

# Efficacy and safety of metyrosine in pheochromocytoma/paraganglioma: a multi-center trial in Japan

Mitsuhide Naruse<sup>1)</sup>, Fumitoshi Satoh<sup>2)</sup>, Akiyo Tanabe<sup>3)</sup>, Takahiro Okamoto<sup>4)</sup>, Atsuhiko Ichihara<sup>5)</sup>, Mika Tsuiki<sup>6)</sup>, Takuyuki Katabami<sup>7)</sup>, Masatoshi Nomura<sup>8)</sup>, Tomoaki Tanaka<sup>9)</sup>, Tadashi Matsuda<sup>10)</sup>, Tsuneo Imai<sup>11)</sup>, Masanobu Yamada<sup>12)</sup>, Tomohiro Harada<sup>13)</sup>, Nobuyuki Kawata<sup>13)</sup> and Kazuhiro Takekoshi<sup>14)</sup>

<sup>1)</sup> Clinical Research Institute for Endocrinology and Metabolic Diseases, National Hospital Organization Kyoto Medical Center, Kyoto 612-8555, Japan

<sup>2)</sup> Division of Nephrology, Endocrinology and Vascular Medicine, Tohoku University Hospital, Sendai 980-8574, Japan

<sup>3)</sup> Department of Diabetes, Endocrinology and Metabolism, National Center for Global Health and Medicine, Tokyo 162-8655, Japan

<sup>4)</sup> Department of Surgery II, Tokyo Women's Medical University, Tokyo 162-8666, Japan

<sup>5)</sup> Department of Endocrinology and Hypertension, Tokyo Women's Medical University, Tokyo 162-8666, Japan

<sup>6)</sup> Department of Endocrinology and Metabolism, National Hospital Organization Kyoto Medical Center, Kyoto 612-8555, Japan

<sup>7)</sup> Department of Metabolism and Endocrinology, St. Marianna University School of Medicine, Yokohama City Seibu Hospital, Yokohama 241-0811, Japan

<sup>8)</sup> Department of Endocrine and Metabolic Diseases, Kyushu University Hospital, Fukuoka 812-8582, Japan

<sup>9)</sup> Department of Molecular Diagnosis, Chiba University Graduate School of Medicine, Chiba 260-8670, Japan

<sup>10)</sup> Department of Urology and Andrology, Kansai Medical University Hospital, Hirakata 573-1191, Japan

<sup>11)</sup> National Hospital Organization, Higashinagoya National Hospital, Nagoya 465-8620, Japan

<sup>12)</sup> Department of Medicine and Molecular Science, Gunma University Graduate School of Medicine, Maebashi 371-8511, Japan

<sup>13)</sup> Ono Pharmaceutical Co., Ltd., Osaka 541-8564, Japan

<sup>14)</sup> Faculty of Medicine, University of Tsukuba, Tsukuba 305-8577, Japan

**Abstract.** To assess the efficacy, safety, and pharmacokinetics of metyrosine (an inhibitor of catecholamine synthesis) in patients with pheochromocytoma/paraganglioma (PPGL), we conducted a prospective, multi-center, open-label study at 11 sites in Japan. We recruited PPGL patients aged  $\geq 12$  years requiring preoperative or chronic treatment, receiving  $\alpha$ -blocker treatment, having baseline urinary metanephrine (uMN) or normetanephrine (uNMN) levels  $\geq 3$  times the upper limit of normal values, and having symptoms associated with excess catecholamine. Metyrosine treatment was started at 500 mg/day and modified according to dose-adjustment criteria up to 4,000 mg/day. The main outcome measure was the proportion of patients who achieved at least 50% reduction in uMN or uNMN levels from baseline. Sixteen patients (11 males/5 females) aged 12–86 years participated. After 12 weeks of treatment and at the last evaluation of efficacy, the primary endpoint was achieved in 31.3% of all patients, including 66.7% of those under preoperative treatment and 23.1% of those under chronic treatment. Sedation, anemia, and death were reported in 1 patient each as serious adverse drug reactions during the 24-week treatment. Metyrosine was shown to be tolerated and to relieve symptoms by reducing excess catecholamine in PPGL patients under both preoperative and chronic treatment.

**Key words:** Metyrosine, Catecholamines, Pheochromocytoma, Paraganglioma, Multi-center clinical trial

## PHEOCHROMOCYTOMAS (PCCs) AND PARAGANGLIOMAS (PGLs) (TOGETHER AS PPGLs) are

Submitted Jul. 3, 2017; Accepted Dec. 10, 2017 as EJ17-0276

Released online in J-STAGE as advance publication Jan. 20, 2018

Correspondence to: Mitsuhide Naruse, MD, PhD, Clinical Research Institute for Endocrinology and Metabolic Diseases, National Hospital Organization Kyoto Medical Center, 1-1, Fukakusa-Mukaihatacho, Fushimi-ku, Kyoto 612-8555, Japan.

E-mail: mnaruse@kyotolan.hosp.go.jp

neuroendocrine tumors arising from chromaffine cells of adrenal medulla and paraganglia of the sympathetic and parasympathetic nervous system, respectively [1]. PPGLs produce and secrete excess catecholamines, leading to various symptoms including hypertension, headache, palpitations, perspiration, and constipation [1]. PPGLs are rare diseases found in 0.05% to 0.1% of patients with hypertension: the annual morbidity in the US is from 500 to 1,600 [2], whereas the estimated num-

ber of Japanese patients is 2,920: 2,600 with benign and 320 with malignant disease [3]. Although most of the patients with PPGL show benign lesions, a considerable portion of these patients show metastatic lesions [2, 3].

For the treatment of PPGL, surgical removal of tumors is the first choice and preoperative treatment with  $\alpha$ -blockers is recommended for controlling blood pressure and heart rate, treating arrhythmia, normalizing the reduced circulating plasma volume, and preventing the perioperative and intraoperative cardiovascular complications caused by excess catecholamine [4, 5]. Treatment with  $\alpha$ -blockers similar to the preoperative treatment is also required for chronic treatment of inoperative and malignant PPGL [4]. Although chemotherapy with a combination of cyclophosphamide, vincristine, and dacarbazine, or radiation brachytherapy with  $^{131}\text{I}$ -meta-iodobenzylguanidine is occasionally indicated for malignant PPGL, effects of these anti-tumor therapies are limited and symptomatic treatment remains important for controlling the symptoms of patients [6-9].

Symptomatic treatment of PPGL starts with  $\alpha$ -blocker followed by additional administration of  $\beta$ -blocker, calcium channel blocker, and metyrosine [4]. While phenoxybenzamine, a noncompetitive, nonspecific  $\alpha$ -blocker, is widely used in the US, only  $\alpha_1$ -blockers such as doxazosin are approved in Japan. Doxazosin selectively blocks the  $\alpha_1$  receptor involved in hypertension in PPGL, but does not suppress  $\alpha_2$ -specific symptoms such as constipation [10]. A therapeutic strategy of blocking catecholamine excess at the upstream process of catecholamine production as well as at the receptor levels by  $\alpha$ -blockers could have clinical significance in the treatment of PPGL.

Metyrosine ( $\alpha$ -methyltyrosine, metirosine) specifically inhibits tyrosine hydroxylase, which catalyzes tyrosine to dihydroxyphenylalanine (DOPA), the first and the rate-limiting step of the catecholamine synthesis pathway, thereby resulting in reduction of catecholamines and their metabolites. Clinical trials have shown metyrosine to suppress catecholamine synthesis and improve symptoms of catecholamine excess, including hypertension [11-16]. Metyrosine (DEMSE<sup>®</sup>) received United States Food and Drug Administration (USFDA) approval in 1979 for preoperative preparation of patients for surgery, management of patients when surgery is contraindicated, and chronic treatment of patients with malignant pheochromocytoma [17]. Clinical studies of that time, however, did not meet the current regulatory standards for evaluating the efficacy and safety [18], and did not pro-

vide sufficient evidence for its efficacy and safety.

