GAS CHROMATOGRAPHY ANALYSIS OF HYDROCARBON CHANGES IN NON-SMOKERS

A thesis submitted in partial satisfaction of the requirements for the degree of Master of Science in Health Science

by

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ABSTRACT

GAS CHROMATOGRAPHY ANALYSIS OF HYDROCARBON CHANGES IN NON-SMOKERS

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With the growing concern about the relationship between smoking and health there has been a commensurate growth in knowledge provided by researchers. Yet there remain many unanswered questions when one attempts to produce scientific causal relationships. Further, there is a need for translating these research findings into appropriate educational programs.

This paper approaches the smoking-health-education relationship from two avenues. The first is an attempt at analyzing the changes that take place with hydrocarbons after having been exposed to lung tissue of non-smokers. Non-smokers were used so that there would be a consistent type of lung tissue exposed to cigarette smoke. Gas chromatography was the method used for measuring these
changes. Atmospheric air, room air, cigarette smoke and expired air samples taken before and after smoking were tested. Various techniques of smoking and collecting expired air samples were also attempted in order to develop the most rational methods needed for this type of research.

The second avenue explores the educational implications of this research. Because the smoking habit is so complex the multi-facets of the problem were explored so that the findings of this paper could be put into its proper place within a larger body of knowledge.

If research confirms a causal smoking-health relationship then the educator must establish appropriate preventive educational programs. These programs can be enhanced in their development and application if they are based on scientific fact. Hopefully this thesis adds to the relationship between basic research and health education programs.

Emphasis in the laboratory was placed on the problems of accurate sampling and precise measurement of air samples. Educational emphasis was placed on the utilization of laboratory research in the development of concepts and objectives.
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CHAPTER I

INTRODUCTION

Man's concern towards the postulated relationships between smoking and health is a continuing area of interest. This concern can be seen in the increased activity in research, from Brosch's (40) observation of epithelial proliferation after applying tobacco "juices" on guinea pigs in 1900, to the Surgeon General Report on Smoking and Health in 1964 (1). Since then a vast amount of clinical, epidemiological, laboratory and behavioral research has added a great deal of knowledge to the subject, but many questions remain unanswered when a direct causal relationship is sought. Even though the World Health Organization Expert Committee on Prevention of Cancer (1964) stated "It is generally accepted that there is a causal connection between cigarette smoking and lung cancer" (44), the mechanisms suggested to explain these relationships are complex requiring further research and simplification if this information is to be integrated into educational programs.

The explanation of the correlation between tobacco usage and pathological changes in man should rest primarily on data from human subjects. Therefore,
epidemiological and laboratory comparisons between smokers and non-smokers may provide this necessary information. The study of smoking practices and physiological effects of these practices may be enhanced if objective, qualitative and quantitative characteristics of cigarette smoke could be identified. The large number of substances existing in cigarette smoke may provide the researcher and educator with important identifying measurements and a source of usable data. Gas Chromatography provides an ideal tool for such measurements by identifying the constituents in cigarette smoke and analyzing expired air samples before and after smoking. This analysis could then provide a basis for understanding the gaseous exchange mechanism within the lungs (11, 14). Important to the relationship between smoking and physiologic response is the identification of smoke constituents that are absorbed by the body. Inhalation studies need to be concerned with determining the types and amount of smoke components that actually reach the lungs. This thesis will concern itself with only one of the many parts of cigarette smoke-hydrocarbons. With a clear differentiation of respiratory volatiles it would seem reasonable to utilize this information in the following manner.

1. Identify specific hydrocarbons in cigarette smoke.

2. Identify hydrocarbon constituents in air samples
taken before and after inhalation of cigarette smoke.

3. Determine if hydrocarbon changes take place within the oral-pulmonary chambers via combination, oxidation, separation or other chemical reactions, thereby explaining that some hydrocarbons may become altered within the lungs and exhaled as a different compound.

4. Provide information related to the immediate effects of smoking that may be applicable to the development of educational programs aimed at affecting smoking behavior.

From an educational point of view, as one studies the literature, it becomes obvious that the act of smoking may be viewed as the interaction of multiple factors in an ecological system, many of which are unknown. The immediate and long-range effects of smoking involve multiple psychological, social and physiological factors. If research findings strengthen or confirm the causal relationship between smoking and harmful physiological responses, then it becomes imperative that health educators establish appropriate preventive educational programs that are based on such findings. It can also be hypothesized, with qualifications, that knowledge of the immediate effects of smoking as a threat to health may assist in altering the smoking behavior of young
people (2, 3, 4, 5). This concept led to the investigation of the immediate effects of smoking in a healthy male population, under 22 years of age, at San Fernando Valley State College, Northridge, California (2). The investigators simultaneously measured multiple variates relating to the immediate effects of smoking. It was from this study that the impetus for this Master's thesis materialized.

Purpose

The purpose of this thesis is to compare the concentration and types of selected hydrocarbons found in the atmosphere, cigarette smoke and expired air samples taken before and after smoking, using non-smokers as subjects. From this research, relevant content could then be used in health education as a means of altering smoking behavior.

Objectives

I. To accomplish these purposes the objectives of the laboratory research is to determine if quantitative differences exist in air samples taken in the following conditions.

1. Atmosphere samples taken in 5 geographical locations.

2. Room air samples taken prior to and after smoking has taken place in the immediate
environment of the subject.

3. Smoke taken directly from a cigarette and diluted to the same smoke-air ratio as found in the average adult lung.

4. Expired air samples taken after the subject has taken one puff, into the mouth only, in an attempt to determine if the oral mucosa absorbs or alters the hydrocarbons present in smoke.

5. Expired air samples taken after the subject "smoked" an unlit cigarette. The cigarette was cut (clipped) regularly at the same rate that a cigarette burns down in length when it is smoked normally.

6. Expired air samples taken before and after smoking and inhaling 2 cigarettes from subjects that are clearly non-smokers. The objective here is to use lung tissue that has not been altered by past smoking thereby providing a constant base line.

7. Expired air samples taken from subjects after
   a. breathing normally
   b. taking a big breath
   c. exhaling one-third of lung capacity.

8. Finally, to generate emperical data that can be combined with other findings related to
the physiological and behavioral aspects of smoking.

II. The educational objectives of this research are to translate and synthesize the laboratory findings into meaningful educational programs.

Null Hypothesis

The null hypothesis of the laboratory experiment in this thesis states, that there is no difference between the true means of the compared samples. Comparisons will be made using a t test.
CHAPTER II

REVIEW OF LITERATURE

The review of literature is divided into two sections.

1. Laboratory research of hydrocarbons in cigarette smoke and expired air.
2. The educational implications of laboratory research and the behavioral aspects of smoking.

1. REVIEW OF LITERATURE PERTAINING TO LABORATORY RESEARCH

To clarify how hydrocarbons were sampled and analyzed in this study, using gas chromatography, an exploration of the progress achieved in these fields will be presented first. A review of the literature revealed a wealth of published information. This material will be "dissected" and presented in the following order:

1. Cigarette Smoke
2. Gas Phase of Smoke
3. Hydrocarbons
4. Gas Chromatography

Cigarette Smoke

Smoke is the visible product of imperfect combustion. Ultimately the smoke will become mixed with air and
cease to be visible. Particles smaller than 10 microns are supported by air molecules and are swept around by air currents. Smoke will not remain permanently in the atmosphere and will eventually settle. Because of the small size of most particles they behave in many ways like gas (29).

Physically cigarette smoke displaces an equal volume of air and introduces particulate matter that may increase airway resistance by 50 per cent when inhaled (21, 31). More than 80 per cent of the particulate matter is retained by smokers who inhale deeply (37).

Chemically two thirds of the compounds found in smoke are not found in the cigarette and can be presumed to be formed by combustion (21). Smoke undergoes rapid changes in temperature as it is pulled through the cigarette. From a burning zone temperature of 800°-900°C during a puff, the smoke exits the butt only slightly warm in less than 0.2 of a second. This rapid cooling plays an important part in aerosol formation. The unburned portion of the cigarette serves both as a filter for aerosol particles and an area for gas combustion. These substances are revaporized as the burning zone passes through and then recondenses further back in the cigarette. Tobacco acts very much like the Gas Chromatograph column (41).
The volume of a cigarette puff is approximately 35 cc, and the volume of the lungs and airways is about 3.5 liters representing a 100-fold dilution of the puff. The intermittent exposure of approximately 1 1/2-2 seconds per puff with one puff per minute, means exposure time while smoking represents only 2-4 per cent of the total time involved in burning one cigarette. The activity of smoke also varies as a function of time from smoke formation to analysis, and hence, to be strictly applicable, studies should be conducted on fresh smoke (21, 30).

