minated for their increased carotene content as well as for their content of other vitamins (*1–5*).

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Deviations from Beer's Law in the Ultraviolet Absorption Spectra of Some Organic Compounds

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In the course of an orientation study of 3-substituted diphenyl ethers, deviations from Beer's law were observed in the absorption spectra of phenols and acetamino compounds derived from diphenyl ether.

Although instrumental deviations occurred at the high-intensity absorption bands, the major part of the deviations resulted from density values below 1.5, which are considered due to fluorescence (*1*). In an effort to trace the chromophore responsible for the deviations, absorption spectra were determined in various concentrations (0.002–0.00001 moles/l) for phenol, nitrobenzene, acetanilide, diphenyl ether, 4-nitrodiphenyl ether, and 3-acetaminodiphenyl ether. The results indicate that some of these substances show deviations from Beer's law in alcoholic solution even at these low concentrations. If instrumental deviations are disregarded, the largest anomalies are observed with phenol (Table 1). The deviations are believed to account at least in part for the discrepancies among the absorption values reported in the literature (λ_{min}, 236–241 mμ; log ε_{min} 1.69–2.0; λ_{max}, 262–274 mμ, log ε_{max} 3.1–3.3) (*2–5*). Acetanilide has λ_{max} 242 mμ (log ε 4.11), and nitrobenzene absorbs maximally at 260 mμ (log ε 3.90), λ_{min} 225 mμ (log ε 3.40). Both compounds show deviations only in high concentrations, and the absorption values agree with the literature (*6*).

TABLE 1

ABSORPTION SPECTRA OF PHENOL IN 95% ALCOHOL

Moles/liter	λ_{max_1}	log ε_{max_1}	λ_{min}	log ε_{min}	λ_{max_2}	log ε_{max_2}
0.00008	219	3.44	No absorption*		275	2.86
.00016	219	3.62	"	"	275	3.08
.00042	220	3.64	238	1.78	275	3.22
.00085	222	3.68	239	1.81	275	3.43
0.00212	225	3.00	240	0.67	272	3.05

* Between 235 and 258 mμ.

Contrary to published data (*7*) diphenyl ether gives absorption curves even in alcohol which show the characteristic fine structure, three sharp maxima at 265, 271, and 278 mμ, and three minima at 252, 267, and 276 mμ. Isosbestic points are observed at 260 and 280 mμ and small deviations from Beer's law in the region of the maxima (log ε_{max_1} 3.18–3.24, log ε_{max_2} 3.26–3.31, log ε_{max_3} 3.21–3.26).

Deviations in 3-acetaminodiphenyl ether (λ_{max} 277 mμ, λ_{min} 276 mμ) occur at the maximum, in 4-nitrodiphenyl ether (λ_{max} 302, λ_{min} 248) only over the region of the minimum.

It appears from the present data that deviations from Beer's law can occur also with diphenyl ethers and phenols in addition to hydrocarbons (*1*), acrylic acids, and esters (*8*), dyes, and other substances (*9*). The increasing number of compounds with concentration-dependent light absorption emphasizes the need for reporting of concentrations, which is unfortunately rarely done in the literature.

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Arginase Activity in Human Skin[1]

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Arginase activity in mammalian skin has been demonstrated by Greenstein (*1*), who found that the normal skin of mice contained about 1/10 the amount found in the liver, and roughly ½ that found in pigmented melanomata of the same animals.

In the present study it was attempted (1) to ascertain the presence of arginase in human skin, and (2) to determine its variations and distribution in normal skin and in cutaneous lesions.

Skin samples from fresh biopsies were crushed in a piston-cylinder apparatus, suspended in normal saline, and then incubated with manganous ion to activate the enzyme (*2*). Incubation was then carried out with arginine as a substrate for ½ hr, and the reaction stopped by the addition of sulfuric acid (*3*). The

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[2] The author is indebted to Stephen Rothman for valuable advice in carrying out the experiments and in preparing this manuscript.

TABLE 1

ARGINASE ACTIVITY OF NORMAL WHOLE SKIN FROM SURGICAL SPECIMENS OF FEMALE BREASTS

Specimen	Arginase activity
1	316
2	490
3	238
4	282
5	562
6	202
7	372
8	277
Average	355

initial and final urea values were determined in the Warburg respirometer by the urease method, first described by Krebs and Henseleit (*4*). The enzyme activity is expressed in mg of urea-N liberated/100 g of wet weight tissue.

