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Varied Fasting Periods Moderate Intestinal Glucose Absorption in Male Wistar Rats

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Abstract: Fasting is abstaining from food and/or drinking water for a while, observed majorly for medical or spiritual purposes. The effect of varied fasting durations on intestinal glucose uptake was investigated in this study. Forty male Wistar rats, weighing between 100 and 120 g were used. They were randomly assigned into four groups (n=10 per group), Group 1: control (no fast), groups 2, 3, and 4, were fasted for 24, 48, and 72 hours respectively. Five rats each from the groups were used for the *in vivo* and *ex vivo* studies. The fasting blood glucose (FBG) level, intestinal glucose uptake (*in vivo and ex vivo* methods), and intestinal luminal electrolytes for sodium and potassium were determined. The FBG and the intestinal glucose uptake of the rats were determined by the glucose oxidase method. Data were represented as Mean±SEM and analysis was done by one-way ANOVA using GraphPad Prism version 7.0. Results were considered significant at p<0.05. The FBG (mg/dl) in the 24H (62.00±5.17), 48H (51.80±4.48) and 72H (75.00±8.32) groups decreased significantly compared with the control (103.80±4.35). Intestinal glucose concentration (mg/dl/g tissue) *in vivo* method decrease significantly in the 48H (11.21±0.38) and 72H (10.39±0.77) groups compared to the control (15.02±0.51) respectively in the jejunum and implied increased glucose absorption. Luminal glucose concentration (mg/dl/g tissue) of the fasted 48H group (27.92±0.66) increased significantly compared to control (13.86±0.75), 24H (17.84±0.65), and 72H (13.56±1.00). The ileum luminal glucose concentration (mg/dl/g tissue) decreased significantly in 48H (11.11±0.63) and 72H (11.02±0.56) groups compared to the control (13.86±0.59). In the assessment of varied fasting durations *in vitro*, intestinal glucose absorption increased while potassium concentration increased and sodium concentration decreased within the mucosa end. The results suggest that varied fasting periods increase intestinal glucose absorption. Conclusively, this study reported an overall increase in intestinal glucose absorption at both the jejunum and ileum ends after 48 and 72 h of continuous fasting.

Keywords: Fasting, Intestinal Glucose Absorption, Electrolyte, Rats

1. Introduction

Fasting generally is referred to as abstinence from food and or drinking water for some time [1]. Fasting is devoid of taking food or fluids containing calories but may allow water intake [2]. Fasting also means eating no food and drinking water for some time [3]. Fasting is done for medical, spiritual, or religious purposes and has been reported to be valuable and beneficial. The benefits of fasting include long life and healthy living [4-7]. The blood glucose level has an established relationship with fasting and that has warranted the use of fasting glucose values as an important screening for glucose derangements [8]. An intensified gluconeogenesis of over 30% of the internally generated glucose after 72 h of this is known to be induced by fasting [9].

The mechanisms of glucose absorption involve release to the systemic circulation through the absorption of carbohydrate-digested products in the gastrointestinal system through the small intestine. Studies from some investigators have provided pieces of evidence that the intestine plays a demanding role in glucose homeostasis [10-12]. Another important strategy of glucose homeostasis includes the reduction of postprandial hyperglycemia by reducing the amount of glucose available for absorption at the intestinal level [13]. It is worth mentioning that the small intestine not only absorbs glucose but also makes good use of it.

Varieties of vital conditions help in the regulation of glucose transporters in the intestine. Thereby inducing the effectiveness

of the essential glucose transporters and the quantity of the absorbed glucose molecules into the plasma. The molecular interaction with the transporters and their ensuing glucose absorption activities is well branded. Rapid up-regulation of SGLT1 has been observed in the jejunal enterocytes at the apical membrane as described previously [14].

Individuals partake in periodic fasts for health, religious or cultural reasons for hours and days in different circumstances. It is known that fasting affects glucose absorption but the effect of change in periods of fasting on intestinal glucose absorption is not known. This study investigated the responsibility of the intestine in the variability of fasting periods on intestinal glucose transports in the *in vivo* and *ex vivo* study in male Wistar rats.

