Effects of Exercise on the Absorption of Insulin Glargine in Patients With Type 1 Diabetes

RAJESH PETER, MRCP[1]
STEPHEN D. LUZIO, PHD[1]
GARETH DUNSEATH, MPHIL[1]
ANDY MILES, PHD[2]
BARRY HARE, PHD[2]
KARIANNE BACKX, PHD[2]
VASSEN PAUVADAY, MRCP[1]
DAVID R. OWENS, MD[1]

OBJECTIVE — To study the effects of exercise on the absorption of the basal long-acting insulin analog insulin glargine (Lantus), administered subcutaneously in individuals with type 1 diabetes.

RESEARCH DESIGN AND METHODS — A total of 13 patients (12 men, 1 woman) with type 1 diabetes on a basal-bolus insulin regimen were studied. ^{125}I-labeled insulin glargine at the usual basal insulin dose was injected subcutaneously into the thigh on the evening (2100) before the study day on two occasions 1 week apart. Patients were randomly assigned to 30 min intense exercise (65% peak oxygen uptake [$V\text{O}_{2\text{peak}}$]) on one of these visits. The decay of radioactive insulin glargine was compared on the two occasions using a thallium-activated NaI gamma counter. Blood samples were collected at regular intervals on the study days to assess plasma glucose and insulin profiles.

RESULTS — No significant difference was found in the ^{125}I-labeled insulin glargine decay rate on the two occasions (exercise vs. no exercise; repeated-measures ANOVA, $P = 0.548$). As expected, a significant fall in plasma glucose was observed over the exercise period (area under curve above fasting [ΔAUC] glucose: -0.39 ± 0.11 vs. -1.30 ± 0.16 mmol · l^{-1} · h^{-1}; nonexercise vs. exercise; $P = 0.001$), but insulin levels did not differ significantly on the two occasions (ΔAUC insulin: -2.1 ± 3.9 vs. 1.5 ± 6.2 pmol·l^{-1}·h^{-1}; nonexercise versus exercise; $P = 0.507$).

CONCLUSIONS — An intense 30-min period of exercise does not increase the absorption rate of the subcutaneously injected basal long-acting insulin analog insulin glargine in patients with type 1 diabetes.

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Despite wide fluctuations in nutritional intake and physical exercise, the concentration of plasma glucose in healthy individuals remains within a narrow range throughout the day (3.5–7.0 mmol/l) (1). In contrast, patients with type 1 diabetes have an absolute deficiency of insulin secretion and are therefore dependent on exogenous insulin to regulate their blood glucose concentrations. The purpose of intensive insulin therapy is to mimic physiological insulin secretion by providing a basal insulin replacement together with insulin boluses to cover prandial glucose excursions. The basal replacement should provide a reproducible supply of insulin into the bloodstream, which remains as stable as possible over 24 h to suppress excess postabsorptive hepatic glucose production and to facilitate the action of the bolus insulin (2,3). The bolus insulin comprising a fast-acting insulin administered immediately before meals should prevent postprandial glycemic surges. The most challenging aspect of insulin therapy is to maintain insulin levels that keep blood glucose levels as close to normal as possible without necessarily increasing the risk of hypoglycemia.

Insulin glargine (21A-Gly-30Ba-L-Arg–30Bb-L-Arg-human insulin; Lantus) is a human insulin analog synthesized by recombinant DNA technology. It results from two modifications of human insulin. The first is the addition of two arginine molecules (two positive charges) to the COOH-terminus of the B-chain, which shifts the isoelectric point from a pH of 5.4 to 6.7, making the molecule less soluble at the physiological pH of subcutaneous tissue. Also, asparagine 21 is replaced with glycine, which is charge neutral. This results in delayed dissociation into monomers providing stability to the resulting human insulin analog. Injected subcutaneously as a clear solution at pH 4.0, insulin glargine forms a microprecipitate in the physiological pH of the subcutaneous space, thereby delaying its absorption and prolonging its duration of action (4–6). Small amounts of zinc (30 μg/ml), a hexamer-stabilizing agent, have been added to the insulin glargine formulation to further extend the duration of action following subcutaneous injection (4). With appropriate once-daily dosing, insulin glargine concentrations in the blood of patients with type 1 diabetes will closely mimic physiological basal insulin concentrations found in people without diabetes, providing a near 24-h duration of action.

