Acta Medica Okayama

Volume 41, *Issue* 6 1987 *Article* 7

DECEMBER 1987

Transamination of L-cysteine sulfinate in the growing rat.

Shuichiro Akahori* Kohei Ejiri† Hirofumi Kanemori‡

Takafumi Kudo** Kaoru Sekiba††

Toshihiko Ubuka‡‡ Reiko Akagi§

*Okayama University,

†Okayama University,

‡Okayama University,

**Okayama University,

††Okayama University,

‡‡Okayama University,

§Okayama Prefectural Junior College,

Transamination of L-cysteine sulfinate in the growing rat.*

Shuichiro Akahori, Kohei Ejiri, Hirofumi Kanemori, Takafumi Kudo, Kaoru Sekiba, Toshihiko Ubuka, and Reiko Akagi

Abstract

The enzyme activities involved in the transamination of L-cysteine sulfinate (L-alanine 3-sulfinic acid), L-aspartate and L-cysteine were examined in fetal, neonatal and maternal rat liver and placenta. In fetal and neonatal rat liver, aminotransferase activity was most active with L-cysteine sulfinate as a substrate and was also active with L-aspartate, while activity with L-cysteine was very low. The activity of transamination of L-cysteine sulfinate in rat liver developed in parallel with that of L-aspartate and L-cysteine. The aminotransferase activity markedly increased after the 19th day of gestation, reaching the same value as adult liver on the 3rd day after birth. The ratios of transamination of L-cysteine sulfinate to that of L-aspartate and to that of L-cysteine were constant during development. These observations suggest that L-cysteine sulfinate, L-aspartate and L-cysteine are transaminated by the same enzyme in the rat liver during development. Since placental aminotransferase activity was extremely low compared with that of the liver, it was suggested that the placenta did not play an important role in the transamination of these amino acids during pregnancy.

KEYWORDS: L-cysteine sulfinate, transamination, rat liver, developmental change, placenta

---

*PMID: 3439482 [PubMed - indexed for MEDLINE]

Acta Med Okayama 41 (6) 279-283 (1987)

Transamination of L-Cysteine Sulfinate in the Growing Rat.

Shuichiro Akahori, Kohei Ejiri, Hirofumi Kanemori, Takafumi Kudo, Kaoru Sekiba, Toshihiko Ubuka* and Reiko Akagi**

*Departments of Obstetrics and Gynecology, and *Biochemistry, Okayama University Medical School, and **Okayama Prefectural Junior College, Okayama 700, Japan*

The enzyme activities involved in the transamination of L-cysteine sulfinate (L-alanine 3-sulfinic acid), L-aspartate and L-cysteine were examined in fetal, neonatal and maternal rat liver and placenta. In fetal and neonatal rat liver, aminotransferase activity was most active with L-cysteine sulfinate as a substrate and was also active with L-aspartate, while activity with L-cysteine was very low. The activity of transamination of L-cysteine sulfinate in rat liver developed in parallel with that of L-aspartate and L-cysteine. The aminotransferase activity markedly increased after the 19th day of gestation, reaching the same value as adult liver on the 3rd day after birth. The ratios of transamination of L-cysteine sulfinate to that of L-aspartate and to that of L-cysteine were constant during development. These observations suggest that L-cysteine sulfinate, L-aspartate and L-cysteine are transaminated by the same enzyme in the rat liver during development.

Since placental aminotransferase activity was extremely low compared with that of the liver, it was suggested that the placenta did not play an important role in the transamination of these amino acids during pregnancy.

Key words : L-cysteine sulfinate, transamination, rat liver, developmental change, placenta

The transaminative pathway of cysteine metabolism functions in various rat tissues, though it is not very active (1). The first step of the pathway is cysteine transamination which is catalyzed by cysteine aminotransferase (EC 2. 6. 1. 3). Cysteine aminotransferase purified from rat liver exhibited the highest activity with L-cysteine sulfinate (L-alanine 3-sulfinate) as a substrate and the second highest with L-aspartate. It was concluded that cysteine aminotransferase was identical with aspartate aminotransferase (EC 2. 6. 1. 1) (2,3).

It has been shown that taurine, the most abundant amino acid in fetal and newborn rats, is not synthesized in these stages of development (4). The present paper deals with the developmental pattern of the transamination of sulfur amino acids, with special reference to L-cysteine sulfinate, in the rat liver and placenta.

