

## LATERAL-LINE INPUT AND STIMULUS LOCALIZATION IN THE AFRICAN CLAWED TOAD *XENOPUS* SP.

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### SUMMARY

1. Intact *Xenopus* sp. responded to a train of surface waves with a single, stimulus-directed motor response which consisted of a turning component and a displacement component. The turning angle increased in a linear fashion with the stimulus angle.

2. The magnitude of the turning angle was not affected by (a) the size of the animal, (b) the way the surface wave was elicited (with a drop of water or by dipping a rod into the water), (c) reflected waves and (d) the number of successively administered stimuli.

3. With no lateral-line organs left intact *Xenopus* could still localize the origin of a surface wave, but with reduced accuracy.

4. With only two stitches left intact on the left and on the right occipital lines, the turning angles were more widely scattered for stimuli placed at angles larger than 90°. The scatter was even larger than that of the equivalent responses from animals without a functional lateral-line system, i.e. the directional responsiveness in the partly lesioned animals was less accurate than in those with their entire lateral-line inoperative.

5. *Xenopus* with four or two ipsilateral occipital stitches left intact were no longer able to orientate accurately. When the animal was stimulated on the lesioned side it frequently turned to the wrong side. These errors were absent or less frequent when the animal was stimulated on the intact side.

6. The lesion experiments indicated that (a) a few organs provide sufficient information for an appropriate turning response, but the turnings were only roughly directed towards the stimulus, and (b) the decision whether *Xenopus* localizes a stimulus on the left or right side depends on whether the central nervous system receives information from one or both sides.

### INTRODUCTION

The functional significance of the lateral-line system in amphibians has been investigated in some detail for the African clawed toad *Xenopus*. The larvae possess well-developed lateral-line organs (Shelton, 1971) which, in the dark, may aid these

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microphagous suspension feeders in aligning side by side to optimize their access to food particles (Katz, Potel & Wassersug, 1981). In the carnivorous adult frog the lateral-line has assumed a different function: blind *Xenopus* are still able to perceive a vibrating glass rod from a distance of 20 cm ('Ferntastsinn': Dijkgraaf, 1947). Furthermore, in shallow water, the frog 'spontaneously' responds to surface waves with stimulus-directed taxis movements (Kramer, 1933; Dijkgraaf, 1947). The stimulus, a glass rod dipped into the water, releases a train of surface waves which are characterized by different phase velocities and frequencies. With an increasing distance of the wave's origin the high frequency components are more rapidly attenuated than the low frequency components (Lang, 1980). The waves also extend into the water column and affect the lateral-line organ. Frogs with all lateral-line organs surgically removed still respond to wave directions (Kramer, 1933; Görner, 1973; Elepfandt, 1982*a,b*), implicating at least one additional sensory system, most probably the labyrinth (Görner, 1976).

This paper will compare the performance of intact animals with that of frogs whose lateral-line organs were partially or totally lesioned. At the same time, the lesion experiments were designed to evaluate the behavioural evidence for a functional relation between the lateral-line organs on the left and right side as suggested in neurophysiological and anatomical studies (Plassmann, 1980; U. Will, G. Luhede & P. Görner, in preparation).

#### MATERIALS AND METHODS

Experimental animals were 31 adult male and female *Xenopus laevis* and one *Xenopus mülleri* ranging in body length from 4.7–6.2 cm, as well as juveniles of about 3.5 cm. They were either obtained through local dealers or taken from our laboratory-bred stock. All experimental animals were blinded by enucleation under anaesthesia (tricaine sulphonate, Sandoz). The animals were kept individually in plastic containers and fed once a week with cow heart. The first series of experiments (method 1) was conducted in a shallow, circular pool 100 cm in diameter. To minimize reflection and to attenuate the surface waves, a gravel bed was placed along the periphery of the pool. Water depth was adjusted before the experiment to cover the frog's dorsal body surface by at least 0.5 cm but was maximally 3.5 cm. The water temperature was between 18 and 25 °C. Prior to testing, the animal was adapted to the experimental conditions for several hours. The stimulus was applied only when the frog was resting on the bottom in a circular central area, 30 cm in diameter. The stimulus consisted of surface waves which were generated at a distance of 12–14 cm from the animal (R–S in Fig. 2) by means of a drop of water released from a stopcock 7.5–8.0 cm above the surface (Fig. 1). To assess the amplitudes of the surface waves we determined the maximum amplitude of the wave train in a Plexiglas container under the same conditions as in the experiment but without the animal present (see Rudolph, 1967). The mean maximum amplitude obtained from 20 measurements was  $139 \pm 5 \mu\text{m}$ . When the frog did not respond to the first stimulus, it was repeated up to 10 times at the same location. Subsequently, the stimulus was set at a different location which was selected by chance. The experiment was terminated when the frog ceased to respond to stimuli from different directions. The turning angle (Fig. 2) was

