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Sympathetic Skin Response and R-R Interval Variation in Chronic Administration of Salmon Calcitonin

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Abstract. The distribution of calcitonin and CGRP-producing cells and pathways in the brain and other tissues suggests functions for the peptide in nociception, ingestive behaviour and modulation of the autonomic and endocrine systems. Sympathetic skin response (SSR), is a reliable indicator of autonomic dysfunctions. The heart rate variability termed as R-R interval variation (RRIV) is another simple and reliable test which can be used to determine vagal autonomic dysfunction. Twenty female patients with active osteoporosis aged between 46-58 and for a control group 20 age-matched healthy female volunteers with no history or evidence of any other disease were included in this study. The study results show that long term therapy with salmon calcitonin does not effect any SSR and RRIV parameters. It can be speculated that though human calcitonin and CGRP have discrete functions in the human autonomic nervous system, replacement therapy with salmon calcitonin does not interfere normal autonomic functions.

Key Words: Calcitonin, SSR, R-R Interval.

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INTRODUCTION

Calcitonin, a hormone secreted from the C-cells of the thyroid, exerts Ca^{++} -regulative effects via bones and kidneys and natriuretic and diuretic effects in the kidney[1, 2)]. However observations suggest that calcitonin has a broader range of actions, including effects on the central nervous system. The possible neural actions of calcitonin include production of analgesia[3)], changes in prolactin release[4)], inhibition of food and water consumption[5)] and behavioral effects[6)]. In the human brain, the highest levels of calcitonin-like peptides and binding sites have been found in the hypothalamus[7)]. Central administration of this peptide decreases appetite[8)] and gastric acid secretion[9)] and increases sympathetic noradrenergic outflow with accompanying hypertension and tachycardia[10)].

Sympathetic skin response (SSR), is a reliable indicator of autonomic dysfunction. The long latency electrical response obtained from the skin surface by supramaximal stimulation of the skin or the peripheral nerve (the so-called sympathetic skin response, SSR) is considered an index of peripheral autonomic nerve fiber function[11–13)]. The SSR is reduced or absent in systemic neuropathies and in dysautonomic syndromes[14, 15)]. The heart rate variability termed as R-R interval variation (RRIV) is another simple and reliable test which can be used to determine vagal autonomic dysfunction. To our knowledge, these two autonomic tests have not been previously applied to patients taking salmon calcitonin (sCT), and the purpose of this study was to evaluate the effects of long term ad-

ministration of sCT on the sympathetic nervous system.

MATERIALS AND METHODS

Twenty female patients with active osteoporosis aged between 46–58 and for a control group 20 age-matched healthy female volunteers with no history or evidence of any other disease were included in this study. The mean duration of the receiving of sCT by the nasal way was 8 ± 2.1 months (3–36 months). In the control group no subject was taking drugs that were likely to interfere with the study and the patient group was receiving only sCT. Patients had to abstain from coffee and alcoholic beverages for at least 12 h prior to the study. Written consent was obtained from each patient and control subjects subsequent to a thorough explanation of the purposes and the methodology to be used in the present study.

SSR recordings were performed in a semi-darkened room, and in the relaxed and supine position. Skin temperatures were not lower than 32°C during SSR recordings. SSR recordings in all of the subjects were performed by a Medelec Premiere electromyograph according to the method described by Shahani et al.[25, 26] Stainless steel disk electrodes of 6 mm diameter were used for the SSR recordings. The active electrodes were attached to both palms and the reference electrode was attached to the dorsum of the hand. Bandpass filters were set at 1 to 1000 Hz and the sensitivity at 0.5 to 2 mV/division. Square wave electrical pulses of 0.1 msec duration and 20 mA intensity were applied to the skin of the right wrist of each subject 10 times, once every 10 sec, and the averaged SSR was recorded. The electrical pulses were applied at low frequency to minimize the motion artifact. Habituation was not encountered during the successive electrical pulses.

The latency and amplitude measurements of SSR were performed manually. The latency was measured in msec from the onset of the stimulus artifact to the onset of the skin response. The amplitude was measured in mV from the peak of the negative component to the baseline. The area under the negative component of SSR was also recorded automatically in mVs.

For the RRIV recordings, with the patient lying supine, one surface disk electrode was placed on the dorsum of each hand, the active electrode being placed on the left hand. A circular ground electrode was placed around one wrist. The bandpass was set at 1 to 100 Hz and the sensitivity at 0.5 to 2 mV/division. By using the triggering mode and the delay line, two QRS complexes were displayed on the screen. Because the first displayed complex is the triggering potential, the variation in timing of the second complex represents the R-R interval variation.

Five groups of QRS complex pairs were recorded at rest and two groups of 20 pairs of QRS complexes were recorded during deep breathing at 5 or 6 breaths per min. The range in the 20 pairs of R-R interval was termed -a-, and the mean of the 20 pairs of R-R intervals was termed -b-. RRIV was expressed as a percentage of the average R-R interval using the formula RRIV= a/b x 100. The average of five recordings at rest was termed as R% and that of two recordings during deep breathing as D%. The difference between D% and R% and the ratio of D% to R% were also calculated.

F wave response study and conduction velocity measurement in two “motor and sensory” nerves (n.medianus and n.ulnaris) were also performed on each subject to exclude the subjects with abnormal findings due to subclinical polyneuropathy using the usual criteria for diagnosis[16].

Statistical analysis was performed using t-test by SPSS for MS Windows.

