Age-matched male and female Long-Evans Agouti (LEA) rats were used as controls. There were 4 rats in each group. The animals were from the domestic line of the Division of Animal Research Resources, Institute of Health Biosciences, the University of Tokushima Graduate School (Tokushima, Japan). Both five-week-old and eight-week-old LEC rats were studied to represent the stage before the onset of acute hepatitis. They show Cu and Fe accumulation in the liver but not in the kidneys [14,15,19,21-24,28-32]. In this early stage several biochemical parameters related to hepatic dysfunction, such as the plasma aspartate transaminase and glutamic-pyruvic transaminase levels, are still within the normal ranges (data not shown [14,15,21,33-35]. All of the rats were fed a laboratory diet (CMF; Oriental Yeast Co. Ltd. Tokyo, Japan) and distilled water *ad libitum*, and were housed in a facility that was maintained at 22°C with a 12-h light-dark cycle. All animal procedures were approved by the Animal Care and Use Committees of Hokkaido Institute of Public Health. All procedures using rats were carried out according to the regulations of the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Extraction of total RNAs from the PBMCs

The 1.5mL blood samples were collected by using retro-orbital bleeding technique under ether-anesthesia. The collected samples were put into 15mL conical tubes (BD Falcon™, Franklin Lakes, NJ, USA) containing 100µL heparin solution (10,000 Units, FUSO Pharmaceutical Industries, Ltd. Osaka, Japan) and were vortexed well for approximately 15 sec. The total RNAs in the blood samples were extracted according to the protocol of QIAamp RNA Blood Mini Kit (Qiagen GmbH, Hilden, Germany). Concentration of each RNA sample was measured using NanoDrop ND-1000 spectrophotometer (NanoDrop Technologies). The RNA samples with 260/280 ratio (an indication of protein contamination) between 1.9 and 2.1 and 260/230 ratio (an indication of reagent contamination) greater than 2.0 were used for the analysis. The total RNA between 50 ng/µL and 500 ng/µL of could be obtained from 1.5 mL blood.

### Extraction of total RNAs from the liver tissue.

The animals were perfused transcardially with a solution of 40 mM Tris-20 mM HCl containing 152 mM NaCl (500 ml/kg) under ether-anesthesia. Their livers were quickly removed, shredded to the scale of about 1.0 × 1.0 × 1.0 mm, and put into RNAlater®. solution (Ambion®. Applied Biosystems). They were stored at 4°C until their total RNAs were extracted. The tissue pieces of the livers were homogenized by a micro-tube homogenizer (Model: 23M, Nippon Genetics, Tokyo, Japan). The total RNAs from the rat livers were extracted using an RNeasy Mini kit (Qiagen GmbH, Hilden, Germany) according to the manufacturer's instructions. The total RNA between 1000 ng/µL and 2000 ng/µL could be obtained from about 20 mg tissues treated with RNAlater® solution.

cDNA synthesis

cDNA syntheses were performed using a TaqMan® Reverse Transcription Reagents kit (Applied Biosystems, Branchburg, NJ, USA) in a total 100 µL reaction mixture containing 1.0 µg of the total RNA samples. The RT-PCR mixtures were incubated at 25°C for 10 min and at 48°C for 30 min, and the reaction were terminated by heating at 95°C for 5 min. The cDNAs were kept at 4°C while quantitative RT-PCR analyses were performed for one week.

Quantitative RT-PCR

The primer and fluorescent-labeled probe sets for Atp7b and metallothionein-1a (Mt-1a) were designed by Biosearch Technologies
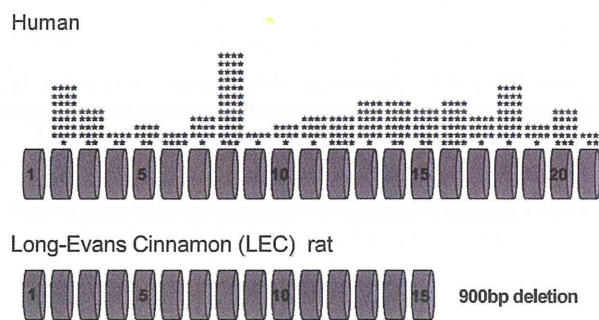
Figure 1: Gene structures on the ATP7B of humans and on the Atp7b of Long-Evans Cinnamon (LEC) rats. In humans, the gene structure is composed of 21 exons. However, 6 exons after exon-15 were completely deleted in the LEC rats. The asterisk (*) marks indicate the mutation points cited from the Wilson disease mutation database in the University of Alberta curated by Bugbee *et. al.* [12].