The problems concerning fresh smoke and dilution are taken into consideration in this study and are explained in the next section, but the analysis and the role of particulate matter is left for other studies and is outside the scope of this paper.

Gas Phase of Smoke

Only 20 per cent of the entire combustion of cigarette smoke is in the gaseous state and 90 per cent of this 20 per cent is carbon monoxide (CO). Within this gaseous state approximately 500 compounds can be found including sixteen carcinogens (6, 28, 39, 40, 45).

The gaseous phase can then be divided into three groups (6, 7, 8, 9, 10, 11).

1. Permanent and inorganic gases and vapors
2. Hydrocarbons
3. Remaining organic compounds.
Of the three groups the hydrocarbon portion of the gas phase of cigarette smoke was selected for this study. Gas Chromatography because of its versatility, sensitivity and speed is ideal for this type of analysis (12, 13, 14, 19).

Because the analysis of hydrocarbons in this study may provide a continuation to the San Fernando Valley State College project (2) by Fodor, Glass, and Weiner, it is obviously imperative that all materials and procedures are duplicated exactly. Researchers familiar with chromatography are fully aware of how difficult it is to duplicate analytical procedures and that each step should be suspect (34). In this study the problems of duplication were easily compensated for by using the same equipment (hardware) and procedures used in the Fodor, Glass, and Weiner project.

Hydrocarbons

The compounds of organic chemistry may be divided into two general classes.

1. Aromatic
2. Aliphatic

Aromatic compounds contain a molecular structure consisting of one or more benzene rings, which has its six carbon atoms arranged in a hexagon with alternate single and double bonds between them. Other groups may replace the hydrogen atoms to form an almost endless
variety of aromatic compounds.

Aliphatic compounds include organic compounds which do not possess a benzene ring. Their basic structure involves chains of carbon atoms which may be either straight, coiled or branched or arranged in rings different than the benzene ring. Again an almost endless variety of compounds of this class are possible. Hydrocarbons fall into the aliphatic class.

From the point of view of composition, one of the classes of organic compounds found in cigarette smoke are the hydrocarbons (26, 27, 28). Hydrocarbons are compounds of carbon and hydrogen of which there are several families. The hydrocarbons identified in this thesis are the short chain gases C₂ to C₆ representing 2.5 mg per cigarette and 0.5 per cent of the total effluent (42). These hydrocarbons include:

1. Ethane
2. Unknown
3. Methyl Chloride
4. Vinyl Chloride
5. Ethylene Oxide
6. Butadiene or Ethyl Chloride
7. Ethanol
8. Acetone or Pentane
9. Isopropanol
10. Unknown
Gas Chromatography

Chromatography has become one of the most widely used methods for separating the components of mixtures since Martin and James (14, 16, 18) successfully experimented with partition gas chromatography in 1952. The name "chromatography" was derived from original experiments of Tswett in 1906 (14, 32). He washed solutions of plant pigments through columns of powdered chalk and observed the mixture separate as they move down the column at different speeds and eventually separate into bands of color (16, 17, 18).

In gas chromatography the compounds in a simple mixture are picked up by an inert carrier gas as it passes the injection point at a constant flow. The carrier gas plus the sample are driven through a column where the components of the sample mixture are separated. The column is a copper, stainless steel, glass or Teflon capillary tube that is about a quarter of an inch in diameter and can be from a few feet to one thousand feet in length. The inside of the tube is coated with an immobile liquid called a partitioner and packed with some pulverized diatomaceous earth. The packing and liquid coating selected largely determine the characteristics of the chromatograph. Samples containing up to 76 different substances as small as one trillionth of an inch have been analyzed at one time (14, 28, 33, 39). The
packing separates the gas mixture as it passes through, slowing up the progress of some substances and allowing others to travel through the column more swiftly. As the gases emerge from the column they pass through a detector which measures the electrical conductivity of the sample as well as that of the carrier gas. The differences between the two measures are recorded on a paper chart called a chromatogram. Another method utilizes the change in ionization - hence electrical conductivity - of the gas stream. The differences in boiling points of various substances results in peaks appearing at different times on the chart. The detector cell measures temperature and conductivity changes of the sensor wire as each hydrocarbon is ionized when passing through the flame. No detector can itself identify substances, but it can measure retention time. This is the point in time when a substance reaches its boiling point and is recorded as a peak on the chart paper (15). The detector can be calibrated by sending samples of known substances into the instrument (14, 15, 16, 22, 23). By measuring the retention times of known substances and comparing these retention times with unknowns, a fairly accurate identifying procedure can be developed. The areas under the peaks formed can be measured and can be said to be proportional to the amounts of each substance in the original sample within a 2% accuracy (14, 23, 25).
2. REVIEW OF LITERATURE PERTAINING TO EDUCATIONAL IMPLICATIONS AND THE BEHAVIORAL ASPECTS OF SMOKING

To place the information derived from this thesis in its proper place within the framework of the educational program, a brief overview of smoking behavior and its relationship to health will be examined. A brief survey of some pertinent factors and their implications on the individual as well as a unifying characteristic between laboratory research and educational research will be presented in the following sequence.

1. Purposes of laboratory and health education research

2. Review of smoking behavior
   a. Reasons for starting smoking
   b. Reasons for continuing smoking
   c. Reasons for stopping smoking

Purposes of Laboratory and Health Education Research

A primary purpose of laboratory research in the field of tobacco and smoking is to determine whether various tobacco smoke products might prove harmful to man, and, if so, which of the many components may be responsible. From the point of view of the tobacco industry, if these components can be identified, then attempts can be made for their removal or reduction (40:3). From the point of
view of the health educator, this information can add to the "armamentarium" of scientific knowledge that must be translated and synthesized into meaningful educational programs.

Health education research has the purpose of discovering meaningful questions and answers through the application of scientific procedures. One of the most interesting endeavors in educational research is the theoretical framework or model for smoking behavior change (2, 4, 51). This undertaking has great implications for health education because it serves as a tool for fitting pieces of information together and provides the possibility of practical application of scientific data. Davis (51) further states that the use of research findings should be made to establish major concepts for identifying behavioral objectives and developing learning opportunities for all school levels.

Much research in health education is operational research rather than basic research. That is, research that emphasizes the application of knowledge gained rather than the development of basic knowledge. Experts tend to differ as to where this emphasis should be placed. Frank (46) states that variables should not be fractionated and studied separately. This does not mean that analytic studies of individual disciplines are not important, but rather, that we recognize their individual limitations in
developing multidimensional approaches to human behavior (2, 47). This multivariate approach does not negate the fundamental contributions of basic research, rather, it is an attempt to translate multiple findings into appropriate unifying application. Larson and Silvette (48:346) justify the need for basic research in complex areas by stating, "the very use of the word 'complex' implies that cigarette smoking may be factored into X number of 'simple' components". Benne (49) and Rosensteck (50) stress the need for both types of research and indicate that though operational research and basic research may serve somewhat different purposes, both can be scientific.

It then becomes obvious that basic research and health education program planning should be intrinsic parts of common objectives.

Review of Smoking Behavior

Since the longitudinal studies related to the psychology of smoking in 1938 at Harvard (53) there has been a great deal of research in the area of smoking behavior. With the overwhelming amount of data that has been accumulated the complexity of the subject being studied becomes clear in that specific answers to some basic questions remain unanswered. It would then seem reasonable to ask researchers for a diagnosis of smoking
determinants with the following questions. (1) Why does a person start smoking? (2) Why does one continue smoking? (3) Why do some smokers stop? A brief review of research that has attempted to answer these questions follows.

Reasons for Starting Smoking. A special report published by the Illinois Medical Journal (54) states that about half of the teen-age smokers interviewed could not explain why they began to smoke. In Hochbaum's (55) opinion, most research reasoning provides little scientific knowledge about the smoking habit, only plenty of speculation, firmly asserted but pseudo-scientific explanations and a fairly large but superficial and often misleading body of statistical data. Though Hochbaum's criticism appears quite harsh in light of other research, he did concede that there is a need for systematic research programs to learn more about the psycho-social aspects of smoking.

