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Relative Carnitine Deficiency in Autism

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A random retrospective chart review was conducted to document serum carnitine levels on 100 children with autism. Concurrently drawn serum pyruvate, lactate, ammonia, and alanine levels were also available in many of these children. Values of free and total carnitine ($p < 0.001$), and pyruvate ($p = 0.006$) were significantly reduced while ammonia and alanine levels were considerably elevated ($p < 0.001$) in our autistic subjects. The relative carnitine deficiency in these patients, accompanied by slight elevations in lactate and significant elevations in alanine and ammonia levels, is suggestive of mild mitochondrial dysfunction. It is hypothesized that a mitochondrial defect may be the origin of the carnitine deficiency in these autistic children.

KEY WORDS: Lactic acidosis; mitochondrial disease; autism; hyperammonemia.

INTRODUCTION

The autistic spectrum disorders are a group of behaviorally defined developmental disorders that all share the same characteristic core deficits in social interaction and verbal and nonverbal communication, with restricted, stereotypic patterns of behaviors or interests (American Psychiatric Association, 1994). The pathophysiology of these disorders has not yet been defined, although strong evidence points to a polygenic etiology of at least some, if not most cases. The majority of autism cases occur in isolation in otherwise healthy-appearing children; however a small proportion of cases are associated with other medical diagnoses, such as neurocutaneous syndromes (e.g., tuberous sclerosis complex), metabolic diseases (e.g., phenylketonuria), chromosomal abnormalities (e.g., 15q inverted duplications) or congenital infections (e.g., rubella).

Extensive laboratory evaluations remain a popular clinical practice advocated by many practitioners in an attempt to "find something wrong" that might cause the disorder. However, the recent "*Practice Parameter: Screening and Diagnosis of Autism*" (Filipek *et al.*, 1999, 2000) reviewed the empiric evidence for such laboratory evaluations in autistic patients and clearly recommended that there is inadequate evidence to support routine clinical testing for trace elements, celiac antibodies, immunological or neurochemical abnormalities, vitamin levels, urinary peptides, mitochondrial disorders, thyroid function, or erythrocyte glutathione peroxidase, or to perform allergy testing, intestinal permeability studies, or stool analyses.

In the author's clinical practice (PAF), serum total and free carnitine levels were routinely obtained at initial presentation of young autistic children with a history of regression, who were scheduled to have an electroencephalogram (Tuchman & Rapin,

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1997) and to potentially begin a trial of valproate therapy (DeVivo *et al.*, 1998). Valproate poses a known risk of metabolic decompensation to individuals with carnitine deficiency (DeVivo *et al.*, 1998; Ohtani, Endo, & Matsuda, 1982), which includes a Reye-like syndrome and fatal hepatotoxicity (Pons & DeVivo, 1995; Triggs, Bohan, Lin, & Willmore, 1990). A preliminary chart review, conducted on 30 subjects, revealed that the majority of carnitine levels in these autistic children were either low, or at the low-normal range. Therefore, total and free carnitine levels on all subsequent clinical patients were routinely obtained to identify those at risk for a deficiency, the results of which are reported here.

METHODS

This study was approved by the University of California, Irvine Institutional Review Board. A random retrospective chart review was conducted on all patients in the author's clinical practice (PAF) to identify those who had total and free carnitine levels assayed as part of their laboratory evaluation at presentation for the diagnosis of autism. All subjects met DSM-IV criteria for an autistic spectrum disorder (e.g., Autistic Disorder, Asperger's or Pervasive Developmental Disorder-Not Otherwise Specified). Exclusionary criteria for this chart review included existing seizure disorder, chromosomal abnormality, concomitant genetic disorder, medications, focal abnormality on neurological examination, or special diets. Many of the children had self-imposed dietary restrictions and/or food aversions, but none were sufficient to produce malnutrition. Other demographic measures were not obtained or recorded as part of routine clinical care, including race, socioeconomic group and psychological test results.

Of these reviewed charts, a total of 145 autistic patients were noted with documented quantitative serum carnitine levels from eight different processing laboratories. To reduce laboratory-based variance, the six laboratories that contributed fewer than eight cases each were eliminated, resulting in a total of 100 patients processed by 2 laboratories. Subjects were 2–13 years old (mean = 5.27 years), with 77 males and 23 females. The reported total and free carnitine levels were recorded for each subject, along with the respective age- and/or gender-specific normative range specified by the two processing laboratories. The carnitine levels were then normalized and transformed into z scores for comparison and analysis. Acyl-carnitine levels were calculated as raw "Total Carnitine minus Free Carnitine" on each patient. Subsequently, acyl/free (A/F) carnitine ratios were calculated and recorded for each subject. In addition to evaluating carnitine levels in our cohort, results from serum chemistry and quantitative amino acid assays were also compiled; all analyses were performed from the same blood draw at clinical presentation. As mentioned for carnitine results, the results were from different laboratories. Initially, 43 subjects had pyruvate levels processed by 2 laboratories, 45 subjects had lactate levels determined by 3 laboratories, 63 subjects had ammonia levels measured by 5 laboratories and 83 subjects had alanine levels evaluated by 3 laboratories. Laboratories with insufficient subjects ($n < 8$) were excluded to arrive at the final number of subjects for analyses in each area of interest. Of the 100 subjects included in the carnitine analyses: 40 subjects had pyruvate levels processed by a single laboratory, 42 had lactate levels determined by 2 laboratories, 50 had ammonia levels measured by 2 laboratories, and 82 subjects had alanine levels evaluated by 2 laboratories. Due to lab-specific criteria for normative range values, data recorded for each subject in each area of interest were transformed to z scores according to the mean of the normative range specified by the corresponding processing laboratory. These z scores were then used for statistical analyses.

