

# Berries Reduce Postprandial Insulin Responses to Wheat and Rye Breads in Healthy Women<sup>1–4</sup>

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## Abstract

Starch in white wheat bread (WB) induces high postprandial glucose and insulin responses. For rye bread (RB), the glucose response is similar, whereas the insulin response is lower. In vitro studies suggest that polyphenol-rich berries may reduce digestion and absorption of starch and thereby suppress postprandial glycemia, but the evidence in humans is limited. We investigated the effects of berries consumed with WB or RB on postprandial glucose and insulin responses. Healthy females ( $n = 13–20$ ) participated in 3 randomized, controlled, crossover, 2-h meal studies. They consumed WB or RB, both equal to 50 g available starch, with 150 g whole-berry purée or the same amount of bread without berries as reference. In study 1, WB was served with strawberries, bilberries, or lingonberries and in study 2 with raspberries, cloudbberries, or chokeberries. In study 3, WB or RB was served with a mixture of berries consisting of equal amounts of strawberries, bilberries, cranberries, and blackcurrants. Strawberries, bilberries, lingonberries, and chokeberries consumed with WB and the berry mixture consumed with WB or RB significantly reduced the postprandial insulin response. Only strawberries (36%) and the berry mixture (with WB, 38%; with RB, 19%) significantly improved the glycemic profile of the breads. These results suggest that when WB is consumed with berries, less insulin is needed for maintenance of normal or slightly improved postprandial glucose metabolism. The lower insulin response to RB compared with WB can also be further reduced by berries. *J. Nutr.* 143: 430–436, 2013.

## Introduction

Starch is one of the main digestible carbohydrates in the diet contributing to blood glucose concentration. Bread is an important contributor to daily starch intake in Western diets. White wheat bread (WB) and rye bread (RB) induce high postprandial glucose responses (1), whereas the insulin response is significantly lower for RB compared with WB (2–6). Repeated post-meal hyperglycemia and hyperinsulinemia may promote insulin resistance,  $\beta$ -cell dysfunction, oxidative stress, inflammation, dyslipidemia, endothelial dysfunction, and repeated feelings of hunger and thus excessive food intake, increasing the risk of obesity, type 2 diabetes, and cardiovascular diseases (7,8).

In vitro and animal studies have demonstrated that polyphenols and polyphenol-rich extracts of foods inhibit digestion and absorption of carbohydrates and suppress postprandial glycemia (9). Polyphenols are a large group of bioactive phytochemicals in plant-based foods and beverages and are abundant in berry fruits (10–14). Polyphenol-rich extracts of several berries have been shown to inhibit  $\alpha$ -amylase and  $\alpha$ -glucosidase activities (15–23) and intestinal uptake and transport of glucose via SGLT1 and GLUT2 in vitro (24). Salivary and pancreatic  $\alpha$ -amylase and intestinal  $\alpha$ -glucosidase (maltase) are the key enzymes in the hydrolysis of starch to glucose. Inhibition of these enzymes has been used as a therapeutic target in the management of hyperglycemia in type 2 diabetic patients (25).

Our previous studies have shown that whole berries and their juice fractions optimize postprandial glucose and insulin responses to sucrose and improve the glycemic profile in healthy participants (26–28). However, the evidence of the effects of berries on postprandial metabolic responses to starch in humans is limited to a small study reporting no effect on glucose response (29). Insulin was not measured in that study.

Therefore, we aimed to investigate the effects of bread ingested with berries on postprandial glucose and insulin responses in healthy females. A series of 3 meal studies were carried out: 1) white WB served with strawberries, bilberries, or lingonberries, i.e., berries that are commonly consumed in the Nordic countries;

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<sup>3</sup> This trial was registered at [clinicaltrials.gov](http://clinicaltrials.gov) as NCT01580150.

<sup>4</sup> Supplemental Figure 1 and Supplemental Table 1 are available from the "Online Supporting Material" link in the online posting of the article and from the same link in the online table of contents at <http://jn.nutrition.org>.