The aim of this study was to investigate the efficacy, safety, and pharmacokinetics of metyrosine in PPGL in a multicenter, open-label clinical trial in accordance with the principles of the Declaration of Helsinki and Good Clinical Practice (GCP [18]) in Japan (MCAP-J Study: Metyrosine reduces excess catecholamines/metabolites and ameliorates symptoms in patients with pheochromocytoma/paraganglioma in Japan).

## Materials and Methods

### Study design

This was a prospective, multicenter, open-label phase I/II study conducted in accordance with the principles of the Declaration of Helsinki and GCP, in which the efficacy, safety, and pharmacokinetics of metyrosine were evaluated in patients with PPGL in Japan. The study comprised a 4-week observation period and subsequent 12-week treatment period followed by a continuation treatment period. The trial registration identification number is JAPIC CTI-152999.

### Patients

Eligible patients were of either sex, aged 12 years or older who met all the following criteria: inoperable patients requiring chronic medication therapy or surgical candidates requiring preoperative treatment; diagnosed as PPGL; with baseline urinary metanephrine (uMN) and/or urinary normetanephrine (uNMN) levels  $\geq 3$  times the upper limit of the corresponding normal value; being treated with  $\alpha$ -blockers; with symptoms associated with excess catecholamine, including hypertension, glucose metabolism disorder, headache, palpitation, sweating, constipation, tachycardia, or tremor. The major exclusion criteria included; patients who received either chemotherapy or radiation therapy within 90 days prior to the study; received radiation brachytherapy within 180 days prior to the study; were newly treated or temporarily treated with a drug or who consumed foods that could affect urinary catecholamines and their metabolites; with difficulty in intestinal absorption of foods and drugs; under severe medication-induced sedation; with estimated glomerular filtration rate (eGFR)  $< 30$  mL/min; with left ventricular ejection fraction  $< 40\%$ ; with uncontrollable complications. All patients provided written consent to participate in the study. The study protocol was approved by the institutional review board of all centers.

### **Treatment**

Metyrosine treatment was started at 500 mg/day, and maintained for 3 days. The dose could be increased stepwise up to 4,000 mg/day, if there was no safety problem, and administered in divided doses once to 4 times daily. For patients with normal renal function or mild renal dysfunction ( $\text{eGFR} \geq 60 \text{ mL/min}$ ) the dose was increased by 250 mg or 500 mg, and for patients with moderate renal dysfunction ( $30 \text{ mL/min} \leq \text{eGFR} < 60 \text{ mL/min}$ ), only a dose increment of 250 mg was allowed. After each dose was decided, two consecutive 24-h urine samples were collected to determine uMN and uNMN. For patients with less than 50% reduction in uMN and uNMN from baseline, dose was increased further, in principle, in the same manner unless there was any safety problem. For patients confirmed with 50% or more reduction from baseline not only in uMN and uNMN, but also in urinary vanillylmandelic acid (uVMA), urinary adrenaline (uA), urinary noradrenaline (uNA), and urinary dopamine (uDA), and with improvement in the catecholamine-induced clinical symptoms, the dosage level could be determined as the maintenance dose. The investigator was to either reduce the dosage level or stop dosing if it was judged necessary for the safety of patients. The dosage level could be reduced to a level at which the safety of the patients had been established or lower, and treatment continued in the same manner as described above. All patients were hospitalized during the dose adjustment until urine examinations were completed. Urinary concentrations were determined by high-performance liquid chromatography with electrochemical detection for MN and NMN, and with fluorescence detection for A, NA, and DA.

### **Pharmacokinetic analysis**

Pharmacokinetic analysis was carried out in patients who met the following conditions: patients who had been on metyrosine treatment with total dosing record and with at least one collected blood sample. Pharmacokinetic parameters including maximum plasma concentration ( $C_{\text{max}}$ ), time to reach  $C_{\text{max}}$  ( $T_{\text{max}}$ ) and area under the concentration-time curve from 0 to 4-h and 9-h after administration ( $\text{AUC}_{4\text{h}}$ ,  $\text{AUC}_{9\text{h}}$ ) were analyzed using a non-compartmental model in Phoenix WinNonlin software ver 6.2 (Certara, Princeton, NJ, USA) on Day 1.  $C_{\text{max}}$ ,  $T_{\text{max}}$ , and  $\text{AUC}_{4\text{h}}$  were also analyzed on Day 7. Pharmacokinetics of multiple administrations of metyrosine were determined by the plasma concentration prior to drug administration on Day 4, 5, 6, and 7. Plasma

concentrations of metyrosine were determined by liquid chromatography tandem mass spectrometry method.

### **Efficacy assessment**

Efficacy of metyrosine was assessed at 84 days post-administration or just before the surgery. The primary efficacy endpoint was the proportion of patients with 50% or more reduction from baseline in uMN or uNMN at the final evaluation. Rationale of the primary endpoint was based on the US package insert, which describes that dosage of metyrosine should be titrated based on uMN and uNMN. Either uMN or uNMN, that with higher ratio of baseline to the upper limit of the reference value, was used as the primary measure for efficacy assessment. If the two ratios were identical, both uMN and uNMN were used. In the latter case, only patients with 50% or more reduction from baseline in both uMN and uNMN were regarded as responders of the primary endpoint. Each of uMN and/or uNMN was measured for two consecutive 24-h urine samples and data were expressed as the mean of the two measurements. Patients who failed to provide two 24-h urine samples were regarded as non-responders of the primary endpoint. Patients who discontinued metyrosine administration during the treatment period were also regarded as non-responders. The secondary efficacy endpoints included 1) the proportions of patients with 50% or more reduction from baseline in uMN, uNMN, uVMA, uA, uNA, and uDA during the treatment period, 2) Clinical Global Impression of Change (CGI-C) of excess catecholamine-induced symptoms (Improvement was assessed by both patient and investigator, independently, using 7-grade relative estimation as markedly worsened, moderately worsened, mildly worsened, no change, mildly improved, moderately improved, greatly improved) [19], and 3) changes in blood pressure, heart rate, blood glucose, and left ventricle ejection fraction (LVEF).

### **Safety assessment**

The safety endpoints included frequency and description of adverse events (AEs), frequency and description of adverse drug reactions (ADRs), vital signs (systolic/diastolic blood pressure, pulse rate, respiratory rate, body temperature), general clinical laboratory tests (hematological examination, blood biochemical tests, and urinalysis), and 12-lead electrocardiogram (ECG). In this report, we describe ADRs that occurred during the first 24-week treatment. Monitoring and evaluation are, however, still ongoing.

### Statistical analysis

No statistical tests were performed since the number of enrolled patients was small. Populations for analysis included a safety set (SAF), full analysis set (FAS), and per-protocol set (PPS, data not shown). The SAF was defined as the population of patients who received metyrosine at least once. The FAS was the population of patients included in the SAF who were evaluated for efficacy at least once. The number of patients to be enrolled was set at least 10 because the population of patients eligible for this study was thought to be very small. Data are presented as mean  $\pm$  SD.

