EVALUATION OF THE HEALTH ASPECTS OF
CANDELILLA WAX AS A FOOD INGREDIENT

1981

Prepared for

Bureau of Foods
Food and Drug Administration
Department of Health and Human Services
Washington, D.C.

Contract No. FDA 223-78-2100
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Life Sciences Research Office
Federation of American Societies for Experimental Biology
9650 Rockville Pike
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NOTICE

This report, one of a series concerning the health aspects of using the Generally Recognized as Safe (GRAS) or prior-sanctioned food substances as food ingredients is being made by the Federation of American Societies for Experimental Biology (FASEB) under contract no. 223-78-2100 with the Food and Drug Administration (FDA), U.S. Department of Health and Human Services. The Federation recognizes that the safety of GRAS substances is of national significance, and that its resources are particularly suited to marshaling the opinions of knowledgeable scientists to assist in these evaluations. The Life Sciences Research Office (LSRO), established by FASEB in 1962 to make scientific assessments in the biomedical sciences, is conducting these studies.

Qualified scientists were selected as consultants to review and evaluate the available information on each of the GRAS substances. These scientists, designated the Select Committee on GRAS Substances, were chosen for their experience and judgment with due consideration for balance and breadth in the appropriate professional disciplines. The Select Committee's evaluations are being made independently of FDA or any other group, governmental or nongovernmental. The Select Committee accepts responsibility for the content of each report. Members of the Select Committee who have contributed to this report are named in Section VII.

Tentative reports are made available to the public for review in the office of the Dockets Management Branch, Food and Drug Administration, after announcement in the Federal Register, and opportunity is provided for any interested person to appear before the Select Committee at a public hearing to make oral presentation of data, information, and views on the substances covered by the report. The data, information, and views presented at the hearing are considered by the Select Committee in reaching its final conclusions. Reports are approved by the Select Committee and the Director of LSRO, and subsequently reviewed and approved by the LSRO Advisory Committee (which consists of representatives of each constituent society of FASEB) under authority delegated by the Executive Committee of the Federation Board. Upon completion of these review procedures the reports are approved and transmitted to FDA by the Executive Director of FASEB.

While this is a report of the Federation of American Societies for Experimental Biology, it does not necessarily reflect the opinion of all of the individual members of its constituent societies.

[Signature]
Kenneth D. Fisher, Ph.D., Director
Life Sciences Research Office
FASEB

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I. INTRODUCTION

This report concerns the health aspects of using candelilla wax as a food ingredient. It has been based partly on the information contained in a scientific literature review (monograph) furnished by FDA (Rogers, 1978), which summarizes the world's scientific literature from 1920 through 1978. To ensure completeness and currency as of the date of this report this information has been supplemented by searches of over 30 scientific and statistical reference sources and compendia that are generally available; use of new, relevant books and reviews and the literature citations contained in them; consideration of current literature citations obtained through computer retrieval systems of the National Library of Medicine; searches for relevant data in the files of FDA; and by the combined knowledge and experience of members of the Select Committee and the LSRO staff. In addition, an announcement was made in the Federal Register of November 7, 1980 (45 FR 74056-74060) that opportunity would be provided for any interested person to appear before the Select Committee at a public hearing to make oral presentation of data, information, and views on the health aspects of using candelilla wax as a food ingredient. The Select Committee received no request.

As indicated in the Food, Drug, and Cosmetic Act [21 USC 321(s)], GRAS substances are exempt from the premarketing clearance that is required for food additives. It is stated in the Act and in the Code of Federal Regulations (Office of the Federal Register, 1980) [21 CFR 170.3 and 170.30] that GRAS means general recognition of safety by experts qualified by scientific training and experience to evaluate the safety of substances on the basis of scientific data derived from published literature. These sections of the Code also indicate that expert judgment is to be based on the evaluation of results of credible toxicological testing or, for those substances used in food prior to January 1, 1958, on a reasoned judgment founded in experience with common food use, and is to take into account reasonably anticipated patterns of consumption, cumulative effects in the diet, and safety factors appropriate for the utilization of animal experimentation data. FDA recognizes further [21 CFR 170.30] that it is impossible to provide assurance that any substance is absolutely safe for human consumption.

The Select Committee on GRAS Substances of LSRO reviewed and evaluated the available information on candelilla wax in full recognition of foregoing provisions. In reaching its conclusions on safety, the Committee, in accordance with FDA's guidelines, relied primarily on the absence of substantive evidence of, or reasonable grounds to suspect, a significant risk to the public health. This report is intended for use of FDA in determining the future status of these substances under the Federal Food, Drug, and Cosmetic Act. The Committee anticipates that its conclusions will be reviewed as new information becomes available.