Researchers commonly state the following reasons for acquiring the smoking habit: exemplary influence of parents and other adults such as teachers, clergymen, athletes and physicians; influence of peers; an assertion for manhood, conformity, curiosity and rebellion (1, 52, 56, 57, 58, 60, 61, 62, 63, 64). Thus, social stimulation or pressure appears to play an important role in a person's early experimentation with smoking. While rebellion has
been implicated by many researchers the Advisory Committee to the Surgeon General (1) did not see much evidence. Interestingly, they did not rule out the existence of predisposing constitutional or heredity factors. Larson and Stilvette (48:269) suggested strongly that "Physiological factors are not involved in the genesis of the smoking habit", but that they do play a strong role in the inability to stop.

As far back as 1934, the oral drive has been implicated in the habit of smoking (5). Levy (65) presented evidence that there is an oral drive, independent of the hunger drive, that is responsible for the smoking habit. Tomkins (5) further elaborated on this theory by stating that the oral sucking mechanism in smoking is different than hunger because it becomes insatiable. Also, that sucking may at first be innate as a positive effect of enjoyment and later in life smoking may be used to reduce distress by providing the positive effect of pleasure. Salber (103) found no differences between breast or bottle feeding and subsequent smoking, nor between thumb-sucking, time of weaning, intervals between feedings and later smoking habits.

The existing research suggests the need for the development of new instruments that can be used from kindergarten to adulthood for a clear determination of why the human organism makes the decision to start smoking.
It would also be advantageous for health educators if techniques could be developed that will predict which youth are likely to become smokers. Educational programs could then be geared to the high-risk groups.

**Reasons for Continuing Smoking.** Smoking behavior is enormously complicated and the reasons given for smoking are many; but the reasons for continuing to smoke, may not be the same reasons for starting. Also, the reasons for continuing may not be the same reasons for not stopping. An example is seen in the question, why do people go on smoking despite what cancer and other researchers tell them about the harmful effects of smoking? Horn (66) called attention to the reasons given by 14% of regular smokers who considered the smoking habit pleasurable, safe and worth the cost. Davidson (67) grouped the reasons for persistence in smoking as: denial of the health hazard; excuse of moderation; the safety effect of filters; diversion of the question; and defiance of the danger. McArthur (68) added personality variables to this list, in that certain persons may be more prone to habit-forming behavior than others.

Tomkins (5) combined innate affects with the learned habit in his comprehensive "psychological model for smoking behavior". On the basis of this theory, he distinguished four general types of smoking behavior, (1) **positive affect smoking** - characterized as stimulating, exciting,
relaxing or enjoyable; (2) **negative affect smoking** - in this case the individual smokes to reduce feelings of fear, shame or disgust; (3) **habitual smoking** - this type of smoking may have started with either or both of the first two types but are no longer connected to the original psychological reason, smoking takes place almost without awareness and is now a habit; (4) **addictive smoking** - here the person smokes both to increase the positive affect and to reduce the negative affect. This may be called psychological addiction and the cigarette becomes an emotional "end-in-itself".

Departing slightly from sole psychological aspects of smoking behavior, Larson and Silvette (48:282) stated that smoking is first a habit and secondly a drug dependency and that the reasons for continuing a drug dependency follows pharmacological rather than physical or psychological laws. They viewed addiction of all types as biological and stated, "In dealing with the biological effects of tobacco smoking, the smoker's attitude towards smoking and its possible consequences is also a biological fact".

Additional reasons given for continuing the smoking practice are; (1) stimulation of the special senses (warmth, touch, aroma, taste, sight of fire and smoke) by tobacco constituents other than nicotine, (2) "craving" in the throat, (3) a bronchial "itch" with smoking acting
as a counter irritant, (4) dulling of appetite, (5) an aid in losing weight, (6) tastes good with coffee or a drink, (7) makes a meal complete and aids in digestion, (8) gives a person something to do with their hands, and (9) it is an act of self-annihilation. With all of these reasons, which is by no means a complete list, the question must be asked, does not the non-smoker have any of these needs? It is possible, then, that researchers should spend more time studying the non-smoker.

The vast variety of reasons for continuing smoking once started, and the different points of view presented by the authors mentioned, only make the quest for answers in smoking behavior more challenging for the health educator. Indeed this quest for answers should be taken into the class room where students themselves might try to answer the question of why they do or do not smoke and why they continue to do so.

Reasons for Stopping. Although a great deal of research in the over-all behavioral aspects of smoking has taken place, Schwartz (3), Mausner and Platt (69) commented on the scarcity of effective research programs that help people to stop smoking. Most important, they found a lack of standardized criteria for measuring success with these programs. Smoking clinics and educational programs have reported a diversity of results in
getting individuals to stop smoking and report an even greater variety of reasons for stopping. This diversity and lack of consensus can be keenly noted in the reports of Ejrup (88), Cartwright et al (89) and Schwartz (3, 94).

Ejrup claimed good results with 1012 patients in withdrawal clinics in Stockholm, giving health reasons as the most prominent reason for stopping. Cartwright found that health was the least frequently mentioned reason for giving up smoking. With so many subjects informed of the health hazards of smoking who do not quit, it becomes apparent that dispensing facts alone is insufficient to influence the smoking habit (70). Podor, Glass and Weiner (2, 82) in their cognitive skills testing of college students, revealed that smokers were more informed about the effects of smoking on health than the non-smokers. They went on to state that merely disseminating information about the bad effects of smoking may not alter behavior. However, they did stress the need for scientific knowledge on the immediate effects of smoking, because young people may not perceive or care about the long-range effects of smoking that do not become evident until the smoker is older.

Carson (80) and Moore (81) also encouraged the dissemination of scientific knowledge and presented strong arguments against the use of the "scare" technique as a long-term motivational tool.
In an attempt to clarify the befuddlement found in numerous research reports, Schwartz (3) evaluated 62 studies of smoking cessation programs, which used over 100 methods, conducted throughout the world. He found only a few "methods" with high success rates. Generally, initial success rates declined rapidly during the first month to the third month, and then tapered off with time. He also mentioned a lack of long-term follow-up as well as some bias in reporting. Part of the reason given for the low-success rate might be explained because the methods used were methods least acceptable to the individual wishing to quit (94). This finding is a good example of why health educators must constantly strive to keep their educational objectives realistically related to the needs and interests of the learner.

Field experiments in schools indicated that educators have not shown a much better success rate (51, 74, 75, 95, 97, 98, 99, 100, 101). Davis (51) stated "we are not yet able to synthesize the findings and say how they can best be used or incorporated into specific education intervention processes." Leventhal (102) contended that intervention techniques must avoid coercion and be consistent with democratic values. Therefore, it is possible that influencing efforts may result in interpersonal psycho-social conflict in a society where smoking is generally an accepted form of behavior. Knight (70)
and Lister (77) contended that in a population propagan-
dized to the pleasure of smoking, anti-smoking propaganda
is likely to be ineffective until cigarette smoking
becomes a socially unacceptable habit. In order to make
a practice socially unacceptable, all types of health
agencies - governmental, medical and lay - have the
responsibility to pursue the educational process
vigorously (78).

The literature cites many reasons for quitting
smoking. Some of these reasons include: scientific
evidence of physical harm, personal health reasons, lack
of satisfaction, self-discipline, economics, esthetics,
fear of cancer, religion, playing the exemplar role,
persuasion and family opinion, and unknown psychological
and/or physiological factors. Considering the diversity
of needs that impel different persons to smoke, no single
approach appears to be satisfactory to persuade or motivate
all people not to smoke (79). Any methodology now avail-
able or yet to be developed to alter smoking behavior
must evolve out of an understanding of the multiplicity
of factors that motivate behavior. An important task
of the health educator then is to identify these factors
and apply them to the solving of individual and community
health problems.
CHAPTER III

METHODOLOGY OF LABORATORY EXPERIMENT

RESEARCH DESIGN

The sample size in this study consisted of 9 subjects, three females and six males ranging in age from 20 to 55. In order to qualify and meet the criteria as a non-smoker the subject should not have smoked a cigarette for two or more years. Seven of the nine subjects never smoked at all.