Materials and Methods

Animals. Female Wistar rats weighing about 200 g were paired with male animals for 12 h (the first day of gestation). Pregnant animals were isolated and allowed commercial rat chow (MF of Oriental Yeast Co. Ltd., Tokyo, Japan) and water ad libitum. The pregnant rats were killed by decapi-

tation at the apropriate gestational age, and the whole fetus, fetal liver, placenta and maternal liver were quickly removed. The liver of the neonatal rats, on the appropriate postnatal day, and of nonpregnant rats weighing about 200 g were removed in the same manner.

Materials. L-Cysteine sulfinate was prepared according to a previously reported method (5). L-Aspartate, L-cysteine, monosodium 2-oxoglutarate and pyridoxal 5'-phosphat (PLP) were obtained from Sigma Chemical Company, St. Louis, Mo., U. S. A. Nicotinamide adenine dinucleotide (NAD^+) and adenosine 5'-diphosphate (ADP) were products of Oriental Yeast Co. Ltd., Tokyo, Japan. Glutamate dehydrogenase (EC 1. 4. 1. 3) (in 50% glycerol) was purchased from Boehringer Manheim GmbH, West Germany.

Enzyme assay. All steps were carried out at 4°C. The fetal, neonatal and maternal rat liver and placenta were washed with cold saline (0.9% NaCl containing 1 mM EDTA) immediately after they were removed. These organs were homogenized with 3 volumes of 20 mM potassium phosphate buffer (pH 7.4) using a Potter-Elvehjem homogenizer equipped with a Teflon pestle. The aminotransferase activities in the homogenate were assayed by determining the amount of L-glutamate (6) formed from 2-oxoglutarate in the presence of L-cysteine sulfinate, L-aspartate or L-cysteine (2).

Results

Developmental patterns of aminotransferase activities with L-alanine sulfinate, L-aspartate and L-cysteine as substrates. Figure 1 shows the aminotransferase activities in rat liver determined with L-cysteine sulfinate, L-aspartate and L-cysteine during development. The transamination of L-cysteine sulfinate was highest among these three substrates, followed by that of L-aspartate, throughout the fetal and neonatal period. The aminotransferase activity with L-cysteine as a substrate was about 0.3% of that with L-cysteine sulfinate.

Changes in the transamination of these three amino acids during development showed a similar pattern. Aminotransferase activity with L-cysteine sulfinate as a substrate increased approximately three-fold through the developmental stages studied. Significant increase in the activity started on the 19th day of gestation reaching the adult level one day after birth. The activity of the transamination of L-aspartate increased about three times during the same period. The ratio of the aminotransferase activity with L-cysteine sulfinate as a sub-

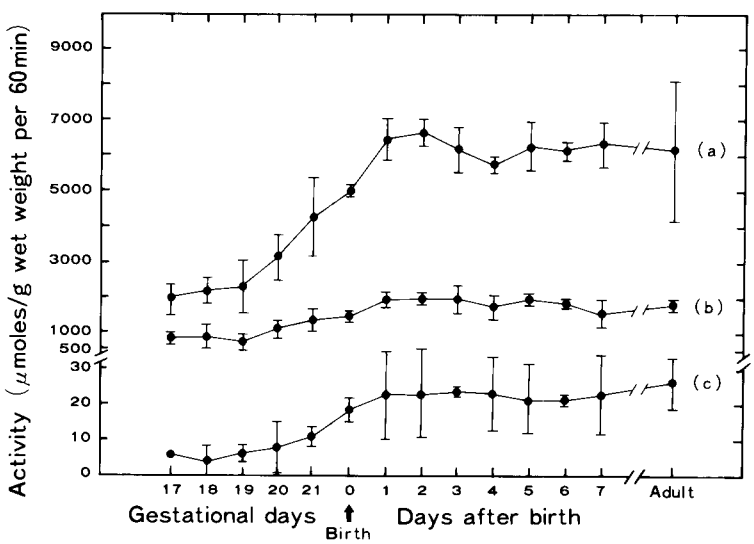

Fig. 1 Developmental change in aminotransferase activity with L-cysteine sulfinate (a), L-aspartate (b) and L-cysteine (c) as substrates in the fetal and neonatal rat liver. Activities are expressed as μmoles of L-glutamate formed/g wet weight/60 min. Values are the means±SEM of 5 determinations of pooled livers from 26 fetal and neonatal rats.

strate to that with L-aspartate did not change from around 3.0 during the period studied.