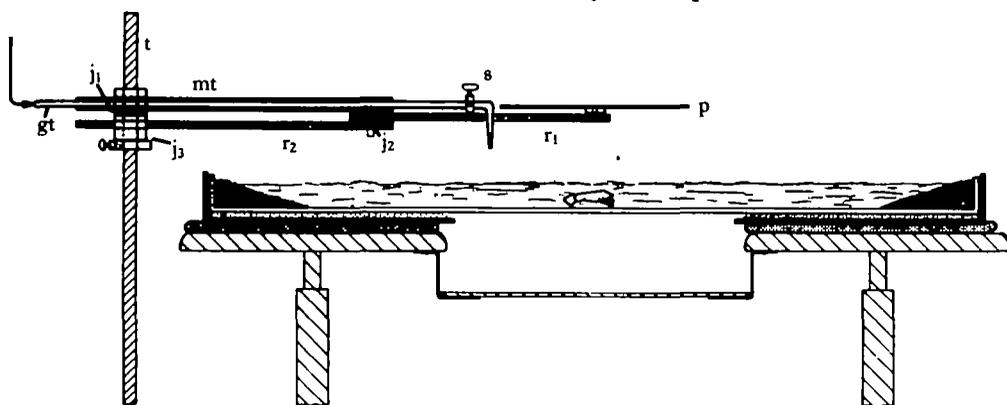


Fig. 1. Experimental set-up for method 1, showing the large pool ( $\Phi = 100$  cm) and the stimulating device. The position of the stopcock, *s*, and the protractor, *p*, can be controlled independently within the horizontal plane, the stopcock by displacing the glass tube, *gt*, within the metal tube, *mt*, or by rotating both tubes around the joint *j*<sub>1</sub>. The protractor can be displaced by bending the two rods *r*<sub>1</sub> and *r*<sub>2</sub> around the two joints *j*<sub>2</sub> and *j*<sub>3</sub>. The arrow indicates the connection of the glass tube to a water hose. *t*, tripod.

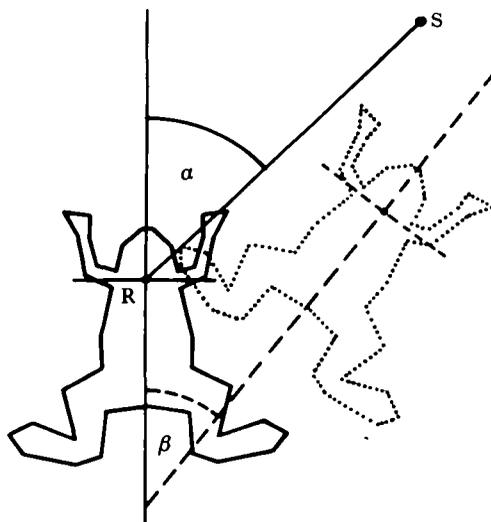


Fig. 2. Definition of stimulus angle ( $\alpha$ ) and turning angle ( $\beta$ ). *S*, stimulus (wave centre); *R*, reference point. Solid contour indicates frog's position before and dotted contour position after turning.

determined by visual inspection with a transparent Plexiglas protractor, the zero-axis of which was aligned with the frog's longitudinal axis prior to and after turning. This was relatively easy to accomplish since the frog remained motionless in the new position for several seconds. Parallax errors were minimized by adjusting the protractor in such a way that the frog's contours were aligned with those of its mirror image. The frog's reaction consisted of two components, a turning response and a spatial displacement of several centimetres, both bringing the mouth of the animal closer to the stimulus origin. The displacement component made it somewhat difficult to determine the exact turning point. Therefore, we used the intercept between the frog's longitudinal axis and a line across the foreleg insertions as reference point (Fig. 2).

The maximum error of the individual measurement was estimated to be  $\pm 5^\circ$ .

The standard procedure (method 1) was modified in the course of the experiments. In method 2 we used experimental Plexiglas containers of  $30 \times 40$  cm. The stimulus, a drop of water which was released from a pipette 1.5–3.0 cm above the water surface, generated, in a distance of 12 cm, surface waves with a maximum amplitude of  $65 \pm 6 \mu\text{m}$ . The stimulus was applied when the frog rested in a central area 10 cm in diameter. Its position before and after stimulation was photographed with a 16 mm overhead movie camera. The turning angles were obtained from single frame analysis. The same procedure was followed in method 3. The stimulus, however, was delivered in an experimental pool 50 cm in diameter by dipping a glass or PVC rod 0.3 cm in diameter into the water. The maximum wave amplitudes were  $68 \pm 15 \mu\text{m}$ .

The lateral-line organs ('stitches') were destroyed by electrocoagulation under anaesthesia. Each animal was subsequently inspected for completeness of the lesion. The stitches were never observed to regenerate.

Stimulus and turning angles to the right were counted positive, those to the left negative.  $0^\circ$  indicated direct frontal stimulation or 'no turning', respectively. The data obtained from animals with their lateral-line organs unilaterally eliminated on the left side were mirrored along the longitudinal axis, and presented as if the frog's right side had been operated.

Statistical analysis: frequency distributions were compared with a chi-square-test when  $N > 20$  and the Fisher-Yates test when  $N < 20$ . For an evaluation of the values obtained with method 1, circular statistics was applied according to Batschelet (1965). Circular distributions were compared with the test of Watson & Williams or the test of Mardia, Watson & Wheeler. The F-test was applied when the data were distributed normally ( $\chi^2$ -test) and/or when  $N > 25$ . For a comparison of the slopes of two linear regression equations Student's test was applied (Zar, 1974). The level of significance is  $P < 0.01$ .