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II. BACKGROUND INFORMATION

Candelilla wax is obtained from several species of Euphorbiaceae, rushlike perennial plants which grow in the semi-arid regions of northern Mexico and southern Texas, and to a limited extent in Arizona and southern California. The plants consist of numerous slender, leafless, cylindrical stalks 2-5 feet high, covered with a powdery wax which gives the plant a blue-green color (Hackett, 1969; Rogers, 1978).

The wax is extracted from the plants by immersing the dried stalks in boiling water acidified with sulfuric acid; the molten product is skimmed off as it rises to the surface. The wax is transferred to lead-lined tanks, kept at the boiling point to remove water, and then allowed to settle to remove any dirt present. The yield of clarified wax is from 1.5-3.0% of the air-dried plant. The sole criterion of purity is usually the color of the product (Hackett, 1969; Rogers, 1978) which varies from light yellow to brown. The wax is hard, brittle, lustrous, and slightly sticky (Hackett, 1969). The physical and chemical properties of the wax vary with the age of the plant. The wax is insoluble in water, but soluble in acetone, chloroform, benzene, and other organic solvents (The Merck Index, 1976). To meet food-grade specifications, candelilla wax must melt between 68.5°C and 72.5°C, have an acid value between 12 and 22, and a saponification value of 43 to 65. It must contain not more than 3 parts per million (ppm) of arsenic or lead, and not more than 40 ppm of heavy metals expressed as lead (National Research Council, 1972). The approximate composition of commercially refined candelilla wax is shown in Table 1.

Table 1. Composition of Candelilla Wax*

<table>
<thead>
<tr>
<th>Component</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydrocarbons</td>
<td>50-51</td>
</tr>
<tr>
<td>Esters</td>
<td>28-29</td>
</tr>
<tr>
<td>Free alcohols, sterols, resins</td>
<td>12-14</td>
</tr>
<tr>
<td>Free acids</td>
<td>7-9</td>
</tr>
<tr>
<td>Minerals</td>
<td>0.7</td>
</tr>
<tr>
<td>Volatiles</td>
<td>0.5-1.0</td>
</tr>
</tbody>
</table>

* Adapted from Warth (1956).

The characteristic features of candelilla wax are an exceptionally high content of hydrocarbons and a relatively small amount of volatile esters. This is evident in Table 2, in which
Table 2. Percent Volatiles of Some Natural Waxes (Tulloch, 1975)

<table>
<thead>
<tr>
<th>Component</th>
<th>Candelilla</th>
<th>Beeswax</th>
<th>Carnauba</th>
<th>Ouricuri</th>
<th>Esparto</th>
<th>Sugarcane</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydrocarbon</td>
<td>41</td>
<td>15</td>
<td>-</td>
<td>1</td>
<td>19</td>
<td>2</td>
</tr>
<tr>
<td>Esters</td>
<td>6</td>
<td>36</td>
<td>36</td>
<td>11</td>
<td>9</td>
<td>6</td>
</tr>
<tr>
<td>Free alcohols</td>
<td>4</td>
<td>-</td>
<td>11</td>
<td>5</td>
<td>8</td>
<td>4</td>
</tr>
<tr>
<td>Free acids</td>
<td>8</td>
<td>12</td>
<td>-</td>
<td>4</td>
<td>9</td>
<td>3</td>
</tr>
<tr>
<td>Hydroxy esters</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>7</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Triterpenes</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>6</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Unidentified</td>
<td>6</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Total volatiles   | 65         | 63      | 47       | 34       | 45      | 15        |
the volatile constituents of various natural waxes are compared (Tullock, 1975).

The waxes were analyzed by gas-liquid chromatography, which determined only compounds volatile between 125°-400°C. About one-third (34%) of candelilla wax was nonvolatile in this temperature range. The composition of this portion has not been completely identified but is believed to include esters of terpenes (Tullock, 1975).