Japan (Tokyo, Japan). The 5'-fluorogenic reporter of the probe was FAM, and the 3'-fluorogenic quencher was Black Hole Quencher (BHQ-1).

The primer sets for Atp7b mRNA were as follows: forward primer: 5'-TGCCTGAACAGGAGAGAAAGGT-3', reverse primer: 5'-TCGTGTGGGCAAAGCAAGT-3', fluorescent-labeled probe: 5'-AGCCAAAGAGGCCAGTCGGAAAATCTTATC-3', respectively. These primer and probe sets were designated with the exon-exon junction between exon-1 and exon-2 of NM_012511 in order to focus on the specific region of Atp7b mRNA using this uniquely designed set. The gene structures of ATP7B in humans and of Atp7b in LEC rats are shown in Figure 1. In humans, the gene structure is composed of 21 exons. However, 6 exons after exon-15 were completely deleted in the LEC rat ([36], Professor T. Agui, unpublished data; personal communications). We focused on the exon-exon junction between exon-1 and -2. This is because any mutation is very rarely detected in the region of human exon-1. Therefore, we hypothesized that it was possible to detect mRNA synthesis around the region of Atp7b even with mutation, especially in LEC rats.

The primer sets for rat Mt-1a mRNA were as follows: forward primers: 5'-GAACTGCAAATGCACCTCCTG -3', reverse primers: 5'-ACACAGCCCTGGGCACAT-3', fluorescent-labeled probe: 5'-AAGAAGAGCTGCTGCTCCTGCTGCC-3', respectively. These primers and probe sets were located at the exon-exon junction between 2 and 3 of NM_138826.

All the primers and probes were made by Biosearch Technologies, Inc. (Novato, CA, USA). All of them were purchased through Biosearch Technologies Japan. The rat ß-actin was utilized as an endogenous standard mRNA (EC-R-3-1000-C, accession No. NM_031144.2) as follows: forward primer: 5'-TGCCCCGAGGCTCTCTT-3', reverse primer: 5'-GATGGAATTGAATGTAGTTTCATGGA-3', fluorescent-labeled probe: 5'- CAGCCTTCCTTCCTGGGTATGGAATCC-3', respectively.

Our quantitative RT-PCR was performed with 2 µL of cDNA sample, 25 µL of 2 X TaqMan Universal PCMaster Mix (Applied Biosystems, Foster City, CA, USA), 900 nM of each primer and 250 nM of probe in a total 50 µL reaction mixture. The reactions were carried out on an ABI PRISM® 7700 Sequence Detection System (Applied Biosystems, Foster City, CA, USA) with the following thermal cycling conditions: 2 min at 50°C, 10 min at 95°C, followed by 40 repeats of 15 sec at 95°C and 1 min at 60°C. The data of RT-PCR were analyzed by the sequence detector software version 1.6.3 (Applied Biosystems, Foster City, CA, USA).

	Male		Female	
	LEA	LEC	LEA	LEC
Atp7b	1.00 ± 0.28	0.31 ± 0.06*	1.00 ± 0.25	0.50 ± 0.14*,+
Mt-1a	1.00 ± 0.36	3.13 ± 0.48*	1.00 ± 0.19	9.18 ± 2.47*,++

*p < 0.001 versus sex-matched LEA rats.
+p < 0.05 and ++p<0.005 versus male LEC rats

Table 1: Relative quantities of mRNA variations in the livers between the LEA and LEC rats at the age of 8 weeks (n=4).

Relative mRNA levels were determined using the comparative C_T (threshold cycle) method. The expression of Atp7b and Mt-1a mRNAs was normalized to ß-actin as an endogenous control gene. C_{TS} was calculated by subtracting the C_T value of the endogenous control (C_{TR}) from the C_T value of the sample (ΔC_T; $\Delta C_{CT} = C_{TS} - C_{TR}$). The relative expression ($2^{-\Delta\Delta C_T}$) to a calibrator was determined by subtracting the $\Delta C_{T(Calibrator)}$ from the ΔC_T value [$\Delta\Delta C_T = \Delta C_T - \Delta C_{T(Calibrator)}$]. The samples and endogenous controls were run in duplicate.

Statistics

In this study, all statistical analyses were performed with the use of statistical software (JMP 7 for Macintosh; SAS Institute Japan Inc., Tokyo, Japan). One-way analysis-of-variance (ANOVA) was employed to identify statistically significant differences between two groups. Differences with P values of less than 0.05 were considered to be statistically significant.

