Effect of SR 49059, a V1a vasopressin receptor antagonist, in Raynaud’s phenomenon

D. Hayoz, G. Bizzini, B. Noël, M. Depairon, M. Burnier, C. Fauveau, A. Rouillon, R. Brouard and H. R. Brunner

Division of Hypertension and Vascular Medicine, CHUV, Lausanne, Switzerland and Sanofi Research, Gentilly, France

Abstract

Objective. To assess whether vasopressin V1a receptor blockade reduces the abnormal vasoactive response to cold in patients suffering from Raynaud’s phenomenon (RP).

Methods. SR 49059, an orally active, non-peptidic vasopressin V1a receptor antagonist, was given orally (300 mg once daily) to 20 patients with RP in a single-centre, double-blind, placebo-controlled, randomized cross-over study with two 7-day periods of treatment separated by 21 days of washout. Bilateral finger systolic blood pressure and skin temperature were assessed before and after immersion of the hand in cold water for 3 min (15°C) during the screening phase and three times (before and 2 and 4 h after drug intake) on days 1 and 7 of each of the two treatment periods. Recovery of digital pressure and skin temperature was measured 0, 10, 20 and 32 min after the end of the cold immersion test.

Results. SR 49059 significantly attenuated the cold-induced fall in systolic pressure by 14.5% (95% confidence interval 0–29; \( P = 0.045 \)) on the most affected hand on day 7 compared with placebo. Temperature recovery after the end of the cold test showed a trend to enhancement 2 and 4 h after SR 49059 on day 7 (\( P = 0.060 \) and \( P = 0.062 \) respectively). The beneficial effects on finger pressure and temperature recovery were obtained without changes in supine blood pressure or in heart rate.

Conclusion. SR 49059 given orally once a day for 7 days to patients with RP showed favourable effects compared with placebo on finger systolic pressure and temperature recovery after cold immersion, without inducing side-effects.

Key words: SR 49059, Raynaud’s phenomenon, Vasopressin, Receptor antagonist, Human.

Exposure to cold and emotional stress causes an abnormally strong vasospastic response in the digital arteries of patients with Raynaud’s phenomenon (RP) [1]. The paroxysmal vasospastic crisis common in younger women produces digital pallor, usually followed by cyanosis and erythema. In spite of considerable research into the pathophysiological mechanisms of RP, there is little definitive data clarifying the underlying abnormal vasospasm. Adrenoreceptors of types \( \alpha \) [2, 3] and \( \beta \) [4, 5], neurotransmitters such as dopamine [6], histamine [7–9], serotonin [10, 11], neuropeptide Y [12, 13], vasoactive intestinal peptide [14], acetylcholine [15, 16], endothelin [5, 17, 18] and more recently calcitonin-gene-related peptide [19, 20], a potent vasodilator, have all been suggested to play a role in RP. Different combinations of these mediators may be involved according to the category of the RP. Indeed, the pharmacological features of RP secondary to vasculitis and to systemic sclerosis in particular might be quite different from those of primary RP.

Independently of the origin of the abnormal vasospastic response, the treatment of RP relies first on conservative measures such as protection from cold, and when necessary on calcium-channel blockers or topical vasodilators. However, the long-term benefit of these agents may be limited because of adverse effects such as headache, flushing and ankle swelling [21–23].

In a recent study we observed that intra-arterial infusion of vasopressin in the brachial artery induced a dual effect on blood flow in the forearm [24]. It reduced blood flow in the skin in a dose-dependent manner, with extreme blanching of the fingers, whereas it increased blood flow in muscles [25, 26]. Furthermore, vasopressin can activate platelet aggregation, which may contribute to the vasospasm by releasing vasoconstrictive compounds [27]. The local reaction of the fingers prompted us to investigate whether vasopressin is involved in the control of vascular tone in the upper extremities and therefore plays a role in RP. The drug SR 49059 is a highly selective, orally active V1a vasopressin antagonist [28]. It is a non-peptide compound with a high affinity...
and selectivity for animal and human vasopressin (V1a) subtype receptors. Oral administration of SR 49059 inhibits AVP-induced platelet aggregation in a time- and dose-dependent manner [28, 32]. It appears to be devoid of the classical side-effects of the vasodilators mentioned previously. The present study was designed to assess whether SR 49059 prevents or reduces the abnormal response in patients with RP.