The Mann–Whitney rank sum test, a nonparametric measure, was used to evaluate the significance of differences ($p < 0.01$) observed in carnitine, pyruvate, lactate, ammonia and alanine levels between our groups of autistic subjects and the corresponding normative ranges.

RESULTS

As shown in Fig. 1, mean total and free carnitine values were significantly reduced in our autistic subjects relative to the normative range specified by the two processing laboratories ($p < 0.001$). Eighty-three percent of the subjects had total and free carnitine levels below the reference mean. In addition, more than one-third (36%) had total carnitine levels, and 27% had free carnitine levels, greater than or equal to 1 standard deviation below the mean. These data from autistic patients indicate a definite shift below the normal reference mean. Carnitine results are summarized in Table I. As

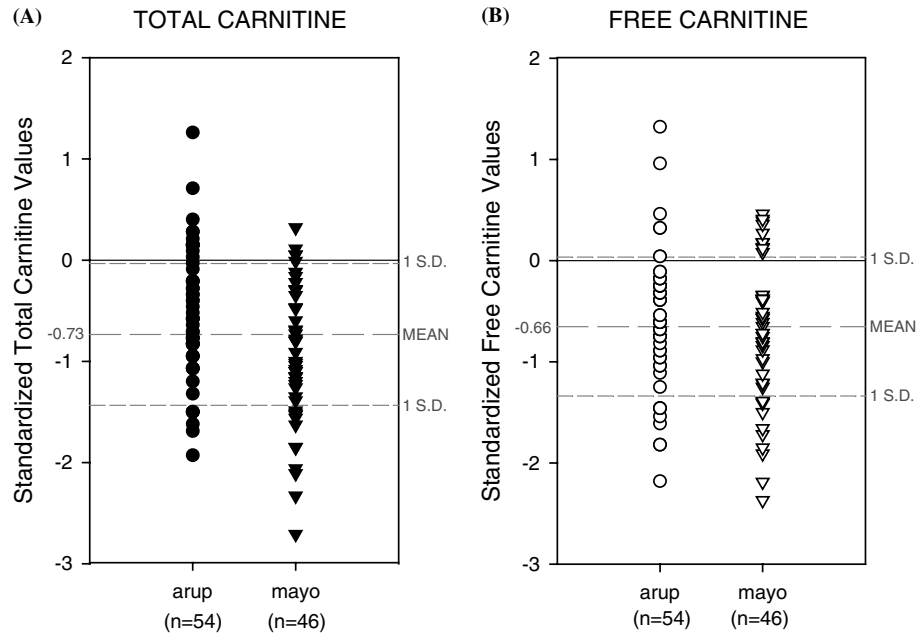

Fig. 1. Distribution of observed total and free serum carnitine levels from both laboratories on our autistic subjects ($n = 100$), after transformation and standardization to z values from respective reference ranges. Dashed horizontal reference lines represent mean and standard deviation values from the combined data of both laboratories. (A) Standardized total carnitine levels (z scores) processed by Associated Regional and University Pathologists (arup) and Mayo Laboratories (mayo). (B) Standardized free carnitine levels (z scores) processed by arup and mayo.

shown in Fig. 2, the A/F ratios we observed predominately fell within the normal range.

Fig. 3 demonstrates that serum pyruvate levels measured concurrently with carnitine in our subjects were also significantly reduced ($p < 0.006$): 85% had values which were greater than 1 standard deviation below the reference range mean. Serum lactate levels, however, were not significantly elevated for the cohort (Fig. 4). Hyperammonemia was observed in the majority of subjects ($p < 0.001$), with 78% greater than 1 standard deviation above the mean (Fig. 5). Alanine, as reported on quantitative serum amino acid profiles, was also significantly elevated in our subjects ($p < 0.001$), with 80% of serum alanine levels greater than 1 standard deviation above the mean (Fig. 6). Results for pyruvate, lactate, ammonia and alanine are summarized in Table II and collectively present a consistent picture of mild mitochondrial dysfunction.