Results and Discussion

The data of quantitative real-time polymerase chain reaction (quantitative RT-PCR) in the livers are indicated in Table 1. Using quantitative RT-PCR, the expression levels of Atp7b and metallothionein 1a (Mt-1a) mRNAs were analyzed in the livers of both the LEC and LEA rats. Concerning the levels of Atp7b mRNA in the LEC rat livers, the two following reports were published. The first, reported by Wu et al. revealed that RT-PCR analyses were detectable as weak bands [17]. The second, reported by Yamaguchi et al. demonstrated that Northern blot analyses indicated that they were absent in the LEC rat livers [18]. Our results support the former because our quantitative RT-PCR analyses detected the expression of Atp7b mRNA. However, the levels in the LEC rat livers at the age of 8 weeks reduced to 0.31 and to 0.50 against the controls in males and females, respectively. These results suggest that a part of mutant Atp7b mRNA exists in the LEC rat livers while it was significantly down-regulated compared to the controls.

Metallothioneins (MTs) are low molecular weight proteins. They can capture heavy metals, such as Cu, zinc, cadmium and mercury. In the LEC rat liver, the abnormal accumulated Cu ions bind to MTs [21]. Our results concerning hepatic Mt-1a, one of the MT isoforms, were reasonable because the mRNA levels of Mt-1a were elevated to 3.13 and to 9.18 against the controls in males and females, respectively. We detected a novel gender difference concerning the induction levels of Mt-1a in the LEC rat livers. Several gender differences concerning the LEC rat were found. One of the most important gender differences was crucial at the acute hepatitis stage in the LEC rat [35]. The onset of acute hepatitis in the female LEC rats occurs at a younger age than in the male rats. The mortality of the female rat is higher than that of the male rat. Regarding the gender differences, the severity observed at the acute hepatitis stage might be related to that of the induction levels of Mt-1a in the LEC rat livers. In human, the gender differences were observed that the prevalence of severe hepatic dysfunction is higher in females than in men [37,38]. The expression of MTs might be related to the tolerance of hepatocytes against Cu toxicity [37]. We also revealed gender differences of porphyrin and heme metabolisms in the LEC rats [39].

The data of quantitative RT-PCR in the peripheral blood mononuclear cells (PBMCs) are indicated in Figures 2 and 3. The expression levels of Atp7b (Figure 2) and Mt-1a (Figure 3) mRNAs in the PBMCs of both the LEC and LEA rats were analyzed. Concerning the levels of Atp7b mRNA in the PBMCs of the LEC rats, our results indicated that the levels of the LEC rats at the age of 5 weeks significantly up-regulated to 7.94 and to 13.0 against the controls in males and females, respectively. Their levels at the age of 8 weeks were significantly increased to 13.7 and to 9.82 against the controls in males and females, respectively. At both ages, novel gender differences were detected concerning the induction of the Atp7b mRNA levels in the PBMCs of the LEC rats. Our findings were the first to report the following: 1) the revelation of the high expression of the Atp7b mRNA levels in the PBMCs of the LEC rats; and 2) the confirmation of the existence of novel gender differences concerning their high expression levels. These results suggest that a part of mutant Atp7b mRNA up-regulates in the PBMCs of the LEC rats, although it is significantly down-regulated in their livers.

In the PBMCs of the LEC rats, the mRNA levels of Mt-1a at the age of 5 weeks were 1.47 and 1.13 against the controls in males and females, respectively (Figure 3). There were no significant differences at this age. Their levels at the age of 8 weeks were significantly increased to 3.32 and to 1.76 against the controls in males and females, respectively. In addition, the gender difference was confirmed concerning the induction levels of Mt-1a in the PBMCs of LEC rats at the age of 8 weeks. Their gender differences suggest the variations in concerning the self-defense mechanisms, for example, against free or loosely binding Cu ions in the blood and/or oxidative stresses in the PBMCs of the LEC rats.
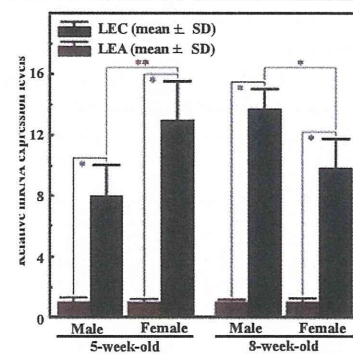
Figure 2: Quantitative RT-PCR analyses of Atp7b mRNA in the peripheral blood mononuclear cells of the LEA and LEC rats (n = 4). The mRNA levels of Atp7b were determined by using real-time RT-PCR and normalized to the expressions of their ß-actin mRNAs. *p < 0.001; **p < 0.05.
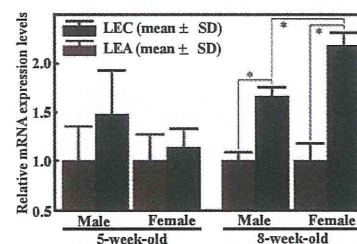
Figure 3: Quantitative RT-PCR analyses of Mt-1a mRNA in the peripheral blood mononuclear cells of the LEA and LEC rats (n = 4). The mRNA levels of Mt-1a were determined by using real-time RT-PCR and normalized to the expressions of their ß-actin mRNAs. *p < 0.001.