RESULTS

Biphasic SSR was present bilaterally in all of the normal subjects. The SSR latencies were considered abnormal if they were not within 95% confidence limits (mean ± 2 SD). The amplitude of SSR varied from a few microvolts to several millivolts even in the same subject but the averaged responses had more consistent waveforms. The interside latency differences, amplitude, and area were statistically insignificant ($p>0.05$). The mean values of the latency, amplitude, and area under the negative component of SSR in the groups are shown in Table 1. Latency, amplitude and area parameters of the groups were not significantly different. Mean values of RRIV for the controls and patients are shown in Table 2. R-R values, D%, D%-R%, and D%:R% did not show any statistically significant differences between the control and the patient groups ($p>0.05$).

Table 1. Group means of sympathetic skin response latencies and amplitudes

	Latencies[a]		Amplitudes[a]		Area[a]	
	Left Hand	Right Hand	Left Hand	Right Hand	Left Hand	Right Hand
	s	s	mV	mV	mVs	mVs
Normal controls	1.3 ± 0.3	1.3 ± 0.2	2.7 ± 1.1	2.6 ± 1.3	7.4 ± 2.1	6.8 ± 2.4
Patients	1.2 ± 0.3[b]	1.3 ± 0.3[b]	2.4 ± 1.5[b]	2.5 ± 1.4[b]	7.1 ± 1.9[b]	7.3 ± 2.7[b]

[a] No significant difference between left and right. [b] Statistically insignificant difference from normal controls ($p>0.5$).

Table 2. Mean value of RRIV

	Hyperventilation			
	Rest (R%)	(D%)	D% - R%	D% : R%
Normal Controls	10.7 ± 3.4	17.6 ± 5.8	6.9 ± 3.7	1.7 ± 0.3
Patients	10.1 ± 2.9[a]	16.9 ± 5.5[a]	6.7 ± 3.2[a]	1.6 ± 0.4[a]

[a] Statistically insignificant difference from normal controls ($p>0.5$).

DISCUSSION

Two mRNAs generated as a consequence of alternative RNA processing events in the expression of the human calcitonin gene encode the protein precursors of either calcitonin or calcitonin gene-related peptide (CGRP). On the basis of sequence comparison, it is suggested that both the calcitonin and CGRP exons arise from a common primordial sequence, suggesting that duplication and rearrangement events are responsible for the generation of this complex transcription unit[17]. The calcitonin mRNA predominates in the thyroid while the CGRP-specific mRNA appears to predominate in the hypothalamus[18]. The distribution of CGRP-producing cells and pathways in the brain and other tissues suggests functions for the peptide in nociception, ingestive behaviour and modulation of the autonomic and endocrine systems[19].

CGRP has potent cardiovascular effects (vasodilation, hypotension, positive chronotropic and inotropic action in the heart). Intracerebroventricular administration of CGRP raises the blood pressure, and both CGRP and calcitonin inhibit gastric acid secretion and food intake[20]. Fisher et al. reported that CGRP acts on the central nervous system to stimulate selectively noradrenergic sympathetic outflow[21].

Lenz and Brown have also shown that intracerebroventricular administration of human calcitonin and human CGRP inhibits meal-stimulated gastric acid secretion in awake dogs. Inhibition of gastric acid secretion by human calcitonin and human CGRP in the dog is not mediated by inhibition of gastrin release or by the vagus nerves. Human calcitonin but not human CGRP appears to inhibit meal-stimulated gastric acid secretion in the dog by activation of the autonomic (sympathetic) nervous system[22]. The inhibition of acid secretion is related primarily to the decrease in vagal efferent activity whereas the inhibition of gastric motor functions involves increases in sympathetic outflow. The central action of CGRP to prevent ethanol-induced lesions is unique to this peptide and not shared by other centrally acting inhibitors of gastric function[23].

Regarding the above findings, we thought that long term salmon calcitonin therapy might be responsible from some autonomic dysfunctions. When autonomic dysfunction is suspected, reproducible, quantifiable and noninvasive tests are needed to detect abnormalities of sympathetic and parasympathetic nervous activity. Traditional tests of autonomic function such as Valsalva ratio, variation of electro-cardiographic R-R interval and effects of posture on blood pressure provide useful but limited information about the function of the autonomic system[24]. Shahani et al.[25, 26] has shown the diagnostic value of two simple tests of autonomic function both of which can easily be performed in an electromyography laboratory.

These tests are the sympathetic skin response (SSR), a reliable indicator of disorders affecting unmyelinated axons, and the heart rate variability termed as R-R interval variation (RRIV), a simple and reliable test of vagal autonomic dysfunction.

The integrity of the parasympathetic system could be assessed with several tests based on cardiovascular reflexes. The heart rate variability termed R-R interval variation has been claimed to be the simplest and the most reliable test of vagal autonomic dysfunction. The neural mechanism depends on a parasympathetic reflex[27]. Cardioinhibitory impulses are transmitted to the heart by thinly unmyelinated vagal fibers from the vagus nucleus. The vagal nucleus is under the control of the tractus solitarius which receives input from peripheral receptors including baroreceptors. Respiration is the most important stimulus for sinus arrhythmia. The increase in heart rate during inspiration results from decreased vagal activity. The vagal afferent fibers that are involved in the reflex innervate thoracic stretch receptors. Many authors concluded that R-R variations are a reliable method for establishing and following autonomic dysfunction[28, 29].

In this study we have shown that long term therapy with salmon calcitonin does not effect any SSR and RRIV parameters. It can be speculated that though human calcitonin and CGRP have discrete functions in the human autonomic nervous system, replacement therapy with salmon calcitonin does not interfere with the normal autonomic functions.

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