Estimation of arginase activity in skin from female breasts, which was either worked up right after it was received from the operating room, or maintained in a frozen state until worked up later, revealed that human skin has a considerable amount of arginase activity (Table 1).

Estimation of the enzyme's activity in isolated epidermis and corium, separated either roughly by mechanical means or quite accurately by the heat method (*5*), showed that most of the enzyme (about 90%) is found in the epidermis (Table 2). In using the heat method of separation the skin loses about 50% of its arginase activity. This loss is corrected for in Tables 3 and 4.

Common warts, which are characterized by considerable proliferation of the prickle cells of the Malpighian layer, showed an arginase activity much higher than normal epidermis. The values found in the warts studied would indicate that the Malpighian cells in the wart have an average eightfold higher arginase content than normal Malpighian cells (Table 3). In patches of psoriasis, also associated with proliferation of the Malpighian layer, high values were obtained, though not as high as those found in common warts.

As contrasted with common warts, the main histologic feature of so-called seborrheic warts, or seborrheic keratoses, is not a proliferation of the prickle cells of the Malpighian layer but a benign proliferation of the basal cells (*6*). In these lesions, as well as in basal cell carcinomas, no such unusually high arginase values were encountered as in common warts.

TABLE 2

ARGINASE ACTIVITY OF EPIDERMIS AND CORIUM (HEAT-SEPARATED) OF NORMAL SKIN FROM FEMALE BREASTS

Epidermis	Corium	Activity ratio of corium/epidermis
1,515	146	1: 10.5
1,672	148	1: 11.5
1,450	178	1: 8.2

On the contrary, in seborrheic warts and in basal cell carcinomas the values were conspicuously low (Table 4).

These results indicate that the arginase activity of the skin is located predominantly in the Malpighian prickle cells of the epidermis and that the basal cells contain much less of the enzyme—if any.

It is generally considered that the prickle cells continuously derive from the basal cells. This never has been clearly demonstrated, however. The two layers become quite distinct early in fetal life; in the human embryo, for example, they are differentiated in the tenth week. The great difference in enzymatic activity suggests that possibly the two layers, after their differentiation, are independent of each other in their perpetual renewal.

TABLE 3

ARGINASE ACTIVITY OF COMMON WARTS

Activity of wart	Activity ratio of normal whole skin/wart	Activity ratio of normal epidermis/wart
16,900	1: 48	1: 5.5
34,400	1: 95	1: 11.0
6,030	1: 17	1: 2.0
43,500	1: 121	1: 14.0
Average 25,207	1: 71	1: 8.0

TABLE 4

ARGINASE ACTIVITY OF MISCELLANEOUS LESIONS

Lesion	Arginase activity	Activity ratio of normal epidermis/lesion	Activity ratio of lesion/wart
Seborrheic keratosis	685	1: 0.22	1: 37
" "	1,223	1: 0.40	1: 20
Basal cell carcinoma	2,370	1: 0.77	1: 10.7
" " "	680	1: 0.22	1: 37
Basal-intermediate cell carcinoma	4,700	1: 1.4	1: 5.4

It has been suggested that urea is formed in the sweat glands by the action of arginase on arginine (*7, 8*). Such mechanism would explain the well-known fact that the urea concentration of sweat is always higher than that of blood. However, with the method employed in this study no arginase activity could be demonstrated in fresh human sweat produced by exposure to heat.

A more detailed report of this work will be published elsewhere.

Addendum: Following the submission of this report for publication the author's attention was called to the work of Mardashev and Semina, which was reported in the Russian literature (*Biokhimiya,* **13**, 236 [1948], quoted by *Chem. Abs.,* **42**, 7806g [1948]). According to the abstract these workers have also demonstrated the presence of arginase in human skin and similarly have noted its concentration in the epidermis.

Recent work in this laboratory indicates that within the epidermis proper the greatest arginase activity is localized in the cells of the upper portions of the Malpighian layer and seems to reach its maximum in the granular layer.

