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Tumor growth inhibition in patients with prostatic carcinoma treated with luteinizing hormone-releasing hormone agonists

(prostate cancer treatment/analogues of luteinizing hormone-releasing hormone)

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ABSTRACT **Ten patients with prostatic carcinoma—six with stage C and four with stage D disease—were treated for 6 weeks to 12 months with agonistic analogues of luteinizing hormone-releasing hormone (LH-RH). [D-Trp^6]LH-RH was given subcutaneously once daily at a dose of 100 μg and [D-Ser(Bu^t)^6]des-Gly-NH$_2$^{10}-LH-RH ethylamide (HOE 766) was given subcutaneously (50 μg once daily) or intranasally (500 μg twice daily). In all patients, mean plasma testosterone levels showed a 75% suppression by the third week of treatment and remained low thereafter. This was followed by a decrease or normalization of plasma acid phosphatase levels by the second month of treatment and a 47% decrease in serum alkaline phosphatase by the 10th week of treatment in all but one patient. In patients with stage C disease presenting with prostatism or urinary outflow obstruction, there was a noticeable clinical improvement. In two such patients, a decrease in the size of the prostate was confirmed by ultrasonography. In patients with stage D disease manifested by diffuse bone metastases, there was relief of bone pain, and in one patient treated for >12 months the improvement was documented by radioisotope bone imaging. It is concluded that superactive agonistic LH-RH analogues hold promise as therapeutic agents in patients with androgen-sensitive prostatic adenocarcinoma. Furthermore, the analogues of LH-RH may be used to assess the responsiveness of patients to surgical castration. Long-term administration of LH-RH analogues could become an alternative to surgical castration and estrogen therapy for the treatment of hormone-dependent prostatic carcinoma.**

There is much evidence that acute administration of superactive analogues of luteinizing hormone-releasing hormone (LH-RH) causes a prolonged release of pituitary gonadotropins, which leads to stimulation of Leydig cell function and an increase in plasma testosterone (T) concentrations (1–8). Chronic administration of large doses of superactive analogues of LH-RH results in suppression of both pituitary and Leydig cell function in animals and men (1, 2, 4, 5, 7, 9–16). This paradoxical antigonadal effect of superactive analogues of LH-RH can result in regression of both mammary and prostatic endocrine-dependent tumors in experimental animals (17, 18). The present report describes the results of administration of two synthetic superactive long-acting LH-RH analogues, [D-Trp^6]LH-RH and [D-Ser(Bu^t)^6]des-Gly-NH$_2$^{10}-LH-RH ethylamide (HOE 766), to 10 patients with prostatic carcinoma.

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PATIENTS, METHODS, AND MATERIALS

Patients. Ten patients with biopsy-proven prostatic adenocarcinoma were studied. Six presented with signs of prostatism and two of the six had urinary outflow obstruction requiring frequent catheterization. None of these six patients (C_1, C_2, C_3, C_4, C_5, C_6; 75, 70, 63, 79, 79, and 81 years old, respectively) had evidence of metastasis and therefore were classified as having stage C disease. The four other patients (D_1, D_2, D_3, D_4; 65, 71, 78, and 63 years old, respectively) presented with extreme back and leg pain which had resulted in severe incapacitation. Patients D_1 and D_3 were practically bedridden. Patient D_2 required lumbar laminectomy. In patients D_1 and D_4, bone survey by conventional radiography and isotopic scanning revealed the presence of widespread osteoblastic metastasis in the entire skeleton. There was no evidence of liver or lung metastasis in any of the patients. All but patient C_3 experienced angina, myocardial infarction, or thromboembolic episodes. D_4 was the only patient who had been treated with estrogens; because painful gynecomastia developed and there was no relief in bone pain, estrogen treatment was discontinued. Estrogen therapy was contraindicated for all other patients because of their medical histories. Surgical castration was an alternative mode of therapy, especially for the patients with stage D disease. However, in view of our previous experience in achieving medical castration with [D-Trp^6]LH-RH in transsexual subjects (15, ††), we suggested this method of treatment. All patients agreed to be so treated and signed an informed consent form approved by the McGill Cancer Center.

