The urea cycle disorders (UCDs) result from inherited molecular defects in metabolism and are prevalent. Since late 1990’s, analysis of ornithine transcarbamylase and amino acids (AA) using ESI-MS/MS has become popular in newborn screening (NBS). This screening is able to detect more than 25 types of inborn errors of metabolism, including two UCDS of argininosuccinic acid synthase deficiency (CIT1) and argininosuccinic aciduria (ASLD). We developed a simplified analytical method of ornithin acid (ORA), a diagnostic marker in ornithine transcarbamylase deficiency (OTCD) and also CIT1 or ASLD, on conventional method of NBS. (Materials and reagents) Oral acid (ORA) standard was purchased from Sigma-Aldrich (St. Louis, MO), and [1,3-15N2]OA of internal standard (IS) was obtained from Cambridge Isotope Laboratories (Andover, MA, USA). NeoBlaze Non-derivatized MS/MS Kit (PerkinElmer) was used for analysis of other acid and acylcarnitine. Other chemicals were obtained from Wako (Japan). (Analytical system) LC/MS-8040 triple quadrupole mass spectrometer (Shimadzu, Kyoto) was used for analysis. Transitory settings: LC: Flow rate of mobile phase: 1 mL/min. MS/MS: Drying Gas Flow: 12 L/min. Nebulizing Gas Flow: 800 mL/min, Capillary: 3.5 kV, Drying Temp: 350 ℃, Exit Temp: 200 ℃, Cone: 30 ℃. (Analytical methods) HPLC: Sample injection volume: 1 µl. Flow rate of mobile phase: 1 mL/min. Drying Gas Flow: 12 L/min. (Materials and reagents) (Analytical methods) Hironori Kobayashi1), Kenji Yamada1), Yuki Hasegawa1), Tomoo Takahashi1), Toshikazu Minohata2), Seiji Yamaguchi1) 1) Department of Pediatrics, Shimane University Faculty of Medicine, Izumo, Japan 2) Shimadzu Corporation, Kyoto, Japan