

## Activation Analysis of Soluble and Fixed Sodium in Mammalian Hair

**Abstract.** *Antelope hair was soaked in sodium-22 to simulate external contamination and then systematically washed to remove all traces of the sodium; progress of the wash was followed by gamma-ray spectrometry. Hair was then activated by neutron bombardment which showed sodium still present as sodium-24. It is concluded that a fraction of sodium in hair can be readily washed away with water and that a second fraction is held in the hair in such a manner that extended washing does not remove it. This suggests that sodium in two states may be associated with hair, one as an external contaminant and the other as a more nearly integral part of the hair.*

Whether trace elements occur in hair and other integumentary derivatives as accidental contamination or as integral parts of the structure of the tissue, or in combinations of the two, still requires definition.

Some investigators (1) suggest that they are present primarily as contaminants, having been absorbed on the hair during contact with materials in the environment, as might result from work with minerals in mines or factories or with trace elements present in perspiration, hair-washing preparations, and so forth. Such trace elements would not, of course, be part of the tissue structure and would be present or not according to the vagaries of environment. Other investigations (2, 3) indicate that certain patterns of trace elements are characteristic of the hair of a given species and, within limits, relatively independent of environment. My experiment was designed to examine more closely the nature of the association between sodium and the hair of the antelope (*Antilocapra americana*).

In order to simulate contamination from the environment, samples of hair were soaked in a solution of radioactive  $^{22}\text{Na}$  ( $1\ \mu\text{C}/\text{ml}$ ) for 10 days. The hair was then washed, starting with distilled water only, in a refluxing water bath with the siphon arranged in such a way that the amount of water used, as well as the length of a washing cycle, could be measured. Results of the  $^{22}\text{Na}$  washing procedure are shown in Fig. 1 which gives the stepwise reduction as the washing progresses. No detectable  $^{22}\text{Na}$  remained after approxi-

mately 16 washing cycles, in which a total of 240 ml of water was used. Washing was allowed to continue for a total of more than 340 cycles (longer than 28 hours) to allow thorough removal of  $^{22}\text{Na}$ . No values above background were recorded throughout this

period (16 to 340 cycles) of washing.

The washed hairs were then activated for 8 hours in a neutron flux of  $1.3 \times 10^{13}$  neutrons  $\text{cm}^{-2} \text{sec}^{-1}$  in the materials testing reactor at the National Reactor Testing Station, Idaho Falls. Activities were recorded and