Three expired air samples were taken from each subject. The first was taken before smoking. These samples are "pre-expired air samples." The second was taken after the investigator lit the cigarette and gave it to the subject who took one drag into the mouth only and blew it out. This second sample will be called "oral air." The third sample was taken after the subject completed smoking two cigarettes within a ten minute period of time. These samples are "post expired air samples." The cigarette used was a filtered Marlboro regular and the subject puffed and inhaled every ten seconds.

Room air samples were taken just above the heads of the subjects before and after smoking. These samples will be labeled "pre and post room air samples."
Atmosphere/air samples were taken at the following five locations:

1. Santa Monica beach
2. West Los Angeles
3. Mid-city Los Angeles, at City Hall
4. San Fernando Valley
5. Lancaster City near the desert

Samples were taken from an apparatus designed to have the same lung capacity of an average size adult. These samples will be called "dilute smoke."

Finally, expired air samples were taken from subjects before and after puffing on an unlit cigarette to consider the possibility that hydrocarbons from the atmosphere may be filtered by the tobacco. These samples will be called "pre and post unlit expired air samples."

In deciding on sampling methods an investigator is faced with many problems. Decisions must be made concerning the following alternatives.

1. Sampling all elements, from particulate matter to gases, in the air or just a select few?
2. Sampling all elements in cigarette smoke or a specified number?
3. Sampling all elements in the expired air of well and sick individuals, male and female, young and old, or before and after activity, or just a few of these?
The major difficulties encountered in each of these approaches is further related to a multitude of influencing variables such as:

1. Are the collected samples representative of the conditions to be studied?
2. Will there be changes in the concentration of the gases as a result of sampling technique?
3. What are the limitations of the testing equipment?
4. Will the investigator provide consistent procedures for all subjects sampled, such as:
   a. intensity and duration of each puff?
   b. length of time between puffs?
   c. degree of inhalation?
   d. how much of the cigarette is smoked?
   e. type of container used to store air samples?
   f. transportation and handling of samples?
   g. temperature that samples are stored which may affect displacement and condensation? (6, 7, 8)

These problems were taken into careful consideration throughout this study. Explanations will be provided as each problem is encountered.

PREPARATION OF AIR SAMPLING MATERIALS

In developing a simple device for the collection of gases, it became necessary to develop a sampling technique by which air components (contamination) could
be excluded from the collected sample. The materials used for collecting and storage of air samples consisted of sterile throw-away 20 and 40cc syringes, 20 gauge needles, and 20cc vacutainers. No special preparation of the syringe and needle was necessary but the vacutainer used for storage of air samples did require special treatment. Each sterile vacutainer test tube was flushed out with Nitrogen gas before being used. A 20 gauge, 1 1/4 inch sterile, disposable needle and Polyethylene tubing was used to run the nitrogen from the tank, through the stopper, into the vacutainer. Another needle was inserted through the stopper to allow the nitrogen to pass out of the test tube. Each tube was flushed approximately three to five minutes. Nitrogen was used because the column in the gas chromatograph was not sensitive to this gas. To be sure that there was no contamination in the flushed tubes, blank samples were run through the gas chromatograph. No contamination was noted on the charts by signal variation.

EXPIRED AIR SAMPLES

Sterile 20cc disposable syringes were used to collect the expired air samples. The subject placed his lips tightly around the hub and blew directly into the syringe until the plunger was pushed back a little past the 20cc mark. After removing the syringe from the subject's mouth, a sterile needle was quickly connected
to the syringe while positive pressure was applied to the plunger reducing the content to exactly 20cc. This positive pressure prevented outside air from entering. The sample was then injected throughout the stopper into a flushed out vacutainer and labeled. The samples were packed in a bucket of ice cubes to prevent pressure build up and the popping of the rubber stopper. To keep the labels from running the samples were first put into plastic baggies, sealed and then packed in the ice. In order to obtain accurate air chemistry it was important that the subjects fasted for at least 4 hours before taking expired air samples.

When developing the procedure for taking expired air samples some important questions were generated. Should the subject take a deep breath before blowing into the syringe? Would the deep breath result in an extreme dilution process or become contaminated from the smoke filled room air? What effect does residual and supplemental air and the respiratory dead air space have on the sample? To answer these questions one must look carefully into the subject of alveolar air.

Normal expired air is a mixture of about two thirds alveolar air and one third air from the dead space. With an average tidal air volume of 500cc, taking in a big breath can add 2500cc of air to the tidal air volume. The combination of these two is called complemental air.
Forced expiration after an ordinary expiration can amount to an additional 1000cc. This is called supplemental air. Yet after the most strenuous expiration there remains 1000-1500cc of residual air (19, 20, 21). In order to get nearly pure alveolar air a forced expiration must be made after a normal expiration (20). Janak (38) divides the respiratory tract into three zones.

1. Upper air passages  
a. Consists of the trachea and large bronchi  
b. Air is the same as atmospheric air  
c. Called the "dead space."

2. Zone of linear mass transfer  
a. Consists of medium sized bronchi and bronchioles  
b. Air is a mixture of atmosphere and alveoli air

3. Alveoli-diffusion zone  
a. Where gas exchange with blood takes place  
b. Represents equilibrium with the blood

With a normal dead space of 150 ml, taking in a big breath results in the first 750 ml of expired air to come from the first two zones; the next 500 ml from the diffusion zone (38). To get alveolar air, then air samples should be taken after the exhalation of at least 1250 ml of air.

In order to take these factors of respiration into account, three different methods of collecting expired air samples were used in this study.

1. Blowing into the syringe while breathing at a normal rate for the subject. That is, without taking a deep breath first.
2. Taking a big breath before taking the sample.

3. Exhaling 2 to 3 seconds before the syringe is placed into the subject's mouth to eliminate dead air and thereby acquire alveolar air. The subject is then blowing into the syringe with approximately the last third or one half of his exhalation capacity.

DILUTE SMOKE: "ARTIFICIAL LUNG" SAMPLES

In an attempt to produce a chromatographic baseline for mainstream smoke taken directly from a cigarette, the problem of dilution had to be taken into consideration. Even with the attenuation control on the chromatographic instrument the peaks were too large, too long and overlapping for proper measurement, making it impossible to identify specific peaks. It also took over an hour to purge the instrument of the highly concentrated sample. To overcome these difficulties an "artificial lung" was designed. It was made of a 4 liter plastic bottle with a rubber stopper. It was flushed with Nitrogen and repeated samples were taken from the container in order to determine if there were leaks or contamination. All tests were negative, indicating no contamination.

In order to get a dilute sample of smoke, a 40cc syringe was attached directly to a lit cigarette and a 35cc quantity of smoke was withdrawn. This quantity falls within the average amount of smoke taken in with one
puff of a cigarette (21, 30). The smoke was then injected into the artificial lung and allowed to mix for a period of one half minute. A 20cc sample was then taken by syringe and injected into a 20cc vacutainer for storage as was all other air samples.

ROOM AIR AND GEOGRAPHICAL AIR SAMPLES

Room air and geographical (environmental) air samples were taken in precisely the same manner as expired air samples except that the technician had to draw the syringe plunger back manually. All room air samples were taken 1 to 2 feet above the heads of the subjects. All geographical samples were taken about 6 feet from the ground at each location within a two hour period of time. Altitude differences were not taken into consideration in this study.

CHROMATOGRAPHIC PROCESSING OF AIR SAMPLES

The samples were processed using a Perkin-Elmer 810 Gas Chromatograph. It was equipped with a flame ionization detector. Prior to processing, the samples were allowed to warm up to room temperature. They were injected through a gas sampling valve in order to get exact 5cc quantities. Each sample was allowed to run for approximately twenty minutes, from the time of injection to the processing of the next sample. The oven temperature of the Chromatograph was kept at a constant 100° Centigrade,
the Hydrogen gas was kept at 15 pounds per square inch, the compressed air was kept at 60 pounds per square inch and the Helium was held at 69 pounds per square inch. Helium was the carrier gas taking the expired air sample through the coiled column containing porapak Q that was 65 inches in length.

The concentration of each hydrocarbon was measured on a continuous graph moving at the rate of one-half inch per minute. The retention times are recorded on the graph paper as peaks. Each retention time is representative of the period of time from injection to the time the peak (concentration) reaches its maximum height. This time period was measured in minutes.

Peak Measurement of Hydrocarbons. Three major methods can be used for measuring the area of a peak (24).