Stransky et al. (1976) reported in a detailed analysis of the hydrocarbon fraction that \( n \)-alkanes constituted 98% and \( 1 \)-alkenes the other 2%. Each \( n \)-alkane between triacosane (C\(_{23}\)) and tetratriacontane (C\(_{34}\)) was detected. By far the most abundant hydrocarbon was hentriacontane (C\(_{31}\)), which constituted over 80% of total \( n \)-alkanes. Tritriacontane (C\(_{33}\)) was the next most abundant, representing approximately 10% of the alkane fraction. Among the unsaturated hydrocarbons, each member of the \( 1 \)-alkene homologous series from C\(_{16}\) through C\(_{34}\) was detected, but over half of the total was present as octacosene (C\(_{28}\)) and triacontene (C\(_{30}\)). Stransky et al. (1976) also demonstrated that the triterpenic alcohol epi-lupeol was present and constituted about 5% of the purified candelilla wax.

Most of the hydrocarbons identified in candelilla wax are natural constituents found ubiquitously in the cuticles of fruit and vegetables. Each of the alkanes (C\(_{27}\)-C\(_{33}\)) reported by Stransky et al. (1976) to be present in more than trace amounts in candelilla wax has been found in significant concentrations by Waldron et al. (1961) in other plant waxes as well: apple peel, brussels sprouts, cocksfoot grass, rye grass, and the leaves of turnip, runner bean, white mustard, green tobacco, and cactus. The most abundant alkane in candelilla wax, hentriacontane (C\(_{31}\)) has been detected in the cuticles of pears, apples, cherries, grapes, oranges, and grapefruit (van Straten, 1977), as well as in the leaves of various vegetables (Waldron et al., 1961). Nonacosane (C\(_{29}\)), another of the hydrocarbons in candelilla wax, comprises 99% of the paraffins in the apple cuticle (Mazliak, 1960) and is one of the most abundant hydrocarbons of all plant waxes thus far analyzed (Waldron et al., 1961).

Candelilla wax is used as a constituent of various polishes, candles, varnishes, sealing waxes, electrical insulations, carbon papers, cosmetics, and other wax-containing preparations (The Condensed Chemical Dictionary, 1977). Its chief food use in the United States is as a component of chewing gum base, and as a glaze for hard candies. It was accorded unpublished GRAS status (Buckley, 1966, 1967; Cassidy, 1961; Checchi, 1959; Orr, 1968) as a chewing gum constituent, as well as a component of food-packaging materials (Orr, 1968; Randolph, 1963). It has been deemed GRAS by the Flavor and Extract Manufacturers' Association as a flavor ingredient (Oser and Ford, 1977). It has been cleared as a regulated additive in adhesives [21 CFR 175.105] (Office of the
Federal Register, 1980); as an adjuvant in resinous and polymeric coatings for polyolefin films contacting food surfaces [21 CFR 175.320]; and as a component of paper and paperboard in contact with dry food [21 CFR 176.180]. It has also been used successfully in Mexico as a coating agent to extend the storage life of citrus fruits (Lakshminarayana et al., 1974). The Select Committee has no information on such a use in the United States.
III. CONSUMER EXPOSURE DATA

The Subcommittee on Review of the GRAS List—Phase II (1972) of the National Research Council (1972) surveyed manufacturers to determine the use of candelilla wax in foods. Six companies reported using this substance in 1970 in hard candies or in chewing gum. The weighted mean for the usual level of addition was 260 mg/kg in the candies and 200 mg/kg in chewing gum. The Expert Panel of the Flavor and Extract Manufacturers' Association (Ozer and Ford, 1977) also suggested the following maximum levels (in mg/kg) for candelilla wax in various foods: beverages (alcoholic and nonalcoholic) 30, frozen desserts 10, and confectionery items 15. There was no evidence from the NRC survey that candelilla wax had, in fact, been added to these foods. The total quantities reported by food processors in 1970 and 1975 (Committee on GRAS List Survey, 1978) are shown in Table 3.

Table 3. Use of Candelilla Wax by Food Processors

<table>
<thead>
<tr>
<th></th>
<th>Total, kg</th>
<th>Per capita daily use, mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1970</td>
<td>650,000</td>
<td>8.7*</td>
</tr>
<tr>
<td>1975</td>
<td>790,000</td>
<td>10.1†</td>
</tr>
</tbody>
</table>

*Estimated U.S. population in 1970, 205,000,000.
†Estimated U.S. population in 1975, 215,000,000.

Since the major food use of candelilla wax is as a constituent of chewing gum base, the average consumption by children would probably be higher than the mean shown in Table 3, which has been estimated for all age groups. Candelilla wax represents about 5% of chewing gum base, which in turn constitutes about 20-25% of the total weight of the stick (Segal, 1979). A package of chewing gum (five sticks) weighs about 15 grams and would thus contain approximately 150-200 mg candelilla wax. The wax is insoluble in water so that little would be extracted in the saliva. However, an unknown amount might be swallowed along with the rest of the gum base. The presence of candelilla wax in certain food packaging materials, e.g., cellophane, represents another possible source of ingestion. The amounts which may migrate into foods from these materials cannot be estimated, but are believed to be very small.