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- 2) WB served with raspberries, cloudberrries, or chokeberries, which were chosen to represent different polyphenol profiles; and  
3) WB or RB served with a berry mixture that previously showed beneficial effects on postprandial responses to sucrose (26,27).

## Participants and Methods

### Participants

For studies 1–3, 13–20 volunteers were recruited by an announcement in a local newspaper in the area of Kuopio, Finland. The inclusion criteria were as follows: healthy women aged 25–69 y, BMI 20–28 kg/m<sup>2</sup>, no diabetes or other chronic disease, no smoking, no antibiotic medication within the past 3 mo, and no blood donation within the past month. At the screening visit, the participants' health status and medical history were assessed by using a structured interview and anthropometric and laboratory measurements (routine hematologic measurements, fasting plasma glucose and lipids, serum thyroid-stimulating hormone, alanine aminotransferase, and creatinine).

This study was conducted according to the guidelines of the Declaration of Helsinki and the study protocol was approved by the Research Ethics Committee of the Hospital District of Northern Savo (Finland). Written informed consent was obtained from all participants.

### Test meals

Studies 1–3 included meals with WB or RB served with berries (150 g, as whole berry purées) and tap water (200 mL). The reference meals consisted of breads served with cucumber (50 g) instead of berries and water (300 mL). The breads were served without crust and each portion provided 50 g available starch. In study 1, WB was served with strawberries, bilberries, or lingonberries, and in study 2 with raspberries, cloudberrries, or chokeberries. In study 3, WB and RB were served with a mixture of 4 berries, i.e., strawberries, bilberries, cranberries, and blackcurrants. The nutrient compositions of the bread and berry portions are given in **Table 1**. The participants were instructed to finish the meals within 10 min and the eating times were recorded.

**Breads.** WB was prepared at VTT pilot bakery. The ingredients were white wheat flour, water, sugar, salt, yeast, and baking margarine. RB was prepared at the pilot bakery of Vaasan. The ingredients were whole-meal rye flour, whole-meal sourdough, water, yeast, and salt. The content of digestible starch in the breads (without crust) was analyzed using the Megazyme total starch assay procedure (30). The bread loaves were frozen and stored at –20°C. Before each study visit, they were thawed overnight in a refrigerator. Before serving to the participants, the breads were sliced after removing the crust.

**Berries.** Frozen strawberries (*Fragaria × ananassa*), bilberries (wild European blueberries, *Vaccinium myrtillus*), lingonberries (*Vaccinium*

*vitis-idaea*), red raspberries (*Rubus idaeus*), cloudberrries (*Rubus chamaemorus*), chokeberries (*Aronia mitschurinii*), cranberries (*Vaccinium oxycoccos*), and blackcurrants (*Ribes nigrum*) of Finnish origin were provided by Aten Marja-Aitta (Puumala, Finland), Joswola Oy (Iisalmi, Finland), and Kiantama Oy (Suomussalmi, Finland). The berry mixture used in study 3 consisted of 37.5 g of each of strawberries, bilberries, cranberries, and blackcurrants. The berries were partially thawed at room temperature, mashed with a household food processor, and the purées were frozen at –20°C in 150-g portions. In the morning of the study visits, these portions were thawed in a microwave oven.

The meals with berries provided a higher amount of available carbohydrate than the reference meals due to the natural sugars (glucose and fructose) present in berries. As calculated according to the Finnish food composition database (31), the berry portions provided 2.4–6.0 g of extra glucose.

### Study design

Three randomized, controlled, crossover studies were carried out single-blinded for the laboratory personnel and analyses. In each study, the volunteers participated in four 2-h meal tests on separate visits, at least 3 d apart. The test meals were served in a randomized order.

The participants were instructed to keep their diet, body weight, and lifestyle constant during the study, abstain from alcohol for 2 d before the test, and refrain from intensive physical activity for 12 h before the test. The day before the study visit, they were advised to not consume any berries or berry products. In the evening before the visit, they were instructed to consume a meal of choice and repeat a similar meal before every visit. Body weight was recorded at every visit.