1. Triangulation
2. Planimeter
3. Cutting and weighing

The triangulation method of measuring peak areas was used in this study. The uncertainty in all three methods is dependent in part on peak shape and size of the area to be measured. This excludes errors in sample injection, dilution and noise in the recording. The relative error is large for extreme shapes. Minimum relative error is achieved for large peaks having a ratio of peak height to width, measured at half height in the
The triangulation method used in this study requires drawing tangents to the sides of the peak at the inflection points and a base line. Multiplying one half the peak height by the length of the base line will provide the area of that peak. Each peak is the relative proportion of the entire sample.

The errors that are difficult to account for with this type of measurement include:

1. Measuring the height from a presumed base line that may be the result of drift and error in setting the pen properly before each sample is run.
2. Errors due to careless drawing of the base line.
3. Errors associated with measuring the length of the base line at the presumed tangents formed at the inflection points.
4. Errors associated with measuring the height at the top of the peak which may be flat, curved or even off the edge of the paper.

The errors mentioned can be insignificant if measuring procedures are carefully performed and carried out to at least three decimal places which was done in this study. When errors beyond the ability of the measuring devices and methods are considered, they are then only academic in nature. This does not excuse the
investigator from being aware of the limitations as they exist. The operator of the gas chromatograph also has some control by means of the sensitivity and attenuation settings of his detector and recorder, with additional control over the width of the peak by means of the chart speed.

**Quantitative Accuracy.** Quantitative accuracy in gas chromatography must be considered. Chemical sources of error can take place because samples may decompose or become altered during analysis or handling. Hardware sources of error may be due to minute alterations in gas flow rate, pressure changes and temperature changes. A 1° Centigrade change in column temperature will cause variations of 3-4 per cent in peak height and 2.5 per cent in retention time.

Sample injection errors can be kept down to a minimum with the use of an injection port. This was done with the use of a three-way injection port that took in exactly 5cc samples from each of the 20cc vacutainers.

**Qualitative Accuracy.** The identification of individual peaks may present major problems if there are too many compounds in the sample, such as particulate matter and nicotine, that the column is sensitive to. Or, if there are materials in the sample that are unknown. In this case other means are necessary for identification
such as mass spectrometry(35). In this study there are two unknowns that require further investigation for positive identification. As for particulate matter, a simple method for removing the majority of particulates would have been the use of a filter at the injection port. A glass tube filled with silanized glass wool would have served as an efficient filter for the analysis of smoke samples (36). However, a filter used for more than one cigarette may cause a concentration build up that may interfere with the consistency required in analyzing large numbers of samples. Therefore, it is suggested that a new filter be used for each cigarette.

Each of the possible errors mentioned above was given careful consideration in designing the procedures used in this study. Taking the possible errors into consideration, a properly functioning, correctly operated chromatographic testing routine is capable of delivering analytical accuracy and precision (23, 25).
CHAPTER IV

STATISTICAL ANALYSIS

To test the null hypothesis, which states that there is no difference between the true means of the compared samples, the t test was used. Fifteen t tests were performed (see list of t tests on page 40). Each test compared ten hydrocarbons (see Hydrocarbons Tested on page 39).

Rational for t Tests. Though it is usually desirable to work with large samples, there are occasions when small samples must suffice. This is the case with this study with an n = 9 and in some cases as small as 2.

When dealing with Post minus Pre differences I am comparing the performance of a single group by testing the significance of a difference between the means of two small correlated samples.

By using a single mean only, the mean of the differences, the problem reduces to a test of whether the mean of the differences is significantly different from zero using -

\[
\bar{D} = \text{mean difference} \\
\bar{s}^2 = \frac{\bar{D}^2}{n} - \bar{D}^2
\]
where \( s^2 \) = variance

\[ \bar{D}^2 = \text{mean difference squared} \]

\( n \) = sample size

\[ s = \sqrt{s^2} \]

\( s \) = standard deviation

\[ S = \frac{s}{\sqrt{n-1}} \]

\( S \) = standard error of the mean

\( \bar{D} \) = difference

\[ t = \frac{\bar{D}}{S} \]

d.f = n-1

d.f = degree of freedom

When testing for the significance of a difference between the means of two small uncorrelated samples such as expired air pre compared to dilute smoke the standard errors of both samples are considered together and the \( t \) is arrived at by the formula

\[ t = \bar{x} - \bar{y} \]

\[ \sqrt{\left( \frac{x^2 + y^2}{nx + ny - 2} \right) \left( \frac{nx + ny}{nx \cdot ny} \right)} \]

where \( \bar{x} \) and \( \bar{y} \) are the sample means

\( nx \) and \( ny \) are the number of cases in each sample

\( x \) and \( y \) are the deviations of the individual scores from the means of
their respective samples.

\[ d.f = nx + ny - 2 \]

Thus if the value of \( t \) is less than .975\% level of significance the null hypothesis is rejected; if larger than .975\% level of confidence the null hypothesis is accepted.

\[ t = (p < .025) \text{ in all tests} \]

**Ten Hydrocarbons Tested:**

Peak 1. Ethane  
Peak 2. Unknown  
Peak 3. Methyl Chloride  
Peak 4. Vinyl Chloride  
Peak 5. Ethylene Oxide  
Peak 6. Butadiene or Ethyl Chloride  
Peak 7. Ethanol  
Peak 8. Acetone or Pentane  
Peak 9. Isopropanol  
Peak 10. Unknown
List of t tests performed:

1. Geography versus Room Air Pre
2. Geography versus Expired Air Pre
3. Geography versus "Artificial Lung" Smoke (dilute smoke)
4. Geography versus Post Expired Air after "Smoking" unlit cigarette clipped.
5. Room Air Pre versus Room Air Post
6. Room Air Pre versus "Artificial Lung" Smoke (dilute smoke)
7. Room Air Pre versus Post Expired air after "smoking" unlit cigarette clipped.
8. Expired Air Pre versus Expired Air Post
9. Expired Air Pre versus "Artificial Lung" Smoke (dilute smoke)
10. Expired Air Post versus "Artificial Lung" Smoke (dilute smoke)
11. Expired Air Pre versus Pre expired air, taking a big breath first
12. Expired Air Pre versus Pre expired air, exhaling 1/3rd lung capacity first
13. Expired Air Pre versus Expired Air Pre, exhaling (big breath first) 1/3rd lung capacity first
14. Oral Smoke versus "Artificial Lung" Smoke (dilute smoke)
15. Pre Expired Air after "smoking" unlit cigarette clipped versus Post Expired Air after "smoking" unlit cigarette clipped.
CHAPTER V

DISCUSSION OF STATISTICAL FINDINGS

Each of the fifteen t tests will be explained. The tables of t values for each of the tests discussed in this section will be found in section IX.

Geography - Environmental Air (See Figure 1).

Because some of the air samples were compared with room air, it was important that the influence of geographical location be considered. To do this five geographical locations in and around the city of Los Angeles were tested. See Figure 1.

Figure 1 shows peak 1 (ethane) and peak 6 (Butadiene or ethyl chloride) to have the largest variations within the 5 geographical areas sampled. To test these differences in per cent, the values at both extremes for each of the two peaks were compared in the following manner:

$$s(p_1 - p_2) = \sqrt{\frac{p_1 q_1 + p_2 q_2}{n_1 n_2}}$$

$$t = \frac{p_1 - p_2}{s(p_1 - p_2)}$$

where $s(p_1 - p_2) = \text{standard error of this sample proportion using percentages}$

$q = 1 - p \text{ in per cent}$

$n = \text{number of peak samples}$
\( t = \text{was interpreted using confidence limits } < 0.025 \)

Using the extreme values for City Hall at .63 (peak 1) as the largest value and San Fernando Valley at .26 (peak 1) as the smallest value in a \( t \) test gave the following:

\[
\frac{t}{s(p_1 - p_2)} = 1.38
\]

A \( t \) value of 1.38 showed no significant difference between any of the five geographical locations for peak 1.

Testing peak 6 which also appeared to have great differences resulted in a \( t \) value of 1.47 which was not significant at 0.025.

Though Figure 1 shows what appears to be interesting differences for peaks 1, 3, 5, 6, and 8, these differences could have occurred by chance and therefore are not different enough for concern when taking room air samples at any of these locations.

A more sensitive measuring device or a gas chromatograph capable of analyzing more than ten hydrocarbons would be in order if one wishes to discern finer differences in hydrocarbon concentrations in various geographical locations.

