mice *Rhodiola kirilowii* aqueous and hydro-alcoholic extracts on their serum angiogenic activity and content of selected polyphenols

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**Abstract**

Angiogenesis plays an important role in many physiological processes, among them the formation of tissues and organs during embryogenesis. A lot of medicinal plants exhibit angiomodulatory properties. This creates the need for a thorough check of whether the plant extracts that we would like to give to pregnant women in order to increase their resistance to bacterial or viral infection will have negative effects on angiogenesis, and consequently on fetal development. This paper seeks to investigate the effect of serum of pregnant and nursing Balb/c mice that received aqueous (Rkw) or hydro-alcoholic (Rkw-a) *R. kirilowii* extracts (20 mg/kg), or epigallocatechin (0.2 mg/kg), on the in vitro proliferation and migration of mouse endothelial cell line HECa10. Of the 15 identified polyphenols in the extracts by HPLC, 8 were present in the sera. Chemical analysis revealed higher salidroside, kaempferol, chlorogenic acid, bFGF and VEGF concentration in Rkw-a sera than in the sera of Rkw group of mice. Rkw-a and EGC sera did not affect migration of endothelial cells, however we noted some increase of migrating cells after Rkw-sera treatment. Rkw and EGC sera did not affect proliferation of endothelial cells. Sera of mothers from Rkw-A group impaired the proliferation of endothelial cells in comparison to other groups. These data allow us to assume that Rhodiola kirilowit hydro-alcoholic extract (Rkw-A) is potentially able to modulate pre- and postnatal angiogenesis what might influence the development of organs in progeny. Sera of Rkw mothers have not harm the proliferation of endothelial cells, despite they also contain antiangiogenic catechins and salidroside. This suggests the existence in Rkw-A extract and in Rkw-A sera of some other, as yet unidentified substances influencing endothelial cells proliferation.

**Key words:** mice, pregnancy, *Rhodiola kirilowii*, polyphenols, angiogenesis.

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**Introduction**

Angiogenesis is the formation of new blood vessels on the basis of those already existing. It is an important part of many phenomena and physiological responses, and together with vasculogenesis is driving the development of tissues and organs during embryonic and fetal life. So, one may expect that remedies containing anti-angiogenic substances might be unsafe in pregnancy [1, 2]. Plants of the *Rhodiola* genus have been used for centuries in Eastern medicine as adaptogens and anti-inflammatory drugs, and in our previous studies they presented immunomodulatory properties [3-8]. Despite their beneficial properties *Rhodiola* species were not recommended for use
during pregnancy and lactation, due to a lack of research into their impact on embryonic angiogenesis.

Our studies on these plants (R. rosea, R. quadrifida and R. kirilowii) showed inhibitory effect of their hydro-alcoholic extracts on tumor angiogenesis [9-11]. However, in studies completed in 2012, we discovered that aqueous extract of roots of Rhodiola kirilowii (stimulating a number of parameters of cellular immunity in mice), did not inhibit L1 sarcoma neovascularization [11]. We could not be sure, of course, if it will not have deleterious effect on fetal development and the health of the offspring of mice fed the extract during pregnancy, because the processes of tumor and fetal angiogenesis do not always run in parallel, and, moreover, the extract contains some potentially anti-angiogenic substances belonging to the group of polyphenols. Then, we decided to see what will be the impact of feeding pregnant mice aqueous and hydro-alcoholic extracts of Rhodiola kirilowii on the angiogenic activity and selected polyphenols content of their sera. We also conducted a determination of proangiogenic growth factors, VEGF and bFGF.

Material and methods

Preparation and analysis of Rhodiola extracts

The roots and rhizomes of Rhodiola kirilowii were collected from field cultivations of Institute of Natural Fibers and Medicinal Plants. The raw material was washed, cut into thick slices, dried in natural conditions and next powdered. **Aqueous extract:** finely powdered roots were extracted two times with water (extraction was performed: first – 2 hour and second – 1 hour long, in the ratio raw material/solvent 1/5), in the temperature of 40°-45°C. The supernatants were mixed together and after centrifugation at 3000 rpm for 15 min were lyophilized. **Hydroalcoholic extract:** finely powdered roots were extracted with ethanol/water solution (1/1, v/v, in the ratio raw material/solvent 1/10) by the percolation method. Then the percolates were lyophilized (preceded by the distilling off the ethanol in the temperature 40°-45°C). Extracts were stored at –70°C until further use.