Previous clinical pharmacological

From the [1]Diabetes Research Unit, Llandough Hospital, Penarth, South Glamorgan, U.K.; and the [2]School of Sport, PE and Recreation, University of Wales Institute, Cardiff, South Glamorgan, U.K.

Address correspondence and reprint requests to Prof. D.R. Owens, Diabetes Research Unit, First Floor, Academic Centre, Llandough Hospital, Penlan Road, Penarth, South Glamorgan CF64 2XX, Wales, U.K. E-mail: owensdr@cf.ac.uk.

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Abbreviations: ΔAUC, area under the curve above fasting.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

studies in healthy subjects have demonstrated that the subcutaneous absorption of radiolabeled drugs occurs significantly earlier for NPH insulin than for 15- and 80-μg/ml insulin glargine formulations (7). In addition, the absorption rates of 30 μg/ml radiolabeled insulin glargine were comparable after subcutaneous injection into the abdominal, leg, and arm regions (7). In patients with type 2 diabetes, the absorption of 30 μg/ml insulin glargine was significantly longer than that for NPH insulin (8).

Previous studies on the effects of exercise on absorption of insulin have been conflicting (9–13). This study was designed to compare the absorption characteristics of insulin glargine injected subcutaneously into the thigh in patients with type 1 diabetes in response to exercise.

RESEARCH DESIGN AND METHODS

— Patients aged between 18 and 45 years with a BMI <32 kg/m^2 were included in the study. All patients had been diagnosed with type 1 diabetes for at least 1 year and had been on a basal-bolus regimen for at least 3 months. Patients with preproliferative or proliferative retinopathy or clinically significant cardiac, hepatic, or renal abnormalities were excluded from the study.

This was a single-center randomized open-label study to compare the effect of exercise on the absorption of subcutaneously injected radiolabeled insulin glargine in type 1 diabetes. The study was approved by the Bro Taf Research Ethics Committee and was conducted in accordance with Good Clinical Practice and complied with the Declaration of Helsinki. Written informed consent was obtained before conducting any of the study-related procedures.

The study consisted of a screening visit to determine eligibility followed by a minimum 2-week titration period during which doses of insulin glargine were titrated to achieve a fasting plasma glucose concentration of 4–7 mmol/l. Patients previously not using insulin glargine as basal insulin were switched over to insulin glargine before the titration period. At the screening visit, all patients had a full medical examination with biochemical and hematological screening and an ultrasound examination of the injection site (midway between the anterior superior iliac spine and the superior aspect of the patella) to determine the depth of subcutaneous tissue to exclude the possibility of intramuscular injection. This was then followed by a visit to determine peak oxygen uptake ($V\text{O}_{2\text{peak}}$).

$V\text{O}_{2\text{peak}}$ measurement

$V\text{O}_{2\text{peak}}$ was measured using an electromagnetically braked cycle ergometer. Patients began cycling with an initial load of 60 W for 2 min, which was increased by 30-W increments every 2 min. The test was continued until the patient could no longer maintain a cycling frequency of 60 rpm. Two 60-s expired air samples were collected between the 5th and 6th min and again just before test termination using Douglas bags (14). The O_2% (Servomex 1440C Oxygen Analyzer) and CO_2% (PK Morgan Carbon Dioxide Analyzer) concentrations in each of the expired air samples were established. Along with the O_2% and CO_2% concentrations, the volume of air expired, room temperature, barometric pressure, and subject mass were entered into a gas analysis computer package (Expair, Cranlea, and Company, Birmingham, U.K.) to determine $V\text{O}_{2\text{peak}}$.