Conclusion

In this study, we focused on the expression level of Atp7b mRNA in the PBMCs of the LEC rats. These analyses were the first trials of the measurement of Atp7b mRNA levels in the blood. At the ages of 5 and 8 weeks, the high expressions of Atp7b mRNA were manifested in the PBMCs of both male and female LEC rats. We consider that the Atp7b mRNA in the PBMCs of the LEC rat is composed of two parts, the normal and deleted parts. However, the synthesis of the normal part in the mutant Atp7b mRNA was accelerated in the PBMCs of the LEC rat. These results are novel and unique because their levels in the livers are significantly lower than those of the LEA rats. These phenomena were discovered for the first time in the LEC rats. At present, we are yet to consider the details of these mechanisms. However, these results suggest the diversity of cell-physiological and endocrinological Cu metabolisms between the PBMCs and the livers of the LEC rats. Our findings indicate the possibility of a novel Cu metabolism in the cardiovascular network that is concerned with Atp7b of the PBMCs. Further studies will need to be done to establish the hypothesis that there is a novel Cu metabolism in the cardiovascular network though Atp7b in the PBMCs.

Upon incubation of PBMCs with the medium containing Cu ions, PBMCs incorporated and accumulated them as Cu-MTs. Our results, showing the increment of Mt-1a mRNAs in the PBMCs of the LEC rats rather than those of the controls, agreed with that phenomenon. The Cu-MTs formation was more pronounced in monocytes than in granulocytes [40]. Recently, unique phenomena were discovered concerning the Cu metabolism in the PBMCs [41]. Those were: the mRNA up-regulation of Atp7a (disease-causing gene of Menkes disease) was revealed in the THP-1 monocytes, suggesting a role in the oxidation of low density lipoproteins. The inductively coupled plasma mass spectrometry (ICP-MS) analyses of Cu in murine PBMCs showed markedly increased intracellular Cu levels in the cells isolated from Atp7a-deficient mice versus control mice [42]. Those results are strongly supported our findings. Therefore, those findings are suggesting a possibility that both Atp7b and Atp7a function as a compensatory mechanism concerning Cu excretion from the PBMCs.

Finally, we believe it is necessary to confirm the expression levels of ATP7B mRNAs in the PBMCs of the patients with WND using the primer and fluorescent-labeled probe set designed at the exon-exon junction between exon-1 and 2.

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XII ライソゾーム病

ゴーシェ病
Gaucher disease

Key words：ゴーシェ病，酵素補充療法，ライソゾーム病

井田博幸

1. 疾患概念

　ゴーシェ病はライソゾーム酵素であるグルコセレブロシダーゼ活性低下により発症するライソゾーム病である．肝脾腫を主症状とし，神経症状の有無とその重症度により1型（慢性非神経型，成人型），2型（急性神経型，乳児型），3型（亜急性神経型，若年型）に臨床分類されている．このような臨床的異質性が存在するが，更にこれら各病型内においても種々の表現型の差異が存在するのが本症の特徴である．

2. 疫　　学

　ユダヤ人に好発し，その頻度は約1,000人に1人である．日本人では50万〜100万人に1人

と報告されている．1992年から現在までに当科で診断されたゴーシェ病は約130人である．

3. 病　　因

　グルコセレブロシダーゼの遺伝的酵素活性低下により，その基質であるグルコセレブロシドが主として肝臓，脾臓，骨髄などの細網内皮系に蓄積することによって発症する．神経症状はグルコセレブロシドのリゾ体であるグルコシルスフィンゴシンの脳内蓄積により引き起こされる．その病態生理を図1にまとめる．

