Investigation of Spontaneous Vasomotor Activity

Relation of the Sympathetic and Somatic Nervous Systems to the Occurrence of Slow Waves (Alpha and Beta) of the Plethysmogram of the Dog’s Paw

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Of the 5 different waves described as appearing in the normal plethysmogram of an extremity, the last 3 (alpha, beta, and gamma waves) are grouped together as the slow or third order waves. Their relative frequencies in the human digit are 2 to 14 per minute, 1 to 2 per minute, and 1 to 8 per hour.1 They were first described by Hewlett and Van Zwaluwenberg (1909)2 as large vasomotor waves “evidently due to marked vasomotor instability.” Since then, these slow waves have been seen in the human digit,3,4 forehead skin, nasal septum,5 and the pinna of the ear.6 Hargis and Mann (1925),7 in plethysmographic studies of the spleen in unanesthetized dogs, reported third order waves which resemble the slow waves of the digital plethysmogram. These were present after denervation of the spleen. Also described was a “splenic reflex” to sensory stimuli (noise, pain) which very much resembles the digital vasconstrictor response. This was abolished by denervation. In addition, we have seen slow waves in plethysmograms recorded from the subarachnoid space; this area also shows a vasconstrictor response to sensory stimuli.

Burton (1939)8 carried out plethysmographic studies on one patient after unilateral cervical ganglionectomy (presumably this was a cervical approach to the stellate ganglion) and reported little correspondence between the volume pulsations in the fingers of the 2 sides. He concluded that “the occurrence of normal rhythmic changes of flow in a limb is dependent on an intact peripheral sympathetic nervous system.” Neumann, Lhamon and Cohn (1944)7 reported that alpha waves disappeared upon cutting the sympathetic nerves. They did not state the number of patients, the extremity studied, the time interval after sympathectomy, nor the extent of the procedure. Burch (1947)9 stated that “sympathectomy or sympathetic nerve blocking will inhibit alpha deflections almost entirely.” He gave no other details. On the other hand, Hertzman and Dillon (1939)5 found that sympathectomy need not abolish these waves and that in some cases the denervated vessels were more active than the normally innervated ones. They further reported that spontaneous waves were almost always synchronous in the skin of both hands, though at times minor waves appeared in one finger and not in an adjacent finger. They concluded “the synchronicity of the waves would imply a vasomotor origin.” Despite this disagreement in findings, much of the current literature carries the statement that alpha and beta waves are dependent upon an intact sympathetic nervous system. The purpose of this investigation was to determine the relation of the nervous system to the alpha and beta waves of the plethysmogram.

Methods

Plethysmograms were recorded on the dog’s paw using the Winsor digital pneumoplethysmograph8 in conjunction with a Brush DC amplifier and direct writing oscillograph or a Sanborn electrocardiograph. The paper speeds were 5 mm. and 2.5 mm. per second, respectively. A preformed
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malleable aluminum tube* was sealed onto the shaved paw with a polysiloxane1 which solidifies at room temperature after mixing with a catalyst. Recordings were often made from 2 sites simultaneously.

The respiratory rate was noted so that respiratory waves were not confused with slow waves. The dog's respiratory rate varies from over 100 during panting to as low as 6 under anesthesia. The rate was recorded using an air-filled plastic tube under a chest strap. Compression of the balloon with inspiration closed an electric microswitch mounted on a rubber diaphragm which activated an event marker pen.

Healthy mongrel dogs weighing 7 to 18 Kg. were selected for study if they were found to be consistent producers of alpha waves, and if they were temperamentally suited to lie quietly for awake experiments. Many tracings were made on unmedicated dogs. Some were sedated with morphone sulfate (1 to 2 mg. per Kg.) subcutaneously, or lightly anesthetized with intravenous pentobarbital. Occasionally intravenous alpha chloralose: 5 per cent (100 mg. per Kg.) in polyethylene glycol (M. W. 200) and saline was used. Dosage was based on effect rather than being administered solely on the basis of the weight of the dog. Pentobarbital dosage varied from 15 to 30 mg. per Kg. Since the pulse-wave amplitude recorded in the plethysmograph varied with the volume of the paw enclosed in the cup and with the circulatory state of the dog, the sensitivity of the plethysmograph was adjusted to give the most satisfactory tracing. No attempt was made to use a standard calibration. Whenever possible, the amplitudes of control and experimental paws were made equal for easier comparison. Volume standardizations were made but in the records presented the relative amplitudes have no meaning. The volume of the paw enclosed in the cup varied from 30 to over 100 ml.

