

Fig. 2. Vibrationssignale eines ♂ von *Panorpa communis*. Zeitmarke am oberen Rand = 1 sec.

reichen Untersuchungen bekannt<sup>6,7</sup> (weitere Literatur siehe <sup>8</sup>), sind die ♂♂ erst 7–10 Tage nach dem Schlüpfen begattungsfähig (Heranwachsen der Speicheldrüsen). Da sich die Tiere in ihrem natürlichen Biotop öfters begegnen, kann so eine schnellere Sortierung erreicht werden. Ausserdem dienen die Signale dazu, das langwierige Liebesspiel zu synchronisieren bzw. zum frühest möglichen Zeitpunkt beginnen zu lassen. Ahmt man die Vibrationen mit einem Sinusgenerator nach, so werden fortpflanzungswillige ♀♀, die ruhig sitzen, plötzlich sehr aktiv, springen in die Höhe und winken lebhaft mit den Flügeln und vibrieren mit dem Abdomen. Diese Aktivität klingt nur allmählich ab und kann danach erneut stimuliert

werden. Trennt man ♂ und ♀, die mit dem Vorspiel schon begonnen haben, einzeln in Gläser, so wird von beiden intensiver als vorher gewinkt und vibriert; beide «suchen» auf diese Weise den Geschlechtspartner. Die Artspezifität dieser Signale steht noch nicht fest. Dabei können kombinierte Film- und Tonbandaufnahmen weiterhelfen.

Vibrationssignale bei der Paarung wurden auch von den Chloropidae/Diptera<sup>2</sup> beschrieben. Diese Ordnung kann nach der phylogenetisch-systematischen Gliederung der Insecta<sup>9</sup> als die nächst verwandte Ordnung der Mecoptera angesehen werden. Daraus ergibt sich aber keine synapomorphe Homologie für die Mecopteroidea, da ähnliche Signale auch von den weiter entfernt stehenden Sialiden/Megaloptera<sup>10</sup> bekannt sind.

*Summary.* Male and female of *Panorpa* find each other for copulation by vibration signals, which they produce through up and downward movements with the end of the abdomen. The frequency is about 100 Hz/sec, the duration 1 to 10 sec.

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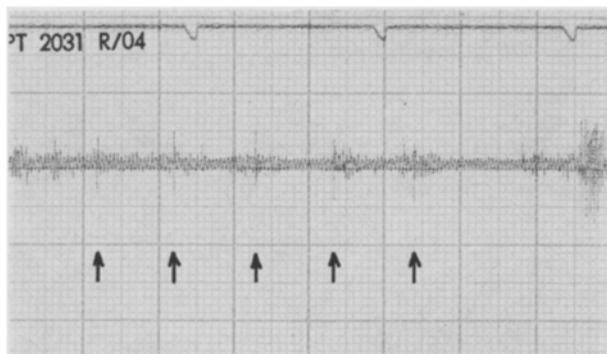


Fig. 3. Klopffolge eines ♂ von *Panorpa communis*, die durch Antippen des Untergrundes mit dem Abdomenende entsteht. Die dadurch ausgelösten Piks sind durch Pfeile markiert. Zeitmarken am oberen Rand = 1 sec.

<sup>6</sup> L. MERCIER, Archs. Zool. exp. gén. 55, 1 (1965).

<sup>7</sup> P. STEINER, Z. Morph. Ökol. Tiere 17, 1 (1930).

<sup>8</sup> P. GRASSÉ, *Traité de Zoologie* (Masson, Paris 1951), Vol. 10, p. 1.

<sup>9</sup> W. HENNIG, *Die Stammesgeschichte der Insekten* (Kramer, Frankfurt 1969).

<sup>10</sup> R. RUPPRECHT, J. comp. Physiol., im Druck (1974).

## Laser-Induced Stimulation of the Vascularization of the Healing Wound. An Ear Chamber Experiment

Various biological reactions of living cells and tissues are considerably influenced by the laser irradiation<sup>1,2</sup>. Some of the effects, including destruction of certain tumors, treatment of detached retinal layers, etc., are of clinical importance.