2. Materials and Methods

2.1. Materials, Chemicals and Solutions

Weighing scales, Plastic Cages, Feeding troughs, Drinkers, Distilled water, Glucometer and glucometer strips, Rats feeds, Filter paper, Syringes (1ml, 5ml, 10ml), Latex Hand Gloves, Dissecting set, Dissecting board, Stopwatch, Methylated spirit, Disinfectants, Meter rule, Krebs solution, Ringer solutions, Measuring cylinder, Cotton wool, Glass Rod, Thread, Wood shavings, Absorptive papers. All chemicals used were of analytical grades.

2.2. Animal Groupings, Husbandry, and Ethics

Forty (40) male Wistar rats, weighing between 100-120g were used. The animals were purchased from the Central Animal House, University of Ibadan, Ibadan where the experiment took place. They were kept in plastic cages bedded with wood shavings and with a wire mesh cover maintained at room temperature. They were acclimatized for two weeks to allow for familiarity with their new environment. The rats had free access to food and water *ad libitum* before the start of the different experimental procedures. The animals were randomly selected for the *in vivo* and *ex vivo* experiment and grouped into 4, with 10 rats per group (5 rats per the mode of study eg., *in vivo* or *ex-vivo*). Group 1, is Control (not fasted), group 2 is the 24 hours fasted group (24H), group 3 is the 48 hours fasted group (48H) and group 4 is the 72 hours fasted group (72H).

The study was conducted in a humane condition and was approved by the University of Ibadan Animal Care and Use Research Ethics Committee with the assigned number: UI-ACUREC/17/0057.

2.3. Determination of Fasting Blood Glucose Level

Food was withdrawn from the animals according to their respective fasting hours (except for the control group that did not fast before the experiment commenced. Groups 2, 3, and 4 fasted for 24, 48, and 72 h respectively. Initial blood samples were taken from the rat tails before the experiment proceeded. The blood glucose concentration was instantly determined through the glucose oxidase method.

2.4. Determination of Intestinal Glucose Absorption Using the in vivo Experiment

To evaluate the effect of varied fasting periods on intestinal glucose absorption, the method of absorption study described by Odukanmi *et al.*, [15] was adopted. The experimental animals had free access to food and water and were maintained at room temperature. Feed was taken from the rats at 24H, 48H, and 72H according to their groups respectively (except for the control group) before the treatment. The animals were treated as described above after which they were anesthetized with ketamine 75mg/kg. Through a midline laparotomy, the intestinal segments about 15 cm long were identified for consistency. The proximal segment was just distal to the ligament of Treitz while the distal end was 3 cm proximal to the ileocecal junction. The segment was opened at both ends and infused with 4 mL cold Krebs buffer (119 mM NaCl, 4,7 mM KCl, 2,5 mM $CaCl_2$, 1,2 mM $MgSO_4$, 1,2 mM KH_2PO_4, 25 mM $NaHCO_3$, Glucose 4g, pH 7.4) [16] with a micropipette. The core body temperature was sustained with a positioned clamp light. The fasting glucose concentration was noted and intraluminal fluid glucose content was subsequently determined from each rat after 60 minutes. The glucose concentrations were analyzed through a quantitative assay (Glucose Oxidase) method using the Fine Test® glucometer and its strips. Each study lasted one hour, at the end of which the two intestinal (ileum and jejunum) segments were measured. The arteriovenous component of the test was as previously described [15].

2.5. Determination of Intestinal Glucose Absorption Using the ex vivo Experiment

The everted sac method was used in this case as previously described [10]. After the allotted duration of fasting according to the experimental groups described above. Tissue preparation and mounting followed after the rats were anesthetized with an overdose of ketamine and the intestines were isolated from 3cm away from the ileocecal junction distally and at the ligament of Treitz proximally. The excised intestine was quickly rinsed and placed in ice-cold Ringer solution (glucose-free). The entire length was divided into 3 equal segments. They were subsequently everted into sacs with a glass rod from where each piece was slipped over the tip of a glass rod. The sleeve of tissues was ligated on one end and 200µL of Ringer solution (Glucose free) was gently released into the serosa end of the sac and then ligated. The sac was then incubated in 5ml Ringer solution (with 10mM Glucose), appropriately gassed at 37°C for 1 hour.

