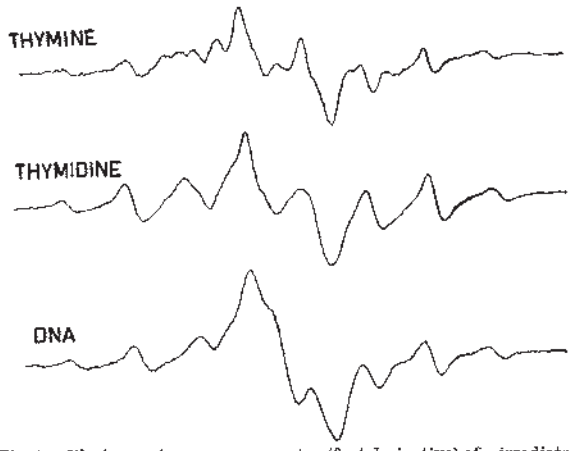

Fig. 1. Electron spin resonance spectra (first derivative) of γ-irradiated, highly purified thymus DNA and the constituents thymine and thymidine. Distance between outermost satellites is about 135 gauss. Doses about 1 Mrad

Fig. 2. Electron spin resonance (—) spectra of γ-irradiated DNA in vacuum and shortly after introduction of air (– – –)

Fig. 1 shows typical electron spin resonance spectra of DNA, thymidine and thymine. A striking similarity between these spectra can be observed. The three outer satellites on each side are present in all three spectra. Also the central regions have features in common. The thymidine spectrum agrees with that published by Gordy[2], disregarding minor details in the centre. The radicals in thymine and thymidine are quite stable, even in air, while the radicals in DNA decay already in vacuum, about 50 per cent remaining after the first 24 h. If the irradiated DNA is exposed to air, most of the radicals disappear leaving a spectrum which can be seen in Fig. 2. The latter type of spectrum is also obtained after irradiation of commercial herring sperm DNA (from L. Light and Co., Ltd.) containing 2–5 per cent protein. The decay *in vacuo* of radiation-induced radicals in the DNA, as well as their reaction with oxygen, seem to be dependent on the water content, but this dependence has not yet been investigated in detail.

In order to gain some knowledge about the radical structure, thymine was treated with heavy water, whereby the dissociable hydrogen was substituted by deuterium. The only spectral change which can be seen is a sharpening of the lines, which may be due to diminishing of the dipolar broadening and/or very weak hyperfine splitting from the exchangeable hydrogens.

The G-value (radicals per 100 eV) for DNA was found to be 0·4 in the dose range 200–600 krad. The order of magnitude agrees with results by A. Müller[10] ($G = 0{\cdot}6$) and P. Alexander *et al.*[4] ($G = 0{\cdot}2$).

Furthermore, electron spin resonance spectra have been recorded for a number of irradiated DNA and RNA constituents, that is, all the pyrimidines and purines and most of their ribose and deoxyribose derivatives, as well as most of the corresponding nucleotides. Of the constituents investigated, only thymine and thymidine (and later also thymidylic acid) gave electron spin resonance spectra similar to that of DNA. Noteworthy is that an irradiated sample of a sodium salt of highly polymeric, pure DNA (from Calbiochem, California) gave the same type of electron spin resonance spectrum as shown for DNA in Fig. 1.

In work on bacteria irradiated by ultra-violet light, Wacker *et al.*[11,12] has shown that the damage of the DNA is associated with a dimerization of thymine. Tokarskaya[13] in a recent publication "gives confirmation to the assumption that thymine is the point of injury after having studied effects of ionizing radiation on dry seeds and isolated DNA".

In view of these results, it is interesting to note that thymine is also the main centre of stabilization of radicals. This is in agreement with data given by Pullman and Pullman[14]. They conclude, after having compared mobile orders for the 4–5 bond in pyrimidines and the 7–8 bond in purines, that "the pyrimidines should thus, from that point of view, be much more reactive towards the fixation of radicals than the purines" and "the site of action of ionizing radiations and the free radicals generated by them on the purine–pyrimidine pairs of DNA is largely located in the pyrimidines".

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A. Ehrenberg
L. Ehrenberg
G. Löfroth

Medicinska Nobelinstitutets biokemiska avdelning, Karolinska Institutet,
and
Strålningsbiologiska Institutionen,
Stockholms Universitet, Stockholm.

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BIOLOGY

Effect of Air Exposure and Occlusion on Experimental Human Skin Wounds

The benefits of special dressings versus air exposure of cutaneous wounds has long been debated. Winter and Scales[1,2] have recently added fresh insight into the problem. In the domestic pig they demonstrated that an occlusive dressing doubles the rate of wound re-epithelization when compared with wounds exposed to the air. In this communication we report parallel studies performed in man.

Our experimental subjects were healthy adult male volunteers. Each subject served as his own control. After intradermal injection of 2 per cent procaine to obtain local anæsthesia and elevate the skin, we horizontally sliced off the epidermis and upper dermis from 0·5 cm^2 marked sites on the inner surface of the arms. We utilized the inner arm, as relatively few hair follicles exist in this area. This simplifies examination of the point of origin of re-epithelization, as the epithelium spreads mainly from the periphery of the wound rather than from the transected hair follicles.