DISCUSSION

Carnitine is an essential co-factor in the utilization of fat reserves from body stores during fasting and stress. It plays a key role in the transport of long chain fatty acids into the mitochondria where they undergo beta oxidation in energy production (Bremer, 1983; Roe & Ding, 2001). Although rare, carnitine deficiency syndromes have been reported for over 25 years (Engel & Angelini, 1973); only recently, however, has its importance as a major risk in early infancy been recognized (Rinaldo *et al*., 1999). Endogenous carnitine biosynthesis occurs in man, with the final synthetic step limited to the liver and occasionally compromised in severe cirrhosis (Roe, 1997). However, the major portion of body carnitine is obtained through the diet (primarily from meat and dairy products). It is well demonstrated that during strictly controlled total parenteral nutrition without carnitine supplementation, a deficiency state ensues (Borum, 1995), which occasionally can also arise in malabsorption syndromes, such as celiac disease (Ceccarelli, Cortigiani, Assanta, Balsano, & Chiaravalloti, 1992; Lerner, Gruener, & Iancu, 1993) and unusually deficient diets (Roe, 1997; Roe & Ding, 2001).

Physiological plasma levels of carnitine are 25–50 μM, but tissue levels are up to 50-fold higher,

Table I. Average Total, Free, and Acyl Carnitine values and their corresponding lab-specific normal reference values for 100 subjects, mean total and free carnitine transformed *z* scores, and percentage of subjects distributed with respect to normal reference range mean and standard deviation

Lab	N(M+F)	N(M)	N(F)	Carnitine	Ref mean (range)	Mean obs (*SD*)	Mean *z* value (*SD*)	*p* Value	# below ref range mean		> 1 *SD* below ref range mean	
Arup	54	39	15									
				Total (uM/dL)	5.35 (2.6–8.1)	4.49 (1.05)	−0.52 (0.64)	0.006	41/54	75.93%	11/54	20.37%
				Free (uM/dL)	4.65 (2.3–7.0)	3.81 (0.92)	−0.60 (0.66)	0.003	47/54	87.04%	12/54	22.22%
				Acyl (uM/dL)	0.95 (0.0–1.9)	0.87 (0.68)						
Mayo	46	38	8									
Total (M+F)												
				Total			−1.98 (0.69)	< 0.001	42/46	91.30%	25/46	54.35%
				Free			−0.72 (0.72)	< 0.001	36/46	78.26%	15/46	32.61%
Males												
				Total (nM/mL)	63.0 (37–89)	46.20 (9.76)						
				Free (nM/mL)	48.5 (28–69)	38.59 (8.46)						
				Acyl (nM/mL)		7.61 (4.50)						
Females												
				Total (nM/mL)	51.5 (30–73)	47.30 (6.53)						
				Free (nM/mL)	39.5 (19–60)	37.49 (4.89)						
				Acyl (nM/mL)		9.81 (5.84)						
All labs	100	77	23									
				Total			−0.73 (0.70)	< 0.001	83/100	83.00%	36/100	36.00%
				Free			−0.66 (0.69)	< 0.001	83/100	83.00%	27/100	27.00%

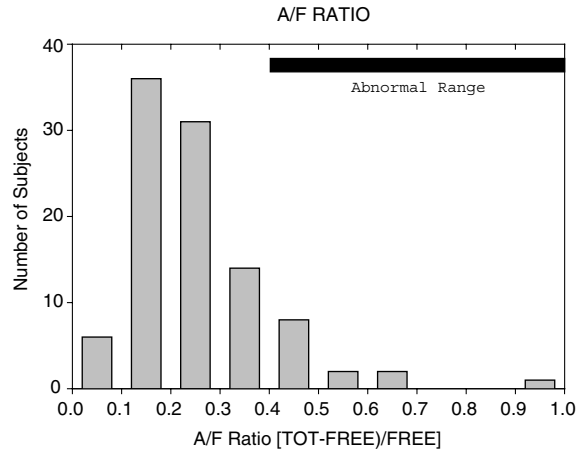

Fig. 2. Number of patients with observed A/F ratio (acyl/free carnitine). Vertical bins separated by increments of 0.1 represent number of patients as a function of A/F ratio ($n = 100$).

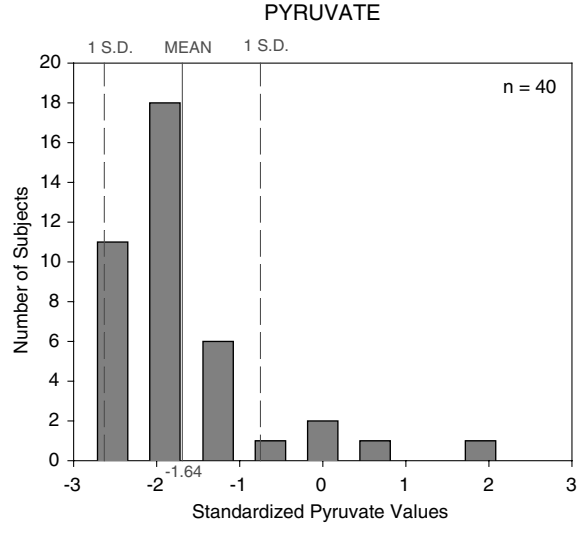

Fig. 3. Distribution of serum pyruvate levels after transformation and standardization to *z* scores. Vertical gray reference lines represent mean and standard deviation values.

