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1   **Title:**

2   Glucose and Fructose Hydrogel Enhances Running Performance, Exogenous Carbohydrate

3   Oxidation and Gastrointestinal Tolerance

4

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17

18   **Running Title:**

19   Carbohydrate Hydrogel Ingestion During Exercise

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29   **ABSTRACT**

30   **PURPOSE:** Beneficial effects of carbohydrate (CHO) ingestion on exogenous CHO oxidation

31   and endurance performance require a well-functioning gastrointestinal (GI) tract. However, GI

32   complaints are common during endurance running. This study investigated the effect of a CHO

33   solution-containing sodium alginate and pectin (hydrogel) on endurance running performance,

34   exogenous and endogenous CHO oxidation and GI symptoms. **METHODS:** Eleven trained

35   male runners, using a randomised, double-blind design, completed three 120-minute steady

36   state runs at 68% $\dot{V}O_2$max, followed by a 5-km time-trial. Participants ingested 90 g·h^{-1} of 2:1

37   glucose:fructose (^{13}C enriched) either as a CHO hydrogel, a standard CHO solution (non-

38   hydrogel), or a CHO-free placebo during the 120 minutes. Fat oxidation, total and exogenous

39   CHO oxidation, plasma glucose oxidation and endogenous glucose oxidation from liver and

40   muscle glycogen were calculated using indirect calorimetry and isotope ratio mass

41   spectrometry. GI symptoms were recorded throughout the trial. **RESULTS:** Time-trial

42   performance was 7.6% and 5.6% faster after hydrogel ([minutes:seconds]19:29±2:24;

43   *p*<0.001) and non-hydrogel (19:54±2:23, *p*=0.002), respectively, versus placebo (21:05±2:34).

44   Time-trial performance after hydrogel was 2.1% faster (*p*=0.033) than non-hydrogel. Absolute

45   and relative exogenous CHO oxidation was greater with hydrogel (68.6±10.8g, 31.9±2.7%;

46   *p*=0.01) versus non-hydrogel (63.4±8.1g, 29.3±2.0%; *p*=0.003). Absolute and relative

47   endogenous CHO oxidation were lower in both CHO conditions compared with placebo

48   (*p*<0.001), with no difference between CHO conditions. Absolute and relative liver glucose

49   and muscle glycogen oxidation were not different between CHO conditions. Total GI

50   symptoms were not different between hydrogel and placebo, but GI symptoms was higher in

51 non-hydrogel compared with placebo and hydrogel ($p<0.001$). **CONCLUSION:** Ingestion of

52 glucose and fructose in hydrogel form during running benefited endurance performance,

53 exogenous CHO oxidation and GI symptoms, compared with a standard CHO solution.

54

55 **INTRODUCTION**

56 It is well established that carbohydrate (CHO) ingestion during prolonged exercise can enhance

57 endurance performance (1). This is associated with the maintenance of plasma glucose

58 concentration and CHO oxidation during the latter stages of prolonged exercise (2). CHO

59 ingestion can also prevent the depletion or attenuate the use of liver glycogen (3, 4) and in

60 some instances, muscle glycogen (5, 6). American College of Sports Medicine (ACSM)

61 guidelines recommend consuming up to 90 $g \cdot h^{-1}$ of CHO during exercise lasting >2.5 hours or

62 where endogenous CHO stores will be depleted (7). However, these guidelines are largely

63 based on the accumulated evidence from studies that used cycle ergometer protocols, and so

64 may not be suitable for adoption by individuals during distance running. In fact, the mean rate

65 of CHO ingestion during marathon running (8, 9) is far below current recommendations (7).

66 This supports anecdotal evidence from practitioners and athletes (10) that the recommendation

67 to consume up to 90 $g \cdot h^{-1}$ (7) is not always practical or tolerable for runners.

68

69 The ergogenic effects of CHO ingestion require a well-functioning gastrointestinal (GI) tract

70 (10), yet surveys suggest individuals experience upper and lower GI symptoms during distance

71 running (11, 12). Moreover, GI symptoms are perceived to negatively affect running

72 performance (13). Symptoms may include nausea, vomiting, stomach cramps, urge for bowel

73 movement, reflux, fullness, bloating and diarrhoea (12). Evidence suggests over a one-month

74 period 78-84% of runners have reported experiencing at least one GI symptom whilst running,

75   and 14% of males, and 22% of females, have encountered moderate-to-severe GI symptoms

76   (14).

77

78   The aetiology of GI symptoms during distance running is likely multifactoral, influenced by

79   exercise intensity and fluid osmolality amongst others (15). The upper range of the exercise

80   intensities (60-75% maximal oxygen consumption; $\dot{V}O_2max$) that are typically achieved by

81   (non)elite distance runners during marathon running events (16) are associated with delayed

82   gastric emptying (17, 18), and the latter is thought to be a main cause of GI symptoms (19).

83   Higher CHO concentrations (>6%) are associated with delayed absorption of CHO (20)

84   resulting in increased residual CHO and water retention in the intestines (21), likely causing

85   elevated GI symptoms. For example, consuming hypertonic CHO beverages in large quantities

86   have been reported to cause a greater prevalence of GI symptoms whilst running (22). Ingestion

87   of multiple transportable CHO (glucose and fructose) can reduce the prevalence of GI

88   symptoms (23), whilst also increasing CHO oxidation rates (24). However, some individuals

89   are still susceptible to GI symptoms (25). Therefore, strategies or methods to increase CHO

90   intake without causing GI symptoms are of significant interest to runners and nutrition

91   practitioners.

92

93   Hydrogel food technology has recently become commercially available in sports nutrition

94   products (26), and may provide a novel means of delivering 90 $g \cdot h^{-1}$ CHO during running

95   whilst potentially reducing the severity of GI symptoms. The addition of sodium alginate and

96   pectin to CHO and water creates a pH-sensitive solution that forms a hydrogel that swells when

97   exposed to the low pH environment in the stomach (27). The hydrogel stays complexed in acid,

98   at this lower pH. Once in the small intestine, the higher alkaline pH causes the breakdown of

99   the gel and the release of the CHO (27).

100   Emerging research suggests that the ingestion of a CHO, sodium alginate and pectin solution

101   can enhance the rate of gastric emptying compared to a standard non-hydrogel CHO solution

102   (28). However, no study to date (29) including running (30, 31), has reported a benefit to

103   exercise performance, total whole-body substrate metabolism, exogenous CHO oxidation, or

104   GI symptoms when CHO was consumed as a hydrogel during endurance exercise. These

105   findings could be related to the mode of exercise, and the exercise intensities (45-60%

106   $\dot{V}O_2$max) studied. As a result, previous CHO hydrogel research using such exercise intensities

107   may not have sufficiently depleted the endogenous CHO stores, or impaired the GI tract to

108   elicit sufficient alterations in CHO absorption or GI symptoms. It is unclear whether the

109   metabolic and GI responses to CHO hydrogel ingestion are different from a non-hydrogel when

110   running at a higher exercise intensity.

111

112   This study aimed to investigate the effects of a multiple transportable CHO hydrogel, against

113   a non-hydrogel solution and placebo, on endurance running performance, substrate utilisation

114   and markers of GI symptoms. Using indirect calorimetry and ^{13}C tracer techniques, this study

115   aimed to assess exogenous and endogenous (liver and muscle) substrate utilisation in runners.

116   It was hypothesised that the addition of sodium alginate and pectin with CHO would improve

117   endurance performance, exogenous CHO oxidation and GI symptoms.

118

119   **METHODS**

120   *Participants*

121   Eleven trained, healthy male runners volunteered to participate in this study (mean ± SD, age

122   29 ± 6 years, body mass 68.7 ± 2.6 kg, body mass index 21.0 ± 1.3 kg/m^2, and $\dot{V}O_2$max 62.6 ±

123   4.2 $mL \cdot kg^{-1} \cdot min^{-1}$). Inclusion criteria required participants to have trained for >4 times per

124   week in running-specific training for at least the last 3 years, completed a marathon within the

125 last 18 months with a time under 2 hour 40 minutes or achieved a $\dot{V}O_2max$ >60 mL·kg^{-1}·min^{-1}

126 . Procedures and potential risks were explained before the study and all participants provided

127 written informed consent. Before commencing, this study gained institutional ethical approval

128 from Leeds Beckett University (Ref No. 57552) and was conducted in accordance with the

129 Declaration of Helsinki.

130

131 *Experimental design*

132 Preliminary testing, conducted 7 days before the first experimental trial, consisted of a

133 submaximal incremental test and maximal exercise test to volitional exhaustion (32) to

134 determine the specific submaximal running speed at 68% $\dot{V}O_2max$ for the experimental trial.