The tests were carried out in the morning after a 10- to 12-h overnight fast. For collection of venous blood samples, an i.v. catheter (BD Venflon Pro 18GA, 1.3 × 32 mm, Beckton Dickinson) was inserted into an antecubital vein of the forearm. Baseline fasting blood samples were drawn before the test meal (0 min) and 6 samples at 15, 30, 45, 60, 90, and 120 min after starting to eat the meal. The samples for plasma glucose measurements were collected in citrate-fluoride tubes and for plasma insulin measurements in EDTA tubes kept on ice. Plasma was immediately separated by centrifugation at 4°C and stored at –70°C until analyzed.

### Laboratory analyses

Plasma glucose concentrations were analyzed with the hexokinase method using Konelab System reagents and a Konelab 20 XTi clinical chemistry analyzer (Thermo Fisher Scientific). The intra- and inter-assay CVs were 2.7 and 1.8%, respectively. Plasma insulin was determined with an immunoluminometric assay using ADVIA Centaur Insulin IRI reagents and a Siemens ADVIA Centaur chemiluminescence immunoanalyzer (Siemens Medical Solutions Diagnostics). The intra- and inter-assay CVs were 2.7 and 5.1%, respectively.

### Calculations and statistical analyses

The data were analyzed by using the SPSS 17.0 for Windows statistical software and are expressed as mean ± SD or ± SEM, as indicated. Linear mixed-effects modeling was used to compare the effects of the test meals. Histograms were used for checking normality of model residuals, and logarithmic transformation was used for non-normally distributed data. Values of  $P \leq 0.05$  were considered significant.

The significance of the overall meal × time interaction was tested in the mixed-model analysis by using participant as a random factor and meal × time and their main effects as fixed factors. When the overall meal × time interaction was significant, the differences between the reference and each berry meal at individual time points were tested by post hoc analysis with Bonferroni adjustment for multiple comparisons.

The maximum increases from the baseline concentrations were calculated by subtracting the fasting value from the highest value. The incremental AUCs were calculated by using GraphPad Prism 5 for Windows, ignoring the area below the baseline (0 min) concentration. This software was also used to calculate the glycemic profile for each participant and test meal according to Rosén et al. (5), by dividing the time (min) during which the plasma glucose was above the fasting concentration with the incremental peak glucose value (mmol/L). The differences

**TABLE 1** Nutrient composition of the bread and berry portions<sup>1</sup>

	WB	RB	Berries <sup>2</sup>	Cucumber <sup>2</sup>
			<i>g</i>	
Portion size	117, <sup>3</sup> 126 <sup>4</sup>	163	150	50
Available carbohydrate				
Starch	50	50		
Sugars <sup>5</sup>			6.2–12.6	0.7
Protein	10	10	0.6–2.1	0.3
Fat	3	2	0.3–1.2	0.05
Total fiber	5	19	2.8–9.4	0.35

<sup>1</sup> RB, rye bread; WB, wheat bread.

<sup>2</sup> Data from (31) (does not provide data for chokeberry).

<sup>3</sup> Studies 1 and 2.

<sup>4</sup> Study 3.

<sup>5</sup> Glucose and fructose.

between the test meals were analyzed using participant as a random factor and meal as a fixed factor in the mixed-model analysis.

## Results

The basic characteristics of the female volunteers enrolled in the 3 studies are presented in **Table 2**. The body weights remained stable throughout the study periods. In study 1, the times spent eating the meals were [mean  $\pm$  SD (min:s)]: WB, 8:27  $\pm$  0:55; WB+strawberry, 8:08  $\pm$  1:21; WB+bilberry, 8:25  $\pm$  0:43; and WB+lingonberry, 8:05  $\pm$  0:57. In study 2, the eating times were (min:s): WB, 7:13  $\pm$  1:23; WB+raspberry, 6:55  $\pm$  1:11; WB+cloudberry, 7:08  $\pm$  1:21; and WB+chokeberry, 8:39  $\pm$  1.28 min:s, which significantly differed from WB. In study 3, the WB meal was eaten in (min:s) 7:42  $\pm$  1:24 and WB+berries in 7:18  $\pm$  1:21 and the RB meal in 8:15  $\pm$  1:12 and RB+berries in 8:19  $\pm$  1:38.