with over 95% of the body stores found in skeletal muscle (Bremer, 1997). A fraction of tissue and plasma carnitine, generally less than 10–15%, is acylated, reflecting its efflux back out of mitochondria with its fatty acid fuel unspent (Roe, 1997). In this form it serves as a fatty acid buffer, freeing up limiting mitochondrial Coenzyme A (to which the acyl chain would be transferred for beta oxidation) for use with other substrates. Carnitine and acyl carnitine derivatives are rapidly lost in the glomerular ultrafiltrate, but free carnitine's ultimate physiological clearance by the kidney is very low because

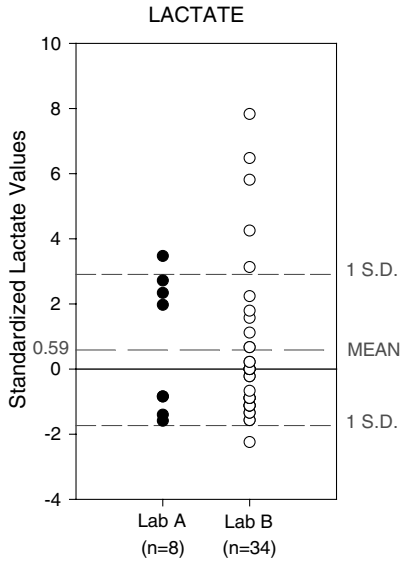

Fig. 4. Distribution of serum lactate levels after transformation and standardization to *z* scores from the appropriate processing laboratory's reference ranges. Dashed horizontal reference lines represent mean and standard deviation values from the combined data of both laboratories.

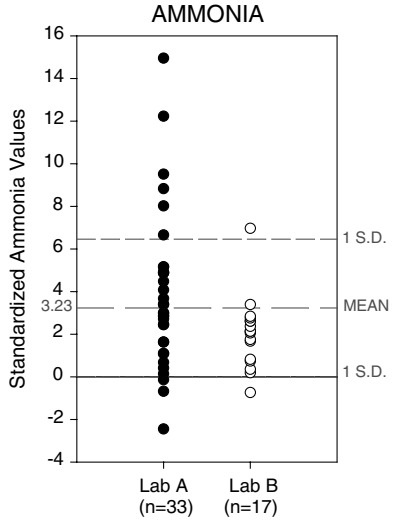

Fig. 5. Distribution of serum ammonia levels after transformation and standardization to *z* scores from the appropriate processing laboratory's reference ranges. Dashed horizontal reference lines represent mean and standard deviation values from the combined data of both laboratories.

of its active, sodium-dependent resorption in the proximal tubule (Scaglia *et al.*, 1998). The acyl carnitines are poor substrates for transport and hence are rapidly lost in the urine. This feature sets the stage for the "secondary carnitine deficiency" syn-

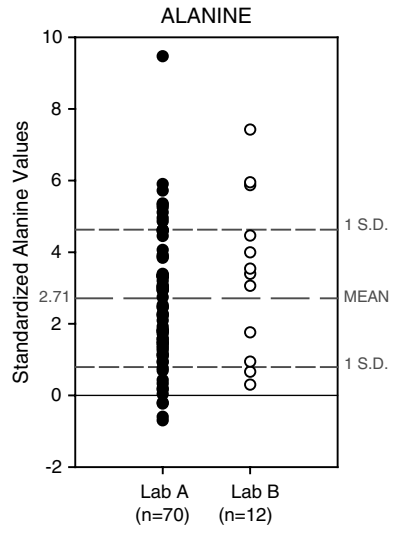

Fig. 6. Distribution of serum alanine levels after transformation and standardization to *z* scores from the appropriate processing laboratory's reference ranges. Dashed horizontal reference lines represent mean and standard deviation values from the combined data of both laboratories.

dromes, those that arise because of some metabolic block that has nothing to do with carnitine itself, but rather predisposes to high concentrations of acyl carnitines, and ultimate depletion of body carnitine stores through renal wasting (Famularo, Matricardi, Nucera, Santini, & DeSimone, 1997; Roe, 1997). Classical examples of inborn errors that produce a secondary carnitine deficiency include the family of diseases caused by mutations in the genes encoding the enzymes involved in fatty acid oxidation (Rinaldo et *al.*, 1991; Roe & Ding, 2001), the family of organic acidemias caused by mutations in the genes encoding the enzymes involved in branched chain amino acid catabolism (Chalmers *et al.*, 1984), and diseases produced by defects in the mitochondrial respiratory complexes (Brenningstall, 1990). Several drugs, most notably valproate, also cause a secondary carnitine deficiency (DeVivo *et al.*, 1998; Farkas, Bock, Cseko, & Sandor, 1996). Finally, because of the cortisol-induced catabolic response of muscle and fat, which release fatty acids and thereby increase the acylcarnitine fraction, carnitine levels are also sensitive to physiological changes brought about by fasting, stress and exercise (Coyle, 2000; Inoue *et al.*, 1999).