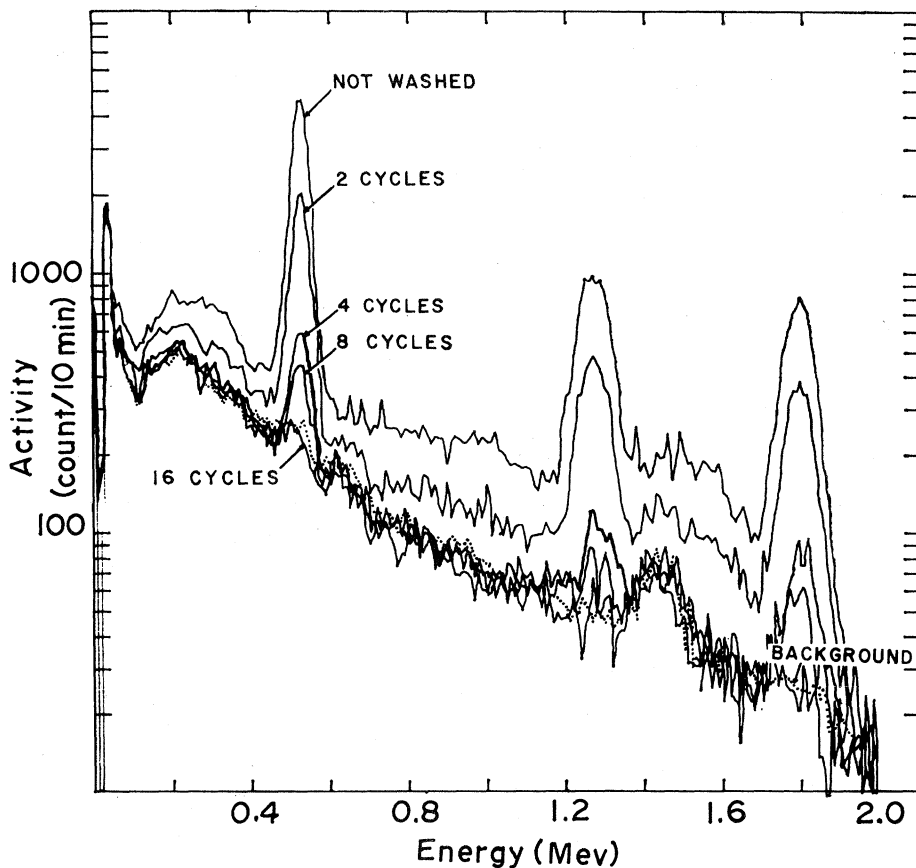


Fig. 1. Gamma-ray spectra of  $^{22}\text{Na}$  showing its removal from antelope hair during washing with water. Each cycle consisted of 15 ml of newly condensed water vapor.

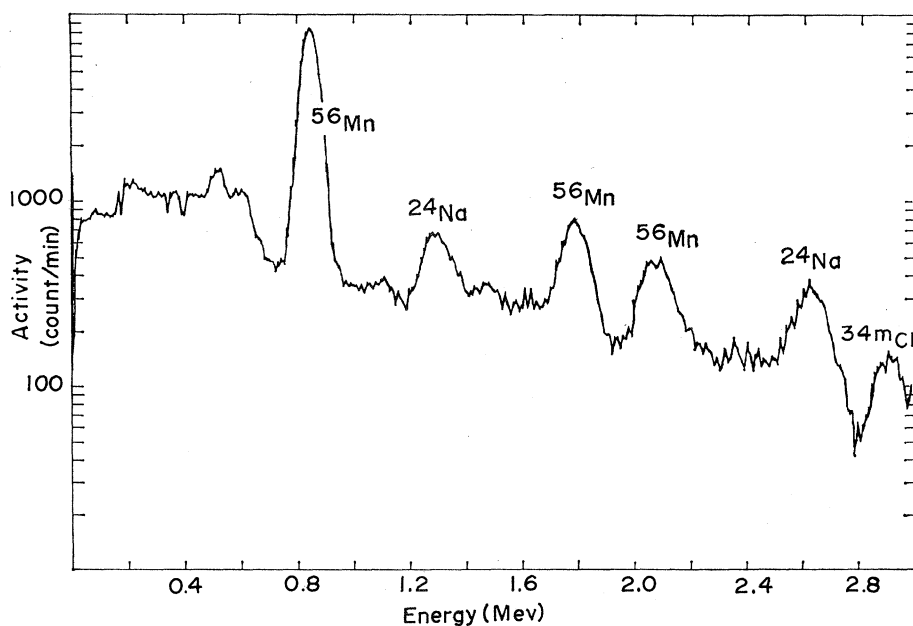


Fig. 2. Gamma-ray spectrum of  $^{24}\text{Na}$  present in hair after extended washing in distilled water.

analyzed by means of a 400-channel pulse-height analyzer (Technical Measurements Corp., model 401C) coupled to a NaI(Tl) scintillation crystal (7.5 by 7.5 cm) and photomultiplier. Crystal and photomultiplier were mounted in a steel cave with walls 27.5 cm thick.

The resulting spectra, shown in Fig. 2, demonstrate that sodium, as activated  $^{24}\text{Na}$ , was present in the washed hair. Parallel tests on hair that had not been treated with  $^{22}\text{Na}$ , but had been subjected to similar washing procedures, showed that approximately 40 percent of the original sodium, in terms of counts per minute, remain in the hair even after the extended period of washing. These findings suggest that part of the sodium associated with hair is a loosely held contaminant which can be washed out. The remaining more tightly held fraction does not wash out with water alone. Furthermore, the removable fraction comes out with relative ease and quickness, while the fixed fraction remains at about the same levels during extended washing, which suggests that there is a qualitative difference involved in the association rather than a simple quantitative difference dependent on, perhaps, depth of the sodium molecule within the hair matrix.