135 This was followed by familiarisation of the GI questionnaire and 5-km time-trial. Following

136 preliminary testing, participants completed three experimental trials (each separated by 7 days)

137 consisting of a 120-minute steady state run at 68% $\dot{V}O_2max$, followed by a 5-km time-trial.

138 During each trial, participants ingested one of three taste-matched solutions (CHO hydrogel,

139 non-hydrogel CHO solution, or placebo) in a randomised, double-blind order. Participants

140 ingested a 200 mL bolus immediately before exercise and then 100 mL of a 18% CHO solution

141 every 15 minutes throughout the steady state run delivering CHO at a rate of 90 g·h^{-1}.

142

143 *Diet and physical activity before testing*

144 Physical activity and food intake during the 48 hours before the first experimental trial were

145 recorded and participants were instructed to repeat the same diet and activity pattern before

146 subsequent trials. Before each experimental trial, participants were required to not undertake

147 any strenuous physical activity and avoid alcohol and caffeine consumption for 24 hours.

148 Before and for the duration of the study, participants were instructed to refrain from ingesting

149 foods with a high natural ^{13}C abundance (i.e. plants with a C4 photosynthetic cycle, or animals

150 fed with such plants) (33). This precaution ensured that background $^{13}CO_2$ abundance was less

151 likely to be perturbed from oxidation of endogenous and dietary substrate stores from naturally

152 "enriched" C4 origin. Prior to each trial, participants consumed a standardised evening meal

153 consisting of a total of ~1196 kcal; 58% CHO, 14% fat, 28% protein (fibre: ~12 g) 10-12 hours

154 before arriving at the laboratory.

155

156 *Experimental trials*

157 After an overnight fast, participants reported to the laboratory at the same time in the morning

158 to avoid any influence of circadian variance. Upon arrival at the laboratory, an in-dwelling

159 catheter (18-gauge Introcan Safety®, B. Braun Medical Ltd, Sheffield, UK) was inserted into

160 an antecubital forearm vein. Resting blood samples were drawn for plasma glucose, plasma

161 lactate, serum free fatty acid (FFA) and serum insulin. Subsequently, a 10-minute resting $\dot{V}O_2$

162 and $\dot{V}CO_2$ measurement was made using an online gas analysis system (Metalyser, Cortex,

163 Germany), calibrated to the manufacturer's instructions. For the measurement of $^{13}CO_2$:$^{12}CO_2$

164 in expired air at rest, 12 ml Exetainers (SerCon Ltd, Crewe, UK) of expired gas were collected

165 in duplicate via a mixing chamber (Jaeger, Germany). Participants then consumed a 200 mL

166 bolus of the experimental drink solution immediately before starting the 120 minutes of

167 running at 68% $\dot{V}O_2$max on a treadmill (Woodway, USA). Additional boluses (100 mL) of

168 each solution were provided every 15 minutes throughout the 120-minute exercise period.

169 Expired gas breath samples were collected, and measurements of $\dot{V}O_2$, $\dot{V}CO_2$, were measured

170 every 15 minutes during the steady state run. Samples of expired gas for $^{13}CO_2$ analysis were

171 collected during the final 60 seconds of each collection period at 60, 90 and 120 minutes.

172 Venous blood samples for the analysis of plasma glucose, plasma lactate, serum FFA and

173 serum insulin were drawn every 15 minutes, and for ^{13}C plasma glucose enrichment at 60

174 minutes and every 30 minutes thereafter. Heart rate measurements were taken every 15 minutes

175 during steady state exercise. In addition, every 30 minutes during the 120 minutes of running,

176 participants completed a GI questionnaire (34) covering three sections (upper, lower &

177 systemic GI symptoms), with a 10-point scale ranging from 1 (no problem at all) to 10 (the

178 worst it has ever been), and a score of ≥ 5 was classified as having severe GI symptoms.

179 Participants were familiarised with the GI questionnaire during preliminary testing. After the

180 steady state run, participants completed a self-paced 5-km time-trial, with a rolling start of a

181 running speed at 68% $\dot{V}O_2$max. Only feedback on distance completed was given at 1, 2, 2.5,

182 3, 4, 4.5, 4.6, 4.7, 4.8, 4.9 and 5-km. Participants were blinded of their finishing times until all

183 three experimental trials were completed.

184

185 *Experimental drinks*

186 The 180 grams of CHO used within the hydrogel and non-hydrogel experimental drinks were

187 a 2:1 ratio of glucose (120 g) (D-glucose; Thornton and Ross Ltd, Huddersfield, UK) and

188 fructose (60 g) (Danisco, Kettering, UK). The natural δ^{13}C abundance of the stock glucose and

189 fructose were measured by isotope ratio mass spectrometry (IRMS, Isoprime, Cheadle, UK),

190 using L-fucose as an isotopic internal standard as previously described (35) and determined to

191 be -25.68 ‰ and -12.27 ‰ respectively. All ^{13}C measurements are quoted with reference to

192 the internationally accepted standard for carbon isotope measurements, Vienna Pee Dee

193 Belemnite (VPDB). Both CHO solutions were enriched with 150 mg per 75 g CHO of

194 universally labelled (U-$^{13}C_6$) glucose and (U-$^{13}C_6$) fructose tracer (2:1 ratio) (Sigma Aldrich,

195 St Louis, MO). The final isotopic enrichment of each ingested CHO solution were 143.81 $\pm$

196 5.18 ‰ for the hydrogel solution and 142.32 $\pm$ 5.32 ‰ for the non-hydrogel solution. The

197 hydrogel solution contained high methoxy pectin and sodium alginate at 0.45 wt% with a ratio

198 of 1.25:1. All formulations contained 2.55 mmol·L^{-1} of NaCl (Saxa, Herts, UK), with the

199 placebo drink containing artificial sweetener (aspartame, Morrisons' plc, Bradford, UK) to

200   blind the participants to each condition. On completion of all three trials participants were

201   asked to stipulate the order of their conditions, only 3 of the 11 determining the order correctly.

202

203   *Analyses*

204   Blood samples were centrifuged and aliquots of plasma and serum were stored at -80°C until

205   analysis. Plasma glucose (glucose oxidase kit; Instrumentation Laboratory, Monza, Italy, Inter-

206   assay CV: 4.9%, Intra assay CV: 2.3%) and plasma lactate (Lactate kit, Randox, County

207   Antrim, UK, Inter CV: 4.5%, Intra CV: 2.7%) concentrations were analysed by

208   spectrophotometry (iLab 300 plus, iLab, UK). Serum insulin was analysed using a

209   chemoilumino-metric immunoassay (ADIVA Centaur, Bayer diagnostics, Berkshire, UK, Inter

210   CV: 3.2–4.6%, Intra CV: 2.6–5.9%). Serum FFA concentration was analysed by an acyl-CoA

211   synthetase and oxidase assay (NEFA-HR2, Wako Chemicals GmbH, Germany, Inter assay

212   CV: 1.5%). Isotope ratio mass spectrometry (IRMS; AP2003, GVI Instruments Ltd,

213   Manchester, UK) were used to determine the $^{13}CO_2:^{12}CO_2$ in expired air as described

214   previously (36). The $^{13}C:^{12}C$ in plasma glucose was determined using liquid chromatography

215   linked-isotope ratio mass spectrometry (LC-IRMS), as previously described (35). Briefly,

216   plasma samples were spiked with an internal standard (L-fucose, Sigma Aldrich, Poole, UK)

217   and prepared by ultrafiltration (30000 MWCO, Amicon Ultra 4, Millipore, Watford, UK) for

218   LC-IRMS analysis of ^{13}C-glucose enrichment ($\delta^{13}C$-glucose).

219

220   *Calculations*

221   Total CHO and fat oxidation (g·min^{-1}) were calculated using the stoichiometric equations

222   (equation 1 and 2) proposed by Jeukendrup & Wallis (37), with protein oxidation during

223   running assumed to be negligible.