Although L-cysteine transamination was very low compared with that of the other two amino acids, the developmental pattern was similar.

Aminotransferase activity with L-cysteine sulfinate as a substrate in the placenta during pregnancy. Aminotransferase activity with L-cysteine sulfinate as a substrate was much lower in the placenta than in the liver.

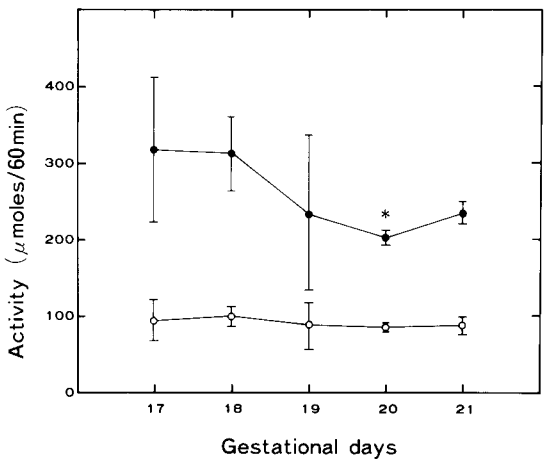

Fig. 2 Changes in aminotransferase activity with L-cysteine sulfinate as a substrate in the rat placenta during pregnancy. Activities are expressed as μmoles of L-glutamate formed/60 min. Values are the means±SEM of 5 determinations of 15 pooled placentas from the pregnant rats. Activity was expressed per gram wet weight (●) or as total activity in the whole placenta (○) *: Significantly different from the activities on 17th day of gestation as evaluated by Student's t test ($p < 0.05$).

The total aminotransferase activity with L-cysteine sulfinate in the whole placenta was constant irrespective of the age of the placenta. However, when expressed with respect to tissue weight, there was a significant decrease on the 20th day of gestation (Fig. 2). The ratio of the aminotransferase activity with L-cysteine sulfinate as a substrate to that with L-aspartate was higher in the placenta (5.0) than the ratio in the liver

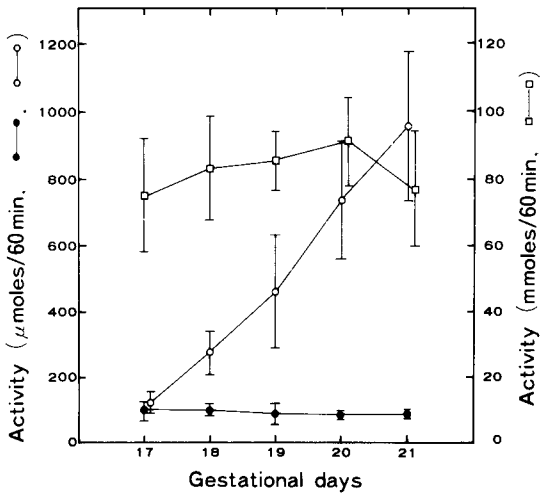

Fig. 3 Changes in the total aminotransferase activity with L-cysteine sulfinate as a substrate in the whole placenta (●), fetal liver (○) and maternal liver (□) of rats during pregnancy. Activities are expressed as μmoles of L-glutamate formed/60 min. Values are the means±SEM of 5 determinations.

of either fetal or pregnant rats.

Total transamination of L-cysteine sulfinate in the placenta and liver during pregnancy. Total aminotransferase activity with L-cysteine sulfinate as a substrate in the placenta, and fetal and maternal liver homogenates is summarized in Fig. 3.

On the 17th day of gestation, the total transamination of L-cysteine sulfinate per fetal liver was equal to that per placenta. Approximately 100 μmol of L-glutamate was formed per organ per 60 min. With the development of the fetus, the total transamination per fetal rat liver significantly increased approximately 8-fold by the 21st day of pregnancy. The increase may have been due to either the increase in the liver weight or the increase per unit weight, or both. The total transamination per placenta did not change during the same period of pregnancy. Likewise, the total transamination of L-cysteine sulfinate in maternal liver did not change during pregnacy.

