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1 **Impulsiveness, postprandial blood glucose and glucoregulation affect**  
2 **measures of behavioral flexibility**

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29 **Keywords:** Glucose; Glucose regulation; glycaemia; impulsivity; behavioral flexibility

## 32 **Abbreviations**

33 ACC= anterior cingulate cortex; BCST= Berg's Card sorting task; BF= behavioural flexibility;

34 BIS-11= Barratt Impulsiveness Scale; BMI= body mass index; CPT= Continuous Performance

35 Task; FBG= fasting blood glucose; GI= glycemic index; IGT= glucose tolerance test; PBG=

36 postprandial blood glucose; RT= reaction time; VIF= variance inflation factor; WCST=

37 Wisconsin Card Sorting Task

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48 **Abstract**

49 Behavioral flexibility (BF) performance is influenced by both psychological and physiological  
50 factors. Recent evidence suggests that impulsivity and blood glucose can affect executive  
51 function, of which BF is a subdomain. Here, we hypothesized that impulsivity, fasting blood  
52 glucose (FBG), glucose changes (i.e. glucoregulation) from postprandial blood glucose (PBG)  
53 following the intake of a 15g glucose beverage could account for variability in BF performance.  
54 The Stroop Color-Word Test and the Wisconsin Card Sorting Test (WCST) were used as  
55 measures of BF, and the Barratt Impulsiveness Scale (BIS-11) to quantify participants'  
56 impulsivity. In Study 1, neither impulsivity nor FBG could predict performance on the Stroop or  
57 the WCST. In Study 2, we tested whether blood glucose levels following the intake of a sugary  
58 drink, and absolute changes in glucose levels following the intake of the glucose beverage could  
59 better predict BF. Results showed that impulsivity and the difference in blood glucose between  
60 time 1 (postprandial) and time 2, but not blood glucose levels at time 2 per se could account for  
61 variation in performance on the WCST but not on the Stroop task. More specifically, lower  
62 impulsivity scores on the BIS-11, and smaller differences in blood glucose levels from time 1 to  
63 time 2 predicted a decrease in the number of total and perseverative errors on the WCST. Our  
64 results show that measures of impulsivity and glucoregulation can be used to predict BF.  
65 Importantly our data extend the work on glucose and cognition to a clinically relevant domain of  
66 cognition.

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## 70        **1. Introduction**

71        Behavioral flexibility (BF) refers to the ability to adaptively modify behaviors when changes in  
72        environmental demands occur, and is one of the core processes of executive function. BF is  
73        made up of several distinct processing mechanisms including the extinguishing of a response,  
74        inhibition, reversal learning, set-shifting and has been associated with creative ability [1, 2]. Two  
75        commonly used tests of BF include the Stroop Color-Word Test (measuring cognitive inhibition)  
76        [3-5] and the Wisconsin Card Sorting task (measuring set-shifting) [1, 6].

77        Impairments in tasks measuring BF have been reported in the clinical domain, for example in  
78        schizophrenics [1], OCD patients [7], stimulant addicts [8], frontal lobe patients [9], and in those  
79        suffering from Williams syndrome [10]. Importantly, many of these individuals have reportedly  
80        high levels of the personality trait impulsiveness [11]. One core feature of impulsive-related  
81        behavior is a deficiency in reversal learning and response inhibition, two specific subdomains of  
82        BF [12, 13].

83        Alongside neuropsychological tools, there have been several attempts to capture impulsivity  
84        using self-report scales. Arguably one of the most commonly adopted and cited scale of  
85        impulsiveness is the Barratt Impulsiveness Scale (BIS-11) [14]. Higher scores on the BIS-11  
86        have been found to be predictive of poorer performance on tests of executive function/BF [15-  
87        18]. Furthermore, causal links have been found between impulsiveness, and biological markers  
88        (e.g. neurotransmitters; [19]), including the brain's primary fuel glucose.