Following previous animal experiments<sup>3</sup>, a new field of clinical application was opened by the observation of MESTER et al.<sup>4–8</sup>, that the laser irradiation has a stimulating effect on wound healing. In various ulcerations, in which disposition for spontaneous healing was never noted and conventional treatment proved to be unsuccessful, intense granulation started after appropriate laser treatment. In 12 clinical cases, complete healing of the ulcer was achieved by repeated irradiation.

The biological mechanism of action of laser irradiation on wound healing is so far not completely understood. According to our previous studies, collagen formation is stimulated by enzymatic processes not yet elucidated<sup>8</sup>. In this study we have investigated one of the most important phases of wound healing: the development of blood vessels, and the influence of laser irradiation on this process. According to our results, the development of blood circulation in the growing tissue is significantly increased by irradiation with laser.

*Method.* Rabbit ear chamber. The ear chamber used in these experiments has been described in detail by SANDERS et al.<sup>9</sup>. It is a modification of the original Sandison-Clark chamber and up to date this chamber has been the most

used. In our present study, however, the valve was missing from the central table, because this was considered superfluous. The thickness of the space, into which the new tissue grew, was 50–55  $\mu\text{m}$ .

**Laser-irradiation.** He-Ne gas laser (5 mW power output, produced by the Hungarian Optical Works) was used. After operation, chambers of 6 rabbits were irradiated daily with laser and other 6 animals (1 chamber in every rabbit) served as controls. For 12 days after implantation, the border area of the central table was irradiated tangentially in 3 spots, with an individual energy-output of 1 Joule. 2 of the 3 spots were near to the central artery, as the vascularization invariably started from this side. Between the 13th and 18th day following implantation, the total surface of the central table was irradiated once a day with an energy of 1 Joule. This was achieved by a scattering lens through which the laser beam covered the entire area (8 mm diameter) of the round central table of the chamber.

During irradiation, the rabbits were kept in stocks, the ear with the chamber was held in the path of the laser beam by hand. Qualitative evaluation was carried out with taking photograph of the growing tissues. The results were expressed also in quantitative form in the following manner: The image of the central table was projected on paper, the vascularized area depicted and after cutting out the weight was measured. The weight obtained in this way was expressed in the percentage of the weight of the total area of the central table (100%). For the photographic work, a blue filter was used for additional contrast.

**Results and discussion.** The field opened up by the transparent chamber is enormous for the pathologist and the pharmacologist as well as for bacteriologists. The rabbit ear chamber technique is most widely used for the microscopic study of living blood vessels. The selected vessel may be observed with the highest power of the microscope. In our present study, the ear chamber technique was used in a new way. Since the study concerned the intensity of vascular development, the experiments could not have been self-controlling. In

this study the result obtained in one chamber was compared to the result observed in the other one. This fact needs some further explanation.

Obviously the success of the ear chamber technique depends mainly on the surgical conditions, implantation technique and after-treatment of animals. According to our experience, however, in the case of identical chambers, dissection technique and postoperative treatment (administration of penicillin), the difference in the intensity of the vascularization of the different chambers was not significant. This observation is the basis of the method in the manner used by us.

Under our uniform conditions, the first capillaries appeared on the edge of the central table 12–18 days after implantation. About 30–40 days was necessary for the complete vascularization of the chamber. In every case, the first vessels originated from the preformed vessels in the region adjacent to the central artery. The entire process itself is described in detail by EBERT et al.<sup>10</sup> Our findings confirmed their observations. The path before the ingrowing vessels is cleared up by macrophages, phagocytosing the bloodclot in the thin observation space.

<sup>1</sup> First annual conference on Biologic Effects of Laser Radiation, Fed. Proc. Suppl. 14 (1965).

<sup>2</sup> S. FINE and E. KLEIN, Adv. biol. med. Phys. 10, 149 (1965).

<sup>3</sup> E. MESTER, G. LUDÁNY, V. FRENYO, M. SELLYEI, B. SZENDE, G. GYENES, M. IHÁSZ, A. F. KISS, A. DÖKLEN and G. J. TOTA, Panminerva med. 13, 558 (1971).