**Chemical analysis of extracts was done** as previously described [1,12]. Total extract polyphenols/flavonoids concentration (16.16 μg/mg and 23.75 μg/mg in RKW and RKW-A, respectively) was assayed by applying the HPLC system (Dionex) equipped in the CoulArray electrochemical detector (ESA Inc.). The content of individual chemical compounds in a dry mass of extract was presented as mean μg/mg ± SEM (mean of 3 independent experiments).

**Epigallocatechin**

Epigallocatechin (EGC) was purchased from Sigma Aldrich (Warsaw, Poland, cat. no. E3768-5MG), dissolved in distilled water and stored at –70°C until used.

**Animals**

Experiments were performed on the adult inbred female Balb/c mice (Mossakowski Medical Research Centre Polish Academy of Sciences), 8-9 weeks old, 20-22 g b.m., mated with adult males from the same strain. Females, since copulatory plug was noted, up to the 28-th day after delivery were fed daily with dissolved in distilled water, lyophilized RKW or RKW-A extract (20 mg/kg b.m.). This dose corresponds to a dose of 100 mg (1.6 mg/kg) given to a person weighing 60 kg with accordance to Shin et al. [13] mouse/human converter. Another group of female mice were supplemented with epigallocatechin (EGC) 0.2 mg/kg b.m. The daily dose of EGC match the total content of epicatechin, epigallocatechin, epicatechin gallate and epigallocatechin gallate present in 400 micrograms (daily dose) of RKW-A extract. Control mothers received the distilled water. Females were housed separately and to avoid stress connected with handling and gavage, tested substances were applied on a corn crisp, and served to the mice in a Petri dish. Control mice received distilled water applied on a corn crisp. On the 28-th day after delivery, mothers were anesthetized, bled and sacrificed. For all performed experiments animals were handled according to the Polish regulation concerning the wellness of laboratory animals (Polish National Institute of Health) standards. All experiments were accepted by the Local Bioethical Committee, (permission 73/2011). Mice were maintained under conventional conditions (room temperature 22.5-23.0°C, relative humidity 50-70%, 12 h day/night cycle) with free access to breeding rodent feed (Labofeed H, Wytównia Pasz “Morawski”) and water.

**Serum**

Mice were bled in anaesthesia (intraperitoneal injection of ketamine 120 mg/kg of b.w. and xylazine 12 mg/kg of b.w. solution) from retro-orbital plexus. Sera were separated by 1-hour clotting (RT), centrifuged at 2000 × g for 20 min and stored at –70°C until analysis.

**HPLC analysis of serum**

Total serum polyphenol/flavonoids concentration from the sera of mothers was assayed by applying the HPLC system (Dionex) equipped with the CoulArray electrochemical detector (ESA Inc.). The extraction procedure of polyphenols/flavonoids was performed as described previously [1,12]. The results are presented as mean ng/ml ± SEM.

**Measurement of growth factors concentration**

The levels of VEGF and bFGF in sera were determined by ELISA tests (R&D Systems) according to the producer’s protocols. The results are presented as mean pg/ml ± SEM.

**Endothelial cell culture**

HECa10 mouse endothelial cell line was kindly provided by Professor Claudine Kieda from the Centre of Mo-
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**Proliferation assay**

Proliferation of endothelial cells was estimated using AlamarBlue® (resazurin), a dye that measures the metabolic activity of living cells.

AlamarBlue assay was performed as described previously [16, 17] with some modifications. Briefly, the cells (in log phase growth stage) were harvested (Accutase PAA), centrifuged, suspended in culture medium (DMEM, 4.5 g/ml glucose with L-glutamine, 10% tested mother-mouse serum, 50 U/ml penicillin, 50 μg/ml streptomycin), to the final density of 0.5 × 10^5 cells/ml and seeded into a 96-well plate (100 μl per well). Next, the plates were incubated at standard culture conditions of 5% CO₂, 37°C, 95% R.H.) with medium renewal every 2-3 days.