89        For example, increasing the level of blood glucose by supplementation can reduce impulsive-  
90        related choice behavior [20-22]. Moreover, hypoglycemia (i.e. low blood glucose) has also been  
91        linked to impulsive related acts such as criminal behavior, sexual promiscuity, behaving

92 recklessly, and the likelihood of initiating and terminating alcohol and nicotine use [23-26].

93 Glucose supplementation has also been used to improve cognitive performance, primarily in the  
94 areas of memory and attention [27-33], but more recently, also in tasks assessing executive  
95 function and BF (indexed by performance on the Stroop) [34]. While glucose supplementation can  
96 improve cognitive performance, unusually low or high fasting blood glucose levels, as observed  
97 in patients suffering from diabetes (type 1 and 2) can have detrimental effects on various aspects  
98 related to executive function, memory, verbal reasoning, attention/vigilance and dual-tasking [35-  
99 46].

100 More recently, postprandial glucose levels (plasma glucose concentrations two hours after eating  
101 [[47]) have also been investigated as possible determinants of cognitive performance. There is  
102 good reason for this, as fasting and postprandial blood glucose concentrations are mediated by  
103 independent physiological mechanisms [48]. Thus far, some of these studies have found that a  
104 low but sustained increase in blood glucose concentrations in the postprandial period is most  
105 beneficial to enhance cognition, achieved by the provision of low GI (glycemic index) meals [49,  
106 50]. Additionally, it is also clear that the ability to utilize glucose (i.e. glucoregulation) is a  
107 contributing factor to cognitive functioning. Studies have shown that when examining changes in  
108 blood glucose from the start of cognitive testing until the end, those individuals who displayed  
109 decreased glucose levels performed cognitively better than individuals whose blood glucose  
110 levels stayed at similar levels or even increased [51, 52]. Moreover, "poor" glucoregulators as  
111 evidenced by blood glucose levels above 7.8 mmol/l following a 75-g oral glucose tolerance test  
112 (IGT), demonstrated impaired cognitive performance in measures of executive function but not of  
113 BF specifically [53-55].

114 Therefore, the objective of this research was to answer the following questions. First, given the

115 association between impulsiveness and executive function, we hypothesized that higher scores on  
116 the BIS-11 would predict impaired BF performance, as measured by the WCST, and the Stroop  
117 Color-Word Test. Second, given the relationship between impulsiveness and blood glucose, we  
118 hypothesized that fasting glucose levels could explain additional variance in BF. Previous  
119 findings have been contradictory with respect to an ‘optimum’ fasting blood glucose level as  
120 many of these have been tested in clinical populations (hence with particularly low or high  
121 fasting concentrations) and have assessed different cognitive functions. Third, while glucose  
122 supplementation has been shown to aid cognitive performance, this has most often been reported  
123 in contexts where fasting blood glucose levels are taken as a point of reference. However, in  
124 more realistic settings, it is likely that individuals perform a variety of cognitive-related tasks  
125 when their blood glucose levels are in a postprandial state. Thus, we took participants'  
126 postprandial state as a point of reference for glucose supplementation instead. Here, we predicted  
127 that glucose supplementation would be unlikely to confer a benefit to BF performance. Fourth,  
128 we hypothesized that individuals with lower changes in blood glucose from postprandial to blood  
129 glucose measured after glucose supplementation (i.e. "better" glucoregulators) would have  
130 superior BF performance.

131 To test this, we administered a more naturalistic dose of glucose (i.e. 15-g or equivalent to a glass  
132 of soda; see [56] for discussion of optimal dose and the inverted U shape curve ) in healthy  
133 populations in their postprandial state. While the IGT has been primarily adopted as a screening  
134 tool to identify individuals with poor glucoregulation (i.e. diabetes), the 75-g glucose drink  
135 provided in the IGT does not represent a typical dosage that an individual would consume prior  
136 to completing a cognitive task.