### WB

**Glucose.** Compared with WB alone, none of the berries ingested with WB had a significant effect on the meal  $\times$  time interaction (**Supplemental Fig. 1A–C**) or the AUC 0–120 min or AUC 0–60 min (**Supplemental Table 1**) of the postprandial glucose responses. The berry mixture (study 3) decreased the AUC 0–30 min by 32% ( $P < 0.001$ ). In addition, the AUC 0–30 min was decreased by cloudberry (23%, study 2), but due to multiple comparisons, this effect was not significant in the post hoc analysis ( $P = 0.16$ ). Strawberries (study 1) and the berry mixture increased the value of the glycemic profile by 36% ( $P < 0.05$ ) and 38% ( $P = 0.005$ ), respectively (**Supplemental Table 1**).

**Insulin.** The meal  $\times$  time interaction for insulin responses was significant ( $P < 0.03$ ) (**Fig. 1A–C**). In study 1, strawberries attenuated the maximum increase of insulin from the baseline by 26% ( $P < 0.05$ ) and reduced the AUC 0–60 min by 24% ( $P < 0.01$ ) (**Table 3**). Bilberries and lingonberries reduced the AUC 0–60 min by 19 and 20% ( $P < 0.05$ ) and AUC 0–30 min by 33 and 32% ( $P < 0.01$ ), respectively. In study 2, chokeberries reduced the AUC 0–60 min by 24% ( $P < 0.05$ ) and AUC 0–30 min by 33% ( $P < 0.001$ ), whereas raspberries and cloudberry had no significant effect on these variables. In study 3, the berry mixture ingested with WB resulted in lower insulin at 15 and 30 min and higher at 120 min (**Fig. 1C**), attenuated the maximum increase from the baseline by 22% ( $P = 0.001$ ), and reduced the AUC 0–120 min by 12% ( $P = 0.041$ ), AUC 0–60 min by 25% ( $P < 0.001$ ), and AUC 0–30 min by 39% ( $P < 0.001$ ) (**Table 3**).

### RB

**Glucose.** In comparison with RB alone, the berry mixture ingested with RB in study 3 had no significant effect on the meal  $\times$  time interaction (**Supplemental Fig. 1D**) or the AUC 0–120 or 0–60 min (**Supplemental Table 1**). However, the berry mixture decreased the AUC 0–30 min by 27% ( $P = 0.026$ ) and increased the value of the glycemic profile by 19% ( $P = 0.05$ ) (**Supplemental Table 1**).

**Insulin.** The meal  $\times$  time interaction for insulin responses was significant ( $P < 0.001$ ) (**Fig. 1D**). In comparison with RB alone, ingestion of the berry mixture with RB resulted in lower insulin at 15 and 30 min and higher at 120 min. It reduced the maximum increase from the baseline by 23% ( $P = 0.001$ ), AUC 0–120 min by 14% ( $P = 0.030$ ), AUC 0–60 min by 25% ( $P < 0.001$ ), and AUC 0–30 min by 40% ( $P < 0.001$ ) (**Table 3**).

## Discussion

In this series of 3 acute clinical studies, healthy female volunteers consumed WB with 6 different berries, or WB or RB with a mixture of 4 berries. The main finding of these studies is the attenuation of the postprandial insulin response to bread by the berries, with little or no effect on the glucose response. The second important finding of our study is that the low insulin response after RB could be further reduced by supplementing the RB meal with berries.

In study 1, strawberries had a pronounced effect on insulin response to WB. The polyphenol composition of strawberries is characterized by ellagitannins (11,32). Therefore, we chose to include 2 other ellagitannin-rich berries, i.e., raspberries and cloudberry, in study 2. Although these berries have a higher content of ellagitannins than strawberries (11,32), they had no clear effect on the insulin response, indicating that the total content of ellagitannins is not a determining factor in lowering the insulin response. Our finding on the effect of strawberries on acute insulin metabolism is supported by a previous study where a strawberry beverage attenuated the postprandial insulin response but not glucose response to a high-carbohydrate meal containing starch from bagel (33).