4. 臨床症状

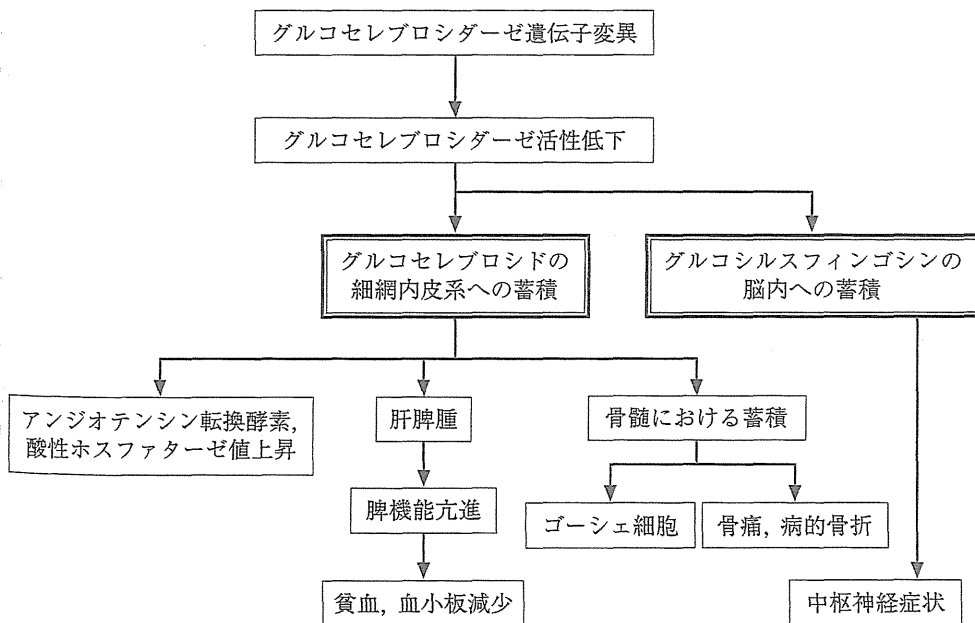　前述したように神経症状の有無とその重症度により，臨床上，1型，2型，3型に分類されて

XII

ライソゾーム病

図1　ゴーシェ病の基本病態

Hiroyuki Ida: Department of Pediatrics, Jikei University School of Medicine　東京慈恵会医科大学　小児科

表1　ゴーシェ病の臨床病型

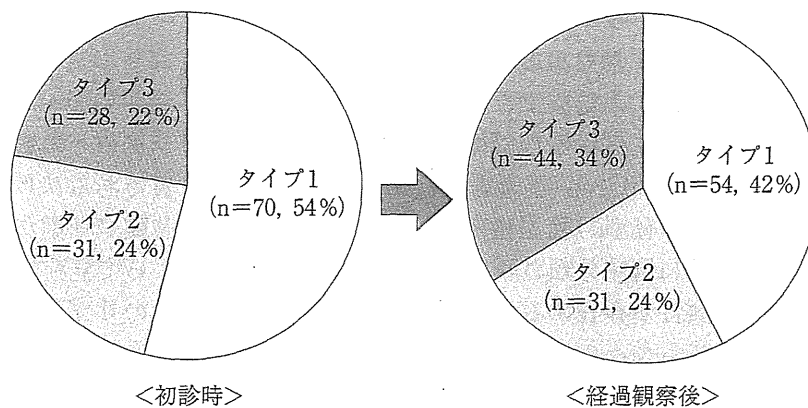|	1型 慢性非神経型	2型 急性神経型	3型 亜急性神経型
発症時期	幼児～成人	乳児	乳児～学童
神経症状	(－)	(＋＋＋)	(＋)～(＋＋)
肝脾腫	(－)～(＋＋＋)	(＋)	(＋)～(＋＋＋)
骨症状	(－)～(＋＋＋)	(－)	(－)～(＋＋＋)
予後	良好	不良	症例により異なる

図2　日本人ゴーシェ病の病型変化(n=129)

いる．これら病型により経過，予後は異なる（表1）．自験例による129例のデータでは日本人ゴーシェ病は1型が42％，2型が24％，3型が34％と神経型ゴーシェ病の頻度が高いことが特徴である（経過観察後のデータによる）．

1型は神経症状を伴わない病型で肝脾腫，血小板減少，骨症状を主症状とする．発症年齢，骨合併症の有無，肝脾腫の程度などの点において臨床的異質性が顕著な病型である．ユダヤ人では極めて慢性に経過するが，日本人では進行性であることが明らかにされている．我が国において酵素補充療法を行っている経過中に神経症状を呈し，3型に再分類される例が1型の約20％報告されているので，1型と診断されても異常眼球運動や精神運動発達遅滞などの神経症状の発症に注意する（図2）．これら1型から3型に移行する症例においてはL444P/L444P，L444P/F213Iという遺伝子型が高頻度に認められる[1]．