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## 138 **2. Methods and materials**

### 139 **2.1. Participants**

140 Sixty undergraduate volunteers (mean age 20.7 years, 38 females and 22 males, S.D. 1.5, study 1)  
141 and forty undergraduate volunteers (mean age 20.3 years, 27 females and 13 males, S.D. 1.4,  
142 study 2) were recruited in the study that was approved by the ethics committee of Sunway  
143 University Department of Psychology and complied with the Declaration of Helsinki. Sample  
144 size was determined using G\*Power to establish a minimum power level of 80% based on linear  
145 multiple regression analyses containing three predictors (study 2) with an estimated large effect  
146 size ( $f^2$ ) of 0.35. The selection of a smaller sample size in study 2 was in line with previous  
147 recommendations on sample size based on number of predictors in the model, size of the effect  
148 and statistical power [57, 58]. Participants were excluded from the study based on a number of  
149 criteria. Approximately 10% of prospective participants who were contacted to volunteer in  
150 taking part in the study did not fulfil the eligibility requirements. Exclusion criteria included  
151 those individuals who declared they were consuming at least two cups of coffee a day on a  
152 regular basis, suffering from diabetes, and/or had other forms of glucose intolerance. After  
153 screening and prior to participation, each volunteer signed an informed consent form.

### 154 **2.2. Cognitive measures**

155 Cognitive testing was carried out using the Psychology Experiment Building Language (PEBL)  
156 test battery [59, 60]. Presentation of tasks occurred via laptop computers using VGA color  
157 monitors and to complete the two tasks, participants took approximately 15 minutes. The  
158 description of the cognitive tasks which follows is based on a previously published paper by our  
159 research group [61].



160        **2.2.1. Stroop Color-Word test**

161        This task is believed to measure selective attention, response inhibition and cognitive flexibility.  
162        Participants were required to determine the color that words appeared in (see Fig.1). In some  
163        trials, the words would correspond to actual color names. When this was the case, participants  
164        had to ignore the written color name and instead select the color of the word. Task measures were  
165        average reaction time (ms) for congruent, incongruent and neutral stimuli and total number of  
166        errors. There were a total of 87 trials. The first 24 were practice trials, while the remaining 63  
167        were made up of congruent (n=20), incongruent (n=24) and neutral (n=19) trials. No other  
168        dependent measures were explored/tested.

169        **2.2.2. Berg's card sorting test**

170        This task is an adaptation of the Wisconsin Card Sorting Test (WCST) and measures complex  
171        executive functioning such as planning, cognitive flexibility, response inhibition, numerical skills  
172        and rules induction [62]. Participants were required to categorize cards based on the pattern  
173        appearing on them (see Fig.1). Each pile of cards had a different color, number and shape. A  
174        sample card would appear on the screen and participants were required to match this with one of  
175        the four piles of cards depending on a rule. Task measures included total number of errors and  
176        perseverative errors. There were a total of 128 trials with rule changing occurring 9 times (in an  
177        variable fashion across participants). No other dependent measures were explored/tested.

178        **2.3. Psychological measures**

179        **2.3.1. Barratt Impulsiveness Scale (BIS-11)**

180        The BIS-11 is a thirty-item self-report questionnaire designed to measure the personality trait of  
181        impulsivity [63]. Each item is rated on a 4-point Likert scale that ranges from 1 (rarely / never) to

182 4 (almost always / always). It is scored to yield a total score, three second-order factors (i.e.  
183 attentional, motor and nonplanning) and six first-order factors (i.e. attention, motor, self-control,  
184 cognitive complexity, perseverance and cognitive instability). Higher scores indicate higher  
185 impulsivity. The Cronbach's alpha for the current sample for total score was .79 and for each  
186 second-order subscales was .65 for attentional, .56 for motor and .67 for non-planning, similar to  
187 those previously reported [64] Test-retest reliability after a month interval for the total score and  
188 subscales scores has been found to be moderate (i.e. 0.61 to 0.83) [64].