<sup>4</sup> E. MESTER, B. SZENDE, T. SPIRY and A. SCHER, Lyon chir. 67, 416 (1971).

<sup>5</sup> E. MESTER, T. SPIRY, B. SZENDE and J. G. TOTA, Am. J. Surg. 122, 532 (1971).

<sup>6</sup> E. MESTER, B. SZENDE, T. SPIRY and A. SCHER, Acta chir. hung. 13, 315 (1972).

<sup>7</sup> E. MESTER and É. JÁSZSÁGI-NAGY, Studia biophys. 35, 227 (1973).

<sup>8</sup> E. MESTER, A. KORÉNYI-BOTH, T. SPIRY, A. SCHER, S. TISZA, Acta chir. hung. 14, 320 (1973).

<sup>9</sup> A. G. SANDERS, L. F. DODSON and H. W. FLOREY, Br. J. exp. Path. 35, 331 (1954).

<sup>10</sup> R. H. EBERT, H. W. FLOREY and B. D. PULLINGER, J. path. Bact. 48, 79 (1939).

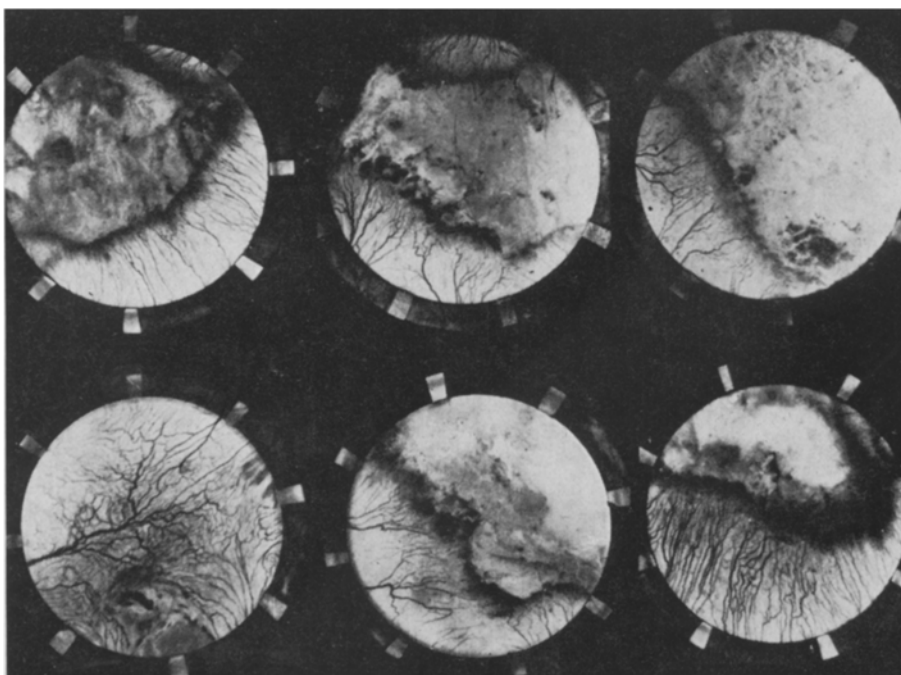


Fig. 1. Photograph of the central table of the ear chambers in rabbits irradiated daily with laser (18th day after implantation of the chambers).