No primary defects in carnitine biosynthesis are known to produce recognized disease (Kerner & Hoppel, 1998), but primary carnitine deficiency is produced by a rare genetic disease caused by muta-

Table II. Average pyruvate, lactate, ammonia and alanine levels, corresponding lab-specific normal reference values, mean transformed *z* scores, and percentage of subjects distributed with respect to normal reference range mean and standard deviation

Pyruvate	Lab	N(M + F)	N(M)	N(F)	Ref Mean [Range] (mg/dL)	Mean Obs (SD) (mg/dL)	Mean z value (*SD*)	*p* value	# < ref range mean		>1 SD below ref range mean	
	LAB A	40	30	10	0.50 [0.30–0.70]	0.24 (0.15)	−1.64 (0.98)	0.006	35/40	87.50%	34/40	85.00%
Lactate	Lab	N(M + F)	N(M)	N(F)	Ref Mean [Range] (meq/L)	Mean Obs (SD) (meq/L)	Mean *z* value (*SD*)	*p* value	# ⩾ ref range mean		>1 SD above ref range mean	
	Lab A	8	6	2	1.35 [0.50–2.20]	1.74 (1.11)	0.73 (2.09)	0.487	4/8	50.00%	4/8	50.00%
	Lab B	34	26	8	1.40 [0.70–2.10]	1.65 (1.07)	0.55 (2.40)	0.983	18/34	52.94%	9/34	26.47%
	LAB A+B	42	32	10			0.59 (2.32)	0.90	22/42	52.38%	13/42	30.95%
Ammonia	Lab	N(M + F)	N(M)	N(F)	Ref Mean [Range] (uM/L)	Mean Obs (SD) (uM/L)	Mean z value (SD)	*p* value	# > ref range mean		>1 SD above ref range mean	
	Lab A	33	24	9	23 [11–35]	51.30 (26.93)	3.85 (3.66)	<0.001	30/33	90.91%	27/33	81.82%
	Lab B	17	14	3	38.5 [17–60]	64.71 (21.64)	2.04 (1.69)	<0.001	16/17	94.12%	12/17	70.59%
	LAB A+B	50	38	12			3.23 (3.23)	<0.001	46/50	92.00%	39/50	78.00%
Alanine	Lab	N(M + F)	N(M)	N(F)	Ref Mean [Range] (nM/mL)	Mean Obs (SD) (nM/mL)	Mean z value (SD)	*p* value	# > ref range mean		>1 SD above ref range mean	
	Lab A	70	59	11	240 [120–360]	420 (128)	2.59 (1.84)	<0.001	66/70	94.29%	57/70	81.43%
	Lab B	12	9	3	225 [100–350]	475 (164)	3.45 (2.26)	<0.001	12/12	100%	9/12	75.00%
	LAB A+B	82	68	14			2.71 (1.92)	<0.001	78/82	95.12%	66/82	80.49%

tions in the *SLC22A5* gene that encodes the sodium-dependent carnitine cotransporter OCTN2 (Huizing *et al*., 1997; Scaglia *et al*., 1998; Wang *et al*., 2001). Thus, these mutations also produce a profound, life-threatening carnitine deficiency through renal wasting, but unlike the secondary deficiency syndromes, do not alter the ratio of free to acyl carnitine (Wang *et al*., 2001).

The mechanism of carnitine deficiency seen in our patients with autism remains to be determined as does its relationship to the underlying etiology of the syndrome. The defect appears to fit more within the pattern of a ''primary deficiency'' since the free and acyl fractions are proportionately reduced, and no recognized abnormal acyl carnitine species is found (Table I). None of the patients, however, have a well-recognized cause of a primary deficiency. None have the profound carnitine deficiency or the clinical picture seen with the classical OCTN2 transport defect (Wang *et al*., 2001). None have a well-defined malabsorption syndrome or are any on parenteral nutrition. On the other hand, the deficiency may be a derivative feature of the autistic phenotype, since it is clear that children with autism do frequently have gastrointestinal difficulties, occasionally including abnormal motility studies and reflux (Horvath, Papadimitriou, Rabsztyn, Drachenberg, & Tildon, 1999). It is also clear that they frequently select a narrow, ritualized diet (Ahearn, Castine, Nault, & Green, 2001; Levin & Carr, 2001). However, in our cases, none of these problems seem to rise to the extraordinary threshold apparently required to produce a deficiency in otherwise healthy individuals (Schafer & Reichmann, 1990), although this requires more extensive systematic study. Further, because of the known ability of chronic stress to deplete carnitine stores (Proulx *et al*., 1997), it is also possible that cryptic stress is a component of the autistic phenotype. Again, this will require further systematic study. Finally, it is unlikely that an isolated carnitine deficiency itself can cause or otherwise underlie the neurodevelopmental abnormalities in autism, since even children with profound primary carnitine deficiency rarely have neurological abnormalities other than those arising as a sequella to a calamitous metabolic decompensation (Wang *et al*., 2001).