At this point, remaining isolated intestinal tissues were left in ice-cold gassed Ringer solution while the solution is replaced at 15 min intervals until the tissues were ready for use. On the expiration of the incubation period of 1 hour, the glucose concentration of the fluid in the serosa and that in the test tube (representing mucosa fluid) were determined through a quantitative assay (Glucose Oxidase) method using the Fine Test® glucometer and its strips. The intestines were dried on a filter paper and weighed to determine the size of tissue used per experiment.

2.6. Statistical Analysis

The outcome was expressed as Mean ± SEM and a one-way ANOVA with Newman-Keuls comparison *post hoc* test was adopted. The results were analyzed on GraphPad Prism (San Diego, CA), version 5.0 Software for Windows. Compared values with p<0.05 were considered significant.

3. Results

3.1. Effects of Varied Fasting Durations on Fasting Blood Glucose Level

The fasting blood glucose level decreased significantly in the fasted 24H (62.00 ± 5.17 mg/dl), 48H (51.80 ± 4.48 mg/dl), and 72H (75.00 ± 8.32 mg/dl) groups compared to the control (103.80 ± 4.35 mg/dl) group (Figure 1).
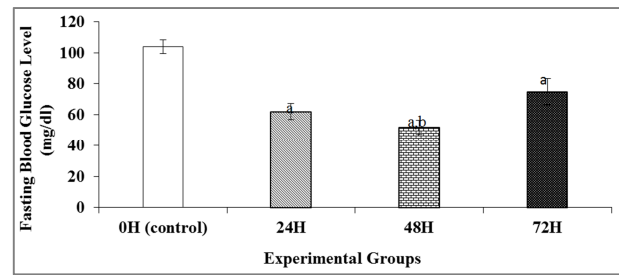

Figure 1. *Effect of varied fasting durations on fasting blood glucose level.*

a = significant decrease in glucose concentration at p<0.05 compared to the control group.
b= significant decrease in glucose concentration at p<0.05 compared to the 72H groups.

Table 1. *Effects of varied fasting durations on Arterio-Venous glucose differences.*

	Control	24H	48H	72H
Abdominal Aorta (mg/dl)	121.40 ± 3.02	104.00 ± 5.75	92.40 ± 6.65	144.50 ± 4.86*
Mesenteric Vein (mg/dl)	111.00 ± 1.44	91.20 ± 3.11[+]	94.60 ± 1.28[+]	146.25 ± 5.07*
Arterio-Venous Difference (mg/dl)	10.40 ± 8.00	12.80 ± 6.61	-2.2 ± 6.89*	-1.75± 24.81*

n= 5 animals per group.
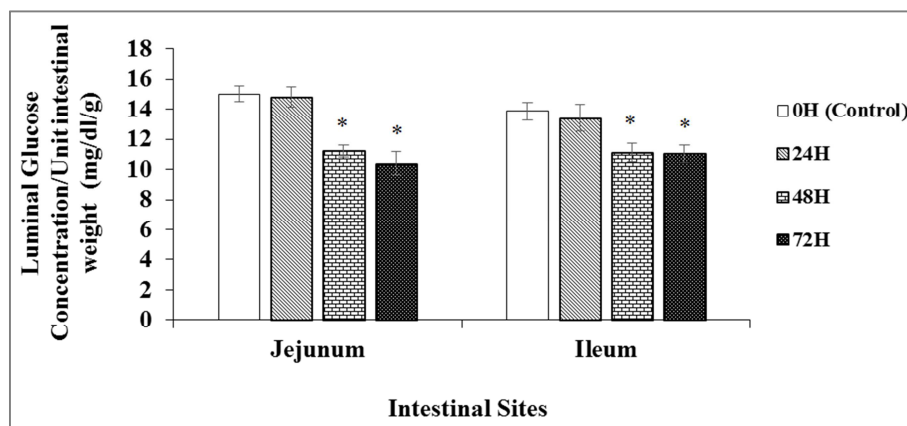*= significant increase in glucose concentration at p<0.05 compared to other groups.
+= significant decrease in glucose concentration at p<0.05 compared to the control groups.