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A Method of Intravenous Injection of Drugs from a Distance in Conditional Reflex Studies

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The study of higher nervous activity through the medium of conditional reflexes usually requires that the animal be isolated from both the experimenter and all irrelevant stimuli. Procedures requiring a series of injections of measured amounts of a drug have not been possible because of the lack of a satisfactory technique. This deficiency had to be overcome before we could investigate cardiac, respiratory, and salivary conditional reflexes in which acetylcholine is used as the unconditional stimulus. In the studies planned, repeated intravenous injections of the drug are necessary, but it is essential to avoid any disturbance of the animal during the course of the injections. The technique described below permits the intravenous injection of measured quantities of a drug while the investigator is separated from the animal, thus avoiding the disturbing effect of the investigator's presence, repeated introduction of the needle, etc.

The authors (*1*) recently described a method for injecting acetylcholine into the superior sagittal cerebral venous sinus, in the isolated, unanesthetized dog. This technique has been modified by the use of plastic tubing inserted into the external jugular or radial vein, as described here. Fitzpatrick, Schnabel, and Peterson (*2*) used plastic tubing for cardiac catheterization, by introducing the tubing into the heart through the external jugular vein. In our experiments a very fine-bore plastic tube about 3 in. long with a steel wire stylet already in place is inserted into the external jugular vein of an anesthetized dog. This is accomplished by first introducing a special 16-gauge needle into the vein and then passing the plastic tubing through the needle. The needle is pulled out over the

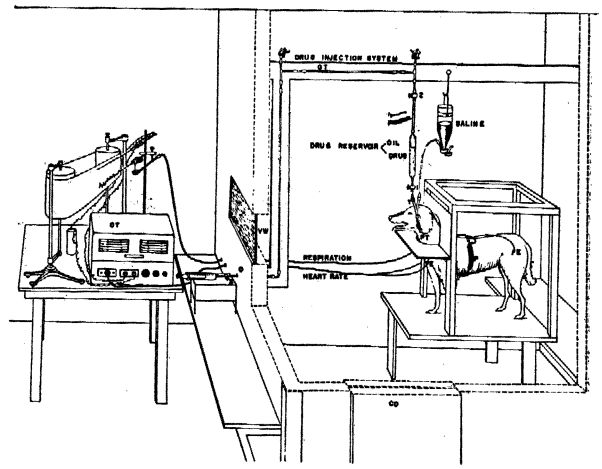

FIG. 1. Arrangement of injection apparatus and dog during experiment: *E*, electrodes for heart rate; *P*, pneumograph; *PT*, plastic (polyethylene) tubing; *T*, Y-tube; *GT*, oil-filled glass tubing; *CT*, cardiotachometer; *CD*, camera door; *O*, outlet for oil-filled tubing; *VW*, viewing window—one-way vision from outside camera; 1, 2, and 3, stopcocks.

tubing, which may be left in the vein for several days. The protruding end of the stylet is then bent at its point of exit so as to prevent accidental movement inward, lest it extend beyond the inner end of the tubing and injure the vein. The bent end of the stylet is sutured to the skin. On the following day, after the dog has recovered completely from the anesthesia, it is placed in the camera under slight restraint and with the injection apparatus applied as illustrated in Fig. 1. A 23-gauge needle with shortened bevel is inserted into the plastic tube (Fig. 1, *PT*) after removal of the stylet, and the needle is fixed to the neck by means of adhesive tape. A metal Y-tube (Fig. 1, *T*), with adapters at the end of the stem and on the arms, is attached to the needle. One arm is connected to a saline solution, and the other to a drug reservoir, which is a glass tube 6 in. long and ½ in. in diameter, tapered at both ends to permit the attachment of

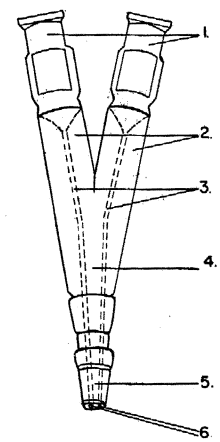

FIG. 2. Y-tube showing details of construction: 1, adapters of arms; 2, arms of Y-tube; 3, inner tubes; 4, stem of Y-tube; 5, adapter; and 6, open ends of inner tubes.