Edited by Charles H Halsted and Barry Shane

Editor's note

As a result of discussion with the entire Editorial Board of .AJCN/, we have decided to initiate a new section, "Highlights from the Literature." The purpose of this section is to highlight articles from other journals that we believe should be of particular interest to readers of .AJCN/. These selected abstracts and comments should indicate the kind of work that could appear in .AJCN/. We have asked the members of the entire Editorial Board to submit two signed selections a year. We would, of course, be interested in receiving exciting contributions from our readership. Each selection should include the official abstract and a comment of no more than 500-1000 words. The plan is to publish 4-5 selections in every other issue of the Journal. Please send your submissions to the .AJCN/ post office box in Berkeley.

Norman Kretchmer
Editor-in-Chief

Tumor necrosis factor stimulates amino acid transport in plasma membrane vesicles from rat liver. .AJ

Pacitti, Y Inoue, WW Souba

ABSTRACT Severe infection is characterized by a translocation of amino acids from the periphery to the liver, an event that is mediated in part by cytokines such as tumor necrosis factor-alpha (TNF). We investigated the activities of Na(+)I-dependent transport systems A, ASC, and N in hepatic plasma membrane vesicles (HPMVs) prepared from rats treated with TNF in vivo. TNF did not alter sodium uptake but resulted in time- and dose-dependent fivefold and 50% maximal increases in system A and system N activity, respectively, in HPMVs secondary to an increase in the transport Vmax. Maximal increases in transport were observed 4 h after exposure to TNF and had returned to basal levels within 24 h. Similarly, system ASC activity was stimulated 80% in HPMVs from rats treated with TNF. Incubation of HPMVs from normal rats in vitro with TNF did not alter transport activity. Pretreatment of animals with the glucocorticoid receptor antagonist RU 38486 attenuated the TNF-induced enhancement in transport activity by 50%. The marked increase in Na(+)I-dependent amino acid transport activity by TNF is mediated in part by the glucocorticoid hormones and represents an important mechanism underlying the accelerated hepatic amino acid uptake that occurs during critical illness. J Clin Invest 1993;91: 474-83.

The acute biological and clinical response to infection and sepsis is mediated in part by cytokines, in particular the interleukins (IL-1, IL-6) and tumor necrosis factor (TNF) that are released from reticuloendothelial cells after translocation of intestinal endotoxin. For example, circulating concentrations of TNF correlate with the severity of illness in septic patients (1), whereas administration of endotoxin to healthy volunteers increased both TNF and IL-6, energy expenditure, serum free fatty acids, and hyperglycemia, with reduced serum amino acids (2). Although altered substrate concentrations and metabolism contribute to illness and mortality in sepsis, the cytokine response may be essential to adaptation by promoting hepatic gluconeogenesis from muscle-derived amino acids, in particular alanine and glutamine.

The present experiments provide a rationale for the gluconeogenic response to endotoxin by showing TNF stimulation of several amino acid-transport systems at the liver plasma membrane. In experiments that used hepatic plasma membrane vesicles (HPMV) from previously treated rats, the data suggest that TNF, in a time- and dose-related fashion, stimulates the hepatic uptake of three separate classes of amino acids. The time course of maximal effectiveness at 2-4 h was consistent with clinical observations on the sequence of endotoxemia. Attenuation of response by a glucocorticoid receptor antagonist suggests an intermediary regulation of amino acid uptake in HPMVs by glucocorticoid hormones, although the data did not include separate experiments with this agent. Because the results could not be reproduced by simple exposure of vesicles to TNF, whereas prior injection of the cytokine increased the Vmax but not K_m of vesicle uptake of several amino acids, the data suggest a primary effect of TNF on the cellular production of amino acid-membrane carriers. Others have shown effects of TNF on gluconeogenesis, including increased hepatic transaldolase and decreased concentrations and transcription of phosphoenolpyruvate carboxykinase (3, 4). The present experiments lend support to the concept that TNF plays a signal role in the gluconeogenic response to sepsis by promoting amino acid uptake at the hepatic plasma membrane. Whether the amino acid uptake effect is primary to TNF or secondary to another cytokine or metabolic event and is on transcription, cellular processing, or membrane modulation of several amino acid carrier proteins are parts of the puzzle to be worked out in awaited future experiments. Others have shown that the cytokine responsiveness of human peripheral blood mononuclear cells can be downregulated by prior dietary fish-oil supplementation together with

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altered membrane phospholipid fatty acids (5). Taken together, these studies point to a complex nutritional regulation of the ability to respond to acute illness.