## 189 **2.4. Physiological measures**

### 190 **2.4.1. Blood glucose**

191 Blood glucose readings were measured via capillary finger prick using Accu-Chek Performa  
192 diagnostic machines and test sticks (Roche Diagnostics, Germany). To minimize discomfort/pain,  
193 finger pricking was performed on the less painful lateral side of the fingertip. This is based on  
194 previous research investigating common practices amongst sufferers of diabetes when taking  
195 blood glucose measurements [65]. Blood glucose was collected once before cognitive tasks began  
196 (study 1). Participants were instructed to refrain from eating and drinking for three hours (i.e.  
197 fasting; for at least 180 minutes and no longer than 195 minutes) before their blood glucose was  
198 sampled (study 1). In study 2, blood glucose measurements were taken from participants having  
199 refrained from eating and drinking for two hours (i.e. postprandial; for at least 120 minutes and  
200 no longer than 135 minutes) instead of three hours as in study 1. A second blood glucose  
201 measurement was taken 15 minutes after having consumed a 15g glucose beverage.

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## 204        **2.5. Procedure**

205    Testing was conducted in research-dedicated laboratories. Testing was carried out in the  
206    afternoon, between 2:00 p.m. and 4 p.m. Participants were first required to complete the BIS-11  
207    questionnaire. Next, the participants' blood glucose levels (fasting) were measured by pricking a  
208    sanitized finger with the glucose meter lancet. After blood glucose levels were recorded,  
209    participants completed two computer based tests of behavioral flexibility, the Stroop Test (ST)  
210    and the Berg's Card sorting task (BCST) (study 1). The two tests were counterbalanced across  
211    participants. The whole experiment lasted approximately 25 to 30 minutes (study 1). Participants  
212    were given Cadbury chocolate bars at the end of testing as compensation. In study 2, following  
213    the first blood glucose measurement (postprandial), all participants received 15g of glucose  
214    dissolved in 200 mL of water flavored with 5 mL of no added-sugar lemon squash. The primary  
215    purpose of administering a glucose drink was to understand whether individual variability in the  
216    way glucose is processed modulated BF performance (i.e. glucoregulation). A secondary purpose  
217    was to capture variability in BF due to increased postprandial blood glucose. To avoid potential  
218    expectation bias of drinking a glucose beverage, we instructed participants that they may receive  
219    either a glucose drink or a placebo, even though this was not the case. To avoid this potential  
220    bias, in a prior small pilot study (i.e. n=20), we administered the same drink used during testing  
221    and found that when participants were asked whether they thought they had consumed a glucose  
222    drink or a placebo, the response rate for the glucose drink was at chance factor (i.e. 54%). Fifteen  
223    minutes after the glucose drink, a second blood glucose measurement was taken and cognitive  
224    testing began. The whole experiment (study 2) lasted approximately 45 minutes (see Fig.2).

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226        **2.6. Statistical analyses**

227        Statistical analyses were performed using SPSS Statistics version 22 (IBM, Armonk, NY, USA).  
228        A *P* value less than .05 was deemed significant. Data are shown as means and SD ±. Several  
229        hierarchical multiple regression analyses were performed to examine the contribution of  
230        psychological and physiological predictors (i.e. impulsivity and FBG in study 1 and impulsivity,  
231        blood glucose 15 minutes following glucose intake [time 2] and changes from PBG [time 1] to  
232        blood glucose following the intake of a glucose drink [time 2] in study 2) to outcomes of BF (i.e.  
233        Berg and Stroop task performance). Examinations of collinearity and independence of errors  
234        were used to rule out potential confounding variables. An independent-sample t test was  
235        conducted to compare fasting and postprandial blood glucose between study 1 and 2.

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## 246 **3. Results**

### 247 **3.1. Study 1**

248 In order to determine the contribution of impulsiveness (as measured by the BIS 11 scale), and  
249 fasting blood glucose levels to measures of behavioral flexibility (i.e. Berg and Stroop), we used  
250 hierarchical multiple regression analyses. Mean and standard deviation scores for both predictors  
251 and outcome variables are presented in **Table 1**. A preliminary examination of collinearity  
252 statistics (i.e. variance inflation factor [VIF] and tolerance) demonstrated that multicollinearity  
253 was not an issue (i.e. VIF= 1.028; Tolerance= 0.98). The data also met the assumption of  
254 independent errors (i.e. Durbin-Watson= 1.52-2.27).