224

225 $$\text{CHO oxidation} = (4.210 \times \dot{V}CO_2) - (2.962 \times \dot{V}O_2) \qquad (1)$$

226

227 $$\text{Fat oxidation} = (1.695 \times \dot{V}O_2) - (1.701 \times \dot{V}CO_2) \qquad (2)$$

228

229 To calculate absolute (g) whole body CHO, exogenous and endogenous (liver and muscle)

230 CHO and fat oxidation the area under the curve technique was applied to the respective rates

231 (g·min^{-1}). Energy expenditure contributions from CHO and fat were calculated from absolute

232 values, by applying their respective energy potentials (4.07 kcal and 9.75 kcal (37)). Total

233 ingested glucose and fructose isotopic enrichment, (R_{exo}), and expired air (R_{exp}) were expressed

234 in standard $\delta^{13}C$ units (‰) relative to VPDB (38). The rate of exogenous CHO oxidation

235 derived from the combined ingestion of glucose and fructose (CHO_{EX}) were computed using

236 equation 4 (39).

237

238 $$CHO_{EX} \ (\text{g·min}^{-1}) = \dot{V}CO_2 \ [(R_{exp} - R_{ref}) / (R_{exo} - R_{ref})] / k \qquad (4)$$

239

240 Where $\dot{V}CO_2$ is in L·min^{-1}, R_{exp} is the isotopic composition of expired CO_2 and R_{ref} is the

241 isotopic composition of expired CO_2 at the same time point with ingestion of placebo. The

242 isotopic composition of the ingested solution computed as R_{exo} and the k (0.747 L·g^{-1}) is the

243 volume of CO_2 provided by the complete oxidation of glucose. The oxidation efficiency,

244 percentage of the ingested CHO utilised was calculated (40).

245

246 Computations were made on the assumption that, in response to exercise, ^{13}C is not irreversibly

247 lost in pools of tricarboxylic acid cycle intermediates and/or bicarbonate, and that lactate

248 produced from either glucose or fructose is either oxidized in muscle or recycled through

249 gluconeogenesis to be used subsequently by complete oxidation. Essentially exogenous CHO

250 oxidation is calculated irrespective of the pathway that finally produces $^{13}CO_2$ that can be

251 measured. The calculations assume that $^{13}CO_2$ recovery in expired gases were complete or

252 almost complete during exercise. Such computation has been shown to underestimate

253 exogenous oxidation rates at the beginning of exercise because of the delay between $^{13}CO_2$

254 production in tissues and expired $^{13}CO_2$ at the mouth (41). Therefore, exogenous CHO

255 oxidation data are presented for the final 60 minutes of the 120-minute running period, where

256 it is expected that there would be isotopic equilibrium in the tissues and at the mouth (42).

257

258 The oxidation rate of plasma glucose was calculated based on the ^{13}C isotopic composition of

259 plasma glucose (R_{glu}) (equation 5, 43):

260

261 $$Plasma\ CHO\ (g \cdot min^{-1}) = \dot{V}CO_2\ [(R_{exp} - R_{ref}) / (R_{glu} - R_{ref})] / k \qquad (5)$$

262

263 Endogenous CHO oxidation is presented as the difference between total CHO oxidation and

264 exogenous CHO oxidation. The oxidation rate of muscle glycogen ($g \cdot min^{-1}$), either directly or

265 through the lactate shuttle (44), were calculated by subtracting plasma glucose oxidation from

266 total CHO oxidation. Finally, glucose oxidation derived from the liver was estimated as the

267 difference between plasma glucose oxidation and exogenous CHO oxidation (43).

268

269 *Statistical analyses*

270 Data evaluation was performed using Prism (8.3.1) (GraphPad Software, La Jolla California

271 USA). Eleven trained male runners were recruited for this study, providing 92% power to

272 detect differences in performance, with an expected mean difference of 1.6% between CHO

273 hydrogel and non-hydrogel (29), assuming a standard deviation of 1.43% at an alpha of 0.05.

274   In addition, eleven male runners would provide 90% power to detect differences in the rate of

275   exogenous CHO oxidation, with an expected mean difference of 0.11 g·min⁻¹ between CHO

276   hydrogel and non-hydrogel, assuming a standard deviation of 0.10 g·min⁻¹ at an alpha of 0.05.

277   Variables were checked for normality prior to performing statistical tests (Kolmogorov–

278   Smirnov test). Differences in $\dot{V}O_2$, $\dot{V}CO_2$, RER, HR, rate of substrate utilisation, plasma and

279   serum metabolites were analysed using a two-factor (time x treatment) analysis of variance

280   (ANOVA) for repeated measures. Time-trial completion time, total energy expenditure,

281   endogenous CHO oxidation and relative substrate data for all three conditions was analysed

282   using a one-way ANOVA. Post hoc analysis was performed for any significant time and

283   condition main effects or interactions using paired sample *t*-tests with Bonferroni adjustment.

284   Only CHO conditions were compared when variables considered $\delta^{13}CO_2$ of expired gas and

285    $\delta^{13}C$ in plasma glucose. A paired sample *t*-test was used to analyse absolute and relative

286   exogenous CHO oxidation and the oxidation of muscle, liver and plasma glucose, as well as

287   the percentage of the exogenous source of CHO utilised. GI symptoms were analysed using

288   Wilcoxon matched-pairs signed rank test. Relationship between time-trial performance and GI

289   symptoms was assessed using a Spearman's rho correlations. Data are presented as mean ± SD

290   and statistical significance was set at $p < 0.05$.

291

292   **RESULTS**

293   *Time-Trial Performance*

294   A one-way ANOVA determined that mean 5-km time-trial performance [minutes:seconds] was

295   significantly different between conditions ($p = < 0.0001$). Post hoc analysis revealed significant

296   improvements in time-trial performance with the ingestion of hydrogel (19:29 ± 2:24, 7.6%, *p*

297   < 0.001) and non-hydrogel (19:54 ± 2:23, 5.6%, *p* = 0.002) compared with placebo (21:05 ±

298   2:34, Figure 1). Time-trial performance for hydrogel was also significantly faster (2.1%)

299   compared with non-hydrogel ($p = 0.033$).

300

301   *$\dot{V}O_2$, $\dot{V}CO_2$, RER and Heart Rate*

302   A two-way ANOVA showed that there was a significant condition and time interaction for

303   $\dot{V}O_2$ ($p = 0.001$), $\dot{V}CO_2$ ($p = 0.003$) and RER ($p = 0.001$) during the 120 minutes of running.

304   Post hoc analysis at each time period during the steady state run revealed $\dot{V}O_2$ was not

305   significantly different between hydrogel and non-hydrogel ($p = 0.33$ to 0.99), but $\dot{V}O_2$ was

306   significantly lower in both CHO conditions compared with placebo ($p = 0.001$ to 0.03; Table

307   1). $\dot{V}CO_2$ was not significantly different between conditions in the first 60 minutes of the steady

308   state run ($p = 0.70$ to 0.99). In the final 60 minutes of the steady state run, $\dot{V}CO_2$ was not

309   significantly different between hydrogel and non-hydrogel ($p = 0.43$ to 0.99), but $\dot{V}CO_2$ was

310   significantly greater in both CHO conditions compared with placebo ($p = 0.001$ to 0.03).

311   Throughout the steady state run, RER was significantly higher in both CHO conditions relative

312   to placebo ($p = 0.001$ to 0.02), with no significant difference in RER between hydrogel and

313   non-hydrogel conditions ($p = 0.09$ to 0.5). In the placebo condition, RER was significantly

314   lower during the final 60 minutes compared with the initial 60 minutes of the steady state run

315   ($p = 0.01$). HR was not significantly different across time ($p = 0.43$) or between conditions (p

316   $= 0.87$) during the steady state run.