A reduced insulin response was observed when WB was consumed with bilberries and lingonberries with anthocyanins and proanthocyanidins as the main polyphenols. Anthocyanins are the dominating polyphenols in bilberries and proanthocyanidins in lingonberries (11,12). Chokeberries are characterized by a very high content of anthocyanins and an extremely high content of proanthocyanidins (11,12). However, the insulin-lowering potency of chokeberries was surprisingly similar to

**TABLE 2** Basic characteristics of the female participants<sup>1</sup>

	Study 1	Study 2	Study 3
<i>n</i>	15	13	20
Age, <i>y</i>	48 $\pm$ 14 (21–64)	50 $\pm$ 12 (23–64)	57 $\pm$ 12 (25–69)
Weight, <i>kg</i>	65.5 $\pm$ 9.0 (52–89)	64.6 $\pm$ 11.3 (51–94)	64.7 $\pm$ 6.1 (54–78)
BMI, <i>kg/m</i> <sup>2</sup>	24.4 $\pm$ 2.7 (20–30)	24.2 $\pm$ 3.2 (20–32)	24.2 $\pm$ 2.0 (21–28)
Plasma glucose, <i>mmol/L</i>	5.5 $\pm$ 0.5 (4.4–6.4)	5.4 $\pm$ 0.6 (4.5–6.1)	5.4 $\pm$ 0.3 (4.9–6.2)
Serum total cholesterol, <i>mmol/L</i>	4.8 $\pm$ 0.9 (3.4–6.3)	4.6 $\pm$ 0.6 (3.6–5.7)	5.3 $\pm$ 1.2 (2.9–7.3)
Serum LDL cholesterol, <i>mmol/L</i>	2.9 $\pm$ 0.8 (1.6–4.4)	2.4 $\pm$ 0.6 (1.6–3.4)	2.8 $\pm$ 1.0 (1.3–4.5)
Serum HDL cholesterol, <i>mmol/L</i>	1.5 $\pm$ 0.4 (1.0–2.2)	1.7 $\pm$ 0.5 (1.0–2.6)	1.8 $\pm$ 0.4 (1.0–2.5)
Serum TGs, <i>mmol/L</i>	1.1 $\pm$ 0.4 (0.5–2.1)	0.9 $\pm$ 0.6 (0.4–2.6)	1.0 $\pm$ 0.4 (0.6–2.2)

<sup>1</sup> Values are mean  $\pm$  SD (range).