255 In the first step of the analysis, we added the measure of BIS-11 total score (i.e. impulsiveness)  
256 as predictor. In the second step of the analysis, we added fasting blood glucose levels (eating and  
257 drinking avoided for 3 hours prior to blood glucose testing). The four dependent variables  
258 consisted of the total number of errors in the Pebl's Berg Card sorting task, perseverative errors,  
259 reaction time (RT) and total errors on the Pebl's Stroop task. The summary of the hierarchical  
260 multiple regression analyses are presented in **Table 2**. Neither BIS-11 total score nor fasting  
261 blood glucose levels contributed significantly to the regression model for any of the four criterion  
262 variables. It has been suggested [66] that the BIS-11 total score may be an imperfect measure of  
263 impulsivity, thus we ran additional analyses exchanging the BIS-11 total score with three  
264 subdomains of impulsivity, namely attention, motor and non-planning (which individually  
265 contribute to the BIS-11 total score). Results of these analyses were also non-significant.

266 To sum up, and contrary to our predictions, neither impulsivity nor fasting blood glucose levels  
267 could account for variability in behavioral flexibility (BF) performance.

## 268 3.2. Study 2

269 An independent sample t-test was conducted to assess whether blood glucose levels were  
270 different between participants in experiment 1 and those in experiment 2 (3 hours fasting versus 2  
271 hours postprandial). This analysis was carried out to ensure that the instructions to refrain from  
272 eating or drinking for either two or three hours did in fact result in differential blood glucose  
273 readings between the studies. Because sample sizes were unequal between the two experiments  
274 (i.e.  $n=60$  vs  $n=40$ ), we randomly selected a sample of 40 participants (out of the total 60) (using  
275 SPSS's Select Cases function) in experiment 1 and compared these with the 40 participants in  
276 experiment 2. Results showed that participants in experiment 2 had significantly higher blood  
277 glucose levels ( $6.23 \pm 1.36$ ) than participants in experiment 1 ( $5.58 \pm 1.02$ )  $t(72.47) = 2.31, p$   
278  $=0.024, d = 0.61$ .

279 As in experiment 1, hierarchical multiple regression analyses were used to determine the  
280 contribution of impulsiveness (as measured by the BIS 11 scale), blood glucose levels after a  
281 glucose drink (time 2) and changes in blood glucose from postprandial blood glucose (time 1) to  
282 time 2 to measures of behavioral flexibility. Mean and standard deviation scores for both  
283 predictors and outcome variables are presented in **Table 1**. A preliminary examination of  
284 collinearity statistics (i.e. variance inflation factor [VIF] and tolerance) demonstrated that  
285 multicollinearity was not an issue (i.e. VIF=1.021-1.064; Tolerance= 0.94-0.98). The data also  
286 met the assumption of independent errors (i.e. Durbin-Watson= 1.49-1.89).

287 In the first step of the analysis, we added the measure of BIS-11 total score (i.e. impulsiveness) as  
288 predictor. In the second step of the analysis, we added blood glucose levels after a glucose drink.  
289 In the third step of the analysis, we added changes in blood glucose from postprandial (time 1) to  
290 time 2 (following the sugary drink). The four dependent variables are the same as in experiment

291 1. The summary of the hierarchical multiple regression results is presented in **Table 3** and **Figure**  
292 **3**. BIS-11 total score, blood glucose levels after a glucose drink, and changes in blood glucose  
293 from time 1 to time 2 did not contribute significantly to the regression model in two of the four  
294 criterion variables (i.e. reaction time (RT) and total errors on the Pebl's Stroop task).