Materials and methods

Patients

Because the diagnosis of RP is exclusively clinical, the patients selected for the study were required to have fulfilled the Allen and Brown criteria [29] (attacks provoked by cold or stress, bilateral expression of the phenomenon, normal radial and ulnar pulses and absence of trophic anomalies of the finger skin) and have a history of paroxysmal vasospastic crises of the upper extremities occurring regularly over a period of >2 yr. After we had explained the purpose of the study, we obtained written informed consent from each patient. The protocol was approved by the institutional ethics committee (CHUV, Lausanne, Switzerland). Patients had to be between 20 and 60 yr old and to have primary RP or RP that was secondary to a connective tissue disease with a duration of >2 yr and without trophic lesions. A screening visit was arranged to assess the patients’ suitability for entry into the study. Before inclusion, all patients underwent a complete physical examination, a detailed medical history was taken and routine laboratory tests were carried out. In order to better appreciate the potential beneficial effect of the V1a receptor antagonist, we arbitrarily enrolled only patients with a digital systolic pressure fall of >30% from the resting value at room temperature after immersion of both hands for 3 min in water at 15 °C. We have reported previously that a 30% reduction in finger pressure after cold immersion permits discrimination of subjects without vasospasm from patients with RP [30], although the specificity and sensitivity of the method reported by others is close to only 90% [31]. We have shown that the intraobserver variability of the method is <5% for measurements performed 7 days apart [30].

Study design

Twenty patients were enrolled between November 1997 and May 1998 in this single-centre, double-blind, two-period randomized sequence cross-over study to receive either placebo or SR 49059 (300 mg micronized formulation, once daily) for a period of 7 days. After a 3-week washout period, the alternate drug was administered for another 7 days. The 3-week washout period was selected to minimize the possible effect of the menstrual cycle on digital vasoreactivity. This allowed all women to be investigated during the same phase of their menstrual cycle, because the duration of their cycle was within the range of 27–29 days. However, no hormonal levels were assessed in this study. Female participants who were of child-bearing age had a pregnancy test before each treatment period. The measurements of finger pressure and temperature were obtained after 1 h of rest in the supine position at room temperature (22 °C) at screening, on day 1 just before treatment (H0), 2 h (H2) and 4 h (H4) after treatment, and on day 7 of each treatment period at H0, H2 and H4. We had shown earlier that the peak effect of SR 49059 on skin blood flow reactivity was reached between 1 and 4 h after oral intake of the drug [32].

Method

The most frequently affected finger from each hand, as shown by the clinical history, was equipped with a mercury-in-Silastic strain gauge (Hokanson, Seattle, Washington, USA) to measure systolic pressure. Finger pressure (opening pressure) was obtained by deflating a finger-cuff inflated 20 mmHg above systolic brachial blood pressure and by detecting the first rhythmic oscillations on the plotter. All finger pressure measurements were performed by a single investigator (G.B.). The cold test consisted of immersion of the hand in cold water for 3 min (15 °C), and was carried out during the screening phase and three times (before and 2 and 4 h after drug intake) on days 1 and 7 of each of the two treatment periods. Finger pressures were measured before the immersion of the two hands in the cold water, immediately after the hands had been removed from the cold water, and 10, 20 and 32 min later. The percentage reduction in finger pressure between the measurement taken before immersion of the hands and the measurement taken immediately after removal of the hands from the water was calculated.

Finger temperature was measured with small thermistors (Aclan, Toulouse, France) attached to the pulp of the 10 fingers with hydrophobic tape and recorded continuously for 45 min throughout the test [33]. Changes in temperature (°C) of the most frequently affected finger between the measurements made before immersion, directly after the 3-min cold test and at the end of the recovery period (32 min) were recorded. The 32-min recovery period was determined by the storage capacity (45 min) of the thermistor system (10 min baseline condition, 3 min cold test, 32 min recovery period).