3.2. Effects of Varied Fasting Durations on Arterio-venous Glucose Differences

Table 1 describes the observed result from the abdominal aorta and mesenteric vein. There was a significant increase in glucose concentration in the fasted arterial and venous glucose concentration 72H (144.50 ± 4.86; 146.25 ± 5.07 mg/dl) group compared to the control (121.40 ± 3.02; 111.00 ± 1.44 mg/dl), 24H (104.00 ± 5.75; 91.20 ± 3.11 mg/dl) and 48H (92.40 ± 6.65 and 94.60 ± 1.28 mg/dl) groups respectively.

3.3. Effects of Varied Fasting Durations on Intestinal (Jejunum and Ileum) Glucose Uptake Using the in vivo Method

In the jejunum, luminal glucose concentration (mg/dl/g tissue) decreased significantly in the 48H (11.21 ± 0.38) and 72H (10.39 ± 0.77) groups compared to the control (15.02 ± 0.51) signifying increasing glucose uptake after the periods of fasting in the jejunum, Figure 2. In a similar trend also in figure 2, the ileum luminal glucose concentration (mg/dl/g tissue) decreased significantly in 48H (11.11 ± 0.63) and 72H (11.02 ± 0.56) groups compared to control (13.86 ± 0.59).
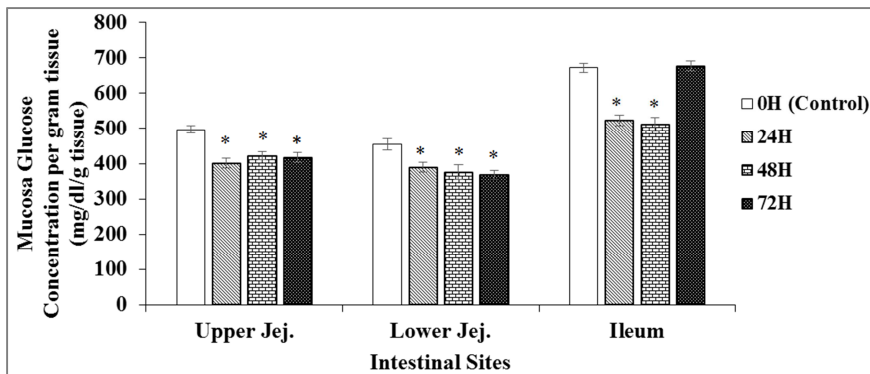

* = significant decrease in luminal glucose concentration at p<0.05 compared to the control

Figure 2. *Effect of varied fasting durations on luminal glucose absorption.*

3.4. Effects of Varied Fasting Durations on Intestinal Glucose Uptake in the Mucosa Using the ex vivo Method

The upper jejunum, lower jejunum and ileum glucose concentration (mg/dl/g tissue) decreased significantly in the 24H (401.31 ± 14.12; 389.4 ± 14.9; 523.23 ± 16.2), 48H (422.26 ± 12.1; 375.52 ± 20.2; 511.22 ± 19.2) groups compared to the control (497.30 ± 14.38; 454.23 ± 16.2; 675.90 ± 6.04), respectively. In the 72H group, there was a significant decrease at the jejunum sites and no significant change at the ileum compared to the control. (Figure 3).

* = significant decrease in glucose concentration at p<0.05 compared to the control group

Figure 3. Effects of varied fasting durations on glucose uptake in the mucosa end of the intestine using the ex vivo method.