295 However, BIS-11 total score entered at step 1 explained 10.6% of the variance in total number of  
296 errors in the Pebl's Berg Card sorting task,  $F(1,38) = 4.51, p=0.040$ . Introducing blood glucose  
297 levels after a glucose drink at step 2 did not produce a significant change in  $R^2$  as it only  
298 explained an additional 8.3% of variation in Berg total errors,  $F(1, 37) = 3.78, p=0.059$ . Finally,  
299 adding changes in blood glucose from time 1 to time 2 produced a significant change in  $R^2$ , as it  
300 explained an additional 19.2% of variation,  $F(1,36) = 11.19, p=0.002$ . Together, the three  
301 independent variables accounted for 38.1% of variance in Berg total errors,  $F(3, 36) = 7.40,$   
302  $p < 0.001$ .

303 We then looked at perseverative errors in the Pebl's Berg Card sorting task, as this represents a  
304 separate measure of behavioral flexibility impairment, namely the repetition of particular  
305 (erroneous) response at least twice consecutively. BIS-11 total score entered at step 1 explained  
306 11.7% of the variance,  $F(1,38) = 5.04, p=0.031$ . Introducing blood glucose levels after a glucose  
307 drink at step 2 did not produce a significant change in  $R^2$  as it only explained an additional 1.8%  
308 of variation in Berg perseverative errors,  $F(1, 37) = 0.76., p=0.387$ . Finally, adding changes in  
309 blood glucose from time 1 to time 2 produced a significant change in  $R^2$ , as it explained an  
310 additional 25.3% of variation,  $F(1,36) = 14.89, p < 0.001$ . Together, the three independent  
311 variables accounted for 38.8% of variance in Berg perseverative errors,  $F(3, 36) = 7.61, p < 0.001$ .

312 Therefore, the lower the blood glucose increases from time 1 to time 2, the better the BF  
313 performance. Moreover, higher postprandial blood glucose levels (time 1) were predictive of

314 lower changes in blood glucose from time 1 to time 2. In fact, participants in the top quartile of  
315 postprandial blood glucose concentrations (7.85 mmol/l) had an average increase in blood  
316 glucose at time 2 of 1 mmol/l, whereas those in the bottom quartile (4.51 mmol/l) an average  
317 increase of 2.3 mmol/l. These differential responses were in turn related to fewer total and  
318 perseverative errors on the WCST (see Fig.4). A simple linear regression analysis confirmed that  
319 blood glucose levels between the postprandial measurement (time 1) and the difference between  
320 time 1 and time 2 were negatively correlated,  $r = -.533$ ,  $n=40$ ,  $p < .001$ . This finding is surprising  
321 given that, for example, fasting blood glucose levels have been reported to have a positive  
322 correlation with postprandial measurements [67].

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## 334 4. Discussion

335 The current investigation had four principal objectives: (1) to further our understanding of the  
336 relationship between impulsiveness and behavioral flexibility (BF) (study 1 and 2); (2) to  
337 explore whether fasting blood glucose levels can be used to predict BF (study 1); (3) to examine  
338 whether glucose levels measured following glucose supplementation from a postprandial state  
339 can explain BF performance (time 2); (4) to investigate whether blood glucose changes from a  
340 postprandial state (time 1) to blood glucose measured following the intake of a sugary drink (time  
341 2) can further be used to predict BF (study 2). To answer these questions, we devised two  
342 separate experiments. In study 1, we found that neither impulsiveness nor fasting blood glucose  
343 levels could account for variation in performance of the BF tasks (WCST and the Stroop task). In  
344 study 2, we found that higher levels of impulsiveness could predict increased number of errors on  
345 the WCST but not on the Stroop task. Moreover, we found that blood glucose levels measured 15  
346 minutes after the sugary drink intake did not explain significant improvements on the WCST nor  
347 on the Stroop. Importantly, however, lower increases in blood glucose from postprandial blood  
348 glucose to 15 minutes after the glucose drink were related to a reduction in the number of errors  
349 on the WCST but not on the Stroop.