317

318   *Energy Expenditure, Total Carbohydrate and Fat Oxidation*

319   A one-way ANOVA showed that there was no main effect of condition for total energy

320   expenditure during the steady state run (hydrogel, 1746 ± 135 kcal; non-hydrogel, 1760 ± 137

321   kcal; placebo, 1818 ± 167 kcal; $p = 0.83$). A two-way ANOVA showed that there was a

322   significant condition and time interaction for both absolute whole-body CHO ($p = 0.001$) and

323   fat oxidation ($p = 0.001$). Post hoc analysis indicated that absolute whole-body CHO oxidation

324   in hydrogel (324.2 ± 17.6 g) and non-hydrogel (318.3 ± 20.7 g) were not significantly different

325   during the 120-minute steady state run ($p = 0.09$), but the absolute CHO oxidation in both CHO

326   conditions was significantly higher than placebo (260.9 ± 16.5 g; $p < 0.00001$ & $p < 0.001$). In

327   addition, absolute whole-body CHO oxidation in hydrogel and non-hydrogel were not

328   significantly different during the first ($p = 0.99$) and second ($p = 0.99$) hour of steady state

329   running, however, both CHO conditions were significantly higher compared with placebo ($p <$

330   0.001; Table 1). Conversely, absolute fat oxidation was significantly lower throughout the 120-

331   minute steady state run for both CHO conditions (hydrogel = 43.5 ± 8.3 g; non-hydrogel, 47.6

332   ± 7.9 g) compared with placebo (77.8 ± 15.2 g; $p < 0.0001$ & $p < 0.0001$) as well as during the

333   first (hydrogel: $p = 0.0003$; non-hydrogel: $p = 0.0003$) and second hour (hydrogel: $p < 0.0001$;

334   non-hydrogel: $p < 0.0001$) (Table 1). Fat oxidation was also significantly lower in hydrogel

335   compared with non-hydrogel during the first ($p = 0.002$) and second hour ($p = 0.001$) of

336   exercise and over the 120-minute steady state run ($p = 0.002$). A one-way ANOVA showed

337   that there was a main effect of condition ($p < 0.0001$) for the relative contribution of CHO and

338   fat to energy expenditure during the 120-minute steady state run (Figure 2). Post hoc analysis

339   showed that the relative contribution of CHO to energy expenditure was significantly greater

340   in both CHO conditions (hydrogel, $76.1 \pm 4.4\%$; non-hydrogel, $74.1 \pm 5.2\%$), compared with

341   placebo ($58.2 \pm 6.5\%$; $p = 0.001$ and $p = 0.001$), with no significant difference between CHO

342   conditions ($p = 0.12$). The relative contribution of fat to energy expenditure was significantly

343   lower for the CHO conditions (hydrogel, $23.9 \pm 2.1\%$; non-hydrogel, $25.9 \pm 1.8\%$), compared

344   with placebo ($41.8 \pm 4.9\%$, $p < 0.0001$ & $p < 0.0001$), with the hydrogel being significantly

345   lower compared with the non-hydrogel ($p = 0.002$).

346

347   *$\delta^{13}CO_2$ in expired gas and $\delta^{13}C$ in plasma glucose*

348 A two-way ANOVA showed that there was a significant main effect of time and condition and

349 time interactions for $\delta^{13}CO_2$ in expired gas ($p < 0.0001$ and $p < 0.0001$). Post hoc analysis for

350 $\delta^{13}CO_2$ in expired gas showed that there was no significant difference between conditions at

351 rest ($p = 0.45$ to 0.99, Figure 3A). In placebo, the $\delta^{13}CO_2$ in expired gas significantly increased

352 over time by 1.28‰ from the start to the end of the steady state run ($p < 0.0001$). These data

353 were then used as the background correction for the calculation of exogenous CHO and plasma

354 glucose oxidation for each CHO condition. The $\delta^{13}CO_2$ in expired gas significantly increased

355 over time from the start of exercise following the ingestion of the ^{13}C enriched CHO solutions

356 ($p = 0.001$) and peak values were reached at 120 minutes. As shown in figure 3A, post hoc

357 analysis indicated that the hydrogel was significantly higher compared with the non-hydrogel

358 at 60 ($p = 0.005$), 90 ($p = 0.009$), and 120 minutes ($p = 0.012$). The isotopic composition of

359 plasma glucose ($\delta^{13}C$) significantly increased by 1.28‰ from 60 to 120 minutes during the

360 steady state run with ingestion of placebo ($p < 0.0001$, Figure 3B). In both CHO conditions,

361 there was a significant main effect of time ($p = 0.001$) for plasma glucose $\delta^{13}C$, with post hoc

362 analysis revealing a significant rise between 60 and 120 minutes ($p = 0.004$). However, there

363 was no significant condition or time interaction for plasma glucose $\delta^{13}CO_2$ between CHO

364 conditions ($p = 0.14$).

365

366 *Exogenous and Endogenous Carbohydrate Oxidation*

367 A two-way ANOVA showed a significant main effect of time for the rate of exogenous CHO

368 oxidation ($p < 0.0001$). Post hoc analysis revealed that the rate of exogenous CHO oxidation

369 increased significantly ($p = 0.001$) in both CHO conditions during the final 60 minutes of the

370 steady state run, peaking at 120 minutes during each condition (hydrogel: 1.27 ± 0.17 g·min^{-1};

371 non-hydrogel: 1.18 ± 0.13 g·min^{-1}, Figure 4A). There was also a condition and time interaction

372 ($p = 0.005$), with post hoc analysis showing that exogenous CHO oxidation was significantly

373   greater in hydrogel compared with non-hydrogel at 60 (1.03 ± 0.19 vs 0.93 ± 0.14 g·min^{-1}, p =

374   0.001), 90 (1.14 ± 0.19 vs 1.06 ± 0.14 g·min^{-1}, p = 0.009) and 120 minutes (1.27 ± 0.17 vs 1.18

375   ± 0.13 g·min^{-1}, p = 0.001). A paired sample t-test showed that absolute exogenous CHO

376   oxidation during the final 60 minutes of the steady state run (Table 2) was significantly greater

377   with hydrogel compared with non-hydrogel (p = 0.003). In addition, relative exogenous CHO

378   (Figure 2) was also significantly greater with hydrogel (31.9 ± 2.69%) compared with the non-

379   hydrogel (29.3 ± 1.96%, p = 0.003). The percentage of the exogenous source of CHO utilised

380   was also significantly greater in the hydrogel (76.2%) compared with the non-hydrogel

381   condition (70.6%, p = 0.003). A two-way ANOVA showed that there was a significant main

382   effect of condition for the rate of endogenous CHO oxidation during the final 60 minutes of

383   the steady state run. Post hoc analysis showed that rate of endogenous CHO oxidation was

384   lower for both CHO conditions (hydrogel 1.64 ± 0.15 g·min^{-1}; non-hydrogel 1.68 ± 0.14 g·min^{-1}

385   ^{1}) compared with placebo (2.03 ± 0.19 g·min^{-1}, p = 0.001), but reporting no significant

386   condition and time interaction (p = 0.07). A one-way ANOVA showed that the absolute

387   endogenous CHO oxidation, during the final 60 minutes of the steady state run was not

388   significantly different between the hydrogel and non-hydrogel conditions (Table 2, p = 0.58).

389   There was also no significant difference in the relative contribution of endogenous CHO

390   oxidation to energy expenditure between the hydrogel (46.0 ± 2.3%) and non-hydrogel

391   conditions (46.8 ± 1.4%, p = 0.46).

392

393   A two-way ANOVA showed a significant main effect of time for plasma glucose oxidation (p

394   < 0.001) during the final 60 minutes of steady state running, reaching peak rates at 120 minutes

395   during each CHO condition (hydrogel: 1.58 ± 0.24 g·min^{-1}; non-hydrogel: 1.49 ± 0.20 g·min^{-1}

396   ^{1}, Figure 4B). However, there was no significant condition and time interaction between the

397   hydrogel and non-hydrogel trials (p = 0.86). In contrast, a paired sample t-test showed that

398   plasma glucose oxidation was significantly higher in hydrogel compared with non-hydrogel

399   when expressed in absolute ($p = 0.001$, Table 2) and relative terms ($41.0 \pm 2.1\%$ vs $38.1 \pm$

400   1.8%, $p = 0.001$).

401

402   The rate of glucose oxidation derived from the liver remained stable in both CHO conditions

403   during the final 60 minutes of steady state run (Figure 4C), with a two-way ANOVA showing

404   that there was no significant condition and time interaction between the hydrogel and non-

405   hydrogel conditions ($p = 0.78$). A paired sample t-test showed that liver glucose oxidation was

406   also not significantly different between the hydrogel and non-hydrogel conditions when

407   expressed in absolute ($p = 0.83$, Table 2) or relative terms ($9.1 \pm 1.5\%$ vs $8.7 \pm 0.9\%$, $p = 0.83$,

408   Figure 2). Muscle glycogen oxidation rates (Figure 4D) remained stable over time, with a two-

409   way ANOVA showing that there was no significant condition and time interaction between

410   CHO conditions ($p = 0.46$). In addition, a paired sample t-test showed that muscle glycogen

411   oxidation was not significantly different between the hydrogel and non-hydrogel when

412   expressed in absolute ($p = 0.26$, Table 2) or relative terms ($36.9 \pm 3.4\%$ vs. $38.1 \pm 1.8\%$, $p =$

413   0.26).