## Results

### Patients

Sixteen patients, 13 under chronic treatment and 3 under preoperative treatment, were enrolled. Nine out of 13 patients under chronic treatment completed the treatment period, and the other 4 patients discontinued the treatment. All 3 patients under preoperative treatment completed the treatment period. The demographic and clinical characteristics of patients are listed in Table 1. Patients consisted of 11 men and 5 women, aged 12 to 86 years. Systolic and diastolic blood pressure was  $126.4 \pm 16.6$  mmHg and  $71.1 \pm 16.1$  mmHg, respectively. Five patients had normal ( $\text{eGFR} \geq 90$  mL/min), 6 mildly reduced ( $60 \text{ mL/min} \leq \text{eGFR} < 90 \text{ mL/min}$ ), and 5 patients had moderately reduced renal function ( $30 \text{ mL/min} \leq \text{eGFR} < 60 \text{ mL/min}$ ). After 24-week treatment, duration of metyrosine treatment was  $124.9 \pm 64.0$  days for patients under chronic treatment and  $53.3 \pm 27.2$  days for patients under preoperative treatment. The duration of the preoperative metyrosine treatment for the 3 patients was 32, 44, and 84 days, respectively. At 84 days post-administration or just before the surgery as the final evaluation time point, the dosage of metyrosine was  $1,027.8 \pm 506.9$  mg/day for patients under chronic treatment and  $1,083.3 \pm 629.2$  mg/day for patients under preoperative treatment.

### Pharmacokinetics

After a single oral dose of 250 mg in the patients with normal, mildly reduced, and moderately reduced renal function,  $C_{\text{max}}$  was  $6,310 \pm 2,140$ ,  $7,500 \pm 2,010$ , and  $7,880 \pm 1,730$  ng/mL, and  $\text{AUC}_{9\text{h}}$  was  $22,600 \pm 4,030$ ,  $30,000 \pm 11,800$ , and  $38,400 \pm 3,930$  ng·h/mL, respectively (Supplementary Table 1). Plasma concentration profiles of metyrosine are shown in Supplementary Fig. 1.

Based on changes in the plasma concentrations after the first dose (trough concentrations), the plasma level of metyrosine in patients with PPGL was considered to have reached a steady state within 3 days after the dosage level was changed at Day 4 (Supplementary Fig. 2).

### Efficacy

The proportion of patients who achieved 50% or more reduction in uMN or uNMN from baseline at the final evaluation time point as the primary endpoint was 31.3% (5/16) overall, 23.1% (3/13) in patients under chronic treatment, and 66.7% (2/3) in patients under preoperative treatment (Table 2). Four patients who underwent chronic treatment failed to continue the study up to the final evaluation point. uMN and uNMN were decreased by  $0.7 \pm 1.1$  mg/day and  $5.1 \pm 10.0$  mg/day from baseline at the final evaluation time point, respectively. The changing rates for uMN, uNMN, and the total amount of uMN and uNMN (shown as uMN + uNMN) were  $-46.8 \pm 24.3\%$ ,  $-42.3 \pm 17.5\%$ , and  $-44.8 \pm 17.4\%$ , respectively (Table 3). The changing rate for uMN or uNMN, that with higher ratio, which was used for the primary measure, was  $-45.4 \pm 18.3\%$ . The CGI-C of excess catecholamine-induced symptoms of each patient at the final evaluation time point is shown in Table 4. The self-assessment by patients reported improvement of symptoms in 8 out of 13 patients (61.5%) and no change in the other 5 patients (38.5%). On the other hand, the investigator judged improvement in 7 patients (53.8%) and no change in 6 patients (46.2%). There was no patient whose symptoms were considered worsened in either evaluation.

At the final evaluation time point, blood pressure, heart rate, blood glucose, and LVEF were  $120.6 \pm 14.5/69.8 \pm 13.8$  mmHg,  $67.4 \pm 21.3$  bpm,  $111.8 \pm 28.7$  mg/dL and  $63.97 \pm 7.13\%$ , respectively.

Fig. 1 shows a representative case with significant improvement in the symptoms of excess catecholamines by metyrosine. The patient was a 40-year-old Japanese male, of height 172.3 cm and weight 44.4 kg, diagnosed as extra-adrenal PGL with metastasis to bone, pleura, and lymph nodes. He had normal kidney function with eGFR of 119.8 mL/min. Urinary excretion of catecholamines and their metabolites uMN, uNMN, uVMA, uA, uNA, and uDA were 0.39 mg/day, 115 mg/day, 202.5 mg/day, 36.7  $\mu$ g/day, 23,600  $\mu$ g/day, and 5,500  $\mu$ g/day, respectively. Systolic/diastolic blood pressure, heart rate, left ventricular ejection fraction, and blood glucose were

**Table 1** Baseline patient characteristics

|  | Total                        | Chronic Treatment            | Preoperative Treatment      |
|--|------------------------------|------------------------------|-----------------------------|
|  | <i>n</i> = 16                | <i>n</i> = 13                | <i>n</i> = 3                |
| Sex, male/female   | 11/5                         | 9/4                          | 2/1                         |
| Age, years, ranges   | 54.8 (24.3), 12 to 86        | 52.9 (26.0), 12 to 86        | 62.7 (15.6), 46 to 77       |
| Age, 65 y or more/less than 65 y                           | 7/9                          | 5/8                          | 2/1                         |
| Diagnosis, Pheochromocytoma/Paraganglioma                  | 9/7                          | 7/6                          | 2/1                         |
| Metastatic PPGL, Yes/No                                    | 8/8                          | 8/5                          | 0/3                         |
| eGFR, mL/min   | 78.6 (40.2)                  | 77.9 (41.6)                  | 81.8 (41.1)                 |
| Systolic blood pressure/<br>Diastolic blood pressure, mmHg | 126.4 (16.6)/<br>71.1 (16.1) | 125.8 (16.7)/<br>69.8 (17.7) | 128.7 (19.7)/<br>76.7 (2.5) |
| Heart rate, bpm  | 70.3 (16.1)                  | 71.9 (17.3)                  | 63.3 (8.1)                  |
| Blood glucose, mg/dL                                       | 112.9 (18.5)                 | 115.5 (19.2)                 | 102.0 (11.5)                |
| LVEF, %  | 65.89 (7.45)                 | 66.02 (7.89)                 | 65.33 (6.51)                |
| uMN, mg/day <sup>a</sup> , (0.05 to 0.20)                  | 4.2 (12.8)                   | 4.6 (14.3)                   | 2.7 (3.2)                   |
| uNMN, mg/day <sup>a</sup> , (0.10 to 0.28)                 | 16.4 (29.9)                  | 19.9 (32.4)                  | 1.0 (0.7)                   |
| uMN + uNMN, mg/day <sup>a</sup>                            | 20.6 (31.3)                  | 24.5 (33.7)                  | 3.7 (2.9)                   |
| uA, µg/day <sup>a</sup> , (1 to 23)                        | 200.4 (547.2)                | 231.5 (606.8)                | 65.3 (48.7)                 |
| uNA, µg/day <sup>a</sup> , (29 to 120)                     | 3,650.8 (5,923.6)            | 4,409.1 (6,365.1)            | 364.5 (376.1)               |
| uDA, µg/day <sup>a</sup> , (100 to 1,000)                  | 1,391.9 (1,339.6)            | 1,326.2 (1,342.0)            | 1,676.7 (1,582.2)           |
| uVMA, mg/day <sup>a</sup> , (1.4 to 4.9)                   | 48.0 (58.9)                  | 57.1 (62.2)                  | 8.8 (3.2)                   |
| Dose of doxazosin, mg, ranges                              | 9.8, 0.5 to 36               | 7.3, 0.5 to 16               | 20.7, 10 to 36              |

Data are presented as means (SD). Reference intervals of catecholamines and their metabolites measurements are appended to their items.

a) Average of 2 consecutive 24-h urine examinations.

PPGL, pheochromocytoma and paragangliomas; eGFR, estimated glomerular filtration rate; LVEF, left ventricular ejection fraction; uMN, urinary metanephrine; uNMN, urinary normetanephrine; uA, urinary adrenaline; uNA, urinary noradrenaline; uDA, urinary dopamine; uVMA, urinary vanillylmandelic acid.