## RESULTS

### *Large pool, Method 1*

#### *Baseline data*

*Adult animals (experiment 1).* Nine adult *X. laevis* were stimulated with surface waves. Single frame analysis of movie records indicated that the animal's overt behaviour in response to a train of surface waves, passing partially or entirely over the animal, consisted of a single (open-loop type) turning response which was not corrected during its execution. As outlined before, the orientation response was accompanied by locomotion toward the source of stimulation (Fig. 2). The swimming angle and the distance moved will not be considered in this paper.

Fig. 3 illustrates the combined data from nine intact animals. The mean turning angle was positively correlated with the stimulus angle, though on the average attenuated by a factor of  $0.83 \pm 0.09$ . On two occasions, at stimulation angles  $-90^\circ$  and  $-150^\circ$ , turning occurred to the contralateral side. These values did not enter into the computation. The accuracy of the turning angle decreased with larger stimulus angles as indicated by an increase in the standard deviation. The difference between standard

Deviations is significant when responses to stimuli at  $30^\circ$  and  $180^\circ$ ,  $30^\circ$  and  $150^\circ$ , and  $-30^\circ$  and  $-180^\circ$  are compared ( $P < 0.01$ ). The increase in standard deviation cannot be attributed to an increase in the measurement error, since it is also significant ( $P < 0.001$ ) in seven *X. laevis*, treated with method 3 (photographic recording). In this case the deviations from the regression line of the turning angles  $0^\circ$  to  $\pm 90^\circ$  were compared with those of  $\pm 91^\circ$  to  $\pm 180^\circ$ .

*Juvenile animals (experiment 2).* In a wave train the wave lengths are in the centimetre to millimetre range. The spatio-temporal excitation of the central nervous system should therefore be different in smaller and larger animals. If the motor response were size-dependent this could lead to different turning angles in smaller and larger frogs. To evaluate this possibility we determined the turning angles of six juvenile *X. laevis* (body length approximately 3.5 cm) and compared them with those from adult frogs (body length approximately 5.5 cm). There were no differences in the turning angles irrespective of the frog's size and age (exception: left turnings at stimulus angle  $180^\circ$ :  $P < 0.01$ ) and there was no difference ( $P > 0.05$ ) between the regression lines of the larger adult and the smaller juvenile animals (Table 1). Here too, we observed two contralateral turns at  $-60^\circ$  and  $+150^\circ$  which did not enter into the computation.

*Single and repeated stimulation.* Frequently, the animal did not respond to the first stimulation but only to repeated stimulation. It is possible that under these conditions *Xenopus*' 'readiness' to turn was somewhat weak, resulting in smaller turning angles. To test this possibility, we combined the turning angles from six groups ( $\pm 30^\circ$ ,  $\pm 60^\circ$ ,  $\pm 90^\circ$ ,  $\pm 120^\circ$ ,  $\pm 150^\circ$ ,  $\pm 180^\circ$ ) and tested whether the turning angle was different when the animal was stimulated only once, two to three times, or four to eleven times. There was no significant difference, neither between any of the mean turning angles

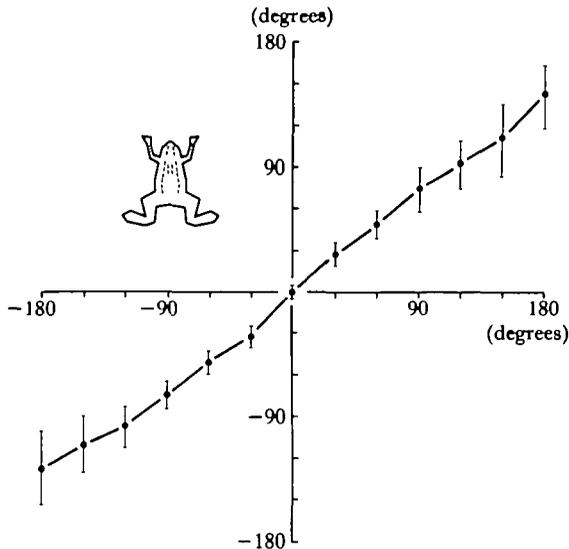


Fig. 3. Responses of nine *Xenopus* to a train of wavefronts which were elicited at a distance of 13.5 cm by a drop of water. Experiments were conducted in the large testing arena (method 1). Vertical bars indicate  $\pm$  standard deviation.

Table 1. *Correlation between stimulus angle and turning angle in 20 Xenopus laevis*

Number of stimuli	a	b	$s_{y \cdot x}$	$r^2$	N
Experiment 1, nine adult <i>Xenopus</i>					
1	0.6	0.77	17.1	0.95	203
2-3	0.8	0.78	10.8	0.97	51
4-11	1.5	0.79	17.7	0.97	132
1-11	0.95	0.78	16.6	0.96	386
Experiment 2, six juvenile <i>Xenopus</i>					
1	2.2	0.85	15.0	0.97	70
2-3	-4.8	0.86	20.7	0.96	48
4-11	-0.8	0.79	25.8	0.92	53
1-11	-0.7	0.83	20.6	0.95	171
Experiment 3, seven adult <i>Xenopus</i> with middle, upper and occipital lines left intact on both body sides					
1-11	0.98	0.76	21.8	0.93	102
Experiment 4, two adult <i>Xenopus</i> with middle, upper and occipital lines left intact on the left body side					
1-11	-2.4	0.80	19.3	0.95	146
Regression equation: $y = a + bx$ . Large pool, method 1.					