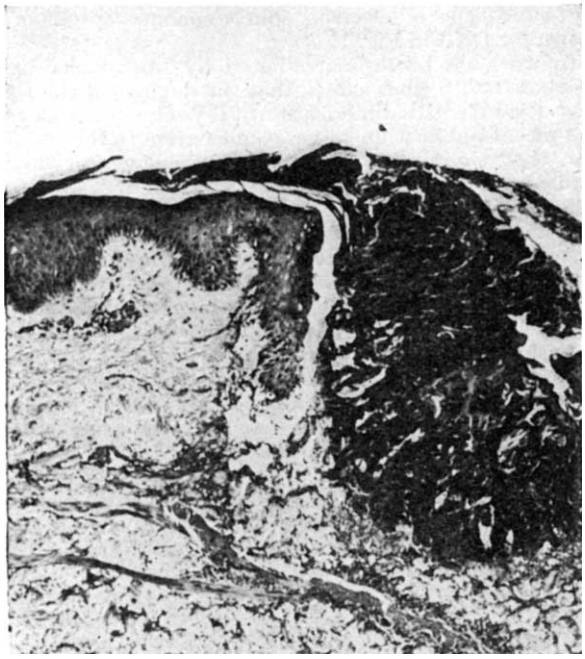

Fig. 1. Air exposed human cutaneous wound (control). Note the angle at which epithelium has grown beneath the eschar

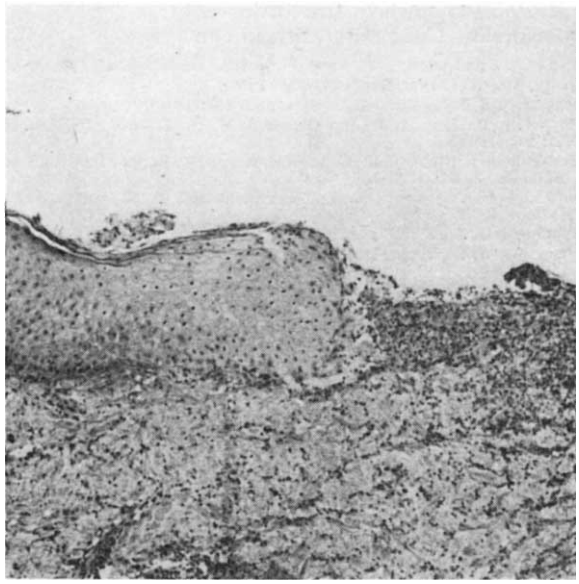

Fig. 2. Occluded experimental human cutaneous wound. Note lack of eschar and straight epithelial growth

After obtaining hæmostasis, we applied 0·1 c.c. of 0·5 per cent neomycin in saline to prevent infection. The control wound was left exposed to the air and the experimental wound occluded with sterile polyethylene film. We excised the wounds with an 8 mm cutaneous punch at three-, five-, seven-, and nine-day intervals respectively. After formalin fixation, the specimens were bisected, paraffin embedded, serially sectioned, and stained with hæmatoxylin and eosin. We examined the slides at 60 times magnification.

With some experience it was possible to determine histologically the site of the original wound as well as the area of re-epithelization. We measured the length of the original wound and that of the new epithelium in every fifth section and totalled for each biopsy. Only growth from the edges was recorded, and that from hair follicles and eccrine sweat gland ducts disregarded.

Table 1. EPITHELIZATION

	Subject	Wound No.		Total length section examined (mm × 10^{-2})	Total new epidermis (mm × 10^{-2})	Per cent new growth
3 days	*A*	51	E*	17,599	2,005	11·4
		52	O*	5,693	1,257	22·0
	B	53	E	22,656	2,450	10·8
		54	O	12,411	5,857	47·4
	C	116	E	8,902	1,424	16·0
		117	O	6,639	1,898	28·5
	D	120	E	7,960	1,183	15·4
		121	O	1,942	1,288	63·9
5 days	*E*		E	3,509	1,014	28·8
			O	3,746	1,606	42·8
	C	118	E	5,187	668	21·1
		119	O	2,551	1,978	78·9
	D		E	2,691	989	36·5
			O	2,730	1,365	50·0
7 days	*F*	105	E			100·0
		106	O			100·0
	G	107	E			100·0
		108	O			100·0
9 days	*F*	103	E			100·0
		104	O			100·0
		109	E			100·0
	G	110	O			100·0

*E, control, air-exposed. *O, occluded.

The results are summarized in Table 1. Most occluded specimens showed so much more epithelization than the air-exposed wounds that the difference was obvious without benefit of measurement. No difference was observed by the seventh day, as both wounds were 100 per cent epithelized at this time. No wound infection occurred.

Histologically, in the air-exposed wounds we noted the epithelium had to grow at right angles to the surface in order to find a plane of cleavage to proliferate under the eschar (Fig. 1). Occluded wounds allowed for no eschar formation, so that the epithelium spread directly across the wound surface (Fig. 2). This alone may explain the difference in rates of epithelization.

We do not know whether these observations will fall in the realm of biological curiosity, or if they will have practical importance in the treatment of cutaneous wounds and burns in man. In the past, infection precluded the practical use of such occlusion, as these dressings provided the moisture necessary for the multiplication of pathogenic organisms. With the introduction of potent anti-bacterial agents for the skin, it may now be practical to take advantage of such an occlusive dressing as was used in this study. Clinical investigation is needed to determine its usefulness in burn and wound therapy.

In summary, experimental split thickness wounds in human volunteers were either air-exposed or occluded with a polyethylene film. Re-epithelization was more rapid in the occluded than in the air-exposed control. This has been verified for wounds of up to duration of five days.

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CAMERON D. HINMAN
HOWARD MAIBACH

Division of Dermatology,
University of California
School of Medicine,
San Francisco.

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Hinman and Maibach have shown that in human skin wounds, as in pig's skin, epidermal regeneration is faster when the wound surface is moist than when exposed to the air and dry. There is little doubt that this is due to the way the scab is formed on an exposed wound surface.