350 At first glance, the findings that impulsiveness could predict BF in experiment 2 but not in 1  
351 seem puzzling, particularly given that mean scores on the BIS-11 were almost identical in both  
352 studies. However, because participants in study 1 and 2 differed on the basis of their fasting  
353 versus postprandial blood glucose profile, and on whether they received additional glucose prior  
354 to cognitive testing, these data should be interpreted taking these methodological differences into  
355 account. A performance comparison on the WCST between study 1 and study 2 participants (30.7  
356 vs 26 errors), does in fact suggest that that a combination of postprandial blood glucose levels

357 and taking additional glucose can alter negatively performance. Therefore, it is plausible that the  
358 BIS-11 scale is capturing variability in BF when cognitive performance declines. Previous  
359 research had demonstrated a relationship between the BIS-11 and measures of BF [15-17].  
360 However, in the above studies no measures of blood glucose concentrations were taken, and  
361 presumably most participants would have performed tasks of BF in a non-fasting and/or non-  
362 postprandial plus glucose intake state. Therefore, our data indicate that high impulsiveness is  
363 predictive of impaired BF performance in individuals who perform the task during their  
364 postprandial blood glucose levels plus glucose supplementation (more naturalistic state) but not  
365 in those in a fasting state.

366 We hypothesized that fasting blood glucose (study 1) could predict BF performance, however,  
367 this was not the case. Previous investigations which have reported a link between executive  
368 function and fasting blood glucose have been based on diabetic patients either hypoglycemic at  
369 fasting (i.e.  $<3.0$  mmol/l) or hyperglycemic (i.e.  $>7.00$  mmol/l). Blood glucose values at fasting  
370 below or above these thresholds negatively impact cognition. Some studies have shown that  
371 fasting blood glucose levels in a healthy, younger population below  $4.1$  mmol/l were detrimental  
372 to executive function (although not BF specifically) [68]. It would thus appear that fasting levels  
373 in the  $5.5$  mmol/l  $\pm 0.9$  range, as in the current study, bring about comparable BF performance  
374 across participants. This is in agreement with a large study in an elderly cohort whereby no  
375 association was found between fasting glucose levels in the  $5.14$  mmol  $\pm 0.78$  and executive  
376 function [69]. In contrast, our findings disagree with a recent study in which older, healthy  
377 participants with higher fasting blood glucose levels in the  $4.91$  mmol/l  $\pm 0.57$ , showed impaired  
378 executive function performance [70]. However, it should be stressed that there are inherent  
379 difficulties in comparing the findings from studies in which young and older adults were

380 employed due to different gluco-regulatory profiles and particularly because we know that  
381 characteristics such as age, BMI (body mass index) and a history of prior disease can negatively  
382 influence cognitive performance [71].

383 In study 2, we also found that blood glucose measured following the intake of a sugary drink  
384 (time 2) from a postprandial state did not account for variability in BF performance, as per our  
385 hypothesis. This finding suggests that once a certain blood glucose threshold has been reached (in  
386 our study  $7.6 \text{ mmol/l} \pm 1.2$ ), BF performance is unaffected. These results are not particularly  
387 surprising given that previous investigations have shown that cognitive improvements in  
388 memory, attention and executive function are only found when participants blood glucose levels  
389 raise to approximately 8.9 to 10 mmol/l, and when contrasted to placebo groups with fasting  
390 blood glucose levels of 4.2 to 5.3 mmol/l [72, 73]. Because all participants in our study 2 did take  
391 the glucose drink, and because their baseline postprandial blood glucose (i.e. pre-glucose  
392 supplementation) was significantly higher, blood glucose variations across participants were  
393 within a much narrower window (i.e.  $7.6 \text{ mmol/l} \pm 1.2$ ) than in previous studies to allow for  
394 cognitive performance differences to be picked up.