Several drugs were used for ganglionic or adrenergic blockade. Tetraethylammonium chloride was given intravenously in doses of 10 to 15 mg. per Kg. depending upon the response of the dog, the drug being infused until no vasoconstrictor response occurred to stimulation with a small spark-gap diathermy. Phenoxybenzamine (Dibenzyline) in doses of 10 to 20 mg. per Kg., intravenously, produced effective blockade. Chlorisondamine (Ecolid, Ciba) was given orally, 10 mg. per Kg., twice daily for 3 or more days to produce a chronic block.

Thoracic sympathectomy was performed through the third interspace, removing the stellate ganglion and 3 to 5 additional thoracic ganglia. Such sympathectomized dogs had marked relaxation of the nictitating membrane, ptosis and obvious warmth of the paw on the operated side. Lumbar sympathectomy consisted of unilateral removal of the ganglia from L-1 or 2 through L-4 or 5. These dogs also had hot paws.

Extremities were denervated by sectioning the sciatic and femoral nerves, and, in some, the obturator nerve as well. The sciatic nerve was cut proximal to its first branch after it emerged from the sciatic notch, the femoral either high in the groin (cutting any branches encountered) or in the abdomen above the inguinal ligament, the obturator in the pelvis.

The adequacy of vascular denervation by surgical procedures or by drug blockade was tested by means of the vasoconstrictor response to sensory stimuli, as seen in the plethysmogram.9 Under light pentobarbital anesthesia, the animal was stimulated with a small spark-gap diathermy (Hyfrecator*). This produced a small superficial burn. In the normally innervated paw there is a prompt vasoconstriction which is recorded as a sharp decrease in volume of the part and a simultaneous decrease in the pulse amplitude (fig. 1B). This response occurs to many stimuli (pain, noise, fear,9 deep breath10). This reaction does not occur in the adequately sympathectomized, denervated or blockaded extremity. No chemically denervated preparation was accepted unless this response was absent to repeated tests when it had previously been present. In operated dogs, it was absent in the experimental extremity but still present in the control. The reaction pattern of each subject was studied so that dogs with consistent responses to auditory stimuli (bell or buzzer) could be selected for awake experiments. If necessary, the dog was anesthetized after waves were found in awake tracings and tested with the Hyfrecator. This stimulus was chosen because it was strong enough to be consistently effective during anesthesia. Only 2 dogs (out of 30) repeatedly failed to respond in a control extremity. Nothing else unusual was observed about these 2 dogs.

Dogs with their aortic and carotid chemoreceptors denervated11 were prepared. The arch of the aorta and its branches were dissected free from all surrounding tissue, including sectioning the left recurrent laryngeal nerve (to denervate the aortic body). The carotid bodies were excised and the sinus nerves sectioned. Four of these animals were also splenectomized. The absence of

*Supplied by Victor Manufacturing Corp., Chico, Calif.
1Special Sili-Gel, manufactured by Dental Perfection Co., Glendale, Calif.
1Matheson Coleman and Bell Division, Matheson Company, Inc., Norwood, Ohio.
the chemoreceptor response was repeatedly tested by intravenous sodium cyanide in the unanaesthetized dog. These dogs were prepared for another investigation but were available for plethysmographic studies.

Alpha waves were not considered to be present unless they could be seen over a considerable length of tracing, preferably several minutes. Vasoconstrictor responses, whether induced or "spontaneous" were not considered as producing alpha waves. Burton apparently often included these responses when he discussed fluctuations in the plethysmogram. Shallow alpha waves were accepted only if they were known to be characteristic of the particular dog. Waves with a frequency of 3 to 15 per minute were considered alpha waves; of less than 3 per minute were considered beta waves. Gamma waves were not recorded because of the limitations of the equipment used.

Dogs varied considerably in their ability to produce slow waves, some doing so consistently, others rarely. This did not appear to be related to a dog's breed or metabolic activity but rather to be a function of its personality or temperament.