Candelilla wax is also present at levels of 12-15% in some lipsticks (Murphy and Lieberman, 1976). It has been estimated that 50-60 mg of lipstick may be ingested daily by users (The Toilet Goods Association, 1959), permitting a possible intake of 5-10 mg/d candelilla wax from this source.
IV. BIOLOGICAL STUDIES

No publications on the digestion, absorption, or toxicity of candelilla wax were found by the Select Committee. The insolvability of waxes in water and their hydrophobic nature suggest that little digestion or absorption of candelilla wax is likely. Lakshminarayana et al. (1974) have stated that the wax is nontoxic, but they offer no supporting documentation. However, the National Association of Chewing Gum Manufacturers provided the Select Committee with several unpublished reports of animal studies in which various gum bases containing candelilla wax were fed or administered by other routes for varying periods of time.

Short-term studies

A mixture of candelilla wax and a gum base (Type II GRS Rubber) was fed for 8 wk to weanling Wistar strain rats at dietary levels of 3% and 5% (Harrisson, 1946). The precise composition of the mixture was not given, but in other gum mixtures, candelilla wax has been reported to constitute 25% or 50% of the final product. The investigators estimated that the daily consumption of the mixture by rats receiving 5% in the diet was 1.25 g/kg body wt at the beginning of the experiment and 0.67 g/kg at its conclusion. The corresponding consumptions at the 3% dietary level were 0.84 and 0.38 g/kg/d, respectively. Presumably, the daily consumption of candelilla wax was one-quarter or one-half of this amount. There was no significant difference from controls in weight gain, food or fluid intake, urinalysis, blood cytology, or gross pathology. Microscopic tissue examinations were not reported.

A similar 8-wk test was conducted by the same laboratory (Harrisson, 1948) utilizing a different admixing vehicle. Weanling rats (Wistar strain) were fed a stock diet containing approximately 1% or 5% of a 1:1 mixture of candelilla wax and Heveatex Polymer N-1017® (butadiene and styrene). The concentrations of the mixture were adjusted periodically to approximate a daily intake of 1 or 5 g/kg body wt of the mixture in the respective diets. The actual intakes fell somewhat short of these objectives; rats receiving the 1% mixture actually consumed approximately 0.75 g/kg/d and those on the 5% mixture, about 3.6 g/kg/d. The daily intakes of candelilla wax were one-half of these amounts. Weight gains, blood cytologies, urinalyses, and organ appearances were all within normal ranges.

In another experiment, this 1:1 mixture was fed for 6 mo at levels of 1% and 5% of the diet (Harrisson, 1949) to 34 rats in each group, equally divided by sex. The estimated intakes of the candelilla wax component of the mixture were 0.65 and 3.5 g/kg body wt/d, respectively. Survivals, urinalyses, blood cytologies, and pathologic findings did not differ significantly from the
control groups. A slight retardation of growth was noted in both
groups of male rats; the effect was more marked among those
receiving the 5% mixture than those on the 1%. At necropsy, four
rats receiving the 5% level of gum base mixture had "somewhat
small" testes and seminal vesicles, one had "spots" on the uterus,
one a small renal retention cyst, and one a slight edematous area
around the salivary gland. No other changes were reported.

More than a score of unidentified gum base formulations
containing candelilla wax were fed to weanling rats for 6 mo.
Each preparation was tested on 12 male and 12 female rats (strain
not stated). The candelilla wax in these preparations represented
4.1-6.1% of the diet for an approximate daily consumption of 2.4
g/kg body wt. All rats survived the experimental period. Food
intakes and weight gains were comparable with control animals
(Hodge, 1973). Urinalyses revealed no significant differences
between test and control animals. One test rat had a lymphoma in
the submucosa of the colon; one experimental and one control rat
exhibited renal calcifications; three test rats (2 males, 1 female)
showed minimal infiltration with mononuclear cells of periportal
spaces in the liver, but the parenchyma appeared normal (Davidsohn,

Groups of four young dogs were maintained for 6 mo on
diets containing either 1% or 10% of two different gum bases. Six
other dogs were kept on the basal diet as controls. The other
constituents of the gums were not revealed, but each base con-
tained 25% candelilla wax. It is estimated that the dogs consumed
about 60 and 600 mg/kg body wt/d of candelilla wax, respectively,
on these diets. Growth, blood and urine indices, and gross and
histopathological findings after 6 mo were within normal limits
(Harrisson, 1953).