Patients returned on two study days 1 week apart after being randomized to exercise on one of these visits. On the evening before each study day, patients came into the investigation unit at 2030. Potassium iodide in 120-mg tablet form was given to each patient to block thyroid radioiodine uptake. A thallium-activated NaI gamma counter was positioned at a net distance of 50 mm from the site of injection to measure the background count rate over 10 min. At 2100, ^{125}I-labeled insulin glargine (iodination on tyrosine at position 14 on the A chain, specific activity 349 mCi/mg) was administered subcutaneously into the thigh at the usual basal insulin dose of the patient at the marked injection site. The injection technique involved inserting the needle into a skinfold at 45° to the skin surface and slowly injecting the insulin. The radioactivity count rate at the injection site was measured for 5 min immediately after the injection. After this count period, the patients were allowed to return home. The following morning, the patients returned to the diabetes investigation unit at 0800 having fasted for 10 h. A cannula was inserted into a forearm vein for repeated blood sampling. The cannula was attached via a three-way tap to a slow running saline infusion to maintain patency of the vein. The normal morning dose of bolus insulin (aspart or lispro) was then given into the patient's abdomen followed (within 2 min) by a 500-kcal standardized mixed meal (58% carbohydrate, 22% fat, and 20% protein) at 0830. Fasting blood samples were collected at −30 and 0 min, with subsequent samples collected at 15-min intervals for the duration of the study day and the last sample collected at 1200. Radioactivity count rate was determined at 0800 and then at 30-min intervals until 1200. Patients remained in a semirecumbent position except during the exercise period. On one of the study days (in random order), the patients carried out exercise 1 h after the standardized meal. The exercise period consisted of 30 min exercise at ~65% $V\text{O}_{2\text{peak}}$ on the cycle ergometer. Patients were monitored for any adverse events including symptomatic and asymptomatic hypoglycemia (plasma glucose <2.5 mmol/l). After the last radioactivity count measurement was obtained, patients were given food and discharged.

Blood samples were taken for the measurement of plasma glucose and insulin levels. After sampling, the blood was centrifuged (2,000*g*, 5 min) in a refrigerated (4°C) centrifuge, and the plasma was aliquoted and frozen at −20°C until assay. Plasma glucose concentrations were measured using a Yellow Springs glucose analyzer (YSI 2300; Yellow Springs Instruments, Aldershot, Hants, U.K.). Plasma insulin was measured by immunoassay (Specific Insulin ICMA; MLT Research, Cardiff, Wales, U.K.) and cross-reactivity of human insulin, insulin glargine, insulin aspart, and insulin lispro of ~100%.

Analysis measures and statistics

Data were analyzed using SPSS for Windows, version 11. Area under the curve above fasting (ΔAUC) for glucose and insulin profiles were calculated by the trapezoidal method and compared by Wilcoxon's test. Repeated-measures ANOVA was used to compare the effects of exercise on the radioactive count decay over the study period. Significance was determined as $P < 0.05$.

RESULTS

— The demographic data and baseline characteristics for the 13 patients (12 men, 1 woman) who par-

Table 1 —*Demographic data and baseline measurements*

Characteristic	Mean ± SD
Age (years)	33.3 ± 6.5
Height (m)	1.78 ± 0.06
Weight (kg)	84.6 ± 12.8
BMI (kg/m^2)	26.8 ± 3.3
HbA_{1c} (%)	7.6 ± 1.3
Systolic/diastolic blood pressure (mmHg)	121.1 ± 14.5/76.8 ± 7.3
Exercise intensity (% of Vo_{2peak})	65.2 ± 10.1
Dose of insulin glargine (units) on the 2 study days (no exercise/exercise)	27.2 ± 9.1/27.2 ± 9.0
Rapid-acting insulin doses (units) on the 2 study days (no exercise/exercise)	10.4 ± 4.1/8.2 ± 3.0

ticipated in the study are presented in Table 1.

Decay of radioactivity

The mean decay of the radioactivity count over time expressed as a percentage of the count rate measured in the first 5 min after administration of ^{125}I-labeled insulin glargine (100%) on the two study days are represented in Fig. 1.

At the end of the study period, 15 h after the injection of radiolabeled glargine, the mean (± SD) percentage radioactivity on the nonexercise study day was significantly lower than that on the exercise study day (73.0 ± 6.5 vs. 76.3 ± 6.9%, $P = 0.046$); however, no significant difference was found in the rate of decay on the two study days (repeated-measures ANOVA, $P = 0.548$).

Plasma glucose and insulin levels

Plasma glucose. Mean plasma glucose profiles for the two study days are represented in Fig. 2. Fasting plasma glucose levels (means ± SE) were similar on both nonexercise and exercise study days (8.2 ± 0.9 and 8.4 ± 0.7 mmol/l, respectively).