The primary end-point, i.e. the attenuation of the reduction in finger pressure at the end of the cold test, was analysed for the main affected hand. This was defined as the hand for which the reduction in finger pressure at the end of the cold test was the most pronounced at the time of screening or, if equal, on day 1 of treatment period 1 at H0. Opening pressures and temperatures were measured on the same finger in all tests.

Capillaroscopy was performed on both hands of all five fingers with a light stereomicroscope (Wild–Leitz), and minimal baseline serological data (anti-nuclear and anti-Scl-70 antibodies) were obtained for all patients.

Plasma levels of arginine vasopressin (AVP) were measured by radioimmunoassay in our laboratory, on
day 1 at H0 and on day 7 at H0, H2 and H4 for each of the two treatment periods [34].

Plasma osmolality was measured with an osmometer as described previously [35].

Statistical analysis

Data were subjected to a cross-over analysis of variance (ANOVA) with terms for subjects as a random effect within sequence, and sequence, period and treatment as fixed effects. The period and treatment effects were tested against the within-subject variability; the sequence effect was tested against the between-subject variability. The SAS PROC MIXED procedure was used. This made it possible to account for incomplete cases by using the appropriate error term to estimate the treatment effect. Estimates of the treatment effect and 95% confidence intervals were calculated from the mixed model.

Results

Patients

A total of 20 patients were enrolled in the study. They were two men and 18 women, with a mean age of 39 yr (range 24–67) and a mean body mass index of 20.5 (range 17–27). They were normotensive, with a systolic pressure of 109 mmHg (range 91–140), a diastolic pressure of 71 mmHg (range 55–90) and a heart rate of 57 b.p.m. (range 55–86). None was a regular consumer of alcohol. Five were smokers (1–40 cigarettes per day), and coffee consumption varied from 0 to 5 cups per day. All patients refrained from smoking and from coffee consumption or any other known caffeine-containing beverages for 12 h before the cold test. One patient suffered from systemic sclerosis, whereas all the others had primary RP. The distribution of the most affected finger was similar between the left and the right sides, and the middle finger was the most frequently affected digit (50% of the patients). At screening, the mean (s.e.m.) reduction in finger pressure at the end of the 3-min hand immersion test was 57.4% (4.9%) for the most affected hand and 43.3% (4.5%) for the opposite hand.

Three patients did not complete the study. One patient receiving placebo discontinued treatment on day 3 of period 1 because of finger oedema and erythema of both hands followed by arthralgia. A second patient, receiving SR 49059, discontinued treatment on day 5 of the second treatment period, 4 days after a pruriginous exanthem. No skin or laboratory test abnormalities were observed at the time the patient informed us about this adverse event, on day 7. A third patient, also being treated with SR 49059, withdrew from the study during period 1 for personal reasons unrelated to the drug. Follow-up information was collected on all patients at least 3 months after randomization. At screening, none of the patients was taking medication that modifies vascular tone, and non-steroidal anti-inflammatory drugs were prohibited throughout the entire treatment period.

There were no statistically significant changes in blood pressure or in heart rate between the time of screening and before dosing (H0) on day 7 and 4 h after dosing (H4) on day 7 (Table 1). Laboratory test results were all in the normal range at screening and did not show any significant alteration throughout the study. Antinuclear antibodies (ANA) were detected in seven patients, but only two of these patients had values above the normal range (<1:80) [one had systemic sclerosis (ANA 1:680) and another had severe RP with possible undifferentiated connective tissue disease (ANA 1:1260)]. No significant change in plasma osmolality, AVP or haematocrit was observed at any time point after treatment, although the vasopressin level was higher at H4 on day 7 in the placebo than in the treated group for obscure reasons (Table 2).

Capillaroscopic examination of the nail-folds of the 10 fingers in all patients did not reveal any megacapillaries. Only the patient with systemic sclerosis was found to have haemorrhagic spots with non-homogeneous diminished capillary density.