2型は乳児期に発症し，肝脾腫に加えて痙攣，後弓反張，喉頭痙攣，異常眼球運動などの神経症状を呈し，これら神経症状が急速に進行する病型である．2型の最重症型として胎児水腫・コロジオンベビーとして発症する新生児型も存在する[2]．

3型は肝脾腫に加えて神経症状を伴うが，その発症は2型に比較して遅く，またその程度や進行も緩徐な病型である．3型は3a，3b，3cの亜型に分類されている．3a型は古典的な3型で肝脾腫に加えて痙攣，ミオクローヌス，小脳失調，眼球運動失行などの神経症状を呈する．3b型は核上性上方注視麻痺を唯一の神経症状とし，それに加えて重篤な臓器症状(巨大脾腫，骨折・骨痛，呼吸器症状など)を呈する病型である．早期発症の1型との鑑別が困難な病型である[3]．3c型は水頭症，角膜混濁，心弁膜石灰化などユニークな臨床症状を呈する病型である[4]．

5. 診断と鑑別診断

肝脾腫と血小板減少を認めた場合，骨髄穿刺

を行い腫瘍性疾患が否定されて，かつゴーシェ細胞が存在すれば診断はほぼ確実である．酸性ホスファターゼ値の上昇，アンジオテンシン変換酵素値の上昇などの所見は本症の診断を支持する．確定診断は培養皮膚線維芽細胞のグルコセレブロシダーゼ活性低下を証明することによってなされる．通常，酵素活性は正常の10％以下である．出生前診断は培養羊水細胞のグルコセレブロシダーゼ活性測定により可能である．鑑別診断としてはNiemann–Pick病A型，B型があるが，骨髄中のNiemann–Pick細胞の存在やスフィンゴミエリナーゼ活性測定により鑑別が可能である．Niemann–Pick病C型は培養皮膚線維芽細胞のフィリピン染色により鑑別が可能である．遺伝子診断は遺伝子変異の集積性が高いユダヤ人では有用であるが，日本人においてはL444P変異が約35％，F213I変異が約15％でcommon mutationのスクリーニングで同定できない変異が約40％存在するため，その有用性は低い[5]．

6.　治療と予後

治療法としては我が国では酵素補充療法と骨髄移植が実地臨床上行われている．1型に対してこれら治療は有効でありその予後は良好である．これに対して2型に対する酵素補充療法や骨髄移植の有効性は乏しく，2型の予後は不良である．3型に対する酵素補充療法は神経症状が重篤でない症例のみに有効である．診断時に神経症状を呈する3型は一般的に酵素補充療法に抵抗性で予後は不良である．これに対して1型から3型に移行した症例では酵素補充療法により生命的予後は比較的良好であるが，神経症状の改善は認められない．

　酵素補充療法としてはグルコセレブロシダーゼcDNAをCHO（Chinese hamster ovary）細胞に導入して作製した酵素製剤（商品名：セレザイム®）が現在，我が国で用いられている．日本ではすべての病型に適応が認められており，初期投与量として病型，年齢にかかわらず1回60単位/kgを2週間ごとに点滴静注する．症状の改善により投与量は漸減できるが骨症状の改善には初期投与量を少なくとも2年間，継続することが推奨されている．この骨症状の評価には単純X線では不十分で，大腿骨頭から膝関節を含めたMRIが必須である．安易な酵素量の減量は骨症状の発症をきたすので注意する[6]．

　骨髄移植は1型にも適応はあるが酵素補充療法が有効かつ安全なので，1型に対しては第一選択ではない．欧米において3型に対する報告が散見される．それによると肝脾腫，血液学的異常，低身長の改善のみならず神経症状の進行停止が認められている[7]．したがって，その危険性や侵襲を考えると，骨髄移植の適応は酵素補充療法を行っても神経症状が進行する全身状態の良好な症例である．

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XII

ライソゾーム病

Gaucher 病

■ SUMMARY

Gaucher 病（OMIM #230800）は，グルコセレブロシダーゼ活性低下により発症する，常染色体劣性遺伝形式をとるライソゾーム病である．その結果，グルコセレブロシダーゼの基質であるグルコセレブロシドが肝臓，脾臓，骨髄などの細網内皮系に蓄積するので肝脾腫を主症状とする．神経症状の有無と重症度により1型，2型，3型に分類される．神経症状はグルコセレブロシドのリゾ体であるグルコシルスフィンゴシンの脳内蓄積により引き起こされる．このような臨床的異質性が存在するが，さらにこれら各病型内においても種々の表現型の差異が存在するのが本症の特徴である．