414

415   *Circulatory Metabolites and Insulin*

416   A two-way ANOVA showed that there were significant condition and time interactions for

417   plasma glucose ($p = 0.04$), plasma lactate ($p = 0.03$), serum FFA ($p < 0.0001$) and serum insulin

418   ($p < 0.0001$). Post hoc analysis showed that plasma glucose concentrations (Figure 5A) during

419   the 120-minute run were significantly greater in both CHO conditions compared with placebo

420   at each time point ($p < 0.001$). Plasma glucose concentrations were not significantly different

421   between hydrogel and non-hydrogel at any time point ($p = 0.74$ to 0.99). Plasma lactate

422   concentration were significantly higher in the CHO conditions compared with placebo ($p =$

423   0.001 to 0.01) throughout the steady state run (Figure 5B). However, plasma lactate was not

424   significantly different between hydrogel and non-hydrogel conditions ($p = 0.66$ to 0.94). Serum

425   FFA concentration (Figure 5C) increased significantly over time throughout the steady state

426   run in the placebo condition ($p < 0.001$). Serum FFA was significantly lower in the CHO

427   conditions at each time point compared with placebo ($p = 0.002$ to 0.01) and was not

428   significantly different between CHO conditions at any time points ($p = 0.34$ to 0.99). Serum

429   insulin concentration (Figure 5D) was significantly higher in both CHO conditions throughout

430   the steady state run compared with placebo ($p < 0.001$) and was not significantly different

431   between CHO conditions ($p = 0.53$ to 0.99).

432

433   *Gastrointestinal Response*

434   Mean GI symptom scores, the minimum and maximum scores, and percentage of participants

435   who reported GI symptom scores ≥ 5 are presented in Table 3. A Wilcoxon signed rank test

436   showed there was no significant differences in total scores of GI symptoms between hydrogel

437   and placebo conditions ($p = 0.19$). However, the total GI symptom scores for the hydrogel and

438   placebo were both significantly lower compared with non-hydrogel ($p = 0.001$ & $p = 0.001$

439   respectively). The prevalence of upper and lower GI symptoms ranged from 0-18% in the

440   placebo, 18-36% in the hydrogel, and 18-64% in the non-hydrogel condition. Upper GI

441   symptom scores were significantly greater during the non-hydrogel compared with hydrogel

442   (Table 3, $p = 0.025$) and placebo ($p = 0.001$) conditions, and was significantly greater in

443   hydrogel compared with the placebo condition ($p = 0.011$). Lower GI symptom scores were

444   significantly greater during the non-hydrogel compared with hydrogel (Table 3, $p = 0.006$) and

445   placebo ($p < 0.001$) conditions, and was significantly greater in hydrogel compared with the

446   placebo condition ($p = 0.007$). Systemic symptom score was significantly greater during the

447   non-hydrogel compared with hydrogel (Table 3, $p = 0.039$) and placebo ($p = 0.002$) conditions

448 and was not significantly different in hydrogel compared with placebo ($p = 0.98$). A Spearman's

449 rank-order correlation showed a strong positive correlation between combined total GI

450 symptom scores and time-trial performance for all three conditions ($r_s = .693$, $p = 0.021$). In

451 the non-hydrogel condition, there was a strong positive correlation between GI symptom scores

452 and time-trial performance ($r_s = .746$, $p = 0.011$). No correlation was shown in the hydrogel

453 and placebo conditions ($r_s = .536$, $p = 0.094$ and $r_s = .560$, $p = 0.067$, respectively).

454

455 **DISCUSSION**

456 This is the first study, to our knowledge, to demonstrate that ingestion of 90 g·h^{-1} of glucose

457 and fructose in hydrogel form whilst running at 68% $\dot{V}O_2$max for 120 minutes improves 5-km

458 time-trial performance in trained runners compared with a non-hydrogel CHO. This improved

459 performance could be attributed to the increase exogenous CHO oxidation, decreased fat

460 oxidation and reduced GI symptoms following hydrogel ingestion, as liver and muscle

461 glycogen oxidation during the last hour of the 120 minutes of running were not different

462 between hydrogel and non-hydrogel. Thus, the CHO hydrogel may allow athletes to consume

463 more adequate amounts of CHO during prolonged running, subsequently improving

464 performance.

465

466 The novel performance effects observed in the present study when ingesting CHO hydrogel

467 whilst running conflict with previous evidence that have failed to detect a performance benefit

468 across cycling, cross-country skiing and running (29). The reasons for the discrepancy are

469 unclear, but it may relate to differences in exercise intensity, duration, CHO type and dose,

470 training status, exercise mode, or the performance test employed. To the authors' knowledge,

471 only one study has investigated the effect of consuming a commercially available CHO

472 hydrogel (90 g·h^{-1}) against a non-hydrogel CHO (90 g·h^{-1}) solution on running performance

473 (30). However, the incremental time to exhaustion treadmill test following a 180-minute run at

474 60% $\dot{V}O_2$max is maximal in nature and may not have been appropriate to detect an effect of

475 CHO hydrogel on running performance.

476

477 In the present study, total CHO oxidation was higher, and fat oxidation was lower when CHO

478 was ingested in either hydrogel or non-hydrogel conditions relative to placebo, which is

479 consistent with the known effects of CHO ingestion on whole body substrate metabolism (45).

480 In addition, lower fat oxidation was observed with hydrogel compared with non-hydrogel

481 during the 120-minute run, which cannot be attributed to between-condition differences in

482 serum free fatty acid and serum insulin concentration. However, there was no difference in

483 whole-body CHO oxidation, liver glucose oxidation and muscle glycogen oxidation between

484 the hydrogel and non-hydrogel conditions during 60-120 minutes of the steady state run. Thus,

485 the improvement in 5-km time-trial performance following the ingestion of hydrogel compared

486 with the non-hydrogel CHO solution is not related to sparing of either liver or muscle glycogen

487 during the final hour of the 120-minute steady state run.

488

489 Exogenous CHO oxidation rates during the final 60 minutes for the non-hydrogel CHO

490 solution (1.06 ± 0.13 g·min^{-1}) are consistent with existing running literature that also

491 administered 90 g·h^{-1} of a 2:1 ratio of glucose and fructose (46). For the first time we show that

492 significantly higher (8.2%) exogenous CHO oxidation rates can be achieved with the ingestion

493 of hydrogel compared with non-hydrogel. The higher exogenous CHO oxidation resulted in a

494 greater utilisation with the hydrogel (76.2%), compared with non-hydrogel condition (70.6%).

495 The elevations in plasma glucose oxidation due to exogenous CHO oxidation may therefore

496 have contributed, at least partially, to the improvement in 5-km time-trial performance

497 following the hydrogel compared with non-hydrogel CHO solution. Of the three studies that

498 previously measured exogenous CHO oxidation following CHO hydrogel ingestion (31, 47,

499 48), only Barber *et al.,* (31) included a comparative CHO condition. In contrast to the present

500 study, Barber *et al.* (31) found no difference in exogenous CHO oxidation between the

501 hydrogel (maltodextrin-fructose) and CHO-matched non-hydrogel conditions in trained

502 runners during 120 minutes of running at 60% $\dot{V}O_2max$. The higher exercise intensity used in

503 our study compared with Barber *et al.,* (31) may be a potential explanation for the disparity, as

504 exogenous CHO oxidation is well accepted to increase with exercise intensity. In addition, a

505 strength of the present study design is that both CHO solutions were enriched with a high dose

506 of universally labelled ^{13}C tracers which enhances the signal to noise-ratio and the ability to

507 definitively detect oxidative differences in ^{13}C labelled substrate metabolism.