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References


β-Carotene has recently gained considerable attention; apart from its provitamin A activity, it has been suggested, on the basis of epidemiologic evidence, to act in cancer prevention, in stimulation of the immune system, in suppression of atherosclerosis, and in prevention of cataract. It could, of course, exert these actions as a result of conversion to retinol or retinoic acid. Compared with the two last-named compounds, β-carotene has the advantage in humans in that it is nontoxic and responds to increasing dietary intake by increased blood concentrations. Alternatively, it could also act as an antioxidant.

The authors of the present paper have previously shown that carotenoids, even those like canthaxanthin that are not convertible to retinoids, can function as anticarcinogens at micromolar concentrations by suppressing the action of methylcholanthrene on mouse fibroblasts in culture. Closely correlated with this anticarcinogenic activity of carotenoids was the stimulation of the formation of gap junctions between cells, resulting in increased cell-cell communication. Carcinogenesis results in a decline in gap-junctional communication and it is thought that growth-inhibitory substances pass across gap junctions to inhibit carcinogenesis. It is known that tumor promoters uncouple gap-junctional communication. The gap junction between two cells consists of a channel made by a protein called connexin43 (Cx43).

The present paper shows that the expression of the Cx43 gene in the form of Cx43 mRNA and subsequently the Cx43 protein, is stimulated by a number of different carotenoids, both with or without provitamin A activity. The importance of this paper is in the demonstration, at least in cell culture systems, that carotenoids can elicit expression of a specific gene for a specific protein intimately involved in suppression of carcinogenesis, without conversion to retinoids, nor by an antioxidant effect. This was done by stimulating Cx43 mRNA and Cx43 protein expression not only by β-carotene, a provitamin A carotenoid, but also by canthaxanthin and lycopene, carotenoids that cannot be converted to retinol or retinoic acid. On the other hand the powerful antioxidant α-tocopherol was inactive. Preliminary results showed that even in human dermal fibroblasts carotenoids up-regulated Cx43 expression.

The importance to clinical nutrition lies in the fact that at last a plausible molecular mechanism for the frequently observed lowering of cancer risk by dietary carotenoids has been proposed. In practical terms, these are substances that are nontoxic and abundantly available in the human diet.

George Wolf

Carotenoids Up-Regulate Connexin43 Gene Expression Independent of Their Provitamin A or Antioxidant Properties. Li-Xin Zhang, Robert V. Cooney, and John S. Bertram

ABSTRACT Epidemiological evidence and studies in whole animals and cell culture have indicated that carotenoids have cancer chemopreventive action. In mouse C3H1OT1/2 cells, this activity is highly correlated with the ability of carotenoids to up-regulate gap junctional intercellular communication. Here, we report that in mouse cells, carotenoids increase the expression of connexin43, a gene that encodes a major gap junction protein. This effect appears unrelated to their provitamin A or antioxidant properties, since carotenoids with and without provitamin A activity increased levels of connexin43 mRNA and protein, whereas the antioxidants methyl-bixin and α-tocopherol were inactive. Moreover, the active carotenoid canthaxanthin did not induce the vitamin A-inducible gene retinoic acid receptor-β. Connexin43 is the first carotenoid-inducible gene described in mammals. By indicating an additional pathway through which carotenoids function, these data provide a mechanistic basis for cancer chemoprevention by carotenoids and may lead to a re-evaluation of carotenoid physiology. Cancer Res 1992;52:5707-12.

References

No Change in Glucose Tolerance and Substrate Oxidation After a High-Carbohydrate, Low-Fat Diet. Isabelle Leclerc, Isabelle Davignon, Denise Lopez, and Dominique R. Garrel