Finger pressure

On day 1, no significant treatment effect was observed at the end of the immersion test on the most affected hand (Table 3). A moderate effect of time on the magnitude of the decline in pressure was noticed. It appeared to be independent of the treatment effect and was observed on days 1 and 7 (Table 3). No difference in baseline mean systolic digital pressure was observed at day 7 between the placebo and SR 49059 groups [97.8 (0.8) and 103 (1.7) mmHg respectively]. However, a significant treatment effect was observed, with a reduction in the size of the decline in pressure of 14.5% (95% CI 0–29, P = 0.0449) at H4 on day 7 in the SR 49059 phase compared with the placebo phase (Fig. 1). A similar trend was observed at H0 and H2 on day 7, but the difference was not significant. No further significant treatment effect was found during the recovery period (Table 3).

Skin temperature

A treatment-independent increase in finger temperature during the study was observed on days 1 and 7, although the trend was more marked in the placebo group on day 7 (Table 4). At the end of the immersion period, no significant treatment effect was detected at any time point on days 1 and 7; finger temperature was between 15.7 and 16.4°C, corresponding closely to the temperature of the water bath. On day 7, 32 min after the cold immersion test, temperature recovery was slightly greater with SR 49059 than for placebo 2 and 4 h after drug intake (Fig. 2). The difference was not significant (P = 0.060 and P = 0.062 respectively); it was probably due to a lower baseline temperature in the active treatment group.

Discussion

Raynaud’s phenomenon is a highly prevalent vasospastic disorder affecting approximately 10% of the adult
population, mainly women in moderate climates [36, 37]. Its prevalence increases to >20% of the population in cool, damp climates [38]. Ethnic differences do not seem to be related to this circulatory disorder [39]. Several neurohumoral factors have been considered as contributors to the abnormal response in blood-vessel tone to cold. Among the different vasomediators, AVP, which is known to be a potent vasoconstrictor agent, has been shown recently to have (i) biphasic effects on forearm blood flow depending on the concentration [25, 36] and (ii) opposite effects on the blood vessels in the skin and muscles [26]. The present study demonstrates that SR 49059, an orally active non-peptide vasopressin V1a receptor antagonist, reduces the cold-induced fall in finger systolic pressure when given at a dose of 300 mg once daily. A 14.5% difference from placebo in pressure reduction was obtained during cold exposure in patients with RP after 7 days of treatment, 4 h after dosing, reflecting a reduction in the vasospastic response. A trough effect is also possible, even though the difference between placebo and SR 49059 was not significant. This result suggests that vasopressin contributes to the abnormal vasospastic response to cold exposure in patients with RP, although the clinical meaning of this reduction in pressure drop remains to be clarified.

With the exception of a slight skin rash 12 h after dosing with SR 49059 in one patient, no side-effects were noticed. No skin or laboratory test abnormalities were detected at day 7. So far, no similar reaction has been observed in any of the clinical studies that have been conducted or that are continuing, in which more than 1000 patients have been treated with SR 49059 for periods ranging between 7 and 28 days (SR 49059 Clinical Investigator’s Brochure, version 0.4. Sanofi Recherche, March 1998).

The reduction in the extent of the fall in blood pressure in the finger during exposure to cold was observed after only 1 week of treatment with a single daily dose SR 49059. No acute effect of the drug was observed on the vasospastic response. Whether higher doses, more frequent dosing or a longer period of administration of the active compound could offer even more protection remains to be determined. However, in a previous study in young volunteers we were unable to show any further blocking effect, as assessed by measuring skin blood flow, for plasma levels greater than those obtained here in subjects receiving 300 mg SR 49059 once daily [32]. No effect of the blockade of the vasopressin V1a receptor was observed on haematocrit, plasma osmolality or plasma AVP levels. This was not surprising, because the active compound does not interfere with renal V2 receptors, and therefore no aquaretic effect should be expected. The patients were not allowed to drink during the cold test. However, they all took a standard light meal comprising 2 dl orange juice, one low-fat yogurt and two pieces of cake after dosing and the first sampling of blood. No statistically significant change in plasma AVP concentration was detected at any time point during the study or between groups, confirming previous observations [41]. It is therefore suggested that in patients with RP, and more particularly in patients with primary RP, AVP at a physiological concentration contributes to the excessive vessel tone in a locally direct or indirect fashion by modulating other vasoactive agents [42].