The metabolic picture of these patients with autism is further complicated by findings additionally suggestive of a mild mitochondrial dysfunction. These include the modest elevations of plasma ammonia (92% above the mean) and anaerobic conversion of pyruvate to lactate (88% of pyruvates below the mean, and 52% of lactates at or above the mean). In those children who had quantitative plasma amino acid analysis, this was reconfirmed by the finding of an elevated alanine (Table I). It is possible that carnitine deficiency and a secondary block in fatty acid oxidation produce the mitochondrial defect, much as occurs in classical medium chain acyl-CoA dehydrogenase deficiency and other fatty acid oxidation disorders. However, none of the children has shown the metabolic hallmark of this deficient state, hypoketotic hypoglycemia (Roe & Ding, 2001). On the other hand, since it is coming to be recognized that classical blocks in the mitochondrial respiratory complexes (slowing CoA recycling because of tight coupling between oxidation and acyl CoA production and metabolism) can secondarily produce excess acylcarnitines and a secondary carnitine deficiency syndrome (Gargus *et al*., 2003), perhaps some more subtle mitochondrial defect may be the origin of the carnitine deficiency in these autistic children.

In this regard, it is intriguing that in autism associated with a 15q inverted duplication, functional defects in respiratory complex III and mitochondrial hyperproliferation in skin and muscle biopsies have also been reported (Filipek *et al*., 2003). The metabolic defect is presumably produced by genes involved in the chromosomal rearrangement that play a still undefined role in mitochondrial biogenesis. This does not appear to be an artifactual finding since a ''tissue bioassay'' of functional impairment, compensatory mitochondrial hyperproliferation (Wiesner *et al*., 1999), is also uniformly found, and has been reported as an incidental finding in other patients with the same chromosomal abnormality (Repetto, White, Bader, Johnson, & Knoll, 1998). Further, it is intriguing that defects in oxidative phosphorylation and beta oxidation produce a secondary defect in the synthesis of docosahexaenoic (22:6n-3) acid (Infante & Huszagh, 2000) and that this same fatty acid has recently been observed to be markedly reduced in children with autism (Vancassel *et al*., 2001), a finding potentially explained by the energetic defect our results suggest. This fatty acid has been noted to be a ligand of the RXR retenoic acid receptor, a receptor known to be essential to normal brain development (De Urquiza *et al*., 2000). Finally, it is now appreciated that minor, primarily craniofacial, dysmorphology is often associated with primary lactic acidosis (Cormier-Daire *et al*., 1996), similar to the mild dysmor-

phology coming to be recognized in a subgroup of patients with autism (Miles & Hillman, 2000).

It is intriguing that a recent report documented elevated death rates for several causes in people with autism (Shavelle, Strauss, & Pickett, 2001). Standardized mortality ratios were reported separately for subjects with no or mild mental retardation, and moderate to profound retardation, respectively: seizures or presumed seizures (22.6 and 36.9), suffocation (5.7 and 51.4), and drowning (3.9 and 13.7) were particularly elevated in comparison to the low rates for other external causes (0.8 and 0.6). Perhaps the present finding of relative carnitine deficiency in autism may be related. Further study on these findings is ongoing.

The implications of these reported findings for practitioners involved in the diagnosis and care of patients with autism, while limited at this time pending further study, are at least that in patients with autism being considered for valproate therapy, a determination of baseline carnitine levels is valuable; if low plasma carnitine be observed, it should not simply be treated with supplements, but should first be followed by studies of lactate, pyruvate, ammonia and an acyl carnitine profile. A standard caveat is that such studies should be obtained from a freely flowing venipuncture. While carnitine supplements are safe, naturally occurring nutrients, it would clearly be imprudent to begin such supplements in a child without a specifically demonstrated deficiency.

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REFERENCES

Ahearn, W., Castine, T., Nault, K., & Green, G. (2001). An assessment of food acceptance in children with autism or pervasive developmental disorder-not otherwise specified. *Journal of Autism Developmental Disorders, 31*(5), 505–511.

American Psychiatric Association. (1994). *Diagnostic and statistical manual of mental disorders* (4th ed.). Washington DC.

Borum, P. (1995). Carnitine in neonatal nutrition. *Journal of Child Neurology 10*(Suppl 2), S25–S31.

Bremer, J. (1983). Carnitine–metabolism and functions. *Physiological Reviews, 63*(4), 1420–1480.

Bremer, J. (1997). The role of carnitine in cell metabolism. In C. DeSimone & G.

Famularo (Eds.), *Carnitine today* (pp. 1–37). Austin, TX: Landes Bioscience.

Brenningstall, G. (1990). Carnitine deficiency syndromes. *Pediatric Neurology, 6*(2), 75–81.

Ceccarelli, M., Cortigiani, L., Assanta, N., Balsano, L., & Chiaravalloti, G. (1992).