**Migration assay**

Migration assays were performed on cells in log phase growth stage as previously described [17]. Briefly, the cells were harvested, centrifuged, suspended in medium with the addition of 10 μg/ml DilC12(3) fluorescent dye (Becton Dickinson Falcon) culture flasks under standard culture conditions (5% CO₂, 37°C, 95% R.H.) with medium renewal every 2-3 days.

**Table 1. HPLC analysis of mothers sera. Mean concentration (ng/ml ± SEM ) polyphenols/flavonoids in the sera of mice-mothers. Statistical differences between RKW and RKW-A sera: salidroside p = 0.03, kaempferol p = 0.0652 (on the border of significance), (+)-catechin and epicatechin p = 0.001. Unpaired t test**

|         | Salidroside | Fisetin | Kaempferol | Ellagic acid | Quercetin | Chlorogenic acid | (+)-catechin | EC    | EGC    |
|---------|-------------|---------|------------|--------------|-----------|-----------------|--------------|-------|--------|
| Control | Not found   | 0.51 ±0.06 | Not found  | 2.51 ±0.21   | 5.32 ±0.46 | 0.62 ±0.03      | 3.36 ±0.27   | 0.62 ±0.14 | 0.55 ±0.08 |
| RKW     | 2.26 ±0.43  | Not found | 3.16 ±0.29 | 2.07 ±0.18   | 4.29 ±0.38 | 0.47 ±0.04      | 4.39 ±0.44   | 1.11 ±0.18 | 0.59 ±0.08 |
| RKW-A   | 4.02 ±0.14  | Not found | 4.20 ±0.44 | 1.76 ±0.08** | 4.46 ±0.40 | 1.26 ±0.38      | 2.03 ±0.16*  | 1.90 ±0.06* | 0.28 ±0.08 |
| EGC     | Not found   | Not found | Not found  | 3.50 ±0.25   | 0.26 ±0.05** | 2.79 ±0.26      | 5.36 ±0.45** | 1.57 ±0.05**|

*Differences from the control: *p < 0.05, **p < 0.01, ***p < 0.001  
EC – epicatechin; EGC – epigallocatechin; RKW – aqueous extract *R. kirilowii*; RKW-A – 50% hydro-alcoholic extract *R. kirilowii*
of (+)-catechin in RKW than in RKW-A group of sera, despite the fact that concentration of this polyphenol in RKW-A extract was almost twice as high than in RKW extract. Described results are presented in Table 1 and on Fig. 1A and Fig. 1B.

Serum analysis of epicatechin and epigallocatechin revealed the highest concentration of both polyphenols in EGC group \((p < 0.001, \text{Fig. 2})\). It has not been observed differences between control, RKW and RKW-A groups except epicatechin level which was significantly increased in RKW-A group.

**Discussion**

Flavonoids represent a wide group of more than 4000 secondary plant metabolites. Food-derived flavonols (quercetin and kaempferol) and flavanols (tannins and catechins) have been reported to exhibit various biological functions, including antioxidant, anti-inflammatory, antimicrobial, anticancer, cardioprotective, neuroprotective, antidiabetic, antiosteoporotic, estrogenic/antiestrogenic, anxiolytic, analgesic and anti-allergic activities [18].

Our studies revealed the presence of salidroside and kaempferol in the sera of mothers belonging to RKW and RKW-A groups, wherein concentration of salidroside was significantly higher in RKW-A mothers sera, than in sera of mothers from RKW group. These substances are also present in other plants of the *Rhodiola* genus, e.g. *Rhodiola sachalinensis* and *Rhodiola rosea* [19]. Sera examined in the present study contained catechins and phenolic acids also. However, comparing various polyphenols/flavonoids content in extracts and in mother mouse sera we did not
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**Fig. 3.** Proliferation of endothelial HECa10 cells in the presence of sera from: control mothers, mothers fed *R. kirilowii* aqueous extract (RKW), hydro-alcoholic extract (RKW-A), or epigallocatechin (EGC). Number of mice in parentheses

|                | Control sera (11) | RKW-A sera (11) | RKW sera (11) | EGC sera (7) |
|----------------|-------------------|-----------------|---------------|--------------|
| **Unpaired t test** |                   |                 |               |              |
| *p* value       | 0.0326            |                 |               |              |
| *p* value summary | *               |                 |               |              |
| Are means significantly different? (*p* < 0.05) | Yes |                 |               |              |
| One- or two-tailed *p* value? | Two-tailed |                 |               |              |