Mean ΔAUC glucose over the total 210-min postprandial period showed no difference between the nonexercise and exercise days (1.69 ± 2.54 vs. −0.61 ± 1.76 mmol · l^{-1} · h^{-1}; $P = 0.345$). However, ΔAUC glucose was significantly lower on the exercise study day during the exercise period (−0.39 ± 0.11 vs. −1.30 ± 0.16 mmol · l^{-1} · h^{-1}; $P = 0.001$) but not the postexercise period (−3.63 ± 0.97 vs. −0.12 ± 0.54 mmol · l^{-1} · h^{-1}; $P = 0.001$).

Plasma insulin. Mean plasma insulin profiles for the two study days are represented in Fig. 3. Fasting plasma insulin levels were similar on both the nonexercise and exercise study days (76.8 ± 12.3 vs. 82.2 ± 12.7 pmol/l, respectively; $P = 0.610$).

Mean ΔAUC insulin over the total 210-min postprandial period showed no difference between the nonexercise and exercise days (600.2 ± 99.9 vs. 483.2 ± 79.6 pmol · l^{-1} · h^{-1}; $P = 0.116$). Also, no differences in ΔAUC insulin were observed between the nonexercise versus the exercise study days during the exercise (−2.1 ± 3.9 vs. 1.5 ± 6.2 pmol · l^{-1} · h^{-1}; $P = 0.507$) and postexercise periods (−162.9 ± 58.5 vs. −122.1 ± 40.6 pmol · l^{-1} · h^{-1}; $P = 0.780$). However, there was a statistically significant difference between the no exercise versus exercise study days at the 105-min time point (330.0 ± 62.2 vs. 232.7 ± 26.1 pmol/l; $P = 0.019$).

Safety

On three occasions, patients had asymptomatic hypoglycemia requiring oral glucose. Two of these incidents were on the study days when there was no exercise,