( $P > 0.05$ ) nor between the regression lines (Table 1), suggesting that the magnitude of the turning angle was independent of the number of stimuli.

### *Lesion experiments*

*Number of functional lateral lines (experiment 3).* The lateral-line organs are unevenly distributed over the animal's body. On the upper side, which is most probably more affected by surface waves than the under side, there are in the head region about 90 stitches whereas there are only about 100 distributed over the much larger remaining upper side of the body, including those organs which are located along the animal's flanks.

Görner (1976) demonstrated that the frog's responsiveness to surface waves was not significantly affected by elimination of the lateral-line organs on the head and on the ventral side. We were now interested whether the turning angle was affected. In this experiment all lateral lines except those on the upper side of the trunk (i.e. the occipital, the upper and the middle row) were eliminated in seven *X. laevis*. There is no significant difference between the regression lines of the lesioned and the intact animals ( $P > 0.05$ , Table 1). However, two of the mean turning angles ( $\pm 60^\circ$  and  $\pm 90^\circ$ ) were significantly different ( $P < 0.001$ , Table 2). Thus, although the lateral-line organs on the trunk with their different directional sensitivities relay unequivocal directional information to the central nervous system, a reduction of about 70% of the stitches on the upper side results in a partial loss of localization accuracy.

*Location of functional lateral lines (experiment 4).* To understand the functional aspects of the system we studied the responses of two frogs whose lateral line organs had been eliminated except those on one trunk side (middle, upper and occipital line). As in the preceding experiment, the stitches of the remaining lateral lines of the trunk

Table 2. Comparison of the turning angles of nine intact animals and seven *Xenopus laevis* whose lateral-line organs (LL) on the head and on the ventral side had been destroyed

Stimulus angle		Mean turning angle and circular standard deviation	<i>N</i>	<i>P</i>
±0°	LL intact	1.6 ± 5.1	33	>0.05
	LL lesioned	1.0 ± 5.2	6	
±30°	LL intact	29.6 ± 7.7	67	>0.01
	LL lesioned	36.2 ± 9.3	8	
±60°	LL intact	51.4 ± 9.6	67	<0.001
	LL lesioned	58.4 ± 8.5	16	
±90°	LL intact	75.8 ± 14.0	67	<0.001
	LL lesioned	69.6 ± 11.7	28	
±120°	LL intact	95.0 ± 15.7	60	>0.05
	LL lesioned	97.1 ± 17.3	23	
±150°	LL intact	111.6 ± 23.3	54	>0.05
	LL lesioned	114.4 ± 22.0	13	
±180°	LL intact	136.8 ± 25.9	38	>0.01
	LL lesioned	107.9 ± 43.7	8	

should provide the central nervous system with unequivocal directional information about each stimulus direction and the frog should accurately localize the stimulus. This assumption seems to be confirmed by the regression equation (Table 1) which is not significantly different from that of intact animals ( $P > 0.05$ ). However, when the stimulus was placed directly in front (stimulus angle: 0°) the frogs localized the wave centre in 85% ( $N = 26$ ) of stimuli with errors made towards the intact side (one turn was toward the lesioned side, on three trials there was no turn at all). When the stimulus was placed directly behind (stimulus angle: 180°) the animals again more often turned toward the intact side (84%,  $N = 25$ ), and only four times contralaterally. (Two additional frogs, one *X. laevis* and one *X. mülleri*, treated with method 3 reacted in the same manner.)

#### Small pool, Method 2

##### Baseline data

*Individual differences (experiment 5)*. In the preceding experiments (using method 1) most of the baseline data and data following lesioning were obtained from different animals. Therefore, individual differences in the turning angles of intact animals were not taken into account. In the following experiments (5–7), controls and successive elimination of lateral line organs were conducted with the same animals. Before lesioning we determined the turning angle for each of the three animals (Fig. 4A). A linear relationship between stimulus and turning angle was justified by a best fit through least squares linear regression with  $r^2 = 0.99$  and a standard deviation of  $s_{y,x} = 13^\circ$  (Table 3). When the animals were stimulated from the rear, slightly off 180°, they occasionally turned contralaterally. These data were not included in the computation.

Table 3. *Regression equations ( $y = a + bx$ ) of three Xenopus laevis in the small pool, method 2, experiment 4 and of three Xenopus laevis in the large pool, method 3, experiment 9*

<i>Xenopus</i> No.	a	b	$s_{y \cdot x}$	$r^2$	N
		Method 2			
8	0.5	0.81	13.1	0.98	192
17	-1.7	0.82	13.3	0.98	203
28	-1.0	0.85	13.3	0.98	197
		Method 3			
78	-0.2	0.81	15.2	0.98	94
82	-3.4	0.84	12.7	0.99	119
86	-3.4	0.74	12.7	0.99	63

There is no significant difference between the slopes of the individual regression lines ( $P > 0.05$ ). The same holds for the three *X. laevis* of experiment 9, three additional *X. laevis* and one *X. mülleri*.