**Fig. 4.** Migration of endothelial HECa10 cells in the presence of sera from: control mothers, mothers fed *R. kirilowii* aqueous extract (RKW), hydro-alcoholic extract (RKW-A), or epigallocatechin (EGC). Number of mice in parentheses

|                | Control sera (11) | RKW-A sera (11) | RKW sera (11) | EGC sera (7) |
|----------------|-------------------|-----------------|---------------|--------------|
| **One-way analysis of variance** |                   |                 |               |              |
| *p* value       | 0.0573            |                 |               |              |
| *p* value summary | ns               |                 |               |              |
| Are means significantly different? (*p* < 0.05) | No |                 |               |              |
| Number of groups | 4                |                 |               |              |
| F              | 2.689             |                 |               |              |
| $r^2$          | 0.1492            |                 |               |              |

**Fig. 5.** VEGF concentration in the sera from: control mothers, mothers fed *R. kirilowii* aqueous extract (RKW), hydro-alcoholic extract (RKW-A), or epigallocatechin (EGC). Number of mice in parentheses

|                | Control (12) | RKW (9) | RKW-A (10) | EGC (8) |
|----------------|--------------|---------|------------|---------|
| **Unpaired t test** |              |         |            |         |
| *p* value       | 0.0051       |         |            |         |
| *p* value summary | **          |         |            |         |
| Are means significantly different? (*p* < 0.05) | Yes |         |            |         |
| One- or two-tailed *p* value? | Two-tailed |         |            |         |

**Fig. 6.** bFGF concentration in the sera from: control mothers, mothers fed *R. kirilowii* aqueous extract (RKW), hydro-alcoholic extract (RKW-A), or epigallocatechin (EGC). Number of mice in parentheses

|                | Control (12) | RKW-A (10) | RKW (9) | EGC (8) |
|----------------|--------------|------------|---------|---------|
| **Unpaired t test** |              |            |         |         |
| *p* value       | 0.0198       |            |         |         |
| *p* value summary | *           |            |         |         |
| Are means significantly different? (*p* < 0.05) | Yes |            |         |         |
| One- or two-tailed *p* value? | Two-tailed |            |         |         |
found equal proportions of those compounds. Some of them exist in proportional concentration (e.g. salidroside, kaempferol) and some in reverse proportions (e.g. ellagic acid or (+)-catechin). The probable mechanism may be connected with bioavailability (absorption from the gastrointestinal tract with possible competition for receptors or formation of conjugates, which will not be absorbed) or biodistribution (change of solubility/affinity by addition of glucoronid, sulfate or other groups; increased absorption in some tissues and organs or increased utilization).

In our research, sera of mice fed during pregnancy and lactation *Rhodiola kirilowii* extracts contained higher concentration of VEGF (RKW-A) and bFGF (both of them) than corresponding controls (Figs. 3-6). Despite of this, we observed antiproliferative activity of sera from mothers fed a hydro-alcoholic extract. This might be associated with a higher, than in mice fed a water extract, serum content of strongly anti-angiogenic substances (salidroside and catechins). Confirmation of anti-angiogenic properties of above polyphenols we obtained in our previous studies on embryonic and tumor angiogenesis in mice [10, 20, 21]. Also Sun et al. [22] showed anticancer effect of salidroside on colon cancer through inhibiting JAK2/STAT3 signaling pathway. In addition Wang et al. [23] demonstrated anticancer effect of salidroside on A549 lung cancer cells through inhibition of oxidative stress and phospho-p38 expression.