508

509 To our knowledge, we are the first to report the effect of CHO hydrogel ingestion during

510 running at exercise intensities that align with (non)elite marathon running (16), and delayed

511 gastric emptying (17, 18). The potential for a lower rate of gastric emptying during running

512 (18, 49) may have been tempered in the present study by the hydrogel solution, since a faster

513 rate of gastric emptying can be achieved when CHO is ingested as a hydrogel compared to a

514 standard CHO solution (28). This may result in a more effective intestinal absorption of CHO

515 (49), and would be consistent with the greater oxidation of exogenous CHO observed in the

516 present study, relative to the non-hydrogel CHO solution. This interpretation is supported by a

517 glucose infusion study which suggested exogenous CHO oxidation to be limited by intestinal

518 absorption (50).

519

520 An improved rate of gastric emptying during running in the hydrogel condition may have also

521 contributed to the lower severity and incidence of GI symptoms reported by our cohort, given

522 that delayed gastric emptying is thought to be one of the main contributors to GI symptoms

523 during exercise (19). However, the lower GI symptoms with hydrogel contrasts with the

524 literature (29). The reasons for the discrepancy between others (29) and the present study may

525 be related to our robust familiarisation of participants to the GI questionnaire, a higher exercise

526 intensity and a different CHO type and dose. In the present study, both CHO conditions used

527 a high concentration of CHO (18%) in the form of glucose:fructose (2:1 ratio) as opposed to

528 maltodextrin:fructose (7.8-15.8%: 1:0.7 ratio) (29). As glucose is monomeric and maltodextrin

529 is polymeric the potential for osmotic differences exists which could also account for the

530 increased prevalence of GI symptoms in the non-hydrogel condition in this study compared

531 with previous literature (29). The high rate of hydrogel ingestion (90 g·h^{-1}) used in the present

532 study did not completely nullify GI symptoms for some individuals, and future research should

533 look to see whether moderate rates of non-hydrogel ingestion (50-60 g·h^{-1}), which are

534 associated with lower GI symptoms (51) are equally efficacious. A limitation of the present

535 study and the literature, is the reporting of how accustomed participants are to consuming CHO,

536 as gut training may alleviate some GI symptoms, such a stomach comfort following CHO

537 ingestion (52). Thus, further research is required to establish whether gut training diminishes

538 the positive effect of hydrogel on GI symptoms seen in this study. In addition, further research

539 is required in measuring the rate of gastric emptying when ingesting CHO in hydrogel form,

540 as the viscosity of the ingested liquid (i.e. CHO hydrogel) could be a limitation of the double

541 sampling gastric aspiration technique previously administered (28). Nevertheless, our data

542 suggest that when men running at exercise intensities consistent with (non)elite marathon

543 running (16) and delayed gastric emptying (17, 18), hydrogel ingestion may be an effective

544 means to increase the rate of CHO ingestion during marathon running (8, 9), in line with ACSM

545 guidelines (7). This would subsequently decrease the incidence and severity of GI symptoms,

546 and improve running performance. However due to females also reporting GI symptoms when

547 running (14), further research is required to establish if CHO hydrogel ingestion is equally

548 efficacious.

549

550 *Conclusion*

551 Ingestion of glucose and fructose (90 $g \cdot h^{-1}$) in hydrogel form whilst running at 68% $\dot{V}O_2max$

552 for 120 minutes improved subsequent 5-km time-trial performance relative to a CHO-matched

553 non-hydrogel solution and placebo. This occurred alongside increased exogenous CHO

554 oxidation, decreased fat oxidation, and a reduction in symptoms of GI when ingesting the

555 hydrogel solution. If individuals choose to ingest a high rate of glucose and fructose (90 $g \cdot h^{-1}$)

556 during prolonged running, they may benefit from ingesting monomeric CHO in hydrogel form

557 as compared to a standard non-hydrogel CHO solution.

558

566

567 **CONFLICTS OF INTEREST**

568 The authors declare no conflict of interest. The results of the present study do not constitute

569 endorsement by the American College of Sports Medicine. The results of the study are

570 presented clearly, honestly, and without fabrication, falsification or inappropriate data

571 manipulation.