All patients were asked to return at weekly intervals for the first month and every other week during subsequent months of treatment. Patients C_4, C_5, C_6, and D_3 were initially treated with [D-Trp^6]LH-RH at a dose of 100 μg subcutaneously once daily. Patient C_4 discontinued the medication for nonmedical reasons after 3 months but, after a 6-week interval, resumed treatment with HOE 766 in doses of 500 μg intranasally twice daily. Patients C_1, C_2, C_3, and D_4 also received 500 μg of HOE 766 intranasally twice daily. Patients D_1 and D_2 were injected subcutaneously with HOE 766 at a dose of 50 μg daily. The

Abbreviations: LH-RH, luteinizing hormone-releasing hormone; T, testosterone; E_2, 17β-estradiol; HOE 766, [D-Ser(Bu^t)^6]des-Gly-NH$_2$^{10}-LH-RH-ethylamide.

†† Tolis, G., Mehta, A., Dimitrakopoulos, C., Stellos, A., Ackman, D., Kinch, R., Comaru-Schally, A. M., Schally, A. V. & Max, D., Annual Meeting, Endocrine Society, Cincinnati, OH, June 17, 1981, p. 91, (abstr.).

duration of treatment varied from 60 days to 14 months.

Methods. Prior to the administration of the daily dose of the medication and at the weekly or fortnightly visits, blood was taken to determine the levels of plasma T, 17β-estradiol (E_2) (19, 20), and acid phosphatase (21) by radioimmunoassay techniques as well as alkaline phosphatase (as part of the SMA-16 performed by Technicon AutoAnalyzer). Prostatic size was assessed by rectal examination and by transabdominal ultrasonography. Bone scanning was performed with ^{99m}Tc-labeled methylene diphosphonate.

Materials. Two superactive long-acting LH-RH analogues, functionally identical and possessing similar physiological and clinical activities (1, 2), were used in this study. The decapeptide (pyro)Glu-His-Trp-Ser-Tyr-D-Trp-Leu-Arg-Pro-Gly-NH_2 ([D-Trp^6]LH-RH) was synthesized by solid-phase methods or by classical synthesis and purified as described (8). During the past 6 years, [D-Trp^6]LH-RH has been evaluated in more than two dozen different studies (1, 2, 8, 18) in laboratory animals and in primates. In toxicological evaluations (including 28-day tests in rats and monkeys) in doses of up to 200 μg/kg, this substance caused no limiting toxic effects except for reversible reductions in the weights of reproductive organs. [D-Trp^6]LH-RH has been used clinically in at least 20 previous clinical studies without displaying any adverse side effects (1–3, 6, 12, 15). The absence of toxicity of [D-Trp^6]LH-RH is also recorded in master drug files (MDF) 3572 and 3576 of Ayerst Laboratories.

The nonapeptide (pyro)Glu-His-Trp-Ser-Tyr-D-Ser(Bu^t)-Leu-Arg-Pro-ethylamide (HOE 766) (Hoechst AG, Frankfurt, Federal Republic of Germany) was prepared by classical synthesis and purified. During the past 5 years, extensive physiological, toxicological (1, 2, 13), and clinical investigations (1, 2, 4–7, 14) have shown this substance to be devoid of adverse effects, except for reversible inhibition of reproductive function on chronic administration. This material was kindly supplied by the Medical Department, Hoechst Canada, Montreal, Quebec, Canada, in the form of an intranasal spray preparation. The injectable form was prepared by dilution in the Pharmacy of the Royal Victoria Hospital, Montreal.

RESULTS

Clinical. By the third week of therapy and continuing thereafter, all patients with stage C disease (patients C_1–C_6) reported relief of symptoms and reversal of the signs of prostatism. Assessment of the size of the prostate by rectal examination was performed independently by three of the authors (G.T., D.A., A.S.) for patients C_1, C_3, and C_4; all agreed that there was a reduction in the size of the prostatic mass. Ultrasonography, performed in two patients (C_3 and D_3), confirmed a reduction in the prostatic size. An image of patient C_3 is shown in Fig. 1. Urinary outflow obstruction was relieved in two patients during their treatment with [D-Trp^6]LH-RH. In patient C_4, cystometry documented a decrease in the residual urine volume from 800 ml to 31 ml, and in patient C_5 the Foley catheter could be removed. Patients C_1, C_2, and C_3 have now been treated for 4 months. Patients C_4, C_5, and C_6 were subjected to orchiectomy after the third month of treatment because they moved out of Montreal to remote areas where they could not be treated. They continue to be in good health, having been followed now for 14–18 months.