Test 1

GEOGRAPHIC SAMPLES COMPARED WITH ROOM AIR PRE:

See Test Table 1
No differences were seen when geographic samples were compared to room air pre samples. These results indicate that a valid base line was being used when testing the possible influence of the environment.

Test 2

**GEOGRAPHIC SAMPLES COMPARED WITH PRE EXPIRED AIR:**

See Test Table 2

No significant differences were seen in this comparison. This non difference may indicate that the tested hydrocarbons from the atmosphere were not altered in any measurable way by their passage into and out of the lungs.

Test 3

**GEOGRAPHIC SAMPLES COMPARED WITH DILUTE SMOKE:**

See Test Table 3

Three peaks, peak 3 (methyl chloride), peak 5 (ethylene oxide) and peak 8 (acetone or pentane), were found to be significantly greater in dilute smoke. It may then be inferred that these hydrocarbons, though found in the atmosphere are increased by combustion of cigarette tobacco.

Of major interest is the presence of large amounts of peak 2 (unidentified) in dilute smoke that is not found in any of the other air samples. This finding is also seen in the Fodor, Glass and Weiner (2) study and presents a question for further investigation.
Test 4

GEOGRAPHIC SAMPLES COMPARED WITH SAMPLES TAKEN AFTER "SMOKING" AN UNLIT CIGARETTE CLIPPED:

See Test Table 4

Peak 5 (ethylene oxide), peak 8 (acetone or pentane) and peak 9 (isopropanol) were found to be significantly less in expired air samples taken after smoking an unlit cigarette. The reduction of hydrocarbons as atmospheric air is drawn through an unlit cigarette may imply that the cigarette acts as a filter.

Test 5

ROOM AIR PRE COMPARED WITH ROOM AIR POST:

See Test Table 5

No significant differences were noted in room air before and after smoking two cigarettes. This finding illustrates that ambient side stream smoke in the room after smoking just two cigarettes should not harm others present in the room. The presence of this small amount of smoke should not have an effect on pre-smoking expired air samples. There are many other factors that must be considered in future studies such as room ventilation, size of the room, function of time, and the number of cigarettes smoked in the room.

Test 6

ROOM AIR PRE COMPARED WITH DILUTE SMOKE:
See Test Table 6

Dilute smoke had significantly less peak 2 (unknown) and peak 6 (butadiene or ethyl chloride) than did room air. Peak 2 (unknown), peak 3 (methyl chloride), peak 5 (ethylene oxide), and peak 9 (unknown) were greater in dilute smoke. These differences could be due to concentration and dilution problems. Or, due to the fact that the dilute smoke in the artificial lung did not come into contact with the moisture found on the mucosal lining of a real lung. This moisture may act as a temporary holding agent to smoke.

Test 7

ROOM AIR PRE COMPARED WITH POST SAMPLES TAKEN AFTER "SMOKING" AN UNLIT CIGARETTE CLIPPED:

See Test Table 7

This is the only test where peak 1 (ethane) is greater in expired air post. It is difficult to explain why it should increase after "smoking" an unlit cigarette.

The unlit post samples showed a reduction in peak 5 (ethylene oxide) and peak 6 (butadiene or ethyl chloride), again indicating a filtering action by the tobacco.

Test 8

EXPIRED AIR PRE COMPARED TO EXPIRED AIR POST:

See Test Table 8

Interestingly no significant differences are seen in a pre and post comparison.
A possible explanation is that the non-smokers used in this study did not inhale as deeply as an experienced smoker and exhaled the entire contents in a rapid fashion. This may point to the learning habit and training required to inhale deeply - an act that requires time to develop and does not appear to come naturally. Future studies based on the length of time smoked may be revealing in this area.

Test 9

EXPIRED AIR PRE COMPARED TO DILUTE SMOKE:

See Table 9

Expired air samples pre compared to dilute smoke showed that dilute smoke has hydrocarbons present in larger amounts. Why all of the hydrocarbons were not found to be greater in dilute smoke still remains to be explained.

Peak 2 (unknown), peak 3 (methyl chloride), peak 5 (ethylene oxide), peak 6 (butadiene or ethyl chloride), and peak 9 (isopropanol) are greater in dilute smoke apparently due to the combustion of the cigarette.

Test 10

EXPIRED AIR POST COMPARED TO DILUTE SMOKE:

See Table 10

This comparison showed that four hydrocarbons were greater in dilute smoke than found in expired air samples
taken after smoking. These were peak 2 (unknown), peak 3 (methyl chloride), peak 5 (ethylene oxide), and peak 9 (isopropanol). Of these 1, 3 and 9 were significantly greater.

The remaining 5 hydrocarbons were greater in expired air post with peak 1, and peak 6, significant.

Why some hydrocarbons are found to be greater in expired air than found in smoke presents a focal point for further exploration. The question of why some smoke hydrocarbons are not found in expired air samples is also important.

It may be speculated that the lungs and oral tissue alters the chemical structure of the hydrocarbons. Through combination, reduction or other chemical changes some hydrocarbons may be changed into other forms. This phenomena again points strongly to the effects of tissue on smoke and indicates further research.

Test 11

EXPIRED AIR PRE (NORMAL BREATHING) COMPARED TO EXPIRED AIR PRE, AFTER TAKING A BIG BREATH FIRST.

See Table 11

No significant differences were seen between samples taken after breathing normally or after taking a big breath first.

These findings seem reasonable considering the facts that the individuals tested were non-smokers and no smok-
ing took place prior to these pre and post test.

Test 12

EXPIRED AIR PRE (NORMAL BREATHING) COMPARED TO EXPIRED AIR PRE AFTER TAKING A SMALL BREATH FIRST.

See Table 12

No significant differences were seen between pre samples taken after breathing normally or after taking a small breath first.

Test 13

EXPIRED AIR PRE (NORMAL BREATHING) COMPARED TO EXPIRED AIR AFTER EXHALING 1/3 TO 1/2 OF LUNG CONTENTS FIRST.

See Table 13

Exhaling 1/3rd of lung air prior to taking a sample did show a significant increase in peak 6 (butadiene or ethyl chloride). This sampling procedure may indicate that the sampled air was taken from deep in the lungs and may confirm the reasoning for using alveolar air in order to get more accurate and consistent findings.

Test 14

DILUTE SMOKE COMPARED WITH AIR SAMPLES TAKEN AFTER THE FIRST PUFF (ORAL AIR) WITH NO INHALATION.

See Table 14

The most dramatic differences were seen with peak 2 (unknown), peak 3 (methyl chloride), peak 6 (butadiene or ethyl chloride) and peak 9 (isopropanol).

Peak 6 increased in the oral air post sample, but
interestingly, peaks 2, 3 and 9 decreased.

The moisture and mucosa may have a holding and altering effect on the hydrocarbons and may release them after a period of time in a new form. Other factors to consider are the pH, saliva, and the length of time the smoke is exposed to the mucosa.

Test 15

EXPIRED AIR SAMPLES PRE COMPARED TO EXPIRED AIR POST SAMPLES TAKEN AFTER "SMOKING" AN UNLIT CIGARETTE CLIPPED AT THE RATE OF A LIT CIGARETTE.

See Table 15

Though peak 8 (acetone or pentane) was significantly smaller in the post sample, all of the hydrocarbons were found to be less after "smoking" the unlit cigarette. These findings indicate that the cigarette itself acts as a filter to some degree.
Statistical Conclusions

The main findings in this study are as follows:

1. Geographical location had no effect on the tests. There were no significant differences between hydrocarbons sampled in 5 geographical locations.

2. Hydrocarbons found in the atmosphere were the same as those found in expired air samples taken prior to smoking.

3. Air samples taken in a room where smoking had taken place did not show any significant differences from room air samples taken before smoking.

4. When subjects drew air through an unlit cigarette there was a filtering effect by the cigarette (tobacco plus filter) which was seen by a reduction of hydrocarbons.

5. There were no significant differences in expired air samples taken before and after smoking and inhaling two cigarettes. This may be due to the non-smokers inability to inhale properly. Also possible is the need for a more sensitive measuring device.

6. There were significant post differences when expired air samples were taken after the subject drew on a lit cigarette once and blew it out without inhaling. Interestingly the oral
cavity is subjected to direct contact with greater concentrations of smoke and at higher temperatures than the lungs, yet the incidence of oral cancer is less than for the lungs. Is it possible that the oral cavity reacts differently to smoke than do the lungs? It would be worthwhile to explore this phenomenon further.