578

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741 **Figure 1.**

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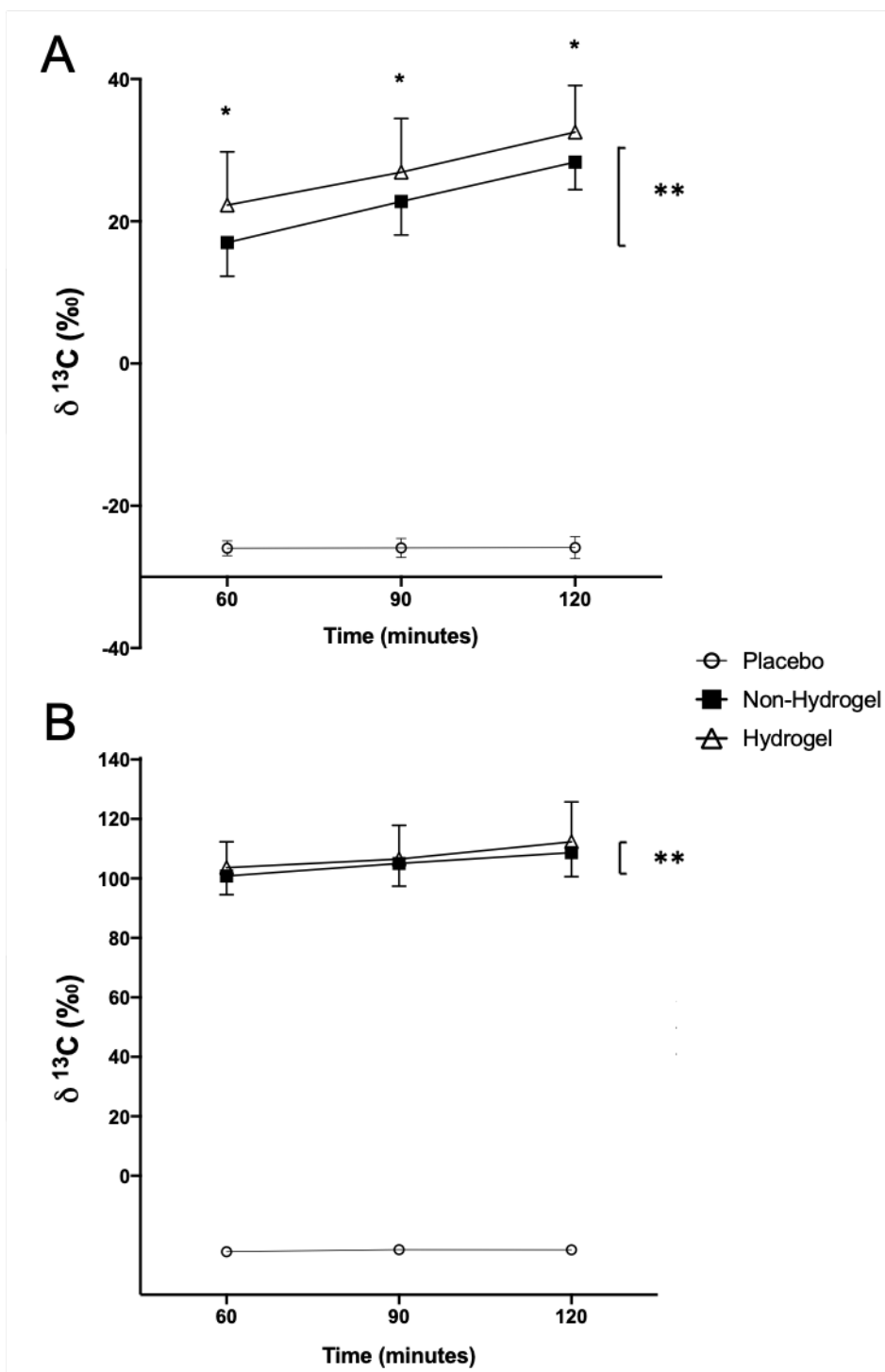751   **Figure 2.**
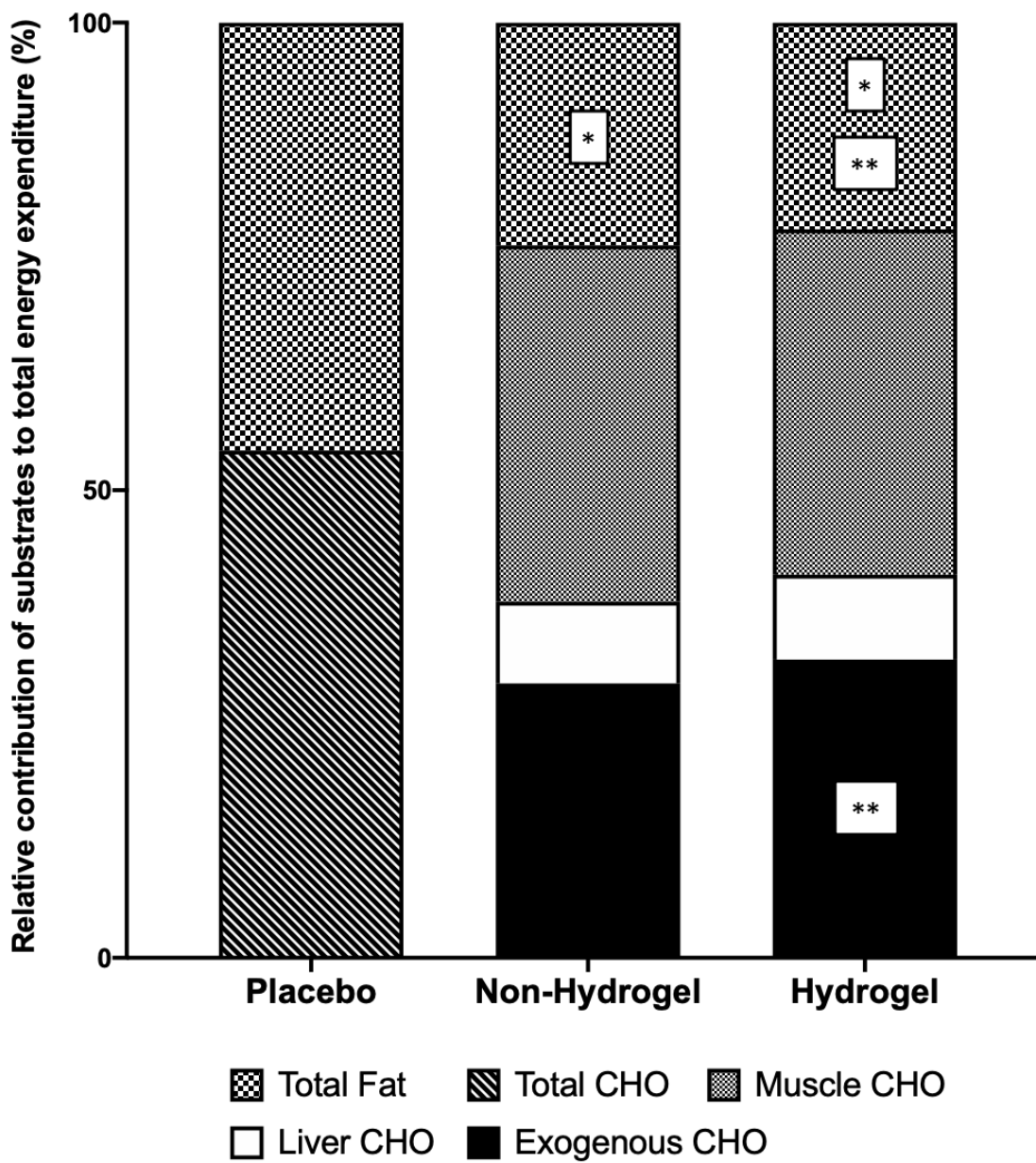
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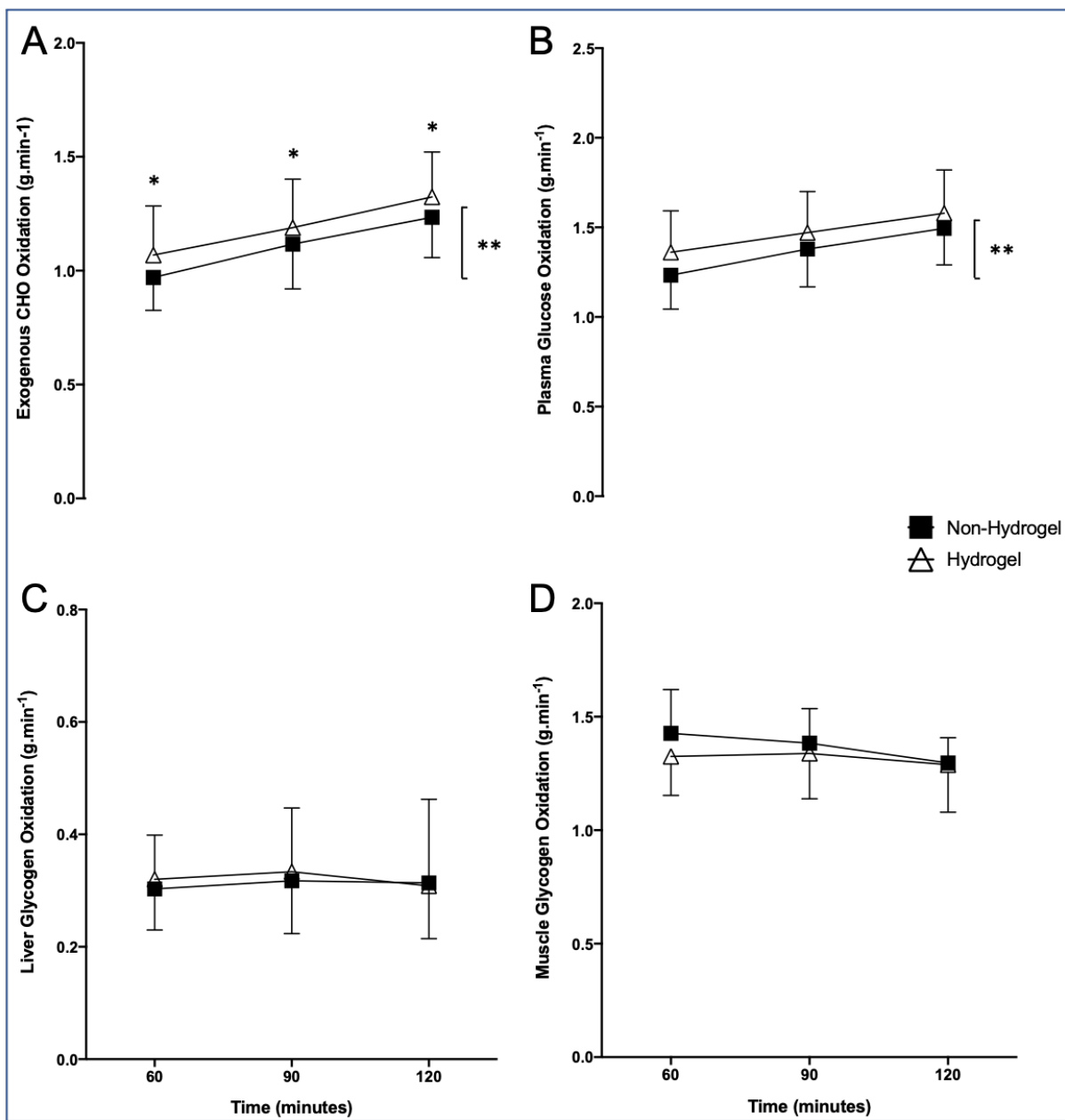
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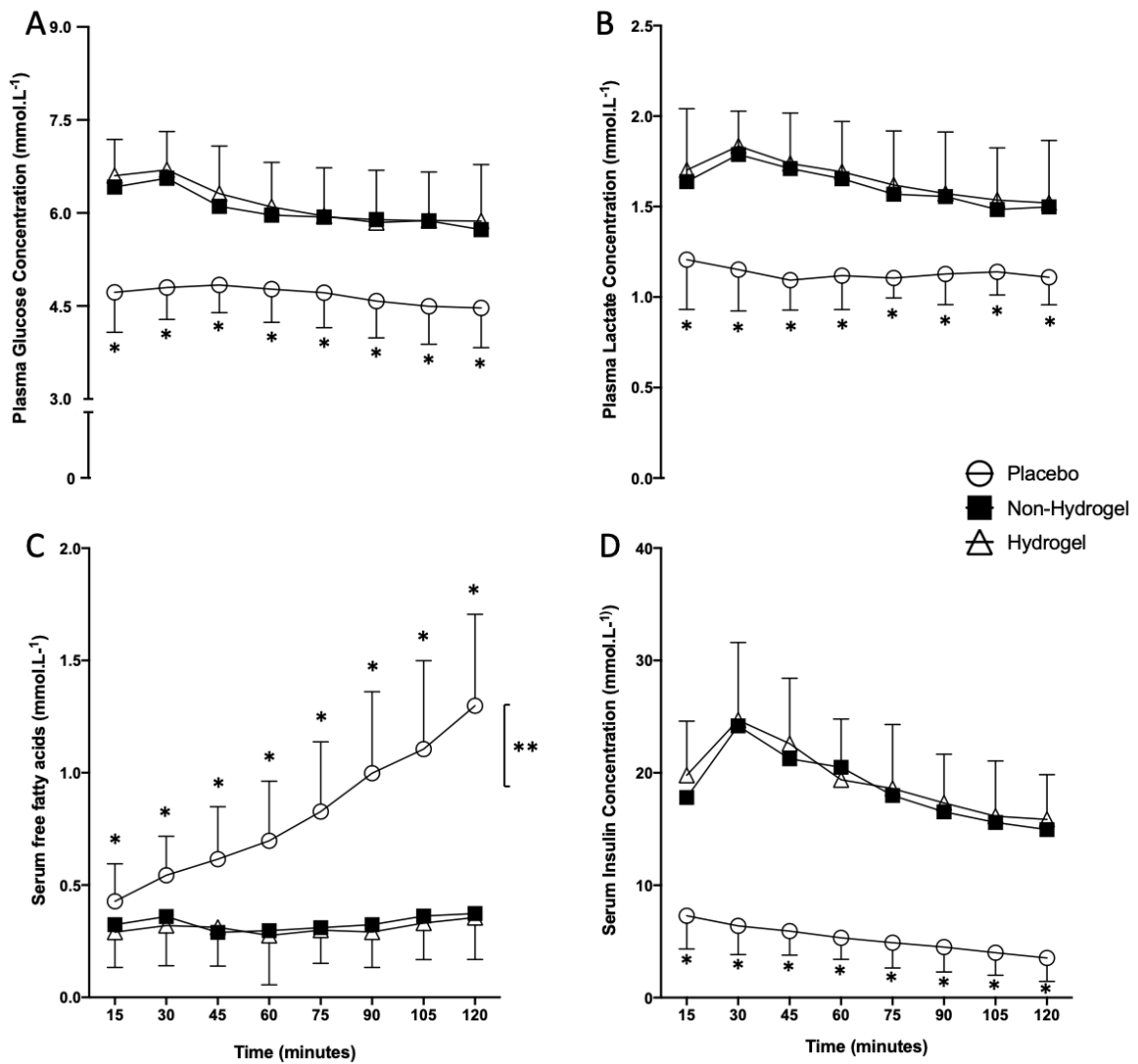