**Table 2** Proportion of patients with 50% or more reduction from baseline in uMN or uNMN at the final evaluation (primary efficacy endpoint)

|                        | Day 6 to 8 <sup>a,b</sup> | Day 28 <sup>b</sup> | Day 56 <sup>b</sup> | Day 84 <sup>a,b</sup> | Final evaluation <sup>a,c</sup> |
|------------------------|---------------------------|---------------------|---------------------|-----------------------|---------------------------------|
| Total                  | 3/15 (20.0%)              | 5/14 (35.7%)        | 5/11 (45.5%)        | 4/10 (40.0%)          | 5/16 (31.3%)                    |
| Chronic Treatment      | 3/13 (23.1%)              | 3/11 (27.3%)        | 4/10 (40.0%)        | 3/9 (33.3%)           | 3/13 (23.1%)                    |
| Preoperative Treatment | 0/2 (0.0%)                | 2/3 (66.7%)         | 1/1 (100.0%)        | 1/1 (100.0%)          | 2/3 (66.7%)                     |

Either uMN or uNMN, that with higher ratio of baseline to the upper limit of the reference value, was used for efficacy assessment.

a) Average of 2 consecutive 24-h urine examinations.

b) All patients who provided 24-h urine samples at each point were included for analysis.

c) The final evaluation point was Day 84 of treatment for chronic treatment and just before the surgery for preoperative treatment. All patients were included for analysis and unevaluated patients were regarded as those without effect.

uMN, urinary metanephrine; uNMN, urinary normetanephrine.

145/96 mmHg, 113 bpm, 72.6%, and 98 mg/dL. At 84 days post-administration, uMN, uNMN, uVMA, uA, uNA, and uDA were 0.095 mg/day (−75.6%), 79.5

mg/day (−30.9%), 155 mg/day (−23.5%), 0.4 µg/day (−98.9%), 26,150 µg/day (+10.8%), and 9,900 µg/day (+80.0%). The reduction of urinary excretion of cate-



**Table 3** Changing rates for catecholamines and their metabolites from baseline

|  | Baseline <sup>a)</sup> | Day 6–8 <sup>a)</sup> | Day 28               | Day 56                | Day 84 <sup>a)</sup> | Final Evaluation <sup>a)</sup> |
|--|------------------------|-----------------------|----------------------|-----------------------|----------------------|--------------------------------|
|  | <i>n</i> = 16          | <i>n</i> = 15         | <i>n</i> = 14        | <i>n</i> = 11         | <i>n</i> = 10        | <i>n</i> = 12                  |
| Changing rate of uMN or uNMN, % <sup>b)</sup><br>(Primary measure) |                        | –27.1 (20.4)          | –34.7 (23.0)         | –41.8 (16.3)          | –42.5 (18.6)         | –45.4 (18.3)                   |
| uMN, mg/day  | 4.2 (12.8)             | 3.9 (12.2)            | 3.5 (11.1)           | 0.4 (0.4)             | 0.2 (0.3)            | 0.5 (0.6)                      |
| Treatment effect, mg/day   |                        | –0.5 (1.1)            | –1.3 (2.7)           | –0.3 (0.4)            | –0.3 (0.4)           | –0.7 (1.1)                     |
| Changing rate, %   |                        | –16.9 (26.4)          | –40.7 (27.2)         | –43.0 (28.4)          | –44.1 (25.9)         | –46.8 (24.3)                   |
| uNMN, mg/day   | 16.4 (29.9)            | 10.6 (18.9)           | 10.0 (26.1)          | 11.1 (26.1)           | 11.2 (24.2)          | 9.4 (22.3)                     |
| Treatment effect, mg/day   |                        | –6.8 (12.0)           | –3.0 (4.7)           | –4.7 (7.7)            | –6.0 (10.8)          | –5.1 (10.0)                    |
| Changing rate, %   |                        | –40.0 (17.2)          | –35.9 (21.6)         | –41.1 (16.1)          | –41.2 (17.9)         | –42.3 (17.5)                   |
| uMN + uNMN, mg/day <sup>c)</sup>                                   | 20.6 (31.3)            | 14.5 (21.1)           | 13.5 (27.8)          | 11.4 (26.0)           | 11.4 (24.2)          | 9.8 (22.2)                     |
| Treatment effect, mg/day   |                        | –7.3 (12.0)           | –4.3 (5.2)           | –5.0 (7.7)            | –6.4 (10.8)          | –5.8 (9.9)                     |
| Changing rate, %   |                        | –30.3 (16.9)          | –35.6 (22.2)         | –41.3 (15.9)          | –42.0 (17.8)         | –44.8 (17.4)                   |
| uA, µg/day   | 200.4 (547.2)          | 139.5 (405.1)         | 232.2 (751.5)        | 25.5 (37.3)           | 15.5 (23.4)          | 20.5 (24.9)                    |
| Treatment effect, µg/day   |                        | –68.6 (163.4)         | 4.5 (185.4)          | –44.7 (74.6)          | –49.5 (75.5)         | –49.1 (69.0)                   |
| Changing rate, %   |                        | –45.4 (31.5)          | –48.6 (36.4)         | –52.9 (57.7)          | –77.7 (16.9)         | –72.9 (20.6)                   |
| uNA, µg/day  | 3,650.8<br>(5,923.6)   | 2,529.2<br>(4,196.3)  | 3,193.8<br>(8,097.7) | 3,345.0<br>(6,577.7)  | 3,914.4<br>(7,950.6) | 3,272.7<br>(7,346.2)           |
| Treatment effect, µg/day   |                        | –1,353.2<br>(1,934.1) | –372.8<br>(2,819.3)  | –1,105.6<br>(1,713.0) | –957.7<br>(2,089.3)  | –812.1<br>(1,920.2)            |
| Changing rate, %   |                        | –42.3 (19.1)          | –37.1 (33.1)         | –46.3 (26.7)          | –44.2 (29.6)         | –46.2 (27.3)                   |
| uDA, µg/day  | 1,391.9<br>(1,339.6)   | 3,004.7<br>(3,626.0)  | 1,937.1<br>(1,726.9) | 3,314.6<br>(5,185.2)  | 2,526.0<br>(3,062.0) | 2,605.0<br>(2,880.0)           |
| Treatment effect, µg/day   |                        | 1,564.3<br>(2,612.5)  | 617.9<br>(908.9)     | 2,092.3<br>(3,845.7)  | 1,256.5<br>(1,883.0) | 1,200.0<br>(1,716.1)           |
| Changing rate, %   |                        | 94.4 (135.2)          | 63.0 (90.5)          | 129.1 (191.7)         | 96.6 (143.4)         | 90.3 (130.9)                   |
| uVMA, mg/day   | 48.0 (58.9)            | 29.4 (35.1)           | 22.0 (39.2)          | 25.9 (54.4)           | 25.1 (46.4)          | 21.4 (42.8)                    |
| Treatment effect, µg/day   |                        | –21.5 (26.2)          | –14.6 (16.0)         | –10.8 (9.1)           | –14.6 (14.8)         | –13.1 (13.9)                   |
| Changing rate, %   |                        | –46.8 (13.9)          | –51.1 (16.6)         | –50.6 (21.2)          | –50.4 (19.2)         | –52.6 (18.3)                   |

Data are presented as means (SD). Treatment effect is the amount of change from baseline.

a) Average of 2 consecutive 24-h urine examinations.

b) Either uMN or uNMN, that with higher ratio of baseline to the upper limit of the reference value, was used for efficacy assessment.

c) Total amounts of uMN and uNMN.

uMN, urinary metanephrine; uNMN, urinary normetanephrine; uA, urinary adrenaline; uNA, urinary noradrenaline; uDA, urinary dopamine; uVMA, urinary vanillylmandelic acid.