**TABLE 3** Insulin variables in healthy women after the WB and RB meals with or without 150 g berries<sup>1</sup>

Study	Treatment				P <sup>2</sup>
	WB	WB + strawberry	WB + bilberry	WB + lingonberry	
Study 1 (n = 15)					
Fasting, <sup>3</sup> mU/L	6.5 ± 1.5	6.5 ± 2.2	7.1 ± 2.8	6.1 ± 3.0	NC
Maximum increase, mU/L	68.8 ± 21.8	50.7 ± 18.4*	60.5 ± 24.9	60.1 ± 18.2	0.035
AUC 0–120 min, <sup>4</sup> mU × min/L	4140 ± 1170	3340 ± 1050	3800 ± 1540	3820 ± 1100	0.06
AUC 0–60 min, mU × min/L	2300 ± 710	1740 ± 640**	1860 ± 800*	1850 ± 500*	0.005
AUC 0–30 min, mU × min/L	620 ± 270	470 ± 210	420 ± 220**	420 ± 210**	0.005
Study 2 (n = 13)					
Fasting, mU/L	6.5 ± 2.5	6.2 ± 2.4	5.6 ± 1.9	6.1 ± 2.6	NC
Maximum increase, mU/L	63.7 ± 33.9	62.9 ± 25.5	67.3 ± 29.0	53.2 ± 26.2	0.049
AUC 0–120 min, mU × min/L	3920 ± 2050	4010 ± 1660	4300 ± 1860	3590 ± 1600	0.08
AUC 0–60 min, mU × min/L	2240 ± 1160	2080 ± 860	2220 ± 1120	1710 ± 860*	0.009
AUC 0–30 min, mU × min/L	630 ± 360	540 ± 250	520 ± 310	420 ± 290***	0.003
Study 3 (n = 20)					
Fasting, mU/L	7.0 ± 3.4	6.0 ± 1.8			NC
Maximum increase, mU/L	68.0 ± 31.5	52.8 ± 24.9			0.001
AUC 0–120 min, mU × min/L	4140 ± 1830	3660 ± 1760			0.041
AUC 0–60 min, mU × min/L	2320 ± 920	1740 ± 840			<0.001
AUC 0–30 min, mU × min/L	640 ± 380	390 ± 230			<0.001
Study 3 (n = 20)					
Fasting, mU/L	5.5 ± 1.9	6.2 ± 2.3			0.06
Maximum increase, mU/L	47.6 ± 25.0	36.4 ± 15.6			0.001
AUC 0–120 min, mU × min/L	2970 ± 1320	2560 ± 1100			0.030
AUC 0–60 min, mU × min/L	1640 ± 740	1220 ± 560			<0.001
AUC 0–30 min, mU × min/L	490 ± 250	300 ± 150			<0.001

<sup>1</sup> Values are mean ± SD. Different from reference (WB or RB): \*P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001 (post hoc analysis with Bonferroni adjustment). NC, no change; RB, rye bread; WB, wheat bread.

<sup>2</sup> Mixed-model analysis for the comparison of the bread and bread+berry meals (after logarithmic transformation).

<sup>3</sup> 1 mU/L = 6.945 pmol/L.

<sup>4</sup> 1 mU × min/L = 6.945 pmol × min/L.

that of bilberries and lingonberries (Table 4) and the much higher content of these polyphenols in chokeberries did not enhance the effect. The comparison of the insulin-lowering profiles presented in Table 4 also shows differences between these 3 berries and strawberries.

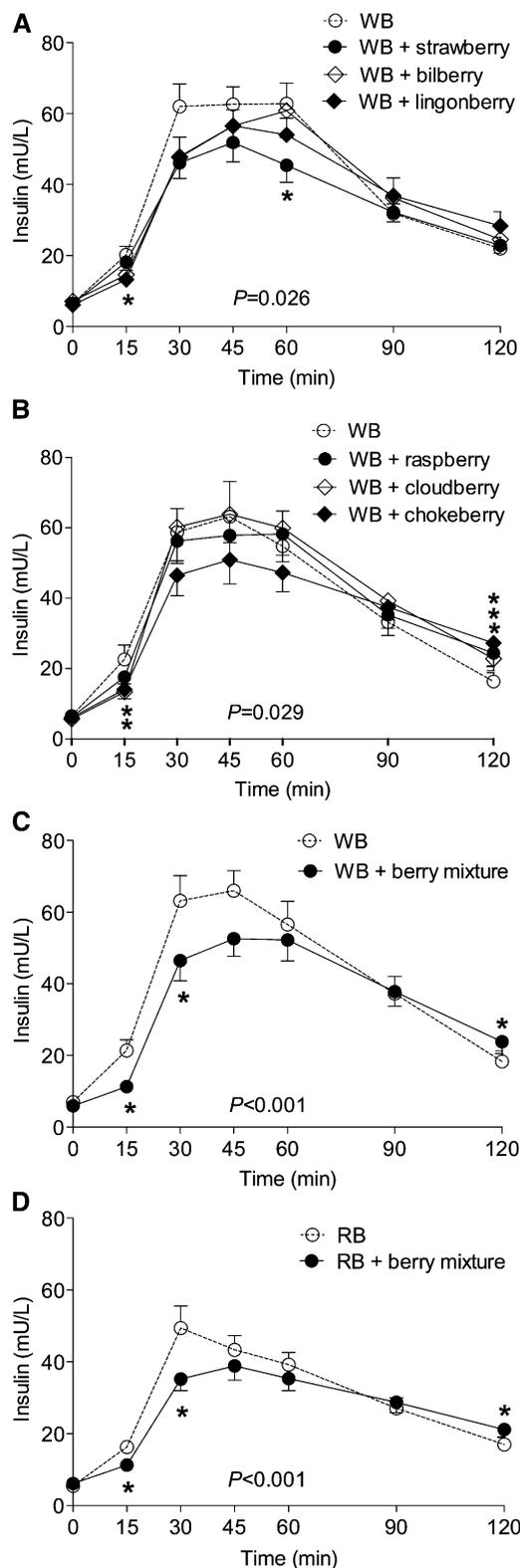
In study 3, a mixture of strawberries, bilberries, cranberries, and blackcurrants was consumed with 2 breads known to have different insulin responses. The extent of the insulin-lowering effect of berries was practically similar on WB and RB (Table 4), in spite of clear differences in their structure (34). Starch in RB is more swollen with amylose leaching out from the granules, which is not the case in WB. RB has a less porous and harder crumb than WB. These structural features are thought to contribute to the lower insulin response of RB compared with WB, possibly through altered secretion of incretin hormones (3,35). It is interesting that berries similarly decreased the insulin responses to both types of bread.