Results

Sympathectomy

Alpha waves were found repeatedly in 3 sympathectomized hindpaws (beta waves in 2) over periods of 2 days to 4 months postoperative, and in 7 sympathectomized forepaws (beta waves in 2) 2 days to 14 months postoperative. All the sympathectomized limbs showed no vasoconstrictor response. One dog did not have a good response in a control paw but the sympathectomized paw was warm and all the eye signs of adequate sympathectomy were present. All sympathectomized extremities showed alpha waves at some time. Figure 1 shows alpha waves in 3 sympathectomized extremities. Each pair of tracings is a simultaneous recording from the sympathectomized paw and from the opposite control paw. Dog no. 20 (fig. 1, A and B) had a left thoracic sympathectomy 12 months previously. Large alpha waves are seen in the records from the left sympathectomized forepaw, but not in those from the control which shows beta waves (A) and 2 sharp vasoconstrictor reactions (B), which are absent on the left. Records from dog no. 32, 3 days after a left thoracic sympathectomy 12 months previously. Large alpha waves are seen in the records from the left sympathectomized forepaw, but not in those from the control which shows beta waves (A) and 2 sharp vasoconstrictor reactions (B), which are absent on the left. Records from dog no. 32, 3 days after a left thoracic sympathectomy, show alpha waves in both forepaws; these are not synchronous (C and D). Tracings from dog no. 48, 2 days after a right thoracic sympathectomy, show shallow

Figure 1

Alpha waves in sympathectomized paws. Paired simultaneous tracings of control and operated extremities. (See text for explanation.) RR, respiratory rate; VC, vasoconstrictor reaction.
synchronous alpha waves in both forepaws (E); these are not respiratory waves since the respiratory rate was in excess of the heart rate.

**Denervation**

Four denervated hind extremities demonstrated alpha waves 6 days to 6 months after operation, and 1 also beta waves. One of these extremities was severely traumatized due to dragging. When the foot was markedly edematous, and later when it was also grossly infected, alpha waves of large amplitude were consistently present (as well as at times when the foot was in good condition). In another dog (no. 40), the right hind extremity was denervated some months after a right lumbar sympathectomy had been performed; alpha waves also occurred in this preparation (fig. 2, C and D).

**Sympathetic Blockade**

Alpha waves were present in the records from dogs during ganglionic blockade with intravenous tetraethylammonium chloride (twice), oral chlorisondamine (3 times), and adrenergic blockade with intravenous dibenzyline (3 times). Beta waves also occurred.

**Miscellaneous**

Unilateral ventral rhizotomy (L-3 through S-2) was done on one dog; alpha waves were present in the operated paw at 7 days. Alpha waves were seen in the paw plethysmograms of 6 dogs with inactivated aortic and carotid body chemoreceptor mechanisms. Since 4 of these dogs were splenectomized, it can also be stated that alpha waves occur in the absence of the spleen. Table 1 summarizes these findings.

In simultaneous recordings of 2 normally innervated paws of a dog, the alpha and beta waves are usually synchronous, as are any vasoconstrictor responses to sensory stimuli. If simultaneous tracings are taken of a denervated or sympathectomized paw and its opposite control, the waves may be synchronous, asynchronous, or absent in 1 paw. The vasoconstrictor response to stimuli does not occur in the operated paw. When waves are present in both paws, they are usually synchronous. One dog with 1 denervated and 1
sympathectomized hind extremity showed periods of synchronous and asynchronous alpha waves. One dog with bilateral thoracic sympathectomy showed synchronous alpha waves in these 2 paws over long periods of time; another showed periods of both synchronous and asynchronous waves. In figure 2, A and B show asynchronous waves in 2 pairs of denervated and control paws; C and D show synchronous waves in 1 hind paw which is denervated and 1 which is both denervated and sympathectomized; and E shows synchronous waves in control forepaw and denervated hindpaw.

Discussion

It became apparent early in these studies that the absence of slow waves could be due to many things other than the experimental conditions. They often disappeared with any change in conditions such as fluctuations in the level of anesthesia, an operative procedure, parenteral medication, or environmental distractions.

The absence of slow waves in a given experiment did not, by any means, indicate that they were always absent under these conditions. Rapid vasodilatation or vasoconstriction usually caused them to disappear. Waves were slow to recur after any procedure which abolishes them. Many acute experiments using parenteral drugs were valueless for this reason. It was more satisfactory to give drugs orally for several days to avoid injections with resultant sudden changes in vasomotion.

Repeated experiments were necessary to verify the presence of waves. With this approach, no experimental situation (with one exception) was encountered in which slow waves were never found. The one exception was with parenteral administration of hexamethonium. This drug was very unsatisfactory because waves disappeared before blockade was adequate, and yet it was difficult to attain an adequate blockade according to our criteria.