cholamine and their metabolites improved the symptoms of excess catecholamines: during the metyrosine treatment, CGI-C was great/prominent both by the investigator and by the patients. In addition to improvement of those symptoms, coldness of peripheral limbs, purple skin color of forearms and hands, severe constipation, nausea and vomiting were improved from Day 1 by the

treatment. However, the patient died while on maintenance dose. The investigator commented on the death as follows: The measured values of catecholamines in this patient kept increasing as the metastatic tumors grew. Although the value was mildly reduced by metyrosine treatment, it was still in a range of 200- to 400-fold above the upper limit of reference value. There was no

**Table 4** Chronological change of the CGI-C of excess catecholamine-induced symptoms assessed by investigators and patients

|                     | Day 8         | Day 14        | Day 28        | Day 56        | Day 84        | Final evaluation |
|---------------------|---------------|---------------|---------------|---------------|---------------|------------------|
|                     | <i>n</i> = 16 | <i>n</i> = 16 | <i>n</i> = 14 | <i>n</i> = 12 | <i>n</i> = 11 | <i>n</i> = 13    |
|                     | Dr/Pts        | Dr/Pts        | Dr/Pts        | Dr/Pts        | Dr/Pts        | Dr/Pts           |
| Greatly improved    | 0/0           | 1/0           | 1/0           | 0/0           | 1/0           | 1/0              |
| Moderately improved | 1/1           | 1/1           | 1/1           | 2/1           | 1/0           | 1/0              |
| Mildly improved     | 6/6           | 5/6           | 6/5           | 4/4           | 5/8           | 5/8              |
| No change           | 9/9           | 9/9           | 6/7           | 6/7           | 4/3           | 6/5              |
| Mildly worsened     | 0/0           | 0/0           | 0/1           | 0/0           | 0/0           | 0/0              |
| Moderately worsened | 0/0           | 0/0           | 0/0           | 0/0           | 0/0           | 0/0              |
| Markedly worsened   | 0/0           | 0/0           | 0/0           | 0/0           | 0/0           | 0/0              |

Data are presented as number of patients.

CGI-C, Clinical Global Impression of Change; Dr, investigator's assessment; Pts, patient's assessment.

effective treatment for the primary disease and the patient was at the terminal stage. Sudden cardiac failure was a typical outcome of this disease, and therefore the death was considered unrelated to the drug treatment.

### Safety

The frequencies of ADRs with their grades during the treatment period and continuation period are summarized in Table 5. All of the 16 patients experienced at least one ADR. Most frequent ADRs were somnolence in 13 patients (grade 1 in 11 patients, grade 2 in 2 patients) followed by sedation in 2 (grade 1 in 1, grade 3 in 1), and weight increased in 2 (grade 1 in 2). Serious ADRs were sedation, anemia, and death (1 patient each). Both sedation and anemia disappeared after cessation of metyrosine treatment. Patients who showed anemia restarted the treatment after the symptom disappeared.

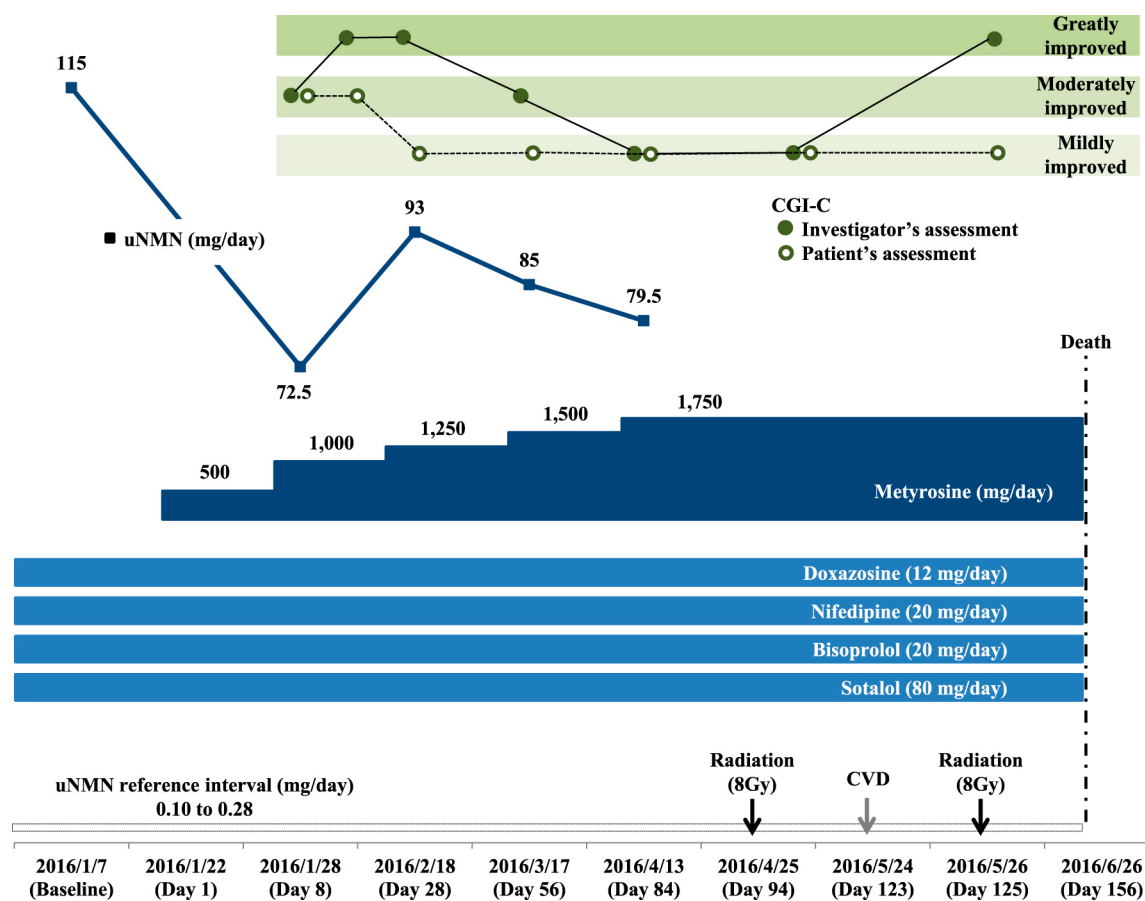
An 84-year-old male patient with paraganglioma and moderate renal dysfunction (eGFR: 43.69 mL/min) was found dead while undergoing treatment with metyrosine as an outpatient. Urinary excretion of catecholamines and their metabolites at baseline was 52 mg/day, 8.2 mg/day, 91.8 mg/day, 2,230 µg/day, 679.5 µg/day, and 860 µg/day for uMN, uNMN, uVMA, uA, uNA, and uDA, respectively. His blood pressure, pulse rate, LVEF, and blood glucose were 136/81 mmHg, 95 bpm, 59%, and 154 mg/dL. His major symptoms were fatigue, tremor, and breathing difficulty. Metyrosine treatment began at 500 mg/day, and the dose was increased to 750 mg/day on Day 4 and decreased to 500 mg/day on Day 38 for the safety of the patient, because his blood pressure had a decreasing trend. The patient was found dead on Day 56.

Although the death was considered by the investigator possibly due to a natural outcome of the underlying malignant paraganglioma, causal relationship to metyrosine was not ruled out.