Analysed murine blood sera samples showed differences in concentrations of polyphenols, which resulted from supplementation of mice diet with various sources of these compounds. Differences between groups RKW and RKW-A were probably caused by using two different extraction methods to extract polyphenols from *Rhodiola kirilowii*. Previous reports have demonstrated that the differences in extraction efficiency are observed mainly in the case of (+)-catechin, p-coumaric acid and naringenin [1]. In the present study sera samples of mother mice from group RKW contained the highest concentration of (+)-catechin when compared with the other groups. Alcohol (RKW-A) extract with significantly higher (+)-catechin, epicatechin and epigallocatechin content did not cause a significant increase in the concentration of these compounds in the serum, except epicatechin. On the contrary water *Rhodiola kirilowii* extract significantly augmented the plasma level of catechins, except epigallocatechin. These observations are not easy to interpret. It is possible, that such effect could be observed due to the analytical method used in this study. Enzymatic hydrolysis conducted during samples preparation for chromatographic analyses aimed to change the glycosidic polyphenols to aglycone forms of these compounds, which could cause accumulation of catechins in the samples. It is also possible that the hydro-alcoholic *Rhodiola kirilowii* extract provides organism with more accessible polyphenol forms – aglycones, which are easier to absorb, thus the metabolisms and excretion may be faster [24]. To verify this hypothesis future experiments should include urine 24-hour volume test and examination of catechins concentration in such samples. Furthermore, in the present study epicatechin and epigallocatechin serum concentrations were the highest in mice receiving diet supplemented with epigallocatechin. High content of pure epigallocatechin in the diet influenced the levels of epigallocatechin and epicatechin (its derivative with lower molecular mass) detected in the serum. Metabolism of these catechins includes lowering of the molecular mass during distribution with blood and metabolism in the liver. Conjugation with glucuronic acid enables their fast excretion from the organism [25]. Although results obtained ambiguously explain the relationship between the type of dietary supplementation with polyphenolic compounds and their serum concentration, the effect of polyphenols on the organism of mother mice was definitely observed, and was significant with reference to investigated parameters.

Conclusions

These data allow to assume that *Rhodiola kirilowii* hydro-alcoholic extract (RKW-A) is potentially able to modulate pre- and post-natal angiogenesis what might influence the development of organs in progeny. Sera of RKW mothers have not harm the proliferation of endothelial cells, despite they also contain antiangiogenic catechins and salidroside. Sera collected from mothers supplemented with epigallocatechin also have not influenced endothelial cells proliferation, despite the highest content of epicatechin and epigallocatechin. This might suggest the existence in RKW-A extract and in RKW-A sera some other, yet unidentified substances influencing endothelial cells proliferation.

It remains to be considered the possibility also that salidroside in lower concentration stimulates the migration of endothelial cells, and in higher inhibits endothelial cell proliferation.

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The authors declare no conflict of interest.