3.5. Effects of Varied Fasting Durations on Intestinal Uptake in the Serosa Using the ex vivo Method

Figure 4 describes the observed glucose concentration (mg/dl/g tissue) results in the serosa fluid of the upper jejunum, the lower jejunum and the ileum, increased significantly in the 72H (392.90 ± 5.77; 392.90 ± 5.77 and 399.70 ± 5.77) compared to the control (134.30 ± 5.99; 145.60 ± 7.78; 208.60 ± 18.28), 24H (275.10 ± 17.43; 275.10 ± 17.43; 281.7 ± 8.82) and 48H (210.00 ± 12.34; 210.00 ± 12.38; 159.6 ± 5.72) groups respectively. The upper and lower jejunum, serosa glucose concentration (mg/dl/g tissue) increased significantly in the 24H (275.10 ± 17.43 and 275.10 ± 17.43), 48H (210.00 ± 12.34; 210.00 ± 12.34) and 72H (392.90 ± 5.77; 392.90 ± 5.77) groups compared to control (134.30 ± 5.99; 145.60 ± 7.78) groups respectively (Figure 4).
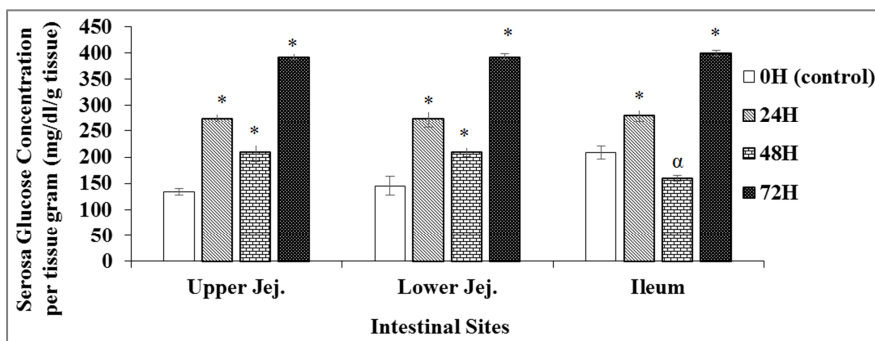

Figure 4. Effects of varied fasting durations on glucose uptake in the serosa of the intestine in the ex vivo method.

*= significant increase in glucose concentration at p<0.05 compared to the control; α= significant decrease in glucose concentration at p<0.05 compared to the control ileum.

3.6. Effects of Varied Fasting Durations on Mucosa Sodium-Ion Flux

The luminal sodium ion concentration (ppm) in the lower jejunum increased significantly in the 72H (502.30 ± 6.18) group, compared to the control (468.00 ± 6.11), 24H (375.70 ± 19.48), and 48H (356.70 ± 1.04) groups. In the ileum, there was a significant decrease in the sodium ion concentration in 24H (400.30 ± 4.21), 48H (450.70 ± 5.67), and 72H (431.00 ± 12.34) groups compared to the control (501.00 ± 4.95 ppm) group, Figure 5. The upper jejunum and ileum, luminal sodium ion concentration decreased significantly in the 24H (359.00 ± 11.39; 400.30 ± 4.21) groups compared to control (470.00 ± 21.33; 501.00 ± 4.95), 48H (478.30 ± 12.04; 450.70 ± 5.67) and 72H (490.70 ± 21.69; 431.00 ± 12.34) groups respectively (Figure 5).
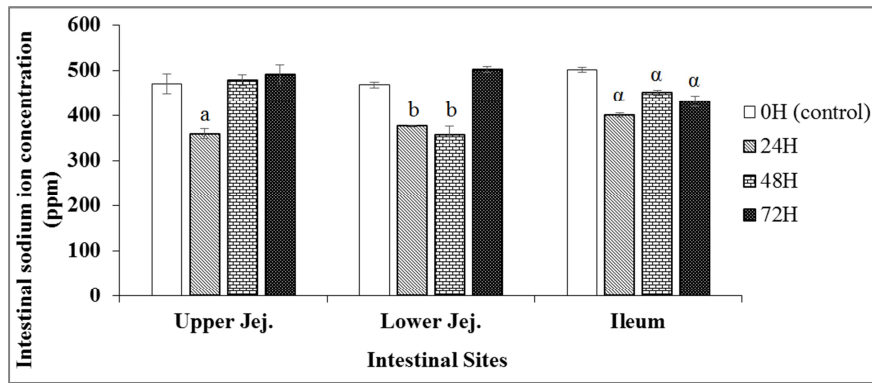
Figure 5. *Effects of varied fasting durations on intestinal sodium ion flux during ex vivo study.*

a= significant decrease in sodium concentration at p<0.05 compared to the control group in the upper jejunum; b= significant decrease in sodium concentration at p<0.05 compared to the control group in the lower jejunum; α= significant decrease in sodium concentration at p<0.05 compared to the control group in the ileum.