Waves were present in dogs sedated with morphine or anesthetized with pentobarbital or chloralose. They were less likely to be present during deep anesthesia.

Our studies in dogs have clearly shown that slow waves are not dependent upon the sympathetic nervous system, nor, for that matter, upon the somatic nervous system. They were present after sympathectomy, after complete peripheral denervation, and after combined sympathectomy and denervation, as well as during ganglionic or adrenergic blockade.

It has been suggested that these waves are the result of the innate rhythmicity of vascular smooth muscle. The periodic contractions and relaxations of metarterioles and precapillary sphincters reported by Zweifach\textsuperscript{12} have a periodicity (30 seconds to 3 minutes) similar to beta waves. This vasomotion is present after denervation. Clark and Clark,\textsuperscript{13} studying the rabbit's ear in a tissue chamber, were struck by the "normal occurrence of spontaneous rhythmic contractions of arteries, involving the main artery down to smaller and smaller branches." However, the different arteries and even parts of the same artery were observed to contract at different tempos. Algire,\textsuperscript{14} also using the transparent chamber technic in mice, found that vasomotor activity ceased when the blood pressure increased or decreased. Golenhofen and Hildebrandt,\textsuperscript{15} using Hensel’s heat-conducting probe, reported spontaneous rhythmic changes in muscle circulation with a period of about 1 minute, accompanied by synchronous contrary fluctuations in the skin. Burch and Murtadha\textsuperscript{16} described a periodic fluctuation in the venous pressure in forearm veins.
with a frequency resembling alpha and beta waves.

Under most conditions slow waves occurred synchronously in 2 extremities of the same animal. This agrees with Hertzman and Dillon, who often found them synchronous in several areas at once (as many as 4 simultaneously), though rarely in all areas recorded at the same time. There would then appear to be some central controlling or coordinating influence. This would, of necessity, have to be either hemohydraulic or humoral in nature. Hertzman's report that the larger arteries of the hand do not usually participate in the spontaneous waves nor in the vasoconstrictor responses is against the former mechanism, unless it is transmitted through the venous circulation. Also, Neumann showed that alpha waves had no relation to spontaneous variations in blood pressure. Among the humoral agents, epinephrine would be a natural possibility, though the appearance of slow waves during dibenzyline blockade argues against this. Some other agent, such as histamine or adenosinetriphosphate, could be involved. The chromaffin-reacting cells demonstrated by Burch and Phillips in human digital skin may have a role in vasomotor control. When these mechanisms are better understood, the return of vasomotor activity following sympathectomy may also be explained.

It is also entirely possible, as Orlov suggests, that the slow waves are the reflection of several changes and are not due to any one factor alone. Their irregularity as to amplitude, form, and frequency favors this concept. It is difficult, however, to explain their appearance in several areas simultaneously without some central coordinating control.

Summary

Spontaneous vasomotor activity, as evidenced by the presence of alpha or beta waves in the paw plethysmogram, was demonstrated in 10 sympathectomized, 4 denervated, and 8 ganglionically or adrenergically blockaded extremities of dogs. Alpha waves occur during natural sleep and during anesthesia. They are present after removal and denervation of the carotid and aortic chemoreceptors. They occur in the splenectomized animal. Alpha waves are usually synchronous in a pair of normally innervated extremities. They may be synchronous, asynchronous, or absent in one paw when one extremity (or both) is denervated or sympathetically blocked. The significance of this information is discussed in relation to the factors which may control spontaneous vasomotor activity.

Summario in Interlingua

Spontanee activitate vasomotori, manifeste per le presentia de undas alpha o beta in le plethysmogramma del pata, esseva demonstrate in 10 sympatectomisate, 4 disnervate, e 8 ganglionica- o adrenalica-mente blocate extremitates de canes. Undas alpha occurre durante le somnio natural e etiam in stato de anesthesia. Illos es presente post le ablation e disnervation del chimoreceptores carotide e aortie. Illos occurre quando le animal es splenectomisate. Le undas alpha es usualmente synchronous in un par de normalmente innervate extremitates. Illos pote esser synchronous o asynchronous o unilateralmente absent quando un del extremitates o ambes es disnervate o sympatheticomisate. Le significacion de iste information es discutite in relation al factores que es possibilemente responsabile pro le regulation del spontanea activitate vasomotori.

References

7. Neumann, C., Lhamon, W. T., and Cohn, A. E.: Study of the factors (emotional) responsible...


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