395 The most noteworthy finding from this study is that the lower the change (i.e. from postprandial  
396 blood glucose) in blood glucose levels following the consumption of a 15-g glucose drink, the  
397 better the performance on the WCST. These data are largely in agreement with previous  
398 investigations on other cognitive functions [51-55] and extend to the domain of behavioral  
399 flexibility. Moreover, however, our study uniquely shows the importance of glucoregulation on  
400 cognitive performance even when a small dose of glucose has been administered to individuals in  
401 their postprandial and not fasting state. Those adopting to track blood glucose and obtain an  
402 estimate of glucose regulation throughout the testing session tend to administer 25g or 50g

403 depending on whether younger or older adults are examined, respectively (see [56] for meta-  
404 analysis; [74] for review). Previous studies have also adopted to administer the glucose tolerance  
405 test (i.e. overnight fasting followed by the ingestion of a 75-g glucose drink) in a separate session  
406 as a measure of glucose regulation. Whilst this method can be used as a diagnostic tool for type 2  
407 diabetes, we aimed to use a smaller glucose dose as a more naturalistic indicator (15-g or  
408 equivalent to a glass of soda) of an individual's intake prior to performing a cognitive related task  
409 in an everyday setting.

410 Further analyses of our data also showed that higher postprandial blood glucose levels were  
411 predictive of smaller changes in blood glucose levels following glucose supplementation. This is  
412 in contrast with a previous study in which high fasting blood glucose levels were predictive of  
413 high postprandial blood glucose [67]. Because we measured glucoregulation from a postprandial  
414 state and not a fasting one, a direct comparison with the above study cannot be made.

415 Importantly, however, our data suggest that glucoregulation is a mechanism that is at least  
416 partially modulated by postprandial glucose levels, rather than being independent from it. Future  
417 studies would need to identify participants with similar postprandial profiles (i.e. within a  
418 1mmol/l range as opposed to over 2mmol/l in this study) to find out whether glucoregulation is  
419 independent from postprandial glucose levels in affecting BF performance.

420 Finally, in both experiment 1 and 2, impulsiveness, fasting blood glucose levels, glucose levels  
421 at time 2 and changes in blood glucose following the intake of a 15-g glucose drink did not  
422 account for variability in Stroop performance. Nevertheless, our findings may be explained by the  
423 observation that although there is great overlap between the neuronal substrates that determine  
424 performance on the WCST and Stroop, there is also some evidence to suggest that performance  
425 on the Stroop task relies more heavily on the anterior cingulate cortex (ACC) [75-77], whereas

426 performance on the WCST on the dorsolateral and ventromedial prefrontal cortex [78-80].  
427 Cognitively there is also good reason to suspect the task tap different processing mechanisms. For  
428 instance, Goshiki & Miyahara [81] in their examination of the tasks within a working memory  
429 framework argue that the WCST recruits both the phonological loop and central executive  
430 components; whereas the Stroop the central executive only.

431 Future studies would need to address some limitations of the current investigation. First, as  
432 participants verbally reported the time from last consumption of a meal, it is possible that the  
433 fasting and postprandial definitions of three and two hours without eating or drinking may have  
434 not been strictly adhered to. However, the blood glucose values for both the fasting group (study  
435 1) and postprandial group (study 2), are largely in line with previously reported studies [47, 82].  
436 Second, as meal composition intake prior to measuring fasting (study 1) and postprandial (study  
437 2) glucose levels was not monitored, there may have been effects of eating food with different  
438 protein, carbohydrate, fat and micronutrients on BF performance unrelated to absolute blood  
439 glucose concentrations per se, but for example due to variation in glucose metabolism, glucagon  
440 to insulin ratio, hormonal and mood effects [83].

441 In conclusion, our findings provide support for a larger body of knowledge which links  
442 impulsiveness and glucose regulation to executive function and extend to the domain of BF  
443 specifically. Additionally, the effect of glucose regulation on BF was mediated using more  
444 naturalistic glucose dosages than in previous investigations, and was partially affected by  
445 participants' postprandial blood glucose profile.

446 -----  
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