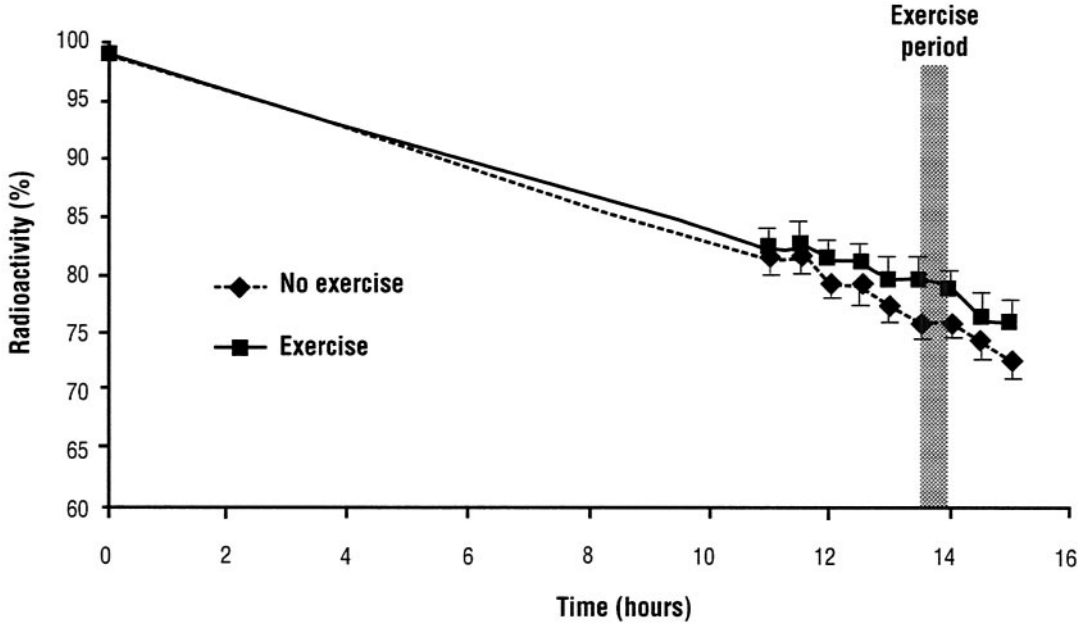

Figure 1—*Mean (± SE) radioactive disappearance.*

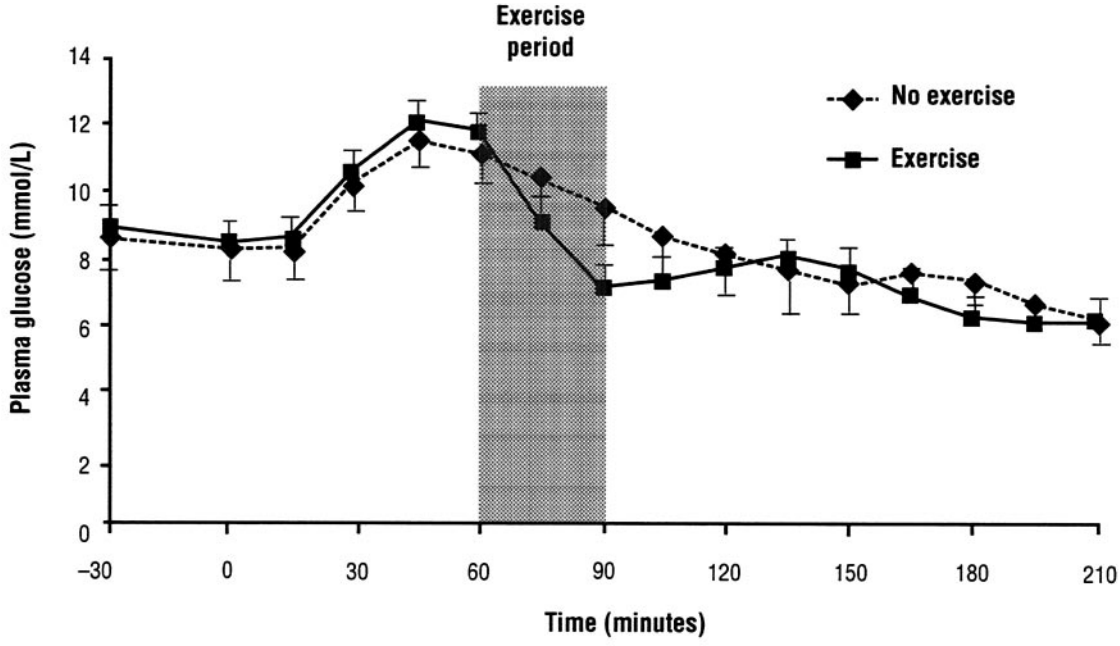

Figure 2—*Mean (± SE) plasma glucose.*

with both episodes occurring ~2 h after the meal. One patient was hypoglycemic on the exercise study day, 3 h after the meal and 90 min after the exercise period.

CONCLUSIONS — The absorption of subcutaneously administered insulin depends on various factors, including the physical state of the insulin (monomers, dimmers, or hexamers) (15,16), injection volume and concentration (15), injection needle used (subcutaneous or intramuscular) (17), injection site (16,18), and local temperature (16,18). The effect of exercise on the absorption of subcutaneously injected insulin will also depend on other factors, such as local blood flow (18) and the massaging effect of the musculature (16,18). Studies using iodinated insulins and related products also show that deiodination can occur after injection because of the presence of ubiquitous deiodinases (19). However, in this study, it was assumed that deiodination of the radiolabeled insulins occurred at a similar order of magnitude on the two study days. Previous studies looking at the in-