Three of the four patients with stage D disease who had bone metastases (patients D_1, D_2, and D_3) experienced a decrease in bone pain which led to a lowering of the dose of analgesic. As a result, these patients were able to resume some outdoor activities. They continue their treatment and have now been followed for 4–12 months. Patient D_4 reported no change in bone pain.

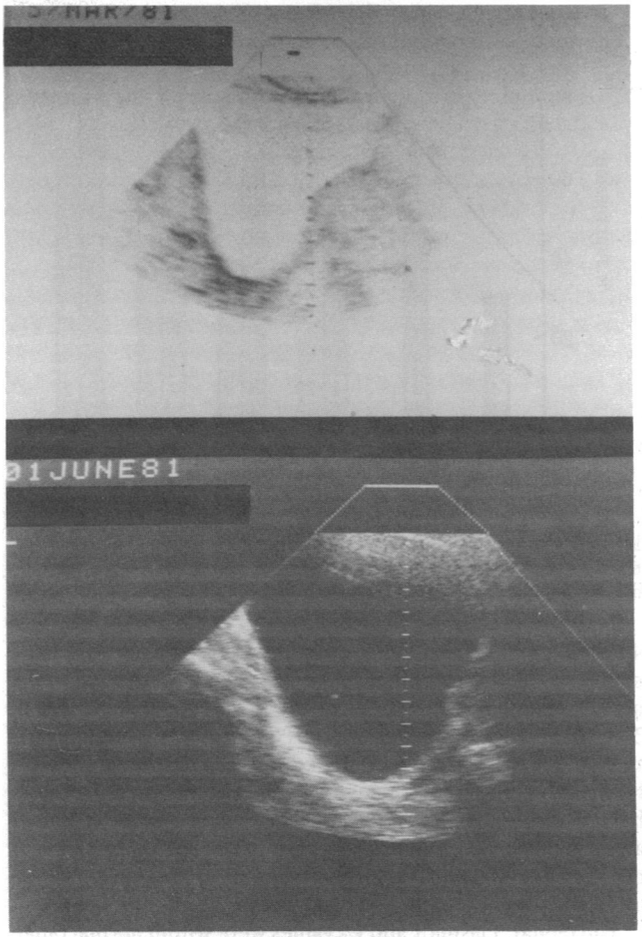

FIG. 1. Prostatic transabdominal ultrasonography in patient C_3 prior to (*Upper*; March 1981) and during (*Lower*; June 1981) treatment with an LH-RH analogue. Note the improvement during treatment illustrated by the creation of space between the midline and a mass on the right side which, on the March evaluation, was protruding and impinging upon the midline.

Both intranasal and subcutaneous administration of analogues were tolerated well by all patients. One patient treated with the intranasal regimen complained of some epigastric burning. Six patients developed climacteric-like vasomotor phenomena consisting of profound perspiration and hot flashes; all complained of a decrease in libido and erectile potency.

Biochemical. Two of the patients with stage C disease had increased serum acid phosphatase levels (C_1, 7.8 ng/ml; C_2, 3.5 ng/ml; normal, <2 ng/ml) which normalized by weeks 8 or 3 of treatment, respectively. These values remained in the normal range up to the last time studied (month 4 follow-up).

All patients with stage D disease had increased acid phosphatase (18.1–98.4 ng/ml) and alkaline phosphatase levels (288–1482 units/liter; normal <150 units/liter). Acid phosphatase levels declined in three of the four patients (D_1, D_2, and D_3) by week 4 of treatment (Fig. 2). In patients D_1 and D_2 levels decreased to normal or close to normal by the second to fourth months (1.5 and 3.7 ng/ml, respectively). In patient D_3, acid phosphatase levels decreased by 82% to 8.0 ng/ml during week 8 of treatment with [D-Trp^6]LH-RH (100 μg/day, subcutaneously). Upon discontinuation of the medication, the level increased to 58.5 ng/ml; it decreased to 10.6 ng/ml by week 8 of treatment with HOE 766 (500 μg twice daily, intranasally). In contrast to these patients, in patient D_4 there was an increase in acid phosphatase levels from 49.9 to 116.3 ng/ml by week