Plasma L-carnitine levels in children with celiac disease. *Minerva Pediatrica, 44*(9), 401–405.

Chalmers, R., Stacey, T., Tracey, B., de Sousa, C., Roe, C., Millington, D., & Hoppel, C. (1984). L-Carnitine insufficiency in disorders of organic acid metabolism: response to L-carnitine by patients with methylmalonic aciduria and 3-hydroxy-3-methylglutaric aciduria. *Journal of Inherited Metabolic Disease, 7*(Suppl 2), 109–110.

Cormier-Daire, V., Rustin, P., Rotiq, A., Chretien, D., Le Merrer, M., Belli, D., Le Goff, A., Hubert, P., Ricour, C., & Munnich, A. (1996). Craniofacial anomalies and malformations in respiratory chain deficiency. *American Journal of Medical Genetics, 66*(4), 457–463.

Coyle, S. (2000). Physical activity as a metabolic stressor. *American Journal of Clinical Nutrition, 72*(Suppl 2), S512–S520.

De Urquiza, A., Liu, S., Sjoberg, M., Zetterstrom, R., Griffiths, W., Sjovall, J., & Perlmann, T. (2000). Docosahexaenoic acid, a ligand for the retinoid X receptor in mouse brain. *Science, 290*(5499), 2140–2144.

DeVivo, D., Bohan, T., Coulter, D., Dreifuss, F., Greenwood, R., Nordli, D. J., Shields, W., Stafstrom, C., & Tein, I. (1998). L-carnitine supplementation in childhood epilepsy: current perspectives. *Epilepsia, 39,* 1216–1225.

Engel, A., & Angelini, C. (1973). Carnitine deficiency of human skeletal muscle with associated lipid storage myopathy: A new syndrome. *Science, 179*(76), 899–902.

Famularo, G., Matricardi, F., Nucera, E., Santini, G., & DeSimone, C. (1997). Carnitine deficiency: primary and secondary syndromes in metabolism. In C. De Simone & G. Famularo (Eds.), *Carnitine Today* (pp. 119–161). Austin, TX: Landes Bioscience.

Farkas, V., Bock, I., Cseko, J., & Sandor, A. (1996). Inhibition of carnitine biosynthesis by valproic acid in rats—the biochemical mechanism of inhibition. *Biochemical Pharmacology, 52*(9), 1429–1433.

Filipek, P. A., Accardo, P. J., Baranek, G. T., Cook, E. H. J., Dawson, G., Gordon, B., Gravel, J. S., Johnson, C. P., 'Kallan, R. J., Levy, S. E., Minshew, N. J., Prizant, B. M., Rapin, I., Rogers, S. J., Stone, W. L., Teplin, S., Tuchman, R. F., & Volkmar, F. (1999). The screening and diagnosis of autistic spectrum disorders. *Journal of Autism and Developmental Disorders, 29,* 437–482.

Filipek, P. A., Accardo, P. J., S, A., Baranek, G. T., Cook, E. H. J., Dawson, G., Gordon, B., Gravel, J. S., Johnson, C. P., Kallan, R. J., Levy, S. E., Minshew, N. J., Ozonoff, S., Prizant, B. M., Rapin, I., Rogers, S. J., Stone, W. L., Teplin, S., Tuchman, R. F., & Volkmar, F. (2000). Practice parameter: Screening and diagnosis of autistic spectrum disorders. *Neurology, 55,* 468–479.

Filipek, P. A., Juranek, J., Smith, M., Ramos, E., Mays, L., Bocian, M. E., Masser-Frye, D., Laulhere, T., Modahl, C., Spence, M., & Gargus, J. (2003). Evidence of mitochondrial dysfunction in autistic patients with 15q inverted duplication. *Annals of Neurology, 53*(6), 801–804.

Gargus, J., Boyle, K., Bocian, M. E., Roe, D. S., Vianey-Saban, C., & Roe, C. (2003). Respiratory complex II defect in sibs associated with a symptomatic secondary block in fatty acid oxidation. *Journal of Inherited Metabolic Disease, 26*, 659–670.

Horvath, K., Papadimitriou, J., Rabsztyn, A., Drachenberg, C., & Tildon, J. (1999). Gastrointestinal abnormalities in children with autistic disorder. *Journal of Pediatrics, 135*(5), 559–563.

Huizing, M., Iacobazzi, V., Ijlst, L., Savelkoul, P., Ruitenbeek, W., van den Heuvel, L., Indiveri, C., Smeitink, J., Trijbels, F., Wanders, R., & Palmieri, F. (1997).Cloning of the human carnitine-acylcarnitine carrier cDNA and identification of the molecular defect in a patient. *American Journal of Human Genetics, 61*(6), 1239–1245.

Infante, J., & Huszagh, V. (2000). Secondary carnitine deficiency and impaired docosahexaenoic (22:6n-3) acid synthesis: A common denominator in the pathophysiology of diseases of oxidative phosphorylation and beta-oxidation. *FEBS Letters, 468*(1), 1–5.