3.7. Effects of Varied Fasting Durations on Mucosa Potassium Ion Flux

In figure 6, there was a significant increase in potassium ion concentration (ppm) at the upper jejunum, in 48H (861.10 ± 8.25) group compared to the 72H group (740.80 ± 26.13). In the lower jejunum, intestinal potassium ion concentration increased significantly in the 24H group (878.70 ± 17.03) compared to the control (795.50 ± 13.80), 48H (776.90 ± 5.77), and the 72H (770.90 ± 28.80) groups. The mucosa potassium concentration in the ileum increased significantly in the 24H group (908.40 ± 4.12) compared to the control (828.90 ± 3.41) and 72H (756.60 ± 23.07) groups.
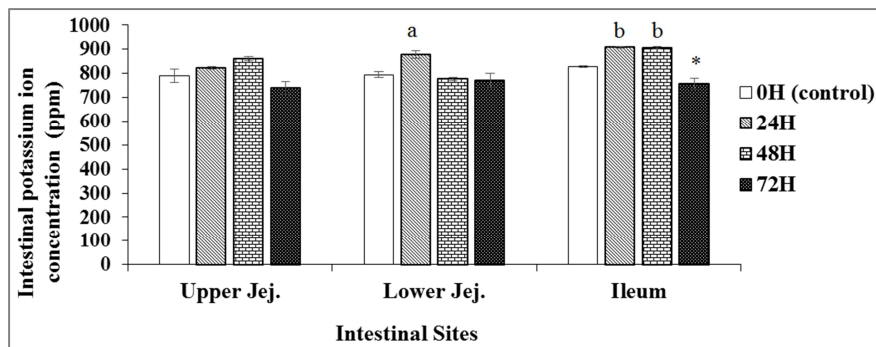

Figure 6. *Effects of varied fasting duration on mucosa potassium ion flux during ex vivo study.*

a= significant increase in potassium ion concentration at p<0.05 compared to the control lower jejunum group; *= significant decrease in potassium ion concentration at p<0.05 compared to the control ileum group; b= significant increase in potassium ion concentration at p<0.05 compared to the control lower jejunum group.

4. Discussion

This study was carried out to evaluate the effects of varied fasting durations on intestinal glucose absorption in male Wistar rats using the *in vivo* and *ex vivo* methods. Fasting is known to affect glucose absorption, and research has shown that fasting could be beneficial to the well-being of man and can improve glucose metabolism [17–19]. It is clear the role of the intestine in glucose homeostasis, however, the role of the intestine in glucose absorption in periodic fasting is not apparent and was investigated in this study.

The decreased fasting glucose level reported in this study during fasting periods buttresses the fact that fasting durations or long-term fasting could cause depletion of the hepatic glycogen stored initially [20], and may tend to increase the rate of gluconeogenesis subsequently. The increased glucose levels in the mesenteric vein in the 48H and 72H groups reveal an active superior mesenteric vein function in the fasting state in a prolonged fast. In a similar study with critically ill patients, luminal glucose uptake was in contrast decreased through the superior mesenteric artery following infusion of normal saline and glucose [21]. The reduction might be because enteric feeding tends to waste nutrients but in a fasting state, there is the need for rapid utilization of glucose to replenish energy. Whether or not this was done in isolation of the endocrine input of insulin was not investigated in this study.

The duration of the experiment which was 1 hour could not have revealed the mobilization of glycogen from the liver,

which could have contributed to the change in the systemic glucose concentration following intestinal infusion with Krebs buffer. Transport of luminal glucose could be through 2-steps pathways, either through the apical sodium-dependent glucose transporter (SGLT 1) into the intracellular and the GLUT 2 translocation into the interstitium [22]. The paracellular possibilities of glucose absorption are still being investigated even though it was reported to have a limited impact on glucose absorption by the intestine [23]. Also, the volume of the epithelial cells was not assessed for the amount of glucose that was retained within it during the interaction.