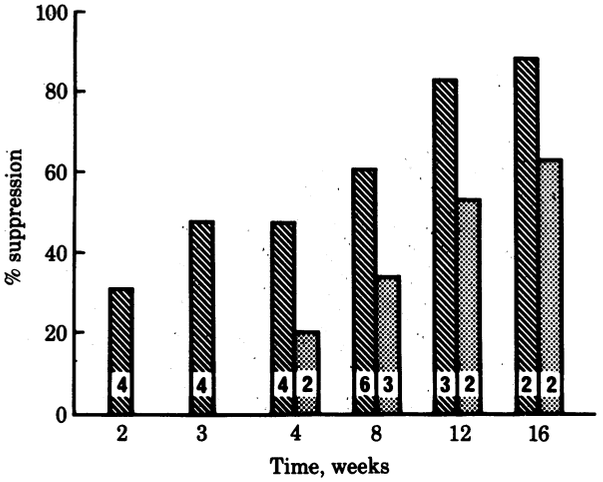

FIG. 2. Percent suppression of acid ▨ and alkaline ▩ phosphatase levels in patients with prostatic carcinoma stage C and D during treatment with agonistic analogues of LH-RH. Numbers inside the bars denote the number of patients tested.

4 of treatment; in this patient, plasma testosterone levels were suppressed in a fashion similar to that seen in patients D_1, D_2, and D_3. Serum alkaline phosphatase levels were decreased to 50% of the initial values by months 3 and 4 of treatment in patients D_1 and D_2 and continue to decrease. In patient D_1, who has had the lengthiest follow-up period (12 months), the levels have decreased from 1482 units/liter to 206. Normalization of alkaline phosphatase levels was seen in patient D_3 by week 10 of treatment. In patient D_4, alkaline phosphatase levels showed an increase at both 4 and 6 weeks of therapy.

Hormonal. Plasma T and E_2 values were within normal range in all patients irrespective of the stage of the disease. Administration of LH-RH analogues led to a significant suppression of both T and E_2 values by week 2 of treatment; this suppression continued during the months of follow-up treatment in all patients (Fig. 3). Discontinuation of the therapy with [D-Trp^6]LH-RH (100 μg/day, subcutaneously) in patient D_3 resulted in a rebound of plasma T and E_2 from 50 and 0.4 ng/dl to 1005 and 3.2 ng/dl, respectively. Administration of HOE 766 (500 μg twice daily, intranasally) induced a 50% suppression by week 2 of treatment; by week 8 of treatment, plasma T levels were decreased to 48 ng/dl, a value similar to the levels achieved by week 4 of treatment with [D-Trp^6]LH-RH.

Radiological. In two patients, C_3 and D_3, the size of the prostate was estimated by ultrasonography after 3 months of treatment with LH-RH analogues. In both, a decrease in size was found—patient C_3, from 5 × 4 cm to 4 × 3 cm; patient D_3, from 4.5 × 5 to 3.5 × 4.5 (width and depth, respectively). An image of patient C_3 is shown in Fig. 1. Repeated bone scans at 6, 13, 22, and 33 weeks of therapy in patient D_1 revealed a significant and continuous decrease in the number and intensity of the bone lesions (Fig. 4); prior to therapy, isotopic bone imaging revealed diffuse osteoblastic lesions involving the dorsal and lumbar spine, sacrum, pelvis, both femora, both humeri, rib cage, both scapulae, and shoulders. By month 10 of treatment, the bone scan was practically normal. The only lesions detected at the time of writing were in the left ischium and right anterior lower rib. (Additional prints of bone scans are available on request.)