The effect on finger temperature showed a trend similar to that for finger pressure after 1 week of treatment with SR 49059, although the differences observed were not significant. Interestingly, as the test was performed between 8:00 a.m. and 2:00 p.m., we observed a progressive time-dependent reduction in the severity of the vasospastic response to the cold immersion test in the two treatment groups (Table 3 and Fig. 1), accompanied by an increase in skin temperature. Therefore, the absolute value of the temperature recovery increased over time. Indeed, at the end of the cold

### Table 1. Haemodynamic characteristics of the patients at different stages of the study

<table>
<thead>
<tr>
<th></th>
<th>Screening</th>
<th>Baseline</th>
<th>D7-H0*</th>
<th>D7-H4*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Systolic pressure (mmHg)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>109 ± 3.6</td>
<td>102.1 ± 3.2</td>
<td>102.9 ± 2.6</td>
<td>100.0 ± 2.6</td>
</tr>
<tr>
<td>SR 49059</td>
<td>104.1 ± 2.9</td>
<td>102.9 ± 3.4</td>
<td>101.2 ± 3.7</td>
<td></td>
</tr>
<tr>
<td><strong>Diastolic pressure (mmHg)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>71 ± 2.5</td>
<td>61.5 ± 2.9</td>
<td>62.7 ± 2.2</td>
<td>61.4 ± 2.4</td>
</tr>
<tr>
<td>SR 49059</td>
<td>64.9 ± 2.6</td>
<td>64.1 ± 2.4</td>
<td>65.0 ± 3.0</td>
<td></td>
</tr>
<tr>
<td><strong>Heart rate (b.p.m.)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>67 ± 2.1</td>
<td>64.9 ± 2.6</td>
<td>64.1 ± 2.4</td>
<td>65.0 ± 3.0</td>
</tr>
<tr>
<td>SR 49059</td>
<td>63.6 ± 2.4</td>
<td>62.7 ± 2.9</td>
<td>62.4 ± 2.9</td>
<td></td>
</tr>
</tbody>
</table>

Values are mean ± s.e.m.  
*D7, day 7 of treatment period.

### Table 2. Effect of SR 49059 on haematocrit, plasma arginine vasopressin and osmolality

<table>
<thead>
<tr>
<th>Time*</th>
<th>Haematocrit (%)</th>
<th>AVP (ng/l)</th>
<th>Osmolality (mosmol/kg H2O)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Placebo</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1-H0</td>
<td>41.10 ± 0.57</td>
<td>0.44 ± 0.07</td>
<td>288.63 ± 0.60</td>
</tr>
<tr>
<td>D7-H0</td>
<td>40.61 ± 0.43</td>
<td>0.40 ± 0.04</td>
<td>288.67 ± 0.82</td>
</tr>
<tr>
<td>D7-H2</td>
<td>39.41 ± 0.74</td>
<td>0.45 ± 0.08</td>
<td>287.11 ± 0.90</td>
</tr>
<tr>
<td>D7-H4</td>
<td>39.27 ± 0.56</td>
<td>0.65 ± 0.18</td>
<td>287.61 ± 1.18</td>
</tr>
<tr>
<td>SR 49059</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D1-H0</td>
<td>40.48 ± 0.49</td>
<td>0.39 ± 0.07</td>
<td>288.37 ± 0.91</td>
</tr>
<tr>
<td>D7-H0</td>
<td>40.56 ± 0.62</td>
<td>0.42 ± 0.07</td>
<td>289.71 ± 1.12</td>
</tr>
<tr>
<td>D7-H2</td>
<td>39.34 ± 0.59</td>
<td>0.46 ± 0.04</td>
<td>288.00 ± 0.92</td>
</tr>
<tr>
<td>D7-H4</td>
<td>39.38 ± 0.55</td>
<td>0.40 ± 0.04</td>
<td>287.06 ± 0.71</td>
</tr>
</tbody>
</table>

Values are mean ± s.e.m.  
*D1, D7, days 1 and 7 of treatment period.
Finger pressure reduction of the main affected hand at the end of the cold immersion test. Mean ± s.e.m. D7, day 7 of the treatment period. *P < 0.05.