治療法としては，日本では酵素補充療法と骨髄移植が実地臨床上行われている．1型に対してこれら治療は有効であるが，2型に対しては有効性に乏しく，3型に対しては神経症状が重篤でない症例のみに有効である．日本では現在まで約130人の患者が診断されている．

■ 代謝障害と病態

図1[1]に Gaucher 病の病態を示す．グルコセレブロシダーゼ遺伝子変異によって引き起こされるグルコセレブロシドとグルコシルスフィンゴシンの蓄積が種々の症状，検査値異常の原因である．

■ 臨床症状，臨床病型

病型により経過，予後は異なる（**表1**）[1]．自験例による129例のデータでは，日本人 Gaucher 病は1型が41.8％，2型が24.0％，3型が44.1％と，神経型 Gaucher 病の頻度が高いことが特徴である．
①1型：神経症状を伴わない病型で，肝脾腫，血小板減少，骨症状を主症状とする．発症年齢，骨合併症の有無，肝脾腫の程度などの点において臨床的異質性

表1 Gaucher 病の臨床病型

	1型 慢性非神経型	2型 急性神経型	3型 亜急性神経型
発症時期	幼児～成人	乳児	乳児～学童
神経症状	－	⧺	＋～⧺
肝脾腫	－～⧺	＋	＋～⧺
骨症状	－～⧺	－	－～⧺
予後	良好	不良	症例により異なる

（井田博幸，2011[1]）

が顕著な病型である．日本人はユダヤ人に比較して重症例が多い．また，日本においては，酵素補充療法を行っている経過中に神経症状を呈し，3型に再分類される例が1型の約20％に報告されているので，1型と診断されても異常眼球運動や精神運動発達遅滞などの神経症状の発症に注意する[2]．
②2型：乳児期に発症し，肝脾腫に加えてけいれん，後弓反張，喉頭けいれん，異常眼球運動などの神経症状を呈し，これら神経症状が急速に進行する病型である．2型の最重症型として，胎児水腫，コロジオンベビーとして発症する新生児型も存在する[3]．
③3型：肝脾腫に加えて神経症状を伴うが，その発症は2型に比較して遅く，またその程度や進行も緩徐な病型である．3型は 3a，3b，3c の亜型に分類されている．

3a型：古典的な3型で，肝脾腫に加えてけいれん，ミオクローヌス，小脳失調，眼球運動失行などの神経症状を呈する．
3b型：核上性上方注視麻痺を唯一の神経症状とし，それに加えて重篤な臓器症状（巨大脾腫，骨折・骨痛，呼吸器症状など）を呈する病型である[4]．早期発症の1型との鑑別が困難な病型である．
3c型：水頭症，角膜混濁，心弁膜石灰化など特徴的な臨床症状を呈する病型である[5]．
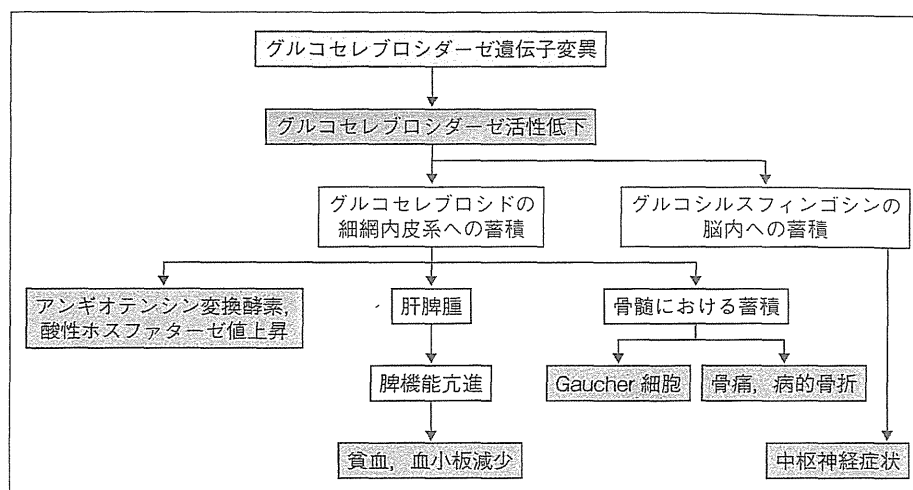
■ 診断

①一般検査：貧血，血小板減少，血清酸性ホスファタ

図1 Gaucher病の基本病態 (井田博幸. 2011[1])