#### *Lesion experiments*

*Number of functional stitches (experiments 6–8).* The three animals of experiment 5 (control) were successively lesioned in the following way: all lateral-line organs were destroyed except (i) two stitches, each on the left and right occipital line (experiment 5), (ii) two stitches on the left occipital line (experiment 6) and (iii) in experiment 7 no stitches were left intact. The results show that, compared with intact animals (Fig. 4A), frogs completely lacking lateral-line input were still able to localize stimuli (Fig. 4B), though less accurately than intact animals. This is indicated by the increase in scatter ( $P < 0.001$ ). Assuming a linear correlation between stimulus and turning angles (as demonstrated with intact frogs) there remained a significant correlation between these two angles in the operated animals ( $r^2 = 0.87$ ). The results from frogs with two or four stitches left intact were particularly interesting. While there was still a correlation between the two angles in the frontal region ( $0-90^\circ$ ) in animals with four stitches left (Fig. 4C,  $r^2 = 0.94$ ), such a correlation was no longer observed in frogs with two stitches left (Fig. 4D,  $r^2 = 0.40$ ). Thus, these animals were no longer able to localize the source of stimulation. A comparison of the frequency of turning responses to the wrong, i.e. the contralateral stimulus side yielded comparable results. In animals with two stitches left these responses amounted to 20% and thus were significantly higher when compared with (i) animals with four stitches left (3%,  $P < 0.001$ ) and (ii) animals without a functional lateral-line (2%,  $P < 0.001$ ). Comparing the turning errors among animals with two stitches left, it became evident that the stimulus direction was more frequently mistaken when the frog was stimulated on the lesioned side than on the intact one ( $P < 0.01$ ). (The data of experiments 6–8 were pooled in Fig. 4B–D because all frogs exhibited similar behaviour.)

The difference between intact animals and those without a functional lateral-line system was also clearly established in a comparison of the variances of the turning angles. The data obtained from animals with their lateral lines rendered inoperative were significantly more scattered than those from intact frogs ( $P < 0.001$ ). However,

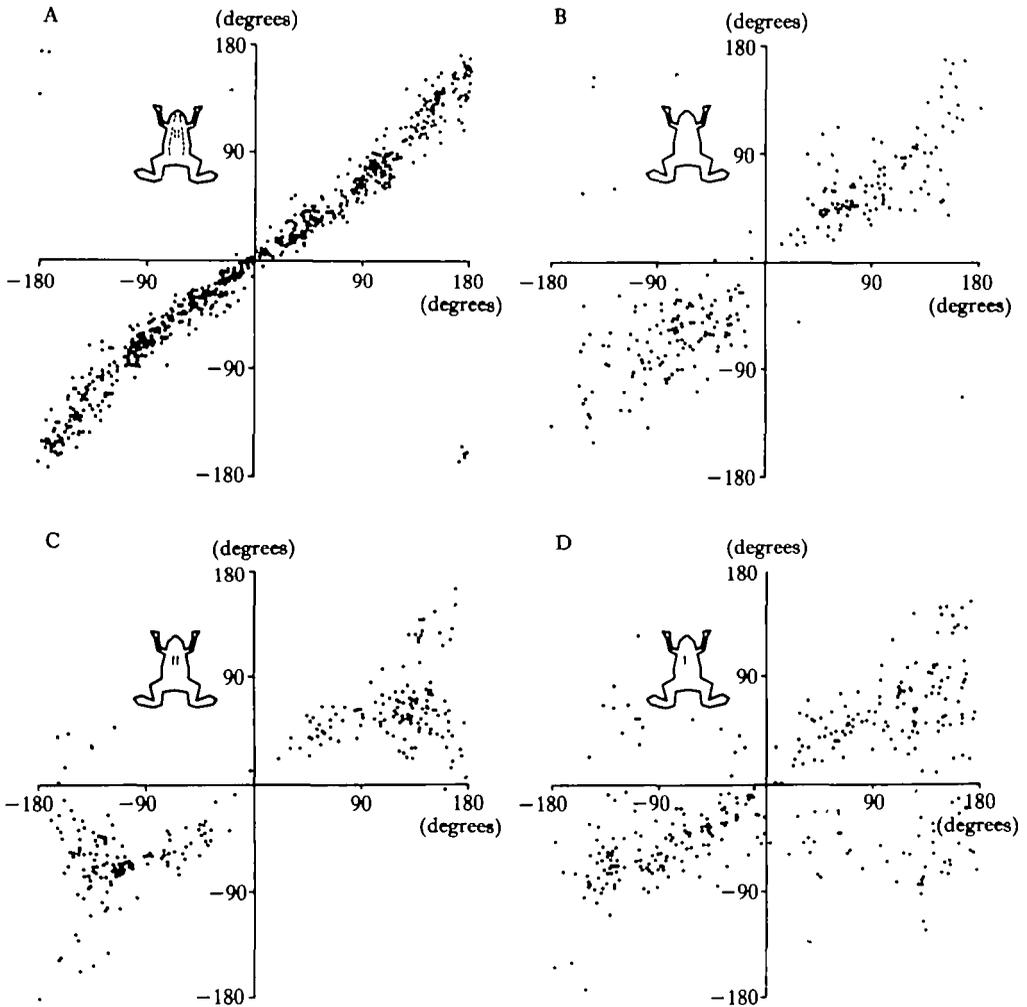


Fig. 4. Responses of three *Xenopus* to a train of wavefronts which were elicited at a distance of 12 cm by a water droplet (method 2). (A) Animals with their lateral-line system intact, (B) animals with no functional lateral-line system, (C) animals with four stitches left intact, two on the left and two on the right occipital line, and (D) animals with only two stitches left intact on the left occipital line.