DISCUSSION

The present data provide evidence that long-term administration of certain agonistic LH-RH analogues can be beneficial to

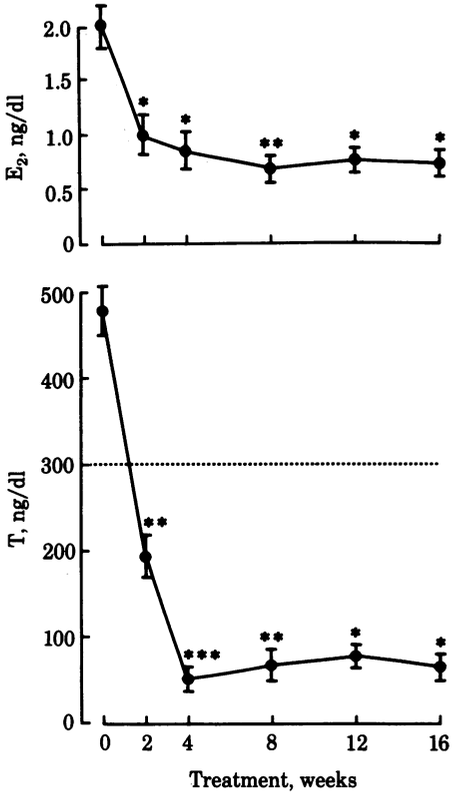

FIG. 3. Effects of agonistic analogues of LH-RH on plasma sex steroids in patients with prostatic carcinoma. Note the lowering and persistent suppression of plasma E_2 (*Upper*) and T (*Lower*) during treatment. Dotted line, lowest limit of normal. *P* values: *, <0.05; **, <0.005; ***, <0.001.

patients with prostatic carcinoma as manifested by clinical, biochemical, and radiological criteria. In addition, persistent suppression of Leydig cell function, as manifested by significant decreases in plasma testosterone and estradiol plasma levels, was also demonstrated. These observations support our previous findings that [D-Trp^6]LH-RH induced medical castration in male transsexual patients (15). Gonadal suppression resulting from long-term administration of LH-RH agonists was also demonstrated in normal men (14, 16). Recently, [D-Trp^6]LH-RH and a closely related agonist have been used effectively for the treatment of true idiopathic precocious puberty (22, 23).

Treatment with LH-RH agonists induced a suppression of acid phosphatase in all but one patient. In this patient, plasma testosterone was suppressed to castrate levels, indicating that his cancer was testosterone insensitive. On the other hand, an androgen-dependent prostate cancer was well demonstrated in patient D_3 in whom a concomitant rebound in plasma testosterone and acid phosphatase levels was found upon cessation of treatment with LH-RH agonist.

The significant regression of prostate size, the decrease in acid and alkaline phosphatase levels, the relief of bone pain, and the normalization of bone isotope scans are probably a result of decreased levels of circulating androgens in the treated patients. In agreement with our present human studies, previous data in rodents have documented the ability of LH-RH agonists, given alone or in combination with anti-androgens, to inhibit normal or tumorous growth of accessory sex organs (9, 18, 24). The mechanism by which LH-RH agonists suppress Leydig cell function is not completely clear (18). Although there appears to be a direct inhibitory effect of LH-RH agonists upon steroidogenesis (25), recent data also indicate a pituitary-dependent