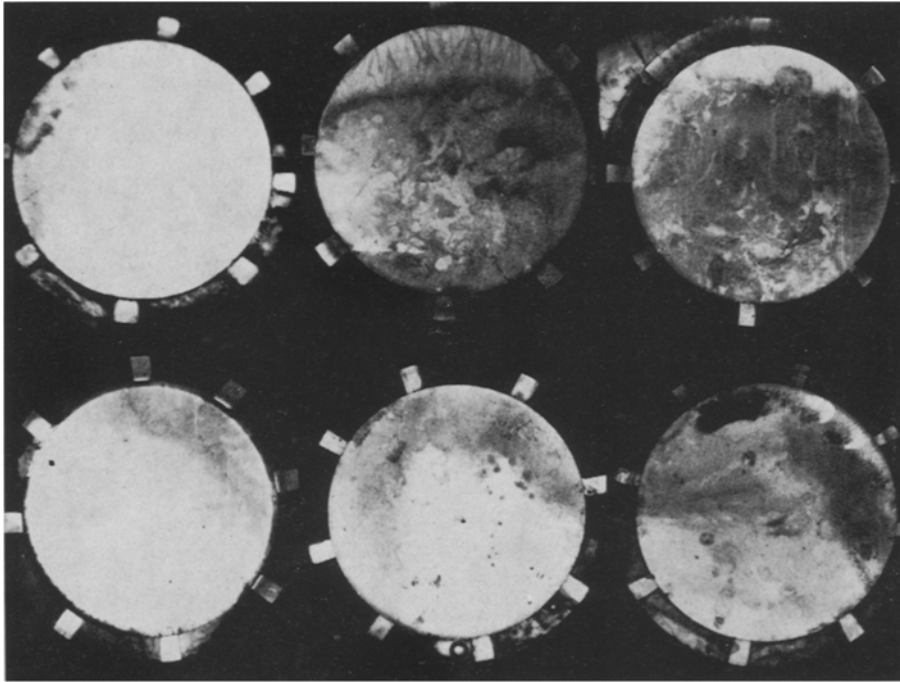


Fig. 2. Photograph of the central table of the ear chambers in the control rabbits 18th day after the implantation.

The endothelium of the new capillaries behind macrophages is still thin and considerable haemorrhage may be observed in front of the growing edge. At the same time, these haemorrhagic edges simplify the identification of vascularized areas on the photograph.

First, the area of the tissue was irradiated where the formation of new vessels had started from. Later we proceeded at the irradiation of the partly vascularized central table. Of the photographs made at various times, we present the pictures taken on the 18th day after the implantation of the chamber, because the difference between vascularization of irradiated and control chambers was the most conspicuous at this time. Figure 1. clearly shows that on the 18th day after operation about one-half (in certain cases two-thirds) of the central table of the chambers irradiated were vascularized. In 1 of the 6 irradiated rabbits vascularization was completed at this time. At the same time, in the control chambers the vascularization was in a very early stage, the first capillaries appeared on the edge of the central table and in one case, one-third of the central table was vascularized (Figure 2). The results are expressed in quantitative form in the Table.

Later on the difference remains, because the arrangement of vessels, the development of arterioles and venules is more rapid in animals receiving laser irradiation.

But, at the same time, the microscopic picture no longer shows so marked a difference from the control animals, because parallel to the progression of the process the number of vessels decreases.

It is difficult, for the time being, to ascertain the mechanism of action by which the laser beam increases the vascularization of the newly formed tissue. Several explanations are feasible. Irradiation may activate the vessels in the region adjacent to the central table, which were injured during the operation (induced fibrinolysis?); it may increase the phagocytic capacity of macrophages, accelerating its activity to clear the way for the advancing vessels; it may loosen the fibrin network of the clot, etc. Considering the small amount of energy used, it is not conceivable that laser irradiation exerts its action by increasing the temperature.

In the case of new tissue formation, the enhancing effect of laser irradiation on vascularization (leading to increase in the number of fibroblasts and thus to increased collagen synthesis) may serve as a basis for the comprehension of its stimulating effect on wound healing. Further investigations are needed to solve this problem.

*Zusammenfassung.* In Ohrkammer Modellexperimenten konnten die Wundheilung wie auch die Gefäßformation mittels Laserstrahlen wesentlich stimuliert werden.

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The effect of repeated laser irradiation on the vascularization of the rabbit ear chamber

Treatment <sup>a</sup>	No. of rabbit	Area vascularized <sup>b</sup> (% ± S.E.)
Control	6	7.7 ± 2.4
Laser-irradiated	6	56.6 ± 9.3

<sup>a</sup> On the 18th day after implantation of the chambers. <sup>b</sup> Total surface of the central table of the chamber was taken to be 100%.

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