Table 4. Finger temperature (°C) on the most affected hand at different stages of the study

<table>
<thead>
<tr>
<th>Time*</th>
<th>Baseline (32 min)</th>
<th>Recovery (32 min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>Placebo SR 49059</td>
<td></td>
</tr>
<tr>
<td>Screening</td>
<td>24.8 ± 0.96</td>
<td>20.6 ± 0.63</td>
</tr>
<tr>
<td>D1-H0</td>
<td>24.1 ± 0.70</td>
<td>20.3 ± 0.72</td>
</tr>
<tr>
<td>D1-H2</td>
<td>27.7 ± 0.89</td>
<td>21.8 ± 1.04</td>
</tr>
<tr>
<td>D1-H4</td>
<td>28.7 ± 1.04</td>
<td>22.2 ± 1.13</td>
</tr>
<tr>
<td>D7-H0</td>
<td>25.3 ± 0.88</td>
<td>20.1 ± 0.81</td>
</tr>
<tr>
<td>D7-H2</td>
<td>27.0 ± 1.01</td>
<td>22.1 ± 1.13</td>
</tr>
<tr>
<td>D7-H4</td>
<td>29.2 ± 0.83</td>
<td>23.2 ± 0.97</td>
</tr>
</tbody>
</table>

Values are mean ± s.e.m. *D1, D7, days 1 and 7 of treatment period.

FIG. 1. Finger pressure reduction of the main affected hand at the end of the cold immersion test. Mean ± s.e.m. D7, day 7 of the treatment period.

FIG. 2. Finger temperature reduction of the main affected hand at the end of the recovery phase. Mean ± s.e.m. D7, day 7 of the treatment period.

Test skin temperature remained close to that of the water bath kept at 15°C.

The reduction in the intensity of the vasospastic response over time seems to be a physiological phenomenon that may be related to nycthemeral hormonal fluctuations rather than to familiarization with the cold test. We saw no such phenomena when assessing the reproducibility of the technique by carrying out two or three cold test challenges within 1 h [30]. However, Wigley et al. [43] found that patients who had become adapted to a cold challenge required a lower temperature before a vasospastic attack could be induced, but this was never observed in our study. A more trivial explanation could be that the fingers warmed up slowly under the effect of the room temperature, which was
maintained constant throughout the duration of the test. However, this explanation seems unlikely because the patients were left supine in a resting position for a minimum of 1 h before the start of the cold test and were asked not to expose their hands to cold on the days of the immersion test. The cold test may also have reset the finger temperature to a lower level, clearing the effect of the ambient temperature. Reduction of the vasospastic response suggests the existence of an inverse relationship between the temperature and the severity of the vasospastic response to cold exposure. Whether a threshold effect between the two variables exists in RP remains to be investigated.

The treatment of most patients with RP remains conservative, and includes advice to protect from cold and stop smoking [37]. When the crisis becomes severe or debilitating, a topical or systemic vasodilator can be prescribed. The calcium channel antagonists (more specifically the dihydropyridine derivative nifedipine) have been the most studied class of drugs in the treatment of abnormal vasospasm in RP [44]. Unfortunately, the beneficial effects of calcium channel blockers are sometimes obtained at the cost of severe side-effects such as headache, flushing and swelling of the ankles [21–23]. These undesirable effects are often so severe that the patient prefers to abstain from a treatment more debilitating than the disease. The adverse vasodilatory effects are the major limitation in patients who tend to have very low blood pressure, as evidenced in the present study. Therefore, SR 49059 could be a real alternative for patients with primary RP, provided it is clinically beneficial, because it is devoid of systemic vasodilator activity, as confirmed in this study.

When assessing the effects of a single oral dose of SR 49059, we did not consider the frequency and duration of the vasospastic reactions. The pressure and temperature changes we observed suggest that benefit may be obtained after only a few days of V1a receptor blockade. Further studies to assess the clinical efficacy of long-term treatment with SR 49059 are warranted.

Acknowledgement
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References
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