

# Application of LC-MS/MS analysis of plasma amino acids profiles in children with autism

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Autism is a neurodevelopmental disability that is usually diagnosed before age 3 years.<sup>(1)</sup> The need to understand the causes of autism and the underlying pathophysiology has become more acute since the number of diagnosed cases has risen markedly.<sup>(2)</sup> Several recent research studies lend support to the hypothesis that abnormalities involving the plasma Amino Acids profiles have been implicated in the etiology of autism.<sup>(3,4)</sup>

We designed a preliminary prospective cohort study to test plasma amino acids profiles in blood samples from patients with autism and normal cases whose age and gender matched. Twenty nonconsecutive patients admitted at 2 hospitals with a diagnosis of autism confirmed by Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSM-IV). All enrolled patients with autism were newly diagnosed. Twenty nonconsecutive control subjects were randomly selected from the eligible outpatients. All the eligible control subjects were defined as children 2–6 years old who were not known to be autistic or to have any other neurodevelopmental that could be related to autism. The Ethics Committee of China Rehabilitation Research Center, China, approved the trial protocol. All subjects' parents provided informed consent. Fasting blood samples were drawn on days 1 (admission) and then were immediately centrifuged and stored at  $-80^{\circ}\text{C}$ . The plasma amino acids profiles were assayed using aTRAQ™ Kits by Liquid chromatography-tandem mass spectrometry. Plasma neurotransmitter (including dopamine, noradrenalin and adrenalin) were also tested using radioimmunoassay. Results were presented as means  $\pm$  standard deviation (SD). Student's unpaired *t* test was used to compare the values in normal and autistic children. Statistical significance was defined as  $p < 0.05$ .

The mean age of the 20 patients was  $3.46 \pm 0.56$  years; 17 were boy. Three patients had family history of autism. The median length of hospital stay was 86 days (18–321 days). The mean hospitalization expense was  $64.6 \pm 5.8$  US dollars per day. On admission, the results indicated that the mean plasma lysine, glutamic acid and homocysteine levels were significantly ( $p < 0.05$ ) higher in autism as compared to controls while plasma leucine, tryptophan, valine, taurine, tyrosine and glutamine levels were lower in autism than control group ( $p < 0.05$ ) (Table 1). However, there was no difference in plasma other amino acids levels between autism and control group. In addition, we observed that the plasma dopamine, noradrenalin and adrenalin were significantly higher in the autistic group compared to controls ( $p < 0.05$ ). Table 1.

These preliminary results confirm an important conclusion: children with autism are significantly more likely to have abnormal in amino acids metabolim. Those may be caused by

**Table 1.** Plasma amino acids profiles and neurotransmitter in children with autism and normal

Plasma amino acids	Autism (SD), $\mu\text{M}$	Control (SD), $\mu\text{M}$	<i>p</i> *
Leucine	102.8 (6.8)	118.5 (8.2)	0.000
Lysine	178.1 (8.2)	170.5 (7.4)	0.004
Tryptophan	42.2 (4.3)	47.5 (4.1)	0.003
Valine	198.5 (10.7)	205.5 (11.5)	0.024
Taurine	43.9 (7.6)	48.2 (8.1)	0.004
Tyrosine	44.8 (3.6)	49.4 (5.2)	0.004
Glutamic acid	45.6 (9.1)	38.9 (7.5)	0.001
Glutamine	540.4 (30.3)	568.2 (33.5)	0.047
Homocysteine	9.6 (1.2)	7.4 (1.1)	0.000
Dopamine <sup>#</sup> (ng/l)	58.5 (6.4)	51.1 (5.7)	0.000
Noradrenalin <sup>#</sup> (ng/l)	455.6 (60.8)	400.5 (35.8)	0.001
Adrenalin <sup>#</sup> (ng/l)	55.7 (7.1)	49.5 (4.5)	0.002

\*Student's unpaired *t* test was used to compare the value in normal and autistic children. <sup>#</sup>Neurotransmitter.

gastrointestinal problems as well as picky eating and selective eating in their children.<sup>(5)</sup> This raises serious concerns regarding the long term effects of supervised restricted diets or nutritional supplement in children with autism. Whiteley *et al.*<sup>(6)</sup> reported that dietary intervention may positively affect developmental outcome for some children diagnosed with autism. Thus, further investigation in Chinese autism required to measure the effectiveness of diet related treatments on improving the clinical symptoms of autism. In addition, elevated levels of various neurotransmitters (including dopamine, noradrenalin and adrenalin) have been observed in autistic group compared to controls. The relationships between autism and plasma neurotransmitter should be further explored.

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## Conflict of Interest

No potential conflicts of interest were disclosed.

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