### Discussion

The present study suggested the efficacy and tolerance of metyrosine in patients with PPGL in compliance with GCP. Metyrosine administration was shown to reduce uMN, uNMN, uA, uNA and uVMA, and improve symptoms related to chronic excess catecholamine in patients with malignant and unresectable PPGL. In addition, metyrosine was very effective in some of patients as shown in Fig. 1. Our study suggested that combination of metyrosine with  $\alpha$ -blocker could be one of the treatment options in patients under preoperative treatment. Steinsapir *et al.* [20] reported that the combination of metyrosine and  $\alpha$ -blocker resulted in better blood pressure control and less need for use of antihypertensive medication or pressor during the surgery, and decrease in surgical morbidity. Perry *et al.* [21] reported less blood loss, the need for intraoperative fluid replacement, and intraoperative morbidity. Engelman *et al.* [13] reported their impression that the combination therapy reduced intraoperative problems with blood pressure control and enabled surgeons to remove tumors with less hazard of hypertensive crisis. In this study, we also were able to successfully perform surgery in patients with PPGL who had preoperative treatment with a combination of metyrosine and  $\alpha_1$ -blocker.

In contrast to urinary excretion of those catecholamine



**Fig. 1** Significant improvement in the symptoms of excess catecholamines by metyrosine in a 40-year-old male patient with extra-adrenal PGL

PGL, paragangliomas; CGI-C, Clinical Global Impression of Change; uNMN, urinary normetanephrine; CVD, chemotherapy with cyclophosphamide, vincristine and dacarbazine

derivatives, uDA, the precursor of NA, A, MN, NMN, and VMA, was increased in some patients. This seemingly problematic observation is actually consistent with a notion by Kuchel *et al.* [22]. These authors demonstrated that metyrosine treatment in malignant PCC resulted in initial increase in plasma DOPA, DOPA sulfate, and dopamine sulfate. uDA progressively increased in the course of metyrosine treatment, and this, along with the increase of dopamine metabolites such as dihydroxyphenylethanol, and plasma dopamine sulfate, occurred in the absence of any change in plasma dopamine. Since we did not measure plasma DA concentration in this study, we are unable to exclude a possibility that plasma dopamine concentration was elevated by metyrosine treatment. Elevated plasma DA concentration may produce serious clinical problems, because excess DAs are able to stimulate not only their own receptors, but also

those for other catecholamines such as NA and A in a variety of tissues/organs, including central nervous system and cardiovascular system [23, 24]. However, a lack of clinically relevant ADRs in this study may be explained by the increase in uDA excretion without elevation of plasma DA concentration as reported by Kuchel *et al.* [22].

All patients experienced at least one ADR, and these were among those previously observed [12-16, 20, 21, 25] and described in the US prescribing information [17]. Sedation and somnolence, reported as AEs specific to metyrosine, also frequently occurred in the Japanese PPGL patients. Most of those ADRs were mild and some were moderate, which were tolerable. Two of three serious ADRs disappeared after cessation of administration.

The US prescribing information [17] reports temporary changes in sleep pattern occur following withdrawal



**Table 5** ADRs with their grades during the treatment period and continuation period

|  | n <sub>total</sub> (n <sub>mild</sub> /n <sub>moderate</sub> /n <sub>severe</sub> ) |                   |                        |
|--|---|-------------------|------------------------|
|  | Total   | Chronic Treatment | Preoperative Treatment |
|  | n = 16  | n = 13            | n = 3                  |
| All ADRs                               | 16 (11/3/2)   | 13 (8/3/2)        | 3 (3/0/0)              |
| Anaemia                                | 1 (0/1/0)   | 1 (0/1/0)         |                        |
| Bradycardia                            | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Diarrhoea                              | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Vomiting                               | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Faeces soft                            | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Death                                  | 1 (0/0/1)   | 1 (0/0/1)         |                        |
| Pyrexia                                | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Blood creatine phosphokinase increased | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Blood pressure decreased               | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Blood triglycerides increased          | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Weight increased                       | 2 (2/0/0)   | 1 (1/0/0)         | 1 (1/0/0)              |
| Protein urine present                  | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Acidosis                               | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Hypokalaemia                           | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Decreased appetite                     | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Dizziness postural                     | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Sedation                               | 2 (1/0/1)   | 1 (0/0/1)         | 1 (1/0/0)              |
| Somnolence                             | 13 (11/2/0)   | 12 (10/2/0)       | 1 (1/0/0)              |
| Anxiety                                | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Depression                             | 1 (1/0/0)   | 1 (1/0/0)         |                        |
| Insomnia                               | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Hypertensive crisis                    | 1 (1/0/0)   |                   | 1 (1/0/0)              |
| Hypotension                            | 1 (1/0/0)   | 1 (1/0/0)         |                        |

Data are presented as number of patients.

Reported ADR terms were replaced by terms from MedDRA ver 19.1J.

ADR: Adverse drug reaction.

of metyrosine, including insomnia, feelings of increased alertness and ambition, and symptoms of psychic stimulation. No such changes occurred in the patients who discontinued treatment in this study.

In addition, the US prescribing information [17] recommends careful administration for patients with renal dysfunction, since metyrosine is excreted *via* kidney and renal dysfunction affects the pharmacokinetics of metyrosine. In support of this, metyrosine exposure as shown by the C<sub>max</sub> and AUC tended to be increased in

PPGL patients with mild as well as moderate renal dysfunction compared to the patients with normal renal function. However, metyrosine was safely used in PPGL patients with these grades of renal function in the present study.

This study has some limitations. First, the number of enrolled patients was small. This is because PPGL are rare diseases. Second, there was no placebo control for the same reason, and thus a randomized clinical trial was not possible. Third, plasma concentrations of free MN

and NMN were not determined. Most uMN and uNMN are conjugated and may be less sensitive to quantification of the inhibition of catecholamine synthesis. However, we had no choice because no such assay system has obtained regulatory approval in Japan. Thus, we used 24-h urine samples for determining the degree of catecholamine synthesis inhibition. The results showed that uA, uNA, and uVMA decreased along with uMN and uNMN, indicating that the efficacy of metyrosine was successfully evaluated. Last, information on the safety of metyrosine was monitored for only 24-week treatment, although the treatment duration is expected to be further extended after the drug is on the market. Fortunately, however, we designed this study so as to allow patients to continue metyrosine treatment until the day of regulatory approval in Japan, and thus the duration of the drug safety monitoring is still ongoing and extending.

In conclusion, this study suggested the efficacy and safety of metyrosine in Japanese patients with PPGL under chronic treatment as well as preoperative treatment. Metyrosine is administered to patients with serious symptoms which are uncontrollable by other medications, thus metyrosine will be considered a useful option added on  $\alpha$ -blocker, such as phenoxybenzamine or doxazosin. This is the first clinical trial of metyrosine conducted in accordance with the principles of the Declaration of Helsinki and GCP, and this will be of importance for approval in Japan and for the treatment of such patients.

Upon the approval of metyrosine for clinical use in Japan, the following researches are expected: (1) long-term durability of the efficacy of metyrosine; (2) efficacy of combination therapy with anti-tumor treatments such as cyclophosphamide, vincristine, and dacarbazine (CVD therapy) or internal radiation therapy with  $^{131}\text{I}$ -metaiodobenzylguanidine (MIBG therapy); (3) metyrosine effect of preoperative treatment on intraoperative events such as blood pressure fluctuation; and (4) efficacy and safety of metyrosine at higher doses. In addition, metyrosine may serve as a new antihypertensive agent with a new mechanism of action, especially in patients with resistant hypertension.

## Acknowledgements

Authors thank all participated patients, their family, and health care professionals, who enabled this study. We thank all the subinvestigators of each 11 institutions for their cooperation to this clinical trial. Medical writing and editorial assistance were provided by ASCA Corporation.

## Disclosure

T.H. and N.K. have been employed by Ono Pharmaceutical Co., Ltd.

## Funding Source

This work was funded by Ono Pharmaceutical Co., Ltd. (Osaka, Japan).