ABSTRACT Possible changes in glucose tolerance and substrate oxidation after a high-carbohydrate, low-fat diet were studied in seven healthy volunteers. Each subject consumed two experimental diets for 1 week after 1 week on a stabilization diet: diet no. 1 11% fat and 64% carbohydrates, and diet no. 2 30% fat and 45% carbohydrates. At the end of each experimental week, plasma levels of glucose, insulin, and free fatty acids were measured before and every 30 minutes for 6 hours after a 75-g oral glucose challenge. At the same time, energy expenditure and substrate oxidation were measured by indirect calorimetry. Plasma lipid and lipoprotein levels were measured at the end of one stabilization period and at the end of each diet. Plasma glucose concentrations and areas under the curve of glucose concentrations were identical after the two experimental periods; the means ± standard deviation for the values at 120 minutes were 6.4 ± 0.3 and 6.4 ± 0.6 mmol/L after diets no. 1 and 2, respectively, and areas under the curve were 1.853 ± 115 and 1.862 ± 211 mmol·min/L after diets no. 1 and 2, respectively. Similarly, plasma concentrations of insulin and free fatty acids after glucose ingestion were unaffected by the dietary changes. Energy expenditure increased after glucose administration, and this thermic effect of glucose was identical after the two experimental diets at 4.2% ± 1.4% and 3.9% ± 1.4% of ingested energy for diets no. 1 and 2, respectively. Substrate oxidation rates were also identical for both the fasted and post-glucose periods after the two diets. Total cholesterol, very-low-density lipoprotein (VLDL), and low-density lipoprotein (LDL) levels showed no significant change between the two diets, but plasma high-density lipoprotein (HDL) concentrations were lower after the high-carbohydrate diet at 1.02 ± 0.10 and 1.15 ± 0.10 mmol/L for diets no. 1 and 2, respectively (P < .05), and triglyceride concentrations were higher after each experimental diet than after the stabilization diet at 1.11 ± 0.04, 1.29 ± 17, and 0.87 ± 0.12 for diets no. 1 and 2 and the stabilization diet, respectively (P < .01). These data do not support the hypothesis of a metabolic adaptation toward increased glucose tolerance and glucose utilization induced by a high-carbohydrate, low-lipid diet in healthy subjects. Furthermore, changes observed in plasma lipoprotein concentrations favor the concept that such a diet may increase the risk for cardiovascular disease. Metabolism 1993;42:365–70.

The paper by Leclerc et al addresses a very old, very controversial, and very important issue, that is, whether or not an increase in dietary carbohydrate improves glucose utilization and glucose tolerance in normal individuals. This has been an important issue since Himsworth (1) first published his eloquent experiments over 55 years ago, and the metabolic impact of variations in the amount of dietary carbohydrate on glucose metabolism has remained controversial to this day. Himsworth’s experiments demonstrated that glucose tolerance that appears abnormal in individuals on restricted carbohydrate intake returns to normal as dietary carbohydrate is increased from 50 to 150 g/d. The data published over a half century ago by Himsworth provided important insights into the role of dietary carbohydrate in maintaining normal glucose tolerance. Indeed, it forms the very cornerstone for the current recommendations that dietary carbohydrate intake needs to be > 150 g/d for ≥ 3 d before the administration of a standard oral glucose-tolerance test. On the other hand, this study also indicated that there was little further improvement in glucose tolerance as dietary carbohydrate was increased from 150 to as much as 300 g/d. Nevertheless, Himsworth’s landmark study has, for the most part, been misinterpreted as evidence that increasing dietary carbohydrate per se improves glucose tolerance. The straightforward and controlled approach taken by Leclerc et al is refreshing and adds clarity and insight into this controversy.

Leclerc et al address the simple and clinically practical question of whether an increase in dietary carbohydrate in diets already adequate in carbohydrate (ie, > 150 g/d) would provide any further improvement in glucose utilization. Specifically, does increasing dietary carbohydrate from 45% to 65% of total energy improve glucose tolerance or enhance glucose and free fatty acid (FFA) oxidation? To address this question plasma glucose and insulin concentrations, as well as glucose and FFA oxidation rates were measured over a 6-h period after a standard 75-g oral glucose challenge at baseline and again at the end of each of two dietary periods. Glucose and insulin concentrations were measured by standard biochemical techniques and glucose and FFA oxidation were measured by indirect calorimetry. The two isoenergetic diets differed only in the percentage of energy as fat and carbohydrate. One dietary period contained 45% of energy as carbohydrates, 30% as fat, and 25% as protein, whereas the other dietary period contained 65% of energy as carbohydrate, 11% as fat, and 25% as protein. The diets were fed on a metabolic ward under defined dietary conditions for 1 wk each with a hiatus of 4 wk between dietary periods.