Despite the pronounced impact on insulin response, the effects of the berries on postprandial glycemia were modest. Only the very early glucose response (AUC 0–30 min) was affected by the berry mixture consumed with WB or RB. The glycemic profile was slightly improved by strawberries consumed with WB and the berry mixture consumed with WB or RB. The glycemic profile is a variable proposed by Rosén et al. (5) for quantitative illustration of the shape of the glucose curve. It is defined as the duration for the incremental postprandial blood glucose response divided with the blood glucose incremental peak. A high value of glycemic profile indicates facilitated postprandial glycemic regulation.

Our results on the effects of berries on postprandial glycemia are in agreement with a previous study reporting that addition of

raspberries and blueberries to starch-based food (pancake) did not alter the postprandial glucose response (29). Insulin was not measured in that study. The findings of these 2 in vivo studies appear to be inconsistent with in vitro data. Strawberries (15,17,22), blueberries (15,22,23), lingonberries (22), raspberries (15,16,22), cloudberry (22), and blackcurrants (15,20,22) have been reported to inhibit pancreatic  $\alpha$ -amylase in vitro, to a varying extent. Strawberries (15,17,18), blueberries (15,23), raspberries (15,16,21), and blackcurrants (15,20) have also been shown to inhibit intestinal  $\alpha$ -glucosidase in vitro. Strawberries (17,18), raspberries (21), and blueberries (23) appear to be more potent inhibitors of  $\alpha$ -glucosidase than  $\alpha$ -amylase. Strawberry polyphenols have also decreased glucose absorption in vitro (24). Although these in vitro findings suggest that the enzymatic digestion of starch and glucose absorption should have been retarded and postmeal glycemia suppressed by the berries included in our study, this was not evident in our results. Interestingly, Eelderink et al. (36) recently proposed that the glycemic response does not always reflect the in vivo starch digestibility. They found that starch meals with different rates of appearance of exogenous glucose into the circulation gave similar glycemic responses.

In our previous studies, lingonberries (28) and the same berry mixture used herein (27) optimized both glucose and insulin responses to sucrose, whereas in the case of starch (the present study), the effects were more pronounced on the insulin than the glucose response. Taken together, the effects of berries on postprandial glycemia are somewhat different for sucrose and starch. Perhaps this could be explained by efficient inhibition  $\alpha$ -glucosidase (sucrase activity) in the case of sucrose and poor inhibition of  $\alpha$ -amylase in the case of starch, as discussed above.