## Appendix

### Study sites

Division of Nephrology, Endocrinology and Vascular Medicine, Tohoku University Hospital; Department of Endocrinology and Diabetes, Gunma University Hospital; Department of Diabetes, Metabolism and Endocrinology, Chiba University Hospital; Department of Breast and Endocrine Surgery, Tokyo Women's Medical University Hospital; Department of Endocrinology and Hypertension, Tokyo Women's Medical University Hospital; Department of Diabetes, Endocrinology and Metabolism, Center Hospital of the National Center for Global Health and Medicine; Department of Metabolism and Endocrinology, St. Marianna University School of Medicine, Yokohama City Seibu Hospital; Department of Breast and Endocrine Surgery, Aichi Medical University Hospital; Department of Endocrinology and Metabolism, National Hospital Organization Kyoto Medical Center; Department of Urology, Kansai Medical University Hospital; Department of Endocrine and Metabolic Diseases/Diabetes Mellitus, Kyushu University Hospital.

**Supplementary Table 1** Pharmacokinetic parameters of metyrosine in plasma after the initial oral administration of metyrosine on Day 1 and 7 in patients with PPGL

|       | Renal failure | Dose, mg              | n | Pharmacokinetic parameters <sup>a)</sup> |                          |                      |                      |
|-------|---------------|-----------------------|---|--|--------------------------|----------------------|----------------------|
|       |               |                       |   | $C_{max}$ , ng/mL                        | $T_{max}$ , h [Min, Max] | $AUC_{4h}$ , ng·h/mL | $AUC_{9h}$ , ng·h/mL |
| Day 1 | Normal        | 500 BID <sup>b)</sup> | 5 | 6,310 (2,140)                            | 1.00 [0.667, 4.00]       | 13,900 (4,600)       | 22,600 (4,030)       |
|       | Mild          | 500 BID <sup>c)</sup> | 6 | 7,500 (2,010)                            | 1.24 [0.967, 1.97]       | 18,500 (6,090)       | 30,000 (11,800)      |
|       | Moderate      | 500 BID               | 5 | 7,880 (1,730)                            | 1.47 [1.00, 1.97]        | 21,000 (2,490)       | 38,400 (3,930)       |
| Day 7 | Normal        | 500 BID               | 1 | 7,310 (NC)                               | 1.52 [1.52, 1.52]        | 16,500 (NC)          | —                    |
|       |               | 1,000 QID             | 4 | 10,400 (1,230)                           | 1.00 [0.700, 1.03]       | 26,700 (3,920)       | —                    |
|       | Mild          | 250 QD                | 1 | 7,820 (NC)                               | 3.08 [3.08, 3.08]        | 22,700 (NC)          | —                    |
|       |               | 1,000 QID             | 5 | 11,500 (1,860)                           | 1.03 [0.667, 2.58]       | 32,400 (6,940)       | —                    |
|       | Moderate      | 500 BID               | 2 | 16,600 (NC)                              | 1.54 [1.08, 2.00]        | 52,600 (NC)          | —                    |
|       |               | 500 BID <sup>d)</sup> | 1 | 9,120 (NC)                               | 1.00 [1.00, 1.00]        | 32,600 (NC)          | —                    |
|       |               | 750 TID               | 2 | 12,300 (NC)                              | 2.01 [1.02, 3.00]        | 39,300 (NC)          | —                    |

Data are presented as means (SD).

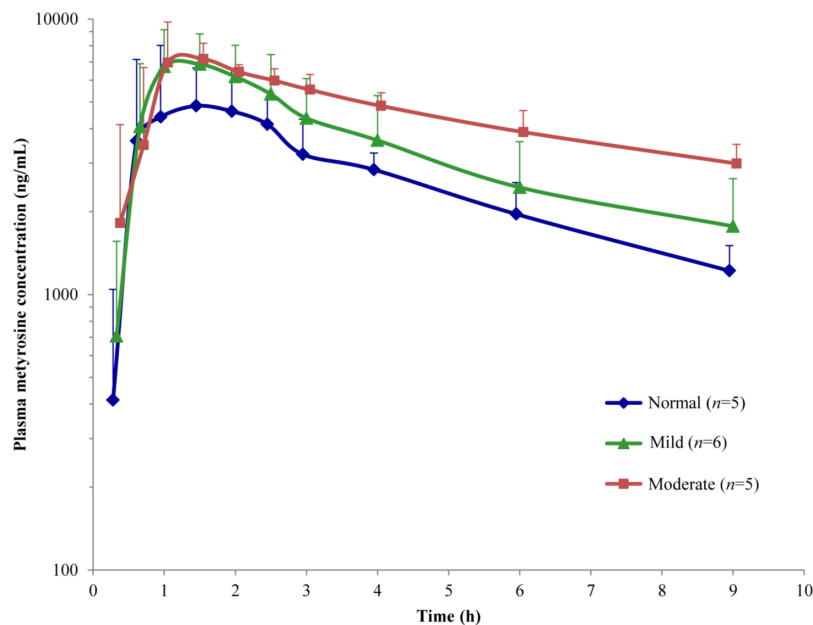
a) Pharmacokinetic parameters were evaluated after a single oral administration of 250 mg metyrosine at Day1 or Day7.

b) 250 mg QD for two patients.

c) 250 mg QD for one patient.

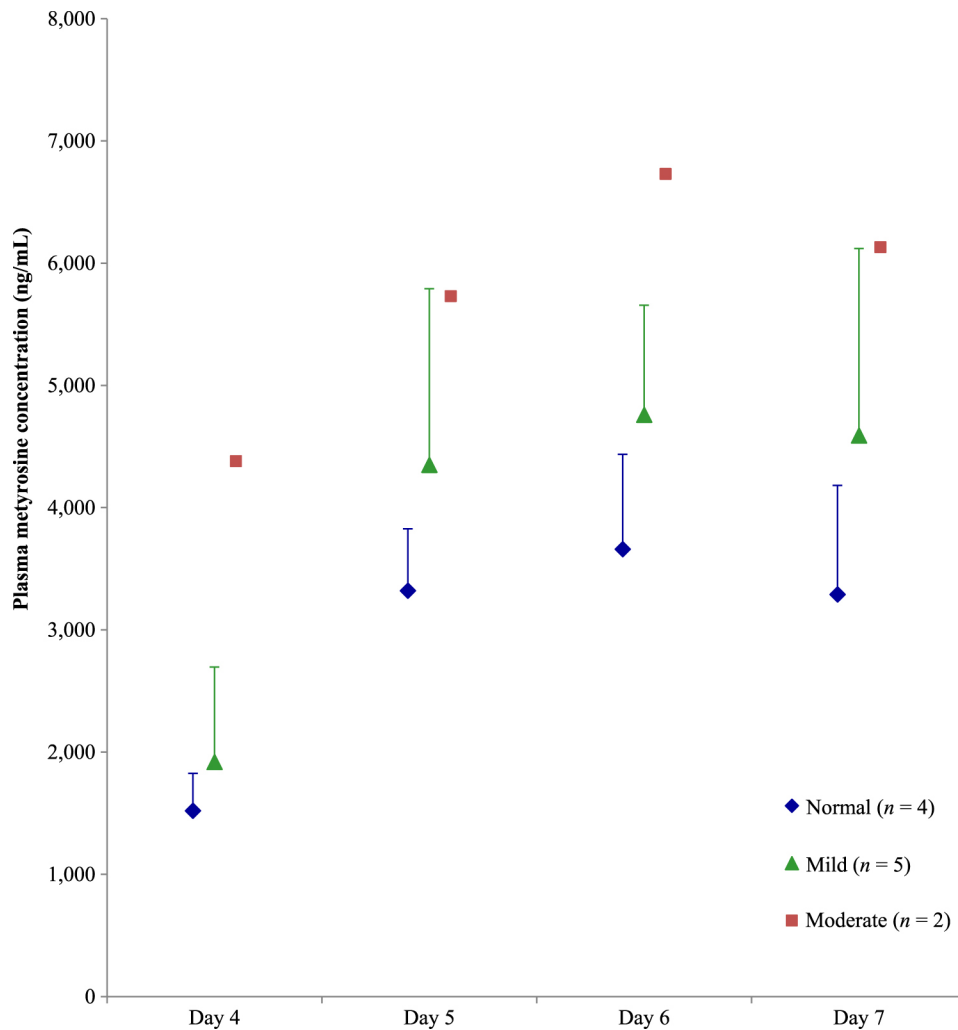
d) The dosage level was reduced from 750 mg/day to 500 mg/day on Day 6.