References

1. Zdanowski R, Lewicki S, Sikorska K, et al. (2014): The influence of aqueous and hydro-alcoholic extracts of roots and rhizomes of Rhodiola kirilowii on the course of pregnancy in mice. Cent Eur J Immunol 39: 471-475.
2. Radomska-Leśniewska D, Skopiński P, Bałan BJ, et al. (2015): Angiomodulatory properties of Rhodiola spp. and other natural antioxidants. Cent Eur J Immunol 40: 249-262.
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3. Siwicki AK, Skopinska-Różewska E, Hartwich M, et al. (2007): The influence of Rhodiola rosea extracts on non-specific and specific cellular immunity in pigs, rats and mice. Cent Eur J Immunol 32: 84-91.
4. Wójcik R, Siwicki AK, Skopińska-Różewska E, et al. (2009): The effect of Chinese medicinal herb Rhodiola kirilowii extracts on cellular immunity in mice and rats. Pol J Vet Sci 12: 399-405.
5. Skopińska-Różewska E, Bychawska M, Bialas-Chromiec B, Sommer E (2009): The in vivo effect of Rhodiola rosea and Rhodiola quadrifida hydro-alcoholic extracts on chemokinetic activity of spleen lymphocytes in mice. Centr Eur J Immunol 34: 42-45.
6. Skopińska-Różewska E, Bychawska M, Bialas-Chromiec B, et al. (2010): The in vivo effect of Rhodiola kirilowii extracts on blood granulocytes metabolic activity in mice. Centr Eur J Immunol 35: 20-24.
7. Siwicki A, Skopińska-Różewska E, Wasiyutyński A, et al. (2012): The effect of Rhodiola kirilowii extracts on pigs blood leukocytes metabolic (RBA) and proliferative (LPS) activity, and on the bacterial infection and blood leukocytes number in mice. Centr Eur J Immunol 37: 145-150.
8. Zdanowski R, Lewicki S, Skopińska-Różewska E, et al. (2014): Alcohol- and water-based extracts obtained from Rhodiola rosea affect differently the number and metabolic activity of circulating granulocytes in Balb/c mice. Ann Agric Environ Med 21: 120-123.
9. Skopińska-Różewska E, Hartwich M, Siwicki AK, et al. (2008): The influence of Rhodiola rosea extracts and rosavin on cutaneous angiogenesis induced in mice after grafting of syngeneic tumor cells. Centr Eur J Immunol 33: 102-107.
10. Skopińska-Różewska E, Malinowski M, Wasiyutyński A, et al. (2008): The influence of Rhodiola quadrifida 50% hydro-alcoholic extract and salidroside on tumor-induced angiogenesis in mice. Pol J Vet Sci 11: 97-104.
11. Zdanowski R, Skopińska-Różewska E, Wasiyutyński A, et al. (2012): The effect of Rhodiola kirilowii extracts on tumor-induced angiogenesis in mice. Centr Eur J Immunol 37: 131-139.
12. Lewicki S, Stankiewicz W, Skopińska-Różewska E, et al. (2015): Spleen content of selected polyphenols, spleenocytes morphology and function in mice fed Rhodiola kirilowii extracts during pregnancy and lactation. Pol J Vet Sci 18: 847-855.
13. Shin JW, Seol IC, Son CG (2010): Interpretation of animal dose and human equivalent dose for drug development. J Korean Oriental Med 31: 1-7.
14. Bizouarne N, Denis V, Legrand A, et al. (1993): A SV-40 immortalized murine endothelial cell line from peripheral lymph node high endothelium expresses a new alpha-L-fucose binding protein. Biol Cell 79: 209-218.
15. Kieda C, Paprocka M, Krawczenko A, et al. (2002): New human microvascular endothelial cell lines with specific adhesion molecule phenotypes. Endothelium 9: 247-261.
16. Skopiński P, Zdanowski R, Grzela T, et al. (2012): The influence of sterilized and non-sterilized amniotic dressings on the proliferation of endothelial cells in vitro. Centr Eur J Immunol 37: 114-118.
17. Rokicki D, Zdanowski R, Lewicki S, et al. (2014): Inhibition of proliferation, migration and invasiveness of endothelial murine cells culture induced by resveratrol. Centr Eur J Immunol 39: 449-454.
18. Irehhiaiy R, Kumar A, Yadav A, et al. (2014): Metabolites in plants and its classification. World J Pharm Pharmac Sci 4: 287-305.
19. Zhou T, Zheng J, Zhou Y, et al. (2015): Chemical Components and Bioactivities of Rhodiola rosea. Int J Trad Nat Med 5: 23-51.
20. Skopiński P, Skopińska-Różewska E, Kamiński A, et al. (2004): Chocolate feeding of pregnant mice resulted in epigallocatechin-related embryonic angiogenesis suppression and bone mineralization disorder. Pol J Vet Sci 7: 131-133.
21. Wasiyutyński A, Siwicki AK, Balan BJ, et al. (2005): Inhibitory effect of cocoa catechins on embryonic and tumor angiogenesis in mice. Pol J Environm Studies 14: 800-805.
22. Sun KK, Xia H, Xia RL (2015): Anticancer effect of salidroside on colon cancer through inhibiting JAK2/STAT3 signaling pathway. Int J Clin Exp Pathol 8: 615-621.
23. Wang J, Li JZ, Lu AX, et al. (2014): Anticancer effect of salidroside on A549 lung cancer cells through inhibition of oxidative stress and phospho-p38 expression. Oncol Lett 7: 1159-1164.
24. Liang J, Xu F, Zhang YZ, et al. (2014): The profiling and identification of the metabolites of (+)-catechin and study on their distribution in rats by HPLC-DAD-ESI-IT-TOF-MS(n) technique, Biomed Chromatogr 28: 401-411.
25. Blount JW, Redan BW, Ferruzzi MG, et al. (2015): Synthesis and quantitative analysis of plasma-targeted metabolites of catechin and epicatechin, J Agric Food Chem 63: 2233-2240.