Although the details are beyond the scope of this report, it is pertinent to point out that certain other trace elements behaved in a manner similar to that of sodium, while still others did not. For example, bromine and nickel were completely removed by the washing procedure described above; manganese, copper, and zinc were not, which suggests that, among other factors, ionic radius and charge may be involved in the control of the distribution of trace elements in hair. These patterns are the subject of continuing studies that will be reported more fully at a later date (4).

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#### References and Notes

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## Volatile Principle from Oak Leaves: Role in Sex Life of the Polyphemus Moth

**Abstract.** *An emanation from oak leaves is necessary for the mating of polyphemus moths under laboratory conditions. This requirement can be satisfied by placing the moths in the presence of oak leaves or aqueous or alcoholic extracts prepared from oak leaves. The active principle is a volatile, heat-stable, polar material which has been partially purified. The oak emanation acts on the female and not on the male, and the sensory receptors are located on the female antennae. The reception of the oak emanation is prerequisite for the female's release of her sex pheromone, which in turn, is necessary for the sexual activation of the male.*

We have long been puzzled by the sex life of the polyphemus moth (*Antheraea polyphemus*). For example, when caged out-of-doors during the proper season, virgin females have routinely attracted the male polyphemus from afar. Yet under laboratory conditions we have (until recently) failed to obtain a single mating among hundreds of these moths when the sexes were placed together in small cages, or in large cages, or even in the Harvard gymnasium (1). The solution of this paradox proves to be far from trivial.

A pair of polyphemus moths was placed overnight in a laboratory cage containing red oak seedlings (*Quercus rubra*) (2). The next morning the pair was mating. But so also was another pair of polyphemus in a nearby cage not containing any plant material! This was the first indication that oak—a favorite food plant of the polyphemus silkworm—produces an emanation prerequisite for the sexual activation of these moths.

To explore the phenomenon in further detail, we standardized the biological assay in the following manner. Each evening, one female and two males, or two females and three males, were placed together in a cage and stored overnight in a darkened laboratory ventilated by a ceiling fan. If mating occurred, the moths ordinarily remained paired until the following evening. If mating did not occur, the sexes were removed to separate cages and stored apart during the day. Unmated females were tested on two successive nights; males on three successive nights. All unmated individuals

were then placed together in one large cage apart from oak leaves.

In control experiments (Table 1), no matings occurred when 30 normal females were tested in the absence of oak leaves. Likewise, none of the older moths mated in the large cage, despite the high density of the population and the frequent collisions between the sexes. By contrast, 12 (33 percent) of the females mated in cages containing oak leaves; 10 (29 percent) mated when oak leaves were only in the vicinity of the cage.

The experiment was repeated with maple, birch, chestnut, horse chestnut, elm, hickory, and beech leaves—all of which are known to be acceptable food plants of polyphemus silkworms. No matings occurred among the 24 females subjected to this treatment.

Does the oak emanation act on the male or the female moth or on both? Answer to this question was sought by separately exposing males and females to oak leaves for specific periods. Then, each of the moths thus treated was caged in separate rooms with untreated moths of the opposite sex in the absence of oak leaves.

The results of these experiments showed that the oak emanation acts solely on the female. In no case did prior exposure of the male provoke mating (Table 1). By contrast this preliminary treatment of females for 4 to 6 hours led to subsequent successful mating in a substantial number of cases. In additional experiments not recorded in Table 1, we found that this preliminary treatment of the females was effective only when the treated females were promptly placed with males at the proper time of day. Thus, a delay of only 30 minutes negated the effects of 24 hours of treatment.

Table 1. Effect of oak leaves on the mating of polyphemus moths.

Treatment	Females assayed (No.)	Mating (%)
Control (no leaves)	30	0
Red oak, in cage	36	33
Red oak, in vicinity	35	29
Prior exposure of males to oak leaves for:		
0.5 hr	2*	0
4 hr	4*	0
6 hr	2*	0
Prior exposure of females to oak leaves for:		
0.5-1 hr	4	0
2-3 hr	5	0
4-5 hr	6	33
6 hr	4	50

\*Number of males tested, with equal numbers of females.