PPGL, pheochromocytoma/paraganglioma;  $C_{max}$ , maximum plasma concentration;  $T_{max}$ , time to reach  $C_{max}$ ; Max, maximum; Min, minimum;  $AUC_{4h}$ , area under the concentration-time curve from 0 to 4-h after administration;  $AUC_{9h}$ , area under the concentration-time curve from 0 to 9-h after administration; BID, twice a day; QID, four times a day; QD, once a day; TID, three times a day; NC, not calculated.

**Supplementary Fig. 1** Plasma metyrosine concentration profiles after a single oral administration of 250 mg metyrosine in PPGL patients with normal, mildly reduced and moderately reduced renal function

Values are means, and bars represent standard deviation.

PPGL, pheochromocytoma/paraganglioma.



**Supplementary Fig. 2** Plasma metyrosine trough concentration after multiple oral administrations of in PPGL patients with normal, mildly reduced and moderately reduced renal function

Values are means, and bars represent standard deviation, while standard deviation was not able to be calculated in patients with moderately reduced renal function. Patients with normal, mildly reduced and moderately reduced renal function were administrated 1,000 mg/day, 1,000 mg/day and 500 mg/day on Day 5 to 7, respectively.

PPGL, pheochromocytoma/paraganglioma.

## References

1. Lenders JW, Eisenhofer G, Mannelli M, Pacak K (2005) Pheochromocytoma. *Lancet* 366: 665–675.
2. Chen H, Sippel RS, O'Dorisio MS, Vinik AI, Lloyd RV, *et al.* (2010) The North American Neuroendocrine Tumor Society consensus guideline for the diagnosis and management of neuroendocrine tumors: pheochromocytoma, paraganglioma, and medullary thyroid cancer. *Pancreas* 39: 775–783.
3. Kimura N, Takayanagi R, Takizawa N, Itagaki E, Katabami T, *et al.* (2014) Pathological grading for predicting metastasis in pheochromocytoma and paraganglioma. *Endocr Relat Cancer* 21: 405–414.
4. NCCN clinical practice guidelines in oncology. Neuroendocrine tumors. Version 2.2016. [https://www.nccn.org/professionals/physician\\_gls/f\\_guidelines.asp](https://www.nccn.org/professionals/physician_gls/f_guidelines.asp). Accessed June 15 2017.
5. Lenders JW, Duh QY, Eisenhofer G, Gimenez-Roqueplo AP, Grebe SK, *et al.* (2014) Pheochromocytoma and paraganglioma: an endocrine society clinical practice guideline. *J Clin Endocrinol Metab* 99: 1915–1942.

6. Averbuch SD, Steakley CS, Young RC, Gelmann EP, Goldstein DS, *et al.* (1988) Malignant pheochromocytoma: effective treatment with a combination of cyclophosphamide, vincristine, and dacarbazine. *Ann Intern Med* 109: 267–273.
7. Shapiro B, Gross MD, Shulkin B (2001) Radioisotope diagnosis and therapy of malignant pheochromocytoma. *Trends Endocrinol Metab* 12: 469–475.
8. Sisson JC (2002) Radiopharmaceutical treatment of pheochromocytomas. *Ann N Y Acad Sci* 970: 54–60.
9. Rose B, Matthay KK, Price D, Huberty J, Klencke B, *et al.* (2003) High-dose <sup>131</sup>I-metaiodobenzylguanidine therapy for 12 patients with malignant pheochromocytoma. *Cancer* 98: 239–248.
10. Mullen JP, Cartwright RC, Tisherman SE, Misage JR, Shapiro AP (1985) Pathogenesis and pharmacologic management of pseudo-obstruction of the bowel in pheochromocytoma. *Am J Med Sci* 290: 155–158.
11. Robinson RG, DeQuattro V, Grushkin CM, Lieberman E (1977) Childhood pheochromocytoma: treatment with alpha methyl tyrosine for resistant hypertension. *J Pediatr* 91:143–147.
12. Amery A, Moerman EJ, Bossaert H, de Schaepdryver AF (1969)  $\alpha$ -methyl-p-tyrosine in malignant pheochromocytoma. *Pharmacologia Clinica* 1: 174–176.
13. Engelman K, Horwitz D, Jéquier E, Sjoerdsma A (1968) Biochemical and pharmacologic effects of alpha-methyltyrosine in man. *J Clin Invest* 47: 577–594.
14. Pyörälä K, Pitkänen E, Toivonen S (1968) Alpha-methyl-p-tyrosine in the symptomatic treatment of patients with malignant phaeochromocytoma. *Ann Med Intern Fenn* 57: 65–73.
15. Jones NF, Walker G, Ruthven CR, Sandler M (1968) Alpha-methyl-p-tyrosine in the management of phaeochromocytoma. *Lancet* 2: 1105–1109.
16. Bagnall WE, Salway JG, Jackson EW (1976) Phaeochromocytoma with myocarditis managed with alpha-methyl-p-tyrosine. *Postgrad Med J* 52: 653–656.
17. DEMSER (metyrosine) capsule. FDA prescribing information. <https://www.drugs.com/pro/demser.html>. Accessed October 31 2016.
18. ICH harmonized tripartite guideline: guideline for good clinical practice E6 (R1). Current step 4 version 1996. [https://www.ich.org/fileadmin/Public\\_Web\\_Site/ICH\\_Products/Guidelines/Efficacy/E6/E6\\_R1\\_Guideline.pdf](https://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Efficacy/E6/E6_R1_Guideline.pdf). Accessed June 15 2017.
19. Busner J, Targum SD (2007) The clinical global impressions scale: applying a research tool in clinical practice. *Psychiatry (Edgmont)* 4: 28–37.
20. Steinsapir J, Carr AA, Prisant LM, Bransome ED Jr (1997) Metyrosine and pheochromocytoma. *Arch Intern Med* 157: 901–906.
21. Perry RR, Keiser HR, Norton JA, Wall RT, Robertson CN, *et al.* (1990) Surgical management of pheochromocytoma with the use of metyrosine. *Ann Surg* 212: 621–628.
22. Kuchel O, Buu NT, Edwards DJ (1990) Alternative catecholamine pathways after tyrosine hydroxylase inhibition in malignant pheochromocytoma. *J Lab Clin Med* 115: 449–453.
23. Missale C, Nash SR, Robinson SW, Jaber M, Caron MG (1998) Dopamine receptors: from structure to function. *Physiol Rev* 78: 189–225.
24. Brunton LL, Chabner BA, Knollman BC (2011) Goodman & Gilman's: the pharmacological basis of therapeutics (12th). McGraw-Hill Medical, 288–289.
25. Wachtel H, Kennedy EH, Zaheer S, Bartlett EK, Fishbein L, *et al.* (2015) Preoperative metyrosine improves cardiovascular outcomes for patients undergoing surgery for pheochromocytoma and paraganglioma. *Ann Surg Oncol* 22: S646–S654.