for both groups, there was no difference between the turning responses to the right and left sides. This also held when we considered the frontal ( $0-90^\circ$ ) and rear ( $91-180^\circ$ ) areas separately. When stimulated from the rear the accuracy of localization of stimuli with four parallel dorsal stitches left intact decreased when compared with intact animals ( $P < 0.001$ ), and decreased even further ( $P < 0.001$ ) when compared with animals lacking lateral-line input altogether.

#### *Large pool, Method 3*

##### *Lesion experiments*

**Location of functional stitches (experiments 9-11).** The higher frequency of turning responses to the wrong, i.e. the contralateral, side in animals with only two stitches

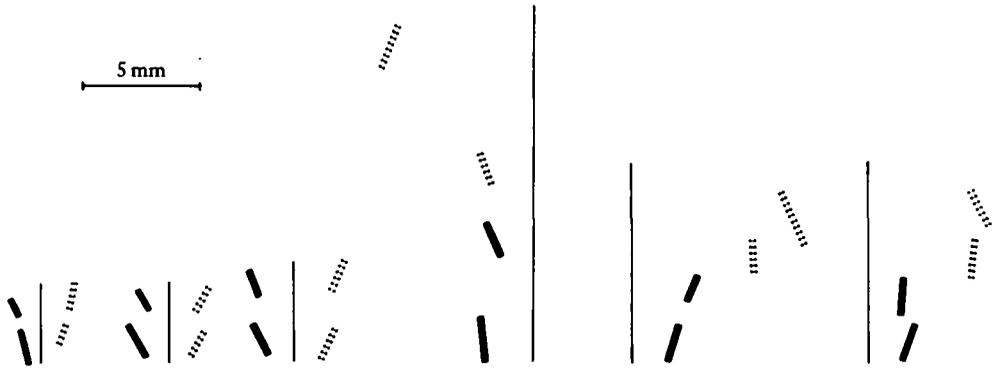


Fig. 5. Location of occipital stitches in three *Xenopus* used in experiment 5 (left) and three *Xenopus* used in experiment 10 (right). Stippled symbols denote stitches which were eliminated in experiment 6 (left) and in experiment 11 (right), respectively. Vertical lines indicate animal's longitudinal axis.

left compared with those in frogs with four stitches may shed some light on stimulus integration. Except for the difference in number, both stitches were ipsilateral. If only the number of organs determined the response direction one would expect animals with four intact ipsilateral stitches to respond as accurately as animals with two stitches on each side. We tested this with three *X. laevis*, leaving four stitches intact either on the right or on the left occipital line, respectively (experiment 10). All four organs were in line but somewhat staggered (Fig. 5) such that a parallel wavefront from the left or right side would not affect all four stitches simultaneously. As Fig. 6B indicates the animals orientated in a similar fashion to those with two intact ipsilateral organs in experiment 6, i.e. correctly when the stimulus was applied on the intact side and frequently to the wrong side when the wavefront first passed the lesioned side. Prior to the operation the animals did not exhibit turning errors (except when stimulated directly in front or from the rear; experiment 9, Fig. 6A). The turning response remained the same when subsequently two of the four stitches were removed, leaving only two dorsal organs intact (experiment 11, Fig. 6C). In Fig. 6 the data of the three frogs were pooled because they all reacted in the same manner.

#### DISCUSSION

Although clawed toads do not possess specific morphological adaptations to perceive surface waves as do surface-feeding fish (Schwartz, 1965, 1967), they respond to surface waves with stimulus-directed movements. We therefore assume that these movements are of adaptive significance in the frog's natural habitat. How the surface-waves are generated, either by a water droplet or by dipping a rod into the water, seems to be of no importance. Under the present conditions, the magnitude of the turning angle was not influenced by the way the surface wave was generated.

At rest, *Xenopus* either remains at the bottom of the tank, or, with the head making contact with the water surface, stays there at an angle, the magnitude of which depends on the water level (see Dijkgraaf, 1967, Fig. 5; Elepfandt, 1982a). Since the higher frequency components of a wave train penetrate less deeply than the lower