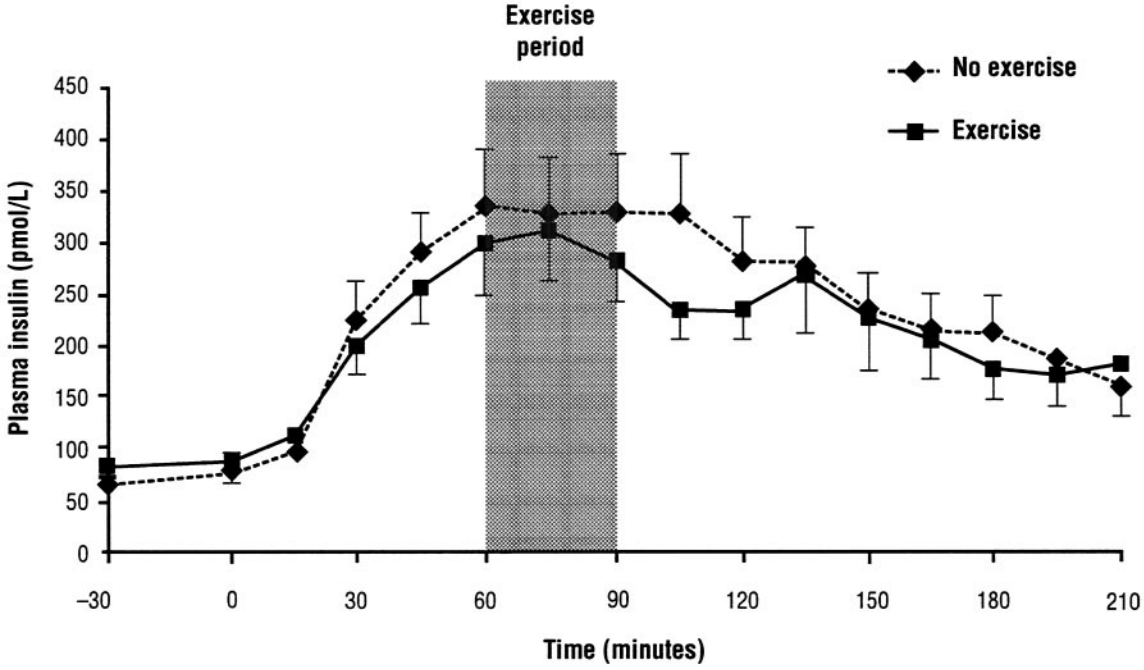

Figure 3—*Mean (± SE) plasma insulin.*

fluence of physical exercise on the absorption of subcutaneously injected insulin have been conflicting. According to some of these studies, subcutaneously injected insulin is absorbed faster from the subcutaneous tissue overlying an exercising muscle than from a remote site (9–13,20,21). These results contrast with the general physiological principle that blood flow is shifted from the subcutaneous tissue of the exercising limbs to the trunk for reasons of thermoregulation and blood supply to the exercising muscle (22–24). Conversely, there are also reports showing no difference in insulin absorption rates in response to exercise (22,25). Under the conditions of this study, we did not find any difference in the disappearance of ^{125}I-labeled insulin glargine from the subcutaneous depot in response to exercise in our cohort of type 1 diabetic patients.

In this study, there was no change in plasma insulin concentrations between the two study days, but there was a fall in plasma glucose levels during the exercise period. This fall in glucose was hence independent of insulin mobilization from the depot site. It has been shown that during the initial minutes of exercise, the rapidly stimulated glycolytic flux is predominantly maintained by muscle glycogenolysis (26), after which glucose homeostasis during exercise is maintained by a precise balance between hepatic glucose production (hepatic glycogenolysis and to some extent gluconeogenesis) and peripheral glucose utilization. The ability of nondiabetic individuals to decrease insulin levels during exercise (27) allows an adequate increase of hepatic glucose production in this situation. In patients with type 1 diabetes, this endogenous regulation is absent and circulating plasma insulin levels can remain static or even be elevated; therefore, hepatic glucose production cannot rise adequately to meet the increased energy needs of the exercising muscle (28). The increase in plasma glucose levels after exercise observed in our study could be due to continuing glucose mobilization combined with a fall in peripheral glucose utilization.

We conclude that, under the conditions of this study, the stated exercise protocol (exercise predominantly to the legs) did not accelerate the absorption of ^{125}I-labeled insulin glargine injected subcutaneously into the thigh. Therefore, the fall in plasma glucose was not related to an increase in insulin glargine absorption but was most likely caused by the relatively persistent hyperinsulinemic state, which was sustained during the exercise and postexercise periods. Increased absorption of fast-acting insulin analog cannot be ruled out, and patients would have to be cautious and make appropriate dose adjustments before commencing exercise (10). This study suggests that insulin glargine can be safely and effectively administered without a dose change during exercise. However, the study did not address late exercise-induced hypoglycemia, which cannot be ruled out from our study, and in some patients, insulin glargine reductions may be warranted depending on individual patient responses. It must also be pointed out that exercise activity may differ from our experimental conditions, and care must be taken not to overinterpret the results of the study. Further studies will elucidate whether the predictability of insulin glargine can be maintained during prolonged periods of exercise.

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