The results of these well-controlled studies clearly demonstrate that neither glucose tolerance nor glucose and FFA oxidation were improved by increasing dietary carbohydrate from 45% to 65% of total energy. Although insulin-mediated glucose disposal was not directly measured in these studies, the authors suggest that plasma glucose and insulin response to the oral glucose challenge suggest that there were no changes in insulin action. Although this conclusion appears to be supported by the fact that plasma glucose and insulin response between the dietary periods were essentially superimposable, the relationship between glucose tolerance and insulin action is not always perfect and caution should be exercised in making this direct comparison.

Finally, although these studies do not provide any new information on the effects of increased dietary carbohydrate on lipoprotein metabolism, they do confirm and add additional data to a growing body of evidence that indicates that increasing dietary carbohydrate results in increases in plasma triglyceride and decreases in plasma high-density-lipoprotein-cholesterol con-
centrations, without any apparent improvement in total plasma or low-density-lipoprotein-cholesterol concentrations.

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Reference

Fructose Transporter in Human Spermatozoa and Small Intestine Is GLUT5. Charles F. Burant, Jun Takenda, Edith Brot-Laroche, Graeme I. Bell, and Nicholas O. Davidson

ABSTRACT We recently reported that the glucose transporter isoform, GLUT5, is expressed on the brush border membrane of human small intestinal enterocytes (Davidson, N. O., Hausman, A. M. L., Ickovics, C. A., Buse, J. B., Gould, G. W., Burant, C. F., and Bell, G. I. (1992) Am. J. Physiol. 262. C795–C800). To define its role in sugar transport, human GLUT5 was expressed in Xenopus oocytes and its substrate specificity and kinetic properties determined. GLUT5 exhibits selectivity for fructose transport, as determined by inhibition studies, with a $K_m$ of 6 mM. In addition, fructose transport by GLUT5 is not inhibited by cytochalasin B, a competitive inhibitor of facilitative glucose transporters. RNA and protein blotting studies showed the presence of high levels of GLUT5 mRNA and protein in human testis and spermatozoa, and immunocytochemical studies localize GLUT5 to the plasma membrane of mature spermatids and spermatozoa. The biochemical properties and tissue distribution of GLUT5 are consistent with a physiological role for this protein as a fructose transporter. J Biol Chem 1992:267:14523–6.

The increased consumption of fructose in the Western diet has been linked to rising incidences of hypertriglyceridemia and hyperinsulinemia. Absorption of fructose in the gut is by a facilitated carrier that is different from the sodium-dependent glucose transporter present on the apical membrane of the intestinal mucosal cells. Facilitated transport of glucose into and out of mammalian tissues is also regulated by a family of sodium-independent carrier proteins that are distinct from the sodium-dependent glucose transporter. The members of the family, known as GLUT1 to GLUT5, were identified by cross hybridization of their cDNAs and they share extensive amino acid homology. The roles of GLUT1 to GLUT4 in glucose transport are now fairly well established. GLUT2, a high $K_m$ transporter found in the liver, the $\beta$ cell, and on the basolateral membrane of mucosal cells while GLUT4 is found in high concentrations in muscle and adipose and its translocation to the plasma membrane is regulated by insulin. GLUT1 is expressed at high concentrations in epithelial cells that line the blood-brain barrier whereas GLUT3 is located in neuronal cells; the major role of these carriers is in the provision of glucose to the brain.

GLUT5 was initially located in the intestine and kidney primarily and more recently in skeletal muscle (1), testes, and spermatozoa, and it was speculated that this carrier may be involved in the transport of hexoses other than glucose. The studies by Burant et al (2) indicate that what was thought to be the GLUT5 glucose transporter is in fact a fructose transporter. Xenopus oocytes expressing GLUT5 cDNA accumulated $[^4C]$ fructose and the uptake of labeled fructose was inhibited by unlabeled fructose but not by glucose and other hexoses. The GLUT5-expressing cells had little ability to transport 2-deoxyglucose. Oocytes expressing the GLUT2 transporter also accumulated labeled fructose, but less effectively than glucose, and accumulation of fructose via the GLUT2 transporter was inhibited by unlabeled glucose and other hexoses as well as by fructose. The GLUT5 transporter is located apically on the intestinal mucosal cell while the GLUT2 protein is solely basolateral. These studies suggest that fructose absorption in the intestine is via the GLUT5 transporter and that this transporter may be specific for fructose. The role of the GLUT2 transporter in intestinal fructose transport may be similar to its role in glucose transport, that is hexose efflux across the basolateral membrane of the mucosal cell.

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Reference