Discussion

Recasens *et al.* reported that L-cysteine sulfinate aminotransferase may play a role in the regulation of the L-cysteine sulfinate level (7). Their findings prompted us to study the transamination of L-cysteine sulfinate during the development of rats.

In the present study, the transamination of L-cysteine sulfinate was detected at a high level in the homogenate of developing rat liver. Aminotransferase activities with L-cysteine sulfinate, L-aspartate or L-cysteine as a substrate were compared. The activity was highest with L-cysteine sulfinate, and was also high with L-aspartate (Fig. 1). The ratio of the aminotransferase activity with L-cysteine sulfinate as a substrate to that with L-aspartate was constant during development, suggesting that the transamination of these amino acids is catalyzed by the same enzyme. The present results agree with other reports (2,3,8). The identity of aspartate aminotransferase and cysteine aminotransferase in rat liver has been reported (2,3). In the present study, aminotransferase activity with L-cysteine as a substrate paralleled that with L-aspartate.

Aminotransferase activity with L-cysteine sulfinate was lower in the placenta than in the liver. The activity in the placenta tended to decrease with age. This result is in agreement with the report that aspartate aminotransferase activity decreased with age (9,10). However, total transamination of L-cysteine sulfinate in the placenta did not change during pregnancy.

The reason why the ratio of the aminotransferase activity with L-cysteine sulfinate as a substrate to that with L-aspartate in the placenta was higher than that in the liver remains unknown. It might be presumed that aspartate aminotransferase in the rat placenta has different properties from that in the liver. Further investigations, especially on the purification of aspartate aminotransferase from the placenta, are necessary.

The total aminotransferase activity with L-cysteine sulfinate as a substrate did not change in the placenta during pregnancy, but in the fetal rat liver, the activity increased significantly during the same period of pregnancy. On the 18th day of gestation, the total aminotransferase activity of the fetal rat liver exceeded that of the placenta. These results suggest that the placenta does not play an important role in the transamination of L-cysteine sulfinate during pregnancy.

References

1. Hosaki Y, Nishina H, and Ubuka T: Metabolism of L-cysteine in guinea pig liver. Acta Med Okayama (1986) **40**, 11-15.
2. Ubuka T, Umemura S, Yuasa S, Kinuta M, and Watanabe K: Purification and characterization of mitochondrial cysteine aminotransferase from rat liver. Physiol Chem Phys (1978) **10**, 483-500.
3. Akagi R: Purification and characterization of cysteine aminotransferase from rat liver cytosol. Acta Med Okayama (1982) **36**, 187-197.
4. Loriette C and Chatagner F: Cysteine oxidase and cysteine sulfinic acid decarboxylase in developing rat liver. Experientia (Basel) (1978) **34**, 981-982.
5. Ubuka T, Kinuta M, Akagi R, Kiguchi S, and Azumi M: Reaction of S-(2-amino-2-carboxyethylsulfonyl)-L-cysteine with sulfite: Synthesis of S-sulfo-L-cysteine and L-alanine 3-sulfinic acid and application to the determination of sulfite. Anal Biochem (1982) **126**, 273-277.
6. Bernt E and Bergmeyer HU: L-Glutamate, UV-assay with glutamate dehydrogense and NAD; in Methods of Enzymatic Analysis, Vol. 4, Bergmeyer eds, Academic Press Inc., New York (1974) pp 1704-1708.
7. Recasens M and Benezra R: An alternate CSA pathway regulationg taurine metabolism ? Adv Biochem Psychopharmacol (1981) **29**, 545-550.
8. Recasens M, Benezra R, Basset P and Mandel P: Cysteine sulfinate aminotransferase and aspartate aminotransferase isoenzymes of rat brain. Purification, characterization, and further evidence for identity. Biochemistry (1980) **19**, 4583-4589.
9. Remesar X, Arola L, Palou A and Alemany M: Activities of enzymes involved in amino acid metabolism in developing rat placenta. Eur J Biochem (1980) **110**, 289-293.

10. Diamant YZ and Shafrir E: Enzymes of carbohydrate and lipid metabolism in the placenta and liver of pregnant rats. Biochim. Biophys Acta (1972) **279**, 424-430.

Received: August 1, 1987
Accepted: September 22, 1987

Correspondence to:
Shuichiro Akahori
Department of Obstetrics and Gynecology
Okayama University Medical School
2-5-1 Shikatacho
Okayama 700, Japan