In support of the initial findings from the *in vivo* component of the study, the *ex vivo* part observed a similar response which suggested an increase in glucose transport across the everted sac with a significant transport in the serosa end of the test tube while the mucosa end decreased in both 48H and 72H groups. This mimick a general increase in absorption in the 48H and 72H fasted states in the *in vivo* study of both the jejunum and ileum. The ileum was opined to have the capacity to transport glucose under the stress of fasting in rats as this is a result of varying the fasting periods in the rats. The increased absorption of glucose is in line with the observation of Shobha et al., [24] in their study after the periods of fasting. With similar findings in both the *in vivo* and *ex vivo* components, the endocrine influence in the increased glucose absorption should be played down. However, the possibility of more efficient absorptive villi surfaces and improved expression of glucose transporters could not be ruled out in this study.

The role of the absorptive surfaces and the role played by the glucose transporters whose capability is influenced by the availability of appropriate electrolytes were embraced by analyzing the sodium and electrolytes within the mucosa end after the *ex vivo* study. This study observed a decreased sodium in the fasting states at the jejunum and ileum, suggesting an improved transport of sodium into the intracellular pathway. The converse was observed with the counterpart potassium ion with increased mucosa value. Thereby indicative of an improved role of SGLT1 across the mucosa end in the 48H and 72H fasted groups in both intestinal sites [25]. The SGLT1 is a mediator of glucose movement across the intestinal lumen and the renal proximal tubule [26]. Impairment of the SGLT1 usually results in malabsorption of glucose and passage of watery stool [27]. However, in this study, the gene expression study to ascertain the role played by this SGLT1 gene was not determined.

5. Conclusion

This study observed an overall increase in intestinal glucose absorption at both the jejunum and ileum ends after 48 and 72 h continuous fasting. The mechanism by which varied fasting durations regulate intestinal absorption was not clarified. Though the electrolyte differences determined in response to intestinal glucose absorption could be a pointer to further evaluating the SGLT 1 gene expression subsequently. It is evident from our findings that the body system requires rapid glucose absorption through the intestine during continuous prolonged fasting durations.

Conflict of Interest

The author declares no conflict of interest.

References

[1]   Patterson RE, Laughlin GA, LaCroix AZ, et al (2015) Intermittent fasting and human metabolic health. J Acad Nutr Diet 115: 1203.

[2]   Mattson MP, Wan R (2005) Beneficial effects of intermittent fasting and caloric restriction on the cardiovascular and cerebrovascular systems. J Nutr Biochem 16: 129–137.

[3]   Obembe AO, Okon VE, Ofutet EO, Ayitu RA (2015) Effect of Fasting on Intestinal Motility and Transit in Albino Wistar Rats. Trends Med Res 10: 63–68.

[4]   Mitchell SJ, Bernier M, Mattison JA, Aon MA, Kaiser TA, Anson RM, Ikeno Y, Anderson RM, Ingram DK, de Cabo R (2019) Daily Fasting Improves Health and Survival in Male Mice Independent of Diet Composition and Calories. Cell Metab 29: 221-228.e3.

[5]   Wei S, Han R, Zhao J, Wang S, Huang M, Wang Y, Chen Y (2018) Intermittent administration of a fasting-mimicking diet intervenes in diabetes progression, restores β cells and reconstructs gut microbiota in mice. Nutr Metab. https://doi.org/10.1186/S12986-018-0318-3

[6]   Buono R, Longo VD (2018) Starvation, Stress Resistance, and Cancer. Trends Endocrinol Metab 29: 271–280.

[7]   Mattson MP, Longo VD, Harvie M (2017) Impact of intermittent fasting on health and disease processes. Ageing Res Rev 39: 46–58.

[8]   Haddadinezhad S, Ghazaleh N (2010) Relation of fasting and postprandial and plasma glucose with hemoglobinA1c in diabetics. Int J Diabetes Dev Ctries 30: 8.

[9]   Mithieux G, Rajas F, Gautier-Stein A (2004) A Novel Role for Glucose 6-Phosphatase in the Small Intestine in the Control of Glucose Homeostasis. J Biol Chem 279: 44231–44234.

[10] Odukanmi OA, Olusegun QB, Olaleye SB (2019) Intestinal Handling of Glucose in Buccholzia coriacea Treated Male Wistar Rats. J Biosci Med 7: 87–98.