**FIGURE 1** Plasma insulin concentrations in healthy women after ingestion of WB or RB with or without 150 g berries. Values are mean  $\pm$  SEM,  $n = 15$  (A; study 1), 13 (B; study 2), or 20 (C,D; study 3). The breads contained 50 g available starch.  $P$  for the overall meal  $\times$  time interaction in the mixed-model analysis (after logarithmic transformation) is indicated in each graph. \*Different from reference (WB or RB),  $P < 0.05$ . Conversion factor for insulin concentration: 1 mU/L = 6.945 pmol/L. RB, rye bread; WB, wheat bread.

**TABLE 4** Relative reductions of the insulin variables in healthy women after the WB and RB meals with berries compared with either bread without berries<sup>1</sup>

	Maximum increase	AUC 0–120 min	AUC 0–60 min	AUC 0–30 min	Study
		%			
WB + strawberry vs. WB	-26	(-19)	-24	(-24)	1
WB + bilberry vs. WB	(-12)	(-8)	-19	-33	1
WB + lingonberry vs. WB	(-13)	(-8)	-20	-32	1
WB + chokeberry vs. WB	(-16)	(-8)	-24	-33	2
WB + berry mixture vs. WB	-22	-12	-25	-39	3
RB + berry mixture vs. RB	-23	-14	-25	-40	3

<sup>1</sup> The percent changes are calculated from the mean values presented in Table 3. Nonsignificant changes (Table 3) are given in parentheses. RB, rye bread; WB, wheat bread.

Our study found no apparent association of the insulin-lowering potency and the polyphenol composition of the berries, and the contribution of different polyphenolic compounds remains to be examined. The postprandial glucose and insulin responses may be reduced by dietary fiber, especially soluble fiber (37). The berry meals provided 2.8–9.4 g total fiber. The fiber content was highest in raspberries and cloudberrries (31) and these berries had no effect on the glucose and insulin responses. The rest of the berries and the berry mixture provided 2.8–5.0 g and 5.4 g total fiber, respectively. Approximately 80% of the total fiber in these berries is insoluble (31) and it is unlikely that the relatively small amount of soluble fiber could solely explain the results. Another factor that should also be considered is the role of acidity. Berries contain citric acid and other organic acids and their pH is  $\sim 3$  (38). Previously, addition of vinegar (acetic acid) to white WB meal reduced glucose and insulin responses during the first 30 min, with no effect on AUC 0–120 min (39). In addition, similar effects have been observed for sourdough bread, which contains lactic acid and has a low pH (40). The 0–30 min and 0–60 min glucose and insulin responses were lower after sourdough bread than after the reference bread. Several mechanisms for the effects of organic acids on glycemic responses have been proposed (39,40). In future studies, the role of organic acids of berries in the modulation of postprandial glucose and insulin responses should be addressed.

The lower insulin response after ingestion of berries implies that less insulin is required and secreted for maintenance of normal or slightly improved postprandial glucose metabolism. It resembles the phenomenon repeatedly demonstrated for RB compared with WB, the mechanism of which still remains unknown (2–6). In the present study, the insulin-lowering effect of RB was enhanced by berries (Table 4). A lower postprandial insulin requirement may be beneficial in the short term and long term. In the short term, a lower insulin response prevents hypoglycemia and inappropriate increases of FFAs and stress hormone concentrations (3,4,35), which are often seen during the late postprandial period after consumption of refined carbohydrates (7). Regular consumption of diets with a low postprandial insulin response may increase first-phase insulin secretion, indicating improved pancreatic  $\beta$ -cell function, and modulate the inflammatory status (4,41,42). Such diets were also shown to down-regulate genes linked to insulin signaling and apoptosis, whereas diets with high postprandial insulin response upregulated genes related to metabolic stress (43). Thus, regular consumption of foods with a lower postprandial insulin requirement, such as RB

and berries, may help in the prevention of type 2 diabetes and can be recommended especially for individuals at high risk of this disease.

We conclude that when white WB is consumed with berries, less insulin is required for maintenance of normal or slightly improved postprandial glucose metabolism. The lower insulin response to RB compared with WB can also be further reduced by consumption of berries. Berries do not suppress the postmeal glucose peak but may slightly optimize the course of the glycemia induced by bread.

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