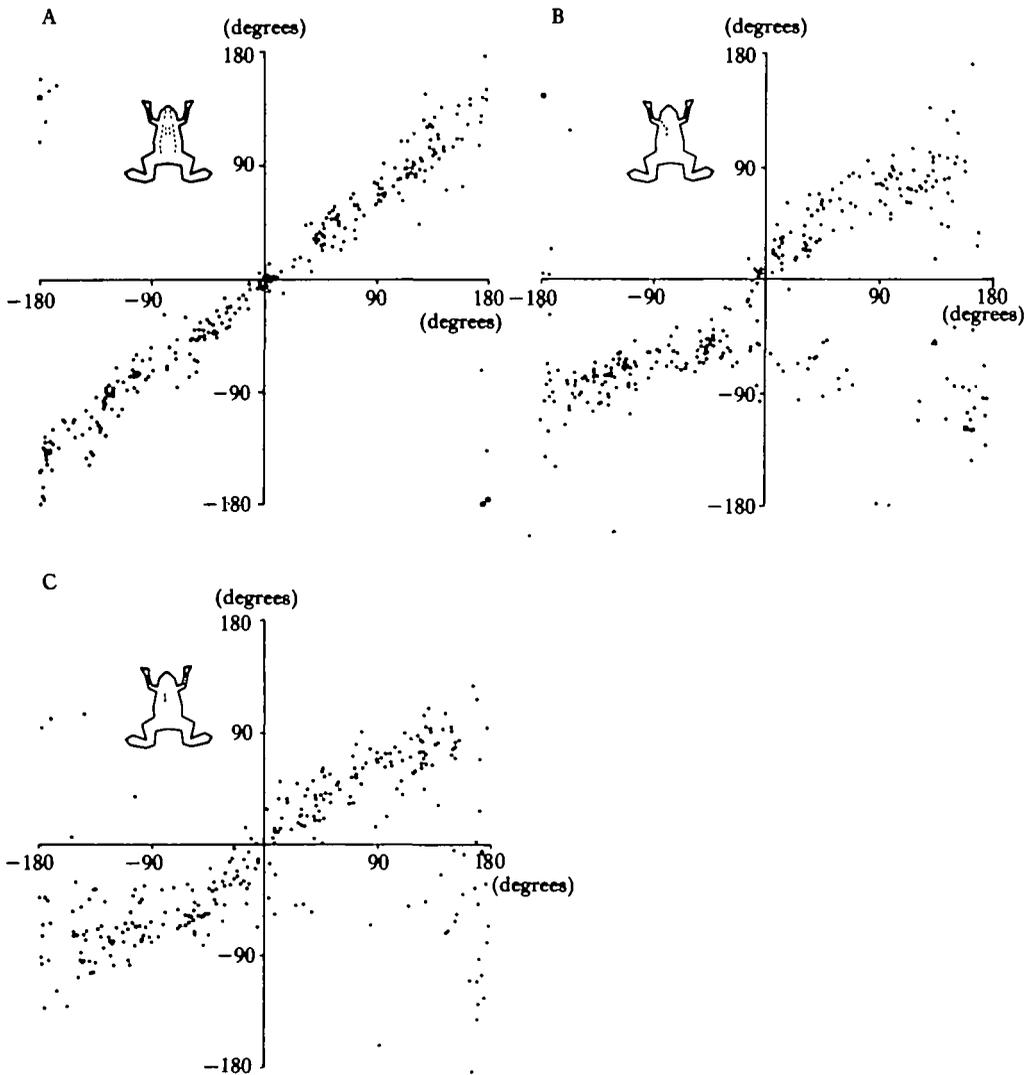


Fig. 6. Responses of three *Xenopus* to surface waves which were elicited by dipping a rod into the water at a distance of 12 cm in the large experimental arena (3 mm rod; method 3). (A) Frogs with their lateral-line system intact, (B) frogs with four stitches on the left occipital line left intact, and (C) frogs with only two stitches on the left occipital line left intact. ■ Turning angles exceeding 180°.

frequency components (Schuijf, 1976; Wiese, Woolnik & Jebram, 1980), the lower-lying lateral-line organs, with the frog in a slanted position, are possibly stimulated by frequency spectra different from those affecting the upper head region. Therefore, only when the frog is resting horizontally on the bottom can one assume that all lateral-line organs of the upper body surface are affected by comparable stimulus conditions (i.e. amplitude and frequency).

The accuracy of the orientation response was surprisingly high. The scatter of the turning angles around the regression line was between  $s_{y,x} = 13$  and  $24^\circ$  (one *X. mülleri*:  $s_{y,x} = 30^\circ$ ). The accuracy decreased somewhat in response to larger stimulus

angles. The close fit suggests a linear relation between stimulus and turning angle in intact *Xenopus*. When the stimulus originated slightly off the labial position ( $180^\circ$ ), *Xenopus* also turned several times to the contralateral, i.e. the wrong, side. Here, the magnitude of the turning angle was correct, but not its direction. In the experiments reported by Elepfandt (1982a) the relation between stimulus angle and turning angle decreased at stimulus angles larger than  $90^\circ$ . Two methodological variables could explain this difference: (i) the water depth in the experimental tank was 7–8 cm and the frogs 'hung' under the water surface. Therefore, stimulus conditions for the different lateral-line stitches and mechanical conditions for the turning movement were different in frogs resting horizontally on the bottom of the tank as in our experiments, and in frogs slanted at angles between  $30$ – $60^\circ$  to the water surface as in Elepfandt's experiments. (ii) The weight of the adult animals used by Elepfandt was considerably greater than that of frogs used in the present study. We measured weight and length of several small and large *Xenopus* and extrapolated that the juvenile *Xenopus* in Elepfandt's experiments had the same weight as our adult frogs. Our frogs never exceeded 8 cm in body length, or 50 g in body weight. It is possible that the relation between stimulus and turning angle changes in large *Xenopus*.