[11] Oyebola DD, Taiwo EO, Idolor GO, Alada AR, Owoeye O, Isehunwa GO (2011) Effect of adrenaline on glucose uptake in the rabbit small intestine. Afr J Med Med Sci 40: 225–233.

[12] Alada AR, Oyebola DD (1996) Evidence that the gastrointestinal tract is involved in glucose homeostasis. Afr J Med Med Sci 25: 243–249.

[13] Inzucchi SE (2002) Oral antihyperglycemic therapy for type 2 diabetes: scientific review. JAMA 287: 360–372.

[14] Kim I-Y, Schutzler SE, Schrader A, Spencer H, Azhar G, Deutz NEP, Wolfe RR (2015) Acute ingestion of citrulline stimulates nitric oxide synthesis in young and older subjects. Am J Physiol - Endocrinol Metab ajpendo.00339.2015.

[15] Odukanmi OA, Oluwole FS and Olaleye SB (2014) Effects of Kolaviron (A Flavonoid Fraction from Garcinia Kola) on Rat Intestinal Glucose Absorption and Alpha-Amylase Amylase Inhibitory Activities | Semantic Scholar. Arch Basic Appl Med 2: 161–167.

[16] Röder P V., Geillinger KE, Zietek TS, Thorens B, Koepsell H, Daniel H (2014) The Role of SGLT1 and GLUT2 in Intestinal Glucose Transport and Sensing. PLoS One 9: e89977.

[17] Harvie M, Wright C, Pegington M, et al (2013) The effect of intermittent energy and carbohydrate restriction v. daily energy restriction on weight loss and metabolic disease risk markers in overweight women. Br J Nutr 110: 1534–1547.

[18] Harvie MN, Pegington M, Mattson MP, et al (2011) The effects of intermittent or continuous energy restriction on weight loss and metabolic disease risk markers: a randomized trial in young overweight women. Int J Obes (Lond) 35: 714–727.

[19] Arguin H, Dionne IJ, Sénéchal M, Bouchard DR, Carpentier AC, Ardilouze JL, Tremblay A, Leblanc C, Brochu M (2012) Short- and long-term effects of continuous versus intermittent restrictive diet approaches on body composition and the metabolic profile in overweight and obese postmenopausal women: a pilot study. Menopause 19: 870–876.

[20] Shrayyef MZ, Gerich JE (2010) Normal Glucose Homeostasis. Princ Diabetes Mellit 19–35.

[21] Sim JA, Horowitz M, Summers MJ, et al (2013) Mesenteric blood flow, glucose absorption and blood pressure responses to small intestinal glucose in critically ill patients older than 65 years. Intensive Care Med 39: 258–266.

[22] Stümpel F, Burcelin R, Jungermann K, Thorens B (2001) Normal kinetics of intestinal glucose absorption in the absence of GLUT2: Evidence for a transport pathway requiring glucose phosphorylation and transfer into the endoplasmic reticulum. Proc Natl Acad Sci U S A 98: 11330–11335.

[23] Lane JS, Whang EE, Rigberg DA, Hines OJ, Kwan D, Zinner MJ, McFadden DW, Diamond J, Ashley SW (1999) Paracellular glucose transport plays a minor role in the unanesthetized dog. Am J Physiol. https://doi.org/10.1152/AJPGI.1999.276.3.G789.

[24] Das S, Kumar R, Nagchoudhuri J (2001) Effect of Fasting on the Intestinal Absorption of D-Glucose and D-Xylose in Rats *in vivo*. Indian J Physiol Pharmacol 45: 451–456.

[25] Wood IS, Trayhurn P (2003) Glucose transporters (GLUT and SGLT): expanded families of sugar transport proteins. Br J Nutr 89: 3–9.

[26] Wright EM, Hirayama BA, Loo DF (2007) Active sugar transport in health and disease. J Intern Med 261: 32–43.

[27] Abdul-Ghani MA, Norton L, DeFronzo RA (2011) Role of Sodium-Glucose Cotransporter 2 (SGLT 2) Inhibitors in the Treatment of Type 2 Diabetes. Endocr Rev 32: 515–531.