7. There were significant differences seen when expired air samples were taken after the subject exhaled 1/3rd of his lung capacity. This finding may indicate that alveolar air should be used in future smoking studies.

In conclusion, one can state that additional and more sophisticated experiments are needed before a correlation between certain smoking parameters and their effect on the human becomes clear. The study of these findings entail many challenges of academic as well as practical significance. Simple answers cannot be expected when dealing with a complex problem. Though there were some insignificant findings in this study, in the educational application of scientific inquiry it is important to discuss failures as well as successes.

The educational implications of this thesis will be discussed in the next chapter.
CHAPTER VI

EDUCATIONAL IMPLICATIONS

EDUCATIONAL IMPLICATIONS OF RESEARCH

Health educators face a unique challenge in developing action research, experimental approaches to teacher training, curriculum development and classroom activities related to tobacco and smoking behavior (51). Necessary for this endeavor are basic scientific facts about tobacco, smoking and teen-age practices and appropriate methods in using this information effectively in meeting the needs of students. Because of the complex interrelationship of smoking knowledge, practices and attitudes, no educational program would be complete without taking the personality and behavioral characteristics of the teacher into consideration. Not to be overlooked is the hypothesis that when the teacher participates in an anti-smoking educational program, they may reflect their own smoking attitudes and habits (90, 91). Educational efforts should then begin with the teacher.

When smoking education should start is as important as how it should be instituted. Van Briesen (71) considered it a waste of time to teach new tricks to old dogs, and called for an effort directed at the teen-ager. He
stated that most teen-agers are intelligent enough to come to sensible conclusions about smoking, even if parents are not. Numerous other researchers advocate anti-smoking campaigns be started at the earliest possible age and stress that the instruction be based on scientific knowledge (4, 61, 72, 74, 75). Truex (76) substantiated the need for scientific information when describing his plan based on the following points:

1. There should be no preaching or moral condemnation of the individual smoker.

2. Care should be taken not to instill the fear of imminent incurable disease or death for all smokers.

3. Youth should be approached as maturing young people and presented with medical and statistical information on smoking and disease for their thoughtful consideration.

The decision of what to do about smoking is, therefore, up to the individual.

Investigation of the literature shows that there is a vast amount of data accumulated on the smoking knowledge, attitudes and practices of students of all ages. Only by application of this data can a realistic evaluation of health education and its effect on behavioral change be made.

Fordor and Dalis (83) presented the following plan for the practical application of specific information into the total health education curriculum.

1. Incorporate research findings into an existing
body of knowledge relative to smoking and health.

2. Ordering and ranking pertinent information, within the body of knowledge, according to importance.

3. Identifying appropriate concepts relative to each content area.

4. Organize the content related to the concepts.

5. Specify instructional objectives related to the concepts and content.

6. Design specific learning opportunities to enable the learner to attain the specified objectives.

7. Construct evaluative procedures that can determine whether or not the student has attained the instructional objectives.

This framework provides a mechanism for the application of scientific theory in health education. Important to this discussion is Fodor and Dalis' (83:26) comment that content should be based on data concerning the health problems of society and the needs and interests of the individual learner. These needs and interests have been determined by a number of authors, Lantagne (84), Havinghurst (85), Sutton and Rich (86) and Sliepcevich (87) to name a few. A review of these findings revealed smoking and health as one of the major subjects of interest, and a problem important enough to be included as content in the health curriculum (84).

It then seems reasonable that the findings provided by laboratory research on hydrocarbons and their dynamic
action on the lung tissue of non-smokers can be part of the school health education curriculum. By ranking detailed content information within the general subject of smoking and health, an interrelationship can be illustrated from which many concepts and objectives can be developed. The following are basic facts to which the knowledge from this research can be added. From these facts basic concepts related to smoking and health must then be developed.

Concepts

- Smoking behavior and tobacco fit into the scope of "Health" as defined by the World Health Organization (92) because they involve physical, mental and social factors.

- Cigarettes are made from a complex biological plant, Nicotiana tabacum, of which there are more than 60 species.

- The constituents of the leaf may be altered by variations in harvesting, processing, curing, aging, fermentation, the addition of fertilizers, insecticides, and additives.

- The tobacco and other substances added to the leaf, plus the paper covering cigarettes are burned (combustion) at a high temperature of 800 to 1000° centigrade.

- Three major actions occur simultaneously during combustion -
  a. pyrolysis - where organic matter is fractionated into smaller molecules
  b. pyrosynthesis - where newly formed components may be formed that were not originally present in the tobacco
  c. decomposition - the distillation of certain compounds

- If complete combustion takes place, no smoke will
be formed, only carbon dioxide and water.

- Complete combustion does not take place, therefore, over 500 substances are formed including nicotine, tars and smoke particles, ammonia, arsenic, acids, oils and pesticides with many other products remaining to be isolated and identified.

- The substances in smoke can be divided into a) particulate matter, b) areasols in which can be found many hydrocarbons, and c) gases.

- Mainstream smoke also has particles of the filter itself, the leaf and stem fragments from nonfiltered cigarettes.

- The human organism purposely draws all of these substances into his lungs when inhaling.

- Up to 90% of particulate matter is retained in the respiratory tract upon deep inhalation.

- 20 - 50% of smoke-particle retention is in the bucal cavity when none of the smoke is inhaled.

- The longer the smoke is retained in the lungs, the greater the absorption, therefore, the greater the potential for harmful effects. The effects are a function of time.

- A great number of hydrocarbons have been identified in the tobacco leaf itself, and in the by-products of combustion such as smoke and tars.

- Gas chromatography is one scientific method used to identify hydrocarbons found in cigarette smoke.

- Some of the hydrocarbons found in cigarette smoke become altered after having been inhaled and blown out by a smoker.

- Hydrocarbons can change form when heated, oxidized, or when in contact with other hydrocarbons.

- Precursors of carcinogenic hydrocarbons and tars are known to be present in tobacco; these are converted by pyrolysis into carcinogens.

- Laboratory studies show the presence of carcinogenic polycyclic aromatic hydrocarbons (PAH) in cigarette smoke such as benzo (a) pyrene, dibenz
(a, h) anthracene, benzo (b) fluorethene, benzo (j) fluoranthene, dibenz (a, l) pyrene, benz(a) anthracene, chrysene, benzo (e) pyrene, and indeno (1, 2, 3, -cd) pyrene.

- Methane and ethylene yield a carcinogen from pyrolysis at 900°C - 1100°C which is in the upper limit that cigarette tobacco burns when a puff is taken. Methane is one of the substances identified in this project.

- Other substances found in tobacco that are used as additives for aroma and flavoring such as humectants, licorice, and sugar, are known to be precursors of carcinogenic hydrocarbons.

- Additives comprise more than 30% of the final product in certain pipe tobaccos.

- Measurable immediate physiological responses indicate that some of the components in cigarette smoke are passed through the tissues into the blood.

- CO has an extreme affinity for hemoglobin. Combined with automobile exhaust, the blood carboxyhemoglobin level can impair biological processes.

- Nicotine, which is a dangerous poison in concentrated form, is strongly implicated in the concept of smoking addiction.

- The concept of smoking addiction may be compared in some respects to drugs and alcohol.

- This addiction and complex habit is difficult to break once started.

The educational implications of considering all of the above factors can aid in understanding the effects of smoking on the human organism. Yet the learning of all these facts would be difficult and of little value unless they were ordered into broader principles or concepts that are significant to individual and community health. Once meaningful concepts are organized, specific objectives
must be developed which provide evidence that the students have internalized the concepts. Such objectives might include practices as well as cognitive skills attained by the learners (83:19,193).

Also important in the utilization of scientific content is the integration and coordination of knowledge with other academic subjects such as chemistry, biology, physiology, statistics, epidemiology and psychology, and any other subject that satisfies the needs and interests of the individual student. It is then the responsibility of the health educator as a communicator to unite theory, research and practice in a manner that is meaningful to the learner.
CONCLUSIONS AND RECOMMENDATIONS

This thesis has explored two important areas: gas chromatography analysis of hydrocarbon exchange in non-smokers and the educational implications of this research. It appears that in the end of this exploration one is left with more questions than answers. This is good.

Hopefully this paper expresses the idea that the complex problem of investigating the effects of smoking on the human organism is far from complete.