Elepfandt reported that his frogs often responded with low accuracy, especially at larger stimulus angles (Elepfandt, 1982a, Fig. 2b). We never obtained such large deviations with any of our frogs.

Whether turns made by intact frogs to the contralateral side, which occurred occasionally in the large pool at stimulus angles less than  $180^\circ$ , were caused by reflection waves cannot be decided. (Elepfandt reported contralateral turnings of about 1%.) The use of a smaller observation tank (in method 2) should result in shorter time lags of any reflected wave and thus affect the turning angle. However, regardless of the size of the experimental tanks used, the response accuracy was quite similar under both conditions. This may suggest that *Xenopus*, though able to discriminate between direct and reflected waves, only responds to the initial wavefront.

Elimination of two-thirds of all lateral-line organs (including all stitches on the ventral side and the head region) did not affect the response frequency (Görner, 1976) but did influence the magnitude of the turning angles. To assess the functional aspects of the lateral-line, a larger number of stitches had to be eliminated. As pointed out before, in evaluating the effects of a lesion, one has to bear in mind that *Xenopus* is able to orientate correctly without an intact lateral-line system (Fig. 4B). Also the specific contribution of the labyrinth and/or any other sensory system (Görner, 1976), and the remaining functional lateral-line organs to a correct and accurate turning response is not known. Recently, Müller & Schwartz (1982) have demonstrated that the surface-feeding fish *Aplocheilichthys lineatus* which, like *Xenopus*, localizes a wave centre with high accuracy by means of its lateral line system, responds to surface waves even after all its neuromasts have been destroyed. After lesioning, however, the turns were only roughly directed towards the stimulus. The receptor system responsible for this reaction is not yet known.

*Xenopus* with four dorsal stitches left intact, two on each side, responded correctly to frontal stimuli ( $0$ – $90^\circ$ ), but exhibited turning errors in response to stimuli from the

feared. We conclude that the afferent information from two parallel stitches on each side is ambiguous about the stimulus direction, but not so about left or right side stimulus origin. At stimulus angles larger than  $90^\circ$  most of the turning angles decrease with an increase in the stimulus angle (Fig. 4C). One may conclude that the sinusoidal course of the turning angles represents the sinusoidal directional sensitivity of the four intact lateral line organs. However, a discrimination among stimulus angles, originating on the left or right side, on the basis of only four intact organs with equal directional characteristics is not to be expected (Görner, 1961, 1963): a strong stimulus from a less effective stimulus angle results in the same excitation as a weak stimulus from the most effective angle (with the wavefront perpendicular to the organ). Thus, correct turning angles with only four intact organs left could be explained in terms of control through the labyrinth, and all other turning angles by an undefined compromise direction between the labyrinth input and the information 'left' or 'right' from the four occipital organs. It remains open why, when stimulated from the front, the animals responded as accurately as intact ones did.

*Xenopus* with only two dorsal ipsilateral stitches left intact almost totally lack directional responsiveness and, in addition, confuse left and right stimulus origin. The fact that *Xenopus* without a lateral-line system are able to localize a stimulus source but are totally confused with an additional input from only two stitches on one side demonstrates convincingly the superior role played by the lateral-line input in stimulus localization.

The left-right errors cannot be attributed to the reduced number of stitches, since – with the stimulus placed on the operated side – they were also observed in animals with four intact organs (experiment 10). Obviously, information to the central nervous system originating from one side only is ambiguous even if it contains a left-right decision. The central nervous system is likely to compare the information from both sides. Neuroanatomical (U. Will, G. Luhede & P. Görner, in preparation) and electrophysiological evidence (Plassmann, 1980) points to such a bilateral interaction in *Xenopus*. Axons of secondary neurones in the nucleus lineae lateralis project *via* external arcuate fibres to the contralateral side and terminate there in the lateral-line neuropile; excitation of the secondary neurones in the medullar nucleus lineae lateralis can be suppressed for about 4–40 ms through prior contralateral electric stimulation of the nucleus lineae lateralis posterior. The distance between two opposed stitches of the left and right occipital lateral-line is at least 0.9 mm. Under the experimental conditions, the wavefront (maximum amplitude) propagated with a speed of about  $20 \text{ cm s}^{-1}$ . Thus, the dorsal stitches were affected by a time difference of more than 4.5 ms, i.e. within the range of the inhibitory phase.

Our results give no indication of a comparable temporal interaction between or among ipsilateral organs; frogs with four ipsilateral stitches intact (separated by the same or larger distances than those of the left and right occipital line) were unable to orientate unambiguously when the stimulus was placed on the lesioned side (experiment 10). These animals responded like frogs with only two ipsilateral organs left intact. This result is in contradiction to findings of Elepfandt (1982a), who reported that *Xenopus* with two or more ipsilateral stitches (in contrast to frogs with only one functional stitch) localized the stimulus 'practically' as accurately as intact animals. Consequently, he concluded that 'temporal comparison of receptor activation seems

to be used for determination of wave directions'. It is difficult to give a convincing explanation for the different results. Only further experiments will provide convincing arguments for the two proposed interpretation.

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