Inoue, F., Terada, N., Nakajima, H., Okochi, M., Kodo, N., Kizaki, Z., Kinugasa, A., & Sawada, T. (1999). Effect of sports activity on carnitine metabolism. Measurement of free carnitine, gamma-butyrobetaine and acylcarnitines by tandem mass spectrometry. *Journal of Chromatography B-Biomedical Sciencs Applications 731*(1), 83–88.

Kerner, J., & Hoppel, C. (1998). Genetic disorders of carnitine metabolism and their nutritional management. *Annual of Review Nutrition, 18,* 179–206.

Lerner, A., Gruener, N., & Iancu, T. (1993). Serum carnitine concentrations in coeliac disease. *Gut, 34,* 933–935.

Levin, L., & Carr, E. (2001). Food selectivity and problem behavior in children with developmental disabilities: Analysis and intervention. *Behavior Modification, 25*(3), 443–470.

Miles, J. H., & Hillman, R. E. (2000). Value of a clinincal morphology examination in autism. *American Journal of Medical Genetics, 91*(4), 245–253.

Ohtani, Y., Endo, F., & Matsuda, I. (1982). Carnitine deficiency and hyperammonemia associated with valproic acid therapy. *Journal of Pediatrics, 101*(5), 782–785.

Pons, R., & DeVivo, D. (1995). Primary and secondary carnitine deficiency syndromes. *Journal of Child Neurology, 10*(Suppl 2), S8–S24.

Proulx, F., Lacroix, J., Qureshi, I., Nadeau, D., Gauthier, M., & Lambert, M. (1997). Acquired carnitine abnormalities in critically ill children. *European Journal of Pediatrics, 156*(11), 864–869.

Repetto, G., White, L., Bader, P., Johnson, D., & Knoll, J. (1998). Interstitial duplications of chromosome region 15q11q13: clincal and molecular characterization. *American Journal of Medical Genetics, 79*(2), 82–89.

Rinaldo, P., Welch, R., Previs, S., Schmidt-Sommerfeld, E., Gargus, J., O'Shea, J., & Zinn, A. (1991). Ethylmalonic/adipic aciduria: effects of oral medium-chain triglycerides, carnitine, and glycine on urinary excretion of organic acids, acylcarnitines, and acylglycines. *Pediatric Research, 30*(3), 216–221.

Rinaldo, P., Yoon, H., Yu, C., Raymond, K., Tiozzo, C., & Giordano, G. (1999). Sudden and unexpected neonatal death: A protocol for the postmortem diagnosis of fatty acid oxidation disorders. *Semin Perinatology, 23*(2), 204–210.

Roe, C. (1997). Clinical experience with carnitine deficiency. *Journal of Rare Disease, 111,* 5–11.

Roe, C., & Ding, J. (2001). Mitochondrial Fatty Acid Oxidation Disorders. In C. Scriver, A. Beaudet, W. Sly, & D. Valle (Eds.), The metabolic and molecular basis of inherited disease (pp. 2297–2326). New York: McGraw Hill.

Scaglia, F., Wang, Y., Singh, R., Dembure, P., Pasquali, M., Fernhoff, P., & Longo, N. (1998). Defective urinary carnitine transport in heterozygotes for primary carnitine deficiency. *Genetics in Medicine, 1*(1), 34–39.

Schafer, J., & Reichmann, H. (1990). Subnormal carnitine levels and their correction in artificially fed patients from a neurological intensive care unit: a pilot study. *Journal of Neurology, 237*(3), 213–215.

Shavelle, R., Strauss, D., & Pickett, J. (2001). Causes of death in autism. *Journal of Autism Developmental Disorders, 31,* 569–576.

Triggs, W., Bohan, T., Lin, S., & Willmore, L. (1990). Valproate-induced coma with ketosis and carnitine insufficiency. *Archives of Neurology, 47*(10), 1131–1133.

Tuchman, R. F., & Rapin, I. (1997). Regression in pervasive developmental disorders: Seizures and epileptiform electroencephalogram correlates. *Pediatrics, 99,* 560–566.

Vancassel, S., Durand, G., Barthelemy, C., Lejune, B., Martineau, J., Guilloteau, D., Andres, C., & Chalon, S. (2001). Plasma fatty acid levels in autistic children. *Prostaglandins Leukot Essent Fatty Acids, 65*(1), 1–7.

Wang, Y., Korman, S., Ye, J., Gargus, J., Gutman, A., Taroni, F., Garavaglia, B., & Longo, N. (2001). Phenotype and genotype variation in primary carnitine deficiency. *Genetics in Medicine, 3*(6), 387–392.

Wiesner, R., Hornung, T., Garman, J., Clayton, D., O'Gorman, E., & Wallimann, T. (1999). Stimulation of mitochondrial gene expression and proliferation of mitochondria following impairment of cellular energy transfer by inhibition of the phosphocreatine circuit in rat hearts. *Journal of Bioenergetics Biomembranes, 31*(6), 559–567.