**Long-term studies**

Weanling Sprague-Dawley rats were fed diets containing
0.8%, 2%, or 5% of two different gum bases (exact composition not
revealed) each containing 25% candelilla wax (Harrisson, 1953).
Each group consisted of 30 animals of each sex. Because no
difference in growth or appearance could be detected between rats
fed the two gums, studies with one gum were discontinued after 89
wk, while the other study continued for 2 yr. It is estimated
that the daily intake of candelilla wax by rats on the various
dietary levels was approximately 125, 300, and 750 mg/kg body wt,
respectively. There were no significant differences from controls
in growth or appearance, or in the blood, urinary, or histopatho-
lological findings at completion of the test.
Carcinogenicity

Thirty weanling Sprague-Dawley rats of each sex in each group were maintained 87 or 104 wk on 0.8%, 2.0%, or 5.0% of gum bases containing 25% candelilla wax (Harrisson, 1952). During this period, a number of spontaneous benign mammary gland tumors and a "few" malignant cancers developed in both control and test animals. Although the detailed protocols of these experiments were not available to the Select Committee, the summary statement indicated that the distribution of these neoplasms throughout the control and treated groups were "about equal".

Black agouti mice (C57) were fed 0.0%, 0.8%, and 5% of the gum base containing candelilla wax (composition not indicated). A fourth group received 0.06% butter yellow in the diet as positive controls. This control group was sacrificed after 9 mo, at which time numerous hepatomas were evident. The other mice were sacrificed at 12-13 mo. No hepatomas were seen and there was no significant difference in neoplasms between control and gum-fed mice (Harrisson, 1952).

Twenty mg pellets of gum base (composition not stated) were implanted in the right axillary areas of 80 young adult rats and 40 guinea pigs, and 5 mg pellets, in 44 C57 mice. The guinea pigs were sacrificed after 11 mo, and the rats and mice, after 13 mo. Encysted pellets were recovered; there was no evidence of vascularization, inflammation, or local tumors (Harrisson, 1952).

Ten mg of each test material were suspended in mineral oil and injected into the axillary region of 40 adult rats equally divided by sex. Five mg in oil were similarly injected into each of 44 young C57 mice. Other groups of rats and mice were injected with 3 mg methylcholanthrene as positive controls or with mineral oil alone as negative controls. All rats and mice receiving methylcholanthrene developed sarcomas within 6 mo. The negative controls and test animals were sacrificed after 13 mo. Necropsy revealed cysts around the injected oil, but no evidence of inflammation or local tumors (Harrisson, 1952).

A 1% benzene solution of gum bases containing candelilla wax was applied three times weekly to the skin of 40 young female C57 mice. A 1% benzene solution of methylcholanthrene was applied to other mice as a positive control and benzene alone as the negative control. All animals receiving methylcholanthrene developed epitheliomas within 6 mo. No epitheliomas were detected after 13 mo in mice receiving either benzene alone or the gum solutions (Harrisson, 1952).

Reproduction studies

No systematic study on reproductive or teratological effects of candelilla wax could be found. The only reported
observations were incidental to the short-term study reported above (Harrisson, 1949) in which various levels of a gum-base mixture of candelilla wax and a polymer of butadiene and styrene had been fed to male and female rats. After the animals had been on these test diets for 5 mo, three males and three females receiving 0%, 1%, and 5% of the mixture, respectively, were mated. Two of the three females from each level conceived on either the first or second mating and produced normal litters.
V. OPINION

Candelilla wax is a complex substance of plant origin which is used as an ingredient in chewing gum base and as a component of food packaging materials. It is virtually insoluble in water. Many of the constituents of candelilla wax are natural components of vegetables and fruits.

Short- and long-term feeding studies of candelilla wax in gum bases up to 5% in the diet revealed no evidence of toxicity in rats, mice, guinea pigs, and dogs. Candelilla wax products also showed no evidence of carcinogenicity when tested in rats, mice, and guinea pigs.

Therefore the Select Committee concludes:

There is no evidence in the available information on candelilla wax that demonstrates, or suggests reasonable grounds to suspect, a hazard to the public when it is used at the levels now current or that might reasonably be expected in the future.
VI. REFERENCES CITED


van Straten, S. 1977. Volatile compounds in food. 4th ed. Zeist, the Netherlands: Central Institute for Nutrition and Food Research TNO.


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