ーゼ高値，アンギオテンシン変換酵素高値が認められる．

②**特殊検査**：肝脾腫と上記の一般検査所見の異常が認められた場合は，骨髄穿刺を行い Gaucher 病の存在を確認する．

③**確定診断**：確定診断は培養皮膚線維芽細胞のグルコセレブロシダーゼ活性低下を証明することによってなされる．通常，酵素活性は正常の 10％以下である．遺伝子診断は遺伝子変異の集積性が高いユダヤ人では有用であるが，日本人においては L444P 変異が約 35％，F213I 変異が約 15％で common mutation のスクリーニングで同定できない変異が約 40％存在するため，その有用性は低い[6]．

治療法

現在，日本では酵素補充療法と骨髄移植が健康保険収載されており，実地臨床の場で用いられている．

グルコセレブロシダーゼ cDNA を CHO（Chinese Hamster Ovary）細胞に導入して作成した酵素製剤（セレザイム®）を点滴静注する治療法が酵素補充療法である．日本ではすべての病型に適応が認められており，初期投与量として病型，年齢にかかわらず1回 60 単位/kg を2週間ごとに点滴静注する．

骨髄移植は1型にも適応はあるが，酵素補充療法が有効かつ安全なので，1型に対しては第1選択では

ない．欧米において3型に対する報告が散見される．それによると，肝脾腫，血液学的異常，低身長の改善のみならず神経症状の進行停止が認められている．したがって，その危険性や侵襲を考えると，骨髄移植の適応は酵素補充療法を行っても神経症状が進行する全身状態の良好な症例である．

予後

酵素補充療法により治療された1型の予後は良好である．2型は治療抵抗性で，予後はきわめて不良である．診断時に神経症状を呈する3型は一般的に酵素補充療法に抵抗性で，予後は不良である．これに対して，1型から3型に移行した症例では，酵素補充療法により生命的予後は比較的良好であるが，神経症状の改善は認められない[2]．

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（井田博幸）

225

IX 造血器腫瘍と類縁疾患

組織球増殖性疾患
リピドーシス

Gaucher病
Gaucher disease

Key words：Gaucher病，酵素補充療法，リソソーム病

井田博幸

1. 疾患概念

Gaucher病はリソソーム酵素であるグルコセレブロシダーゼ活性低下により発症するリソソーム病である．肝脾腫を主症状とし，神経症状の有無とその重症度により1型（慢性非神経型），2型（急性神経型），3型（亜急性神経型）に臨床分類されている．

血液領域では貧血と血小板減少を主症状とする1型が問題となるので，本稿では1型を中心に概説する．

2. 疫学

ユダヤ人に好発し，その頻度は約1,000人に1人である．しかも，その大多数が1型である．これに対して，日本人では全病型で50万〜100万人に1人と頻度は極めて低い．更に後述するように日本人では欧米人，特にユダヤ人に比較して1型の頻度が少ないことが特徴である．

3. 病因

グルコセレブロシダーゼ遺伝子変異によりグルコセレブロシダーゼ活性が低下する．その結果，その基質であるグルコセレブロシドが主として肝臓，脾臓，骨髄などの細網内皮系に蓄積することによって発症する．日本人とユダヤ人のグルコセレブロシダーゼ遺伝子変異分布を図1に示す．ユダヤ人Gaucher病ではN370S変異が全変異の約70％を占めている．これに対して日本人Gaucher病ではN370S変異は全く認められない[1]．N370S変異は1型にリンクする．また，N370S変異を有する1型Gaucher病患者は軽微な臨床症状を呈することが知られている[2]．このgenotype/phenotype相関のため，後述するように日本人Gaucher病1型はユダヤ人に比較して異なる臨床表現型を呈する．

4. 臨床症状

1型，2型，3型の経過，予後は異なる（表1）．日本人Gaucher病129例の診断時の病型は1型が54％，2型が24％，3型が22％と神経型Gaucher病の頻度が高いことが特徴である（図2）．日本人Gaucher病の1型の中には経過観察中に3型に移行する例もあり最終的に1型の頻度はこれより更に低くなる[3]．

1型は神経症状を伴わない病型で肝脾腫，血小板減少，骨症状を主症状とする．発症年齢，骨合併症の有無，肝脾腫の程度などの点において臨床的異質性が顕著な病型である．病因で記載したように日本人Gaucher病ではN370S変異が存在しないため，日本人1型Gaucher病はユダヤ人Gaucher病に比較して臨床症状は重症で，かつ進行性である[4]（表2，3）．

2型は乳児期に発症し，肝脾腫に加えて痙攣，後弓反張，喉頭痙攣，異常眼球運動などの神経症状を呈し，これら神経症状が急速に進行する病型である．

3型は肝脾腫に加えて神経症状を伴うが，その発症は2型に比較して遅く，またその程度や進行も緩徐な病型である．古典的な3型は肝脾

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