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775   **Figure 5.**

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Table 1. Comparisons of oxygen uptake, carbon dioxide production, total carbohydrate oxidation, total fat oxidation and heart rate over the first and second 60 minutes of the 120 minute steady state run.

	60-min period	Condition		
		Placebo	Non-Hydrogel	Hydrogel
VO_2 ($L \cdot min^{-1}$)	First	$3.04 \pm 0.31^{*}$	2.95 ± 0.26	2.91 ± 0.25
	Second	$3.08 \pm 0.27^{*}$	2.94 ± 0.21	2.93 ± 0.21
VCO_2 ($L \cdot min^{-1}$)	First	2.69 ± 0.24	2.69 ± 0.22	2.67 ± 0.21
	Second	$2.64 \pm 0.20^{*}$	2.73 ± 0.22	2.73 ± 0.18
RER	First	$0.89 \pm 0.02^{*}$	0.91 ± 0.01	0.92 ± 0.01
	Second	$0.86 \pm 0.02^{*\dagger}$	0.93 ± 0.01	0.93 ± 0.01
CHO_{ox} (g)	First	$138.8 \pm 7.8^{*}$	153.9 ± 11.4	156.9 ± 9.5
	Second	$122.1 \pm 10.8^{*\dagger}$	$164.7 \pm 9.8^{\dagger}$	$167.3 \pm 8.3^{\dagger}$
Fat_{ox} (g)	First	$34.8 \pm 7.7^{*}$	26.1 ± 5.4	$23.6 \pm 4.8^{**}$
	Second	$43.0 \pm 8.2^{*\dagger}$	$21.6 \pm 3.3^{\dagger}$	$19.8 \pm 3.8^{**\dagger}$
HR ($b \cdot min^{-1}$)	First	152 ± 11	153 ± 12	152 ± 13
	Second	155 ± 12	154 ± 13	154 ± 13

VO_2: Oxygen consumption. VCO_2: Carbon dioxide production. RER: Respiratory Exchange Ratio. CHO_{ox}: CHO oxidation. Fat_{ox}: Fat oxidation. HR: Heart rate. All values are mean $\pm$ SD. * significantly different from hydrogel and non-hydrogel. ** significantly different from non-hydrogel. † significantly different to first 60 minute period.

796   **Table 2**. Comparison of carbohydrate oxidation from various sources between non-hydrogel

797   and hydrogel during the final 60 minutes of the 120-minute steady state run

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	Non-hydrogel (g)	Hydrogel (g)	*p-value*
Exogenous CHO oxidation	63.4 ± 8.1	68.6 ± 10.8	$p = 0.003$
Endogenous CHO oxidation	101.2 ± 6.5	98.9 ± 9.1	$p = 0.58$
Plasma glucose oxidation	82.3 ± 11.7	88.1 ± 13.1	$p = 0.001$
Glucose oxidation from liver	18.8 ± 4.6	19.5 ± 6.5	$p = 0.83$
Muscle glycogen oxidation	82.4 ± 7.5	79.4 ± 10.8	$p = 0.26$

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Table 3. Comparison of GI symptoms between placebo, non-hydrogel and hydrogel conditions

	Symptoms	Placebo			Non-Hydrogel			Hydrogel		
		Score	Range	%	Score	Range	%	Score	Range	%
Upper-gastrointestinal symptoms	Belching	2 ± 1	1 - 5	9	4 ± 1**	2 - 7	64	3 ± 1**	1 - 6	36
	Stomach Burn	1 ± 1	1 - 5	9	4 ± 1*	1 - 7	46	2 ± 1**	1 - 5	27
	Urge to Vomit	1 ± 0	1 - 4	0	2 ± 1**	1 - 5	36	2 ± 1**	1 - 5	27
	Bloatedness	3 ± 1	1 - 5	9	4 ± 1*	1 - 7	55	3 ± 1	1 - 5	36
	Nausea	3 ± 1	1 - 5	18	3 ± 1*	1 - 6	36	2 ± 1	1 - 5	27
Mean Score		**2 ± 1**			**4 ± 1***			**3 ± 1****		
Lower-gastrointestinal symptoms	Stomach Problems	2 ± 1	1 - 5	9	4 ± 1*	1 - 7	64	3 ± 1**	1 - 5	27
	Flatulence	1 ± 1	1 - 4	0	4 ± 1*	1 - 7	55	2 ± 1**	1 - 5	27
	Urge to Defecate	2 ± 1	1 - 5	9	3 ± 1**	1 - 6	36	2 ± 1	1 - 6	36
	Side Ache (Left)	1 ± 1	1 - 3	0	2 ± 1**	1 - 5	18	1 ± 0	1 - 5	18
	Side Ache (Right)	1 ± 1	1 - 4	0	4 ± 1*	1 - 6	64	1 ± 1	1 - 5	27
	Stomach Cramps	2 ± 1	1 - 4	0	4 ± 1*	1 - 7	64	1 ± 1	1 - 5	27
Mean Score		**1 ± 1**			**4 ± 1***			**2 ± 1****		
Systemic	Dizziness	2 ± 1	1 - 5	9	2 ± 1	1 - 5	34	2 ± 1	1 - 6	27
	Headache	2 ± 1	1 - 4	0	3 ± 1*	1 - 6	27	2 ± 1	1 - 5	18
	Urge to Urinate	3 ± 2	1 - 8	55	3 ± 2	1 - 7	55	3 ± 2	1 - 5	36
Mean Score		**2 ± 1**			**3 ± 1***			**2 ± 1**		
Total Mean Score		**2 ± 1**			**3 ± 1***			**2 ± 1**		

Score, mean ± SD; Range. min minimum and maximum score, %, percentage of participants who reported scores of ≥5. * significantly different from placebo and hydrogel. ** significantly different from placebo.