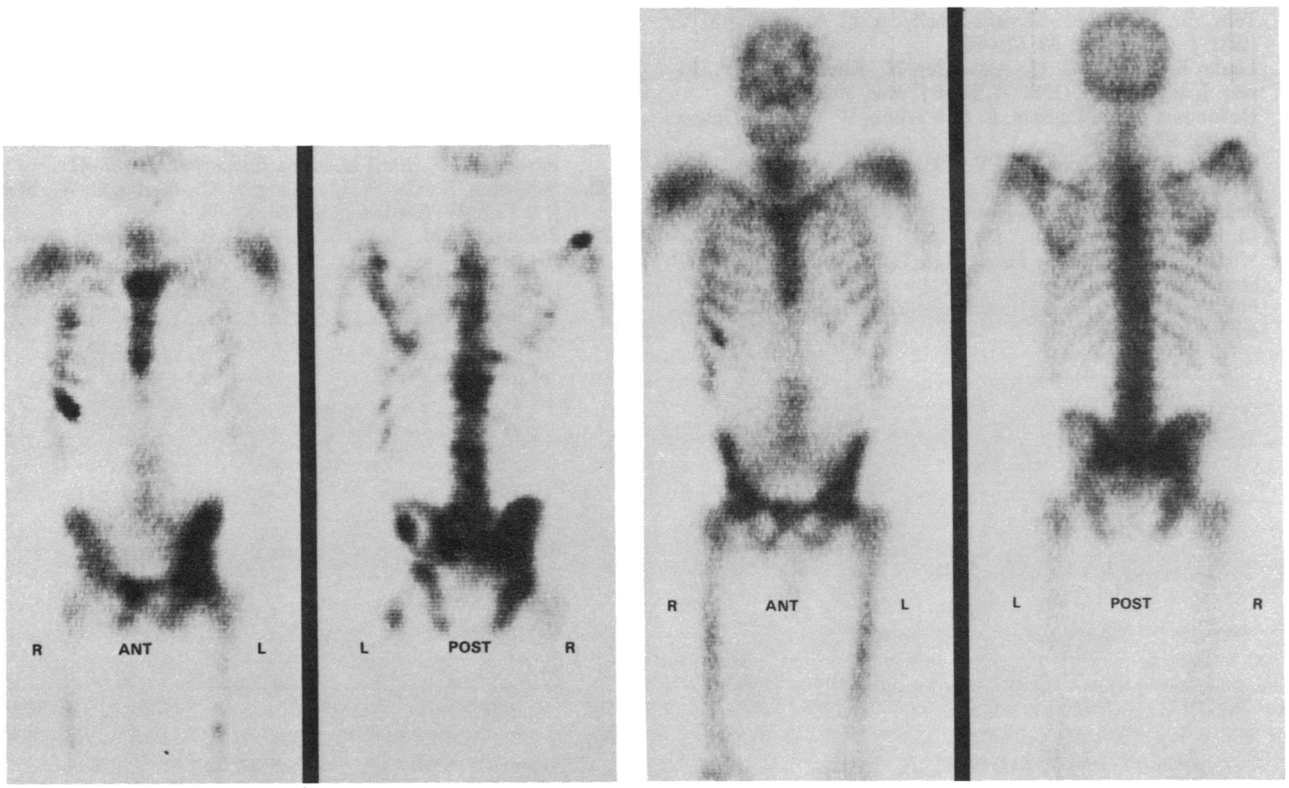

FIG. 4. Bone scans, with ^{99m}Tc-labeled methylene diphosphonate, of patient D_1 treated with HOE 766, 50 μg/day, subcutaneously. (*Left*) Nov. 18, 1980, showing extensive and intensive metastatic disease involving dorsal and lumbar spine, sacrum, pelvis, both femora, both humeri, rib cage, both scapulae, and shoulders. Details: treatment, 6 weeks; T, 15 ng/dl; alkaline phosphatase, 834 units/liter; acid phosphatase, 1.5 ng/ml. (*Right*) May 26, 1981. Note persistence of widespread metastases, but the foci show less-intense activity, indicating significant improvement. Details: treatment, 33 weeks; T, 12 ng/ml; alkaline phosphatase, 247 units/liter; acid phosphatase, 1.0 ng/ml.

mechanism (26). On the other hand, the possibility of a direct effect of LH-RH agonists upon hormone-sensitive prostatic cancer has to be considered because [D-Trp^6]LH-RH has been reported to blunt the action of estrogens and androgens on the female and male reproductive tracts in rodents (2, 25).

Regardless of the mechanism of action of LH-RH analogues on prostate, the regression of prostate size and metastatic disease manifested clinically, radiologically, and biochemically is encouraging. Analogues of LH-RH may open new vistas for the treatment of some hormone-dependent carcinomas. Confirmation of our findings may decrease or eliminate the need for surgical orchiectomy. Furthermore, a positive response to LH-RH analogues by the methods described herein may identify those patients bearing hormone-sensitive prostatic adenocarcinomas. Conversely, a lack of response to LH-RH agonists could detect patients with hormone-insensitive prostate cancer so that unnecessary procedures (such as castration) or medications (such as estrogens) which are associated with morbidity may be avoided. In this respect, it is of interest that the LH-RH analogues used induced tumor regression despite a concomitant decrease in endogenous estrogens during therapy. LH-RH analogues may provide physicians with an estrogen-free treatment for patients suffering from prostatic carcinoma. This would prove particularly advantageous for patients who, because of cardiovascular or thromboembolic disease, cannot tolerate estrogen therapy. Our findings suggest the merit of further therapeutic trials of LH-RH analogues in the management of prostatic carcinoma.

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