In the analysis of hydrocarbons there was no attempt to show a direct causal relationship between smoking and disease. Before causality can be definite it must be inferred that smoking antecedes the pre-clinical stages of disease. It must also be assumed that these stages occur after the smoking habit is acquired. Further, one must ask, does smoking induce the process in susceptible persons or does it make the person susceptible? Clarification of the means by which these relationships develop is necessary for the understanding of the smoking-health problem. Thus the need for scientific research. This means going back to the beginning of a multiple chain of events--cigarette smoke. After the constituents that
comprise smoke are clearly identified quantitatively and qualitatively then the next logical step is to examine carefully what happens to smoke when it is inhaled into the lungs. Somehow all or part of the constituents come in contact with the respiratory tract for varying lengths of time. At this point in the dynamic sojourn of smoke, many additional questions arise. Do any of the constituents change qualitatively when they come into contact with tissue? Do they change because of mere contact with the other substances of smoke? Does the substance passing through lung tissue into the blood stream become altered once in the blood? Do other changes take place as the absorbed material is exposed to "target" tissue in various parts of the body? What changes take place when the absorbed substances leave the blood and enter the tissue cells? What happens when the cellular by-products leave the cell and go back into the blood stream to be eliminated by the excretory organs? What effect does all of this have on the excretory organs? What actually comprises this "pipe line" from the lungs to the distant organs that result in immediate observable and measurable physiological changes?

The basic research of this thesis is only a small attempt at answering some questions. It can only hope to provide a small contribution to the vast laborious task that lies ahead in smoking research. It also appears
that the more basic the research the more unrelated or fragmented the results appear in the scheme of the whole problem—assuming one knows what the problem is. Also, the value of any research may not be known for a long time after completion. It is also possible that the value may never be known.

The relationship between scientific information acquired through valid research and health education is essential. As there still remains much room for developing new and better methods in research, so is there a need for new and better methods of utilizing research information in the educational process.

Important to health education is that it be taught with scientific facts. Unfortunately not all research results in "facts" and many text books and research findings go unchallenged by professional educators (104). The health educator has the responsibility of understanding and questioning research methods used in science as well as the research methods used in education.

Further research is indicated along several broad lines.

1. Verification and refinement of this study.
2. Provide research opportunities for educators within the elementary and secondary level schools. If educational goals include relevancy to the needs of the student then the educator
should have first hand experience in understanding these needs.

3. Ideally research possibilities should be made available to students as an educational experience, with the student selecting his own problem.

4. Finally, further research is needed in the evaluation of the educational process. More specifically, evaluative procedures should be designed that will determine whether instructional objectives have been attained.
FIGURE I

Geographical Locations

<table>
<thead>
<tr>
<th></th>
<th>Peaks in Per Cent</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>San Fernando Valley</td>
<td>.26</td>
</tr>
<tr>
<td>West Los Angeles</td>
<td>.58</td>
</tr>
<tr>
<td>Santa Monica Beach</td>
<td>.53</td>
</tr>
<tr>
<td>Lancaster (desert)</td>
<td>.50</td>
</tr>
<tr>
<td>Los Angeles City Hall</td>
<td>.63</td>
</tr>
</tbody>
</table>

CODE: San Fernando Valley ————
West Los Angeles ————
Santa Monica Beach ————
Lancaster ————
City Hall ————
Tables 1 through 15 list the t values for each Pre minus Post test for each of the ten hydrocarbons. The mean value ($\bar{X}$) and the standard deviation ($X$) are also given for each hydrocarbon. The ten hydrocarbons are designated as peaks. (see page 37 for a list of what hydrocarbon each peak represents)

* = Significant t at $\alpha = .025$ level are marked with an asterisk.
### TABLE I

Geography ($n_x = 5$) versus Room Air Pre ($n_y = 8$)

<table>
<thead>
<tr>
<th>Peak</th>
<th>$t$ value</th>
<th>$\bar{X}$</th>
<th>$S$</th>
<th>$\bar{X}$</th>
<th>$S$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-.351</td>
<td>50.00</td>
<td>14.26</td>
<td>52.66</td>
<td>12.51</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>-1.230</td>
<td>2.26</td>
<td>1.68</td>
<td>3.06</td>
<td>.55</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>.054</td>
<td>3.23</td>
<td>.75</td>
<td>3.20</td>
<td>1.07</td>
</tr>
<tr>
<td>6</td>
<td>.744</td>
<td>35.94</td>
<td>13.80</td>
<td>31.27</td>
<td>8.36</td>
</tr>
<tr>
<td>7</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>.017</td>
<td>3.60</td>
<td>.563</td>
<td>3.58</td>
<td>2.57</td>
</tr>
<tr>
<td>9</td>
<td>2.10</td>
<td>4.96</td>
<td>6.80</td>
<td>no peak</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>-.791</td>
<td>no peak</td>
<td>-</td>
<td>4.90</td>
<td>13.51</td>
</tr>
</tbody>
</table>

$d.f. = 11$

$*=\text{Statistically significant at } \alpha = .025$
TABLE II

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>X</th>
<th>S</th>
<th>X</th>
<th>S</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>.324</td>
<td>50.00</td>
<td>14.26</td>
<td>52.78</td>
<td>15.84</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>3</td>
<td>.000</td>
<td>2.26</td>
<td>1.68</td>
<td>2.26</td>
<td>1.06</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>5</td>
<td>.550</td>
<td>3.23</td>
<td>.75</td>
<td>2.88</td>
<td>1.22</td>
</tr>
<tr>
<td>6</td>
<td>2.52</td>
<td>35.94</td>
<td>13.80</td>
<td>21.45</td>
<td>8.09</td>
</tr>
<tr>
<td>7</td>
<td>1.11</td>
<td>no peak</td>
<td>–</td>
<td>6.34</td>
<td>12.83</td>
</tr>
<tr>
<td>8</td>
<td>.69</td>
<td>3.60</td>
<td>.56</td>
<td>9.69</td>
<td>18.67</td>
</tr>
<tr>
<td>9</td>
<td>2.14</td>
<td>4.96</td>
<td>6.80</td>
<td>.23</td>
<td>.69</td>
</tr>
<tr>
<td>10</td>
<td>-1.46</td>
<td>no peak</td>
<td>–</td>
<td>4.44</td>
<td>6.50</td>
</tr>
</tbody>
</table>

d.f. = 12

* = Statistically significant at $\alpha = .025$
TABLE III

Geography ($n_x = 5$) versus Dilute Smoke ($n_y = 2$)

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>$\bar{X}$</th>
<th>S</th>
<th>$\bar{X}$</th>
<th>S</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1.93</td>
<td>50.00</td>
<td>14.26</td>
<td>29.25</td>
<td>1.25</td>
</tr>
<tr>
<td>*2</td>
<td>-15.29</td>
<td>no peak</td>
<td>-</td>
<td>17.80</td>
<td>2.20</td>
</tr>
<tr>
<td>*3</td>
<td>-7.56</td>
<td>2.26</td>
<td>1.68</td>
<td>12.85</td>
<td>1.15</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>*5</td>
<td>-4.92</td>
<td>3.23</td>
<td>.75</td>
<td>8.35</td>
<td>1.65</td>
</tr>
<tr>
<td>6</td>
<td>2.88</td>
<td>35.94</td>
<td>13.80</td>
<td>6.15</td>
<td>.15</td>
</tr>
<tr>
<td>7</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>1.45</td>
<td>.05</td>
</tr>
<tr>
<td>*8</td>
<td>-6.36</td>
<td>3.60</td>
<td>.56</td>
<td>6.40</td>
<td>.10</td>
</tr>
<tr>
<td>9</td>
<td>-1.91</td>
<td>4.96</td>
<td>6.80</td>
<td>14.70</td>
<td>.30</td>
</tr>
<tr>
<td>10</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

d.f. = 5

* = Statistically significant at $\alpha = .025$
### TABLE IV

Geography ($n_x = 5$) versus Unlit Clipped Cigarette Post Expired Air ($n_y = 2$)

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>$\bar{X}$</th>
<th>$S$</th>
<th>$\bar{X}$</th>
<th>$S$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-3.11</td>
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<td>14.26</td>
<td>86.65</td>
<td>9.35</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>3</td>
<td>-.09</td>
<td>2.26</td>
<td>1.68</td>
<td>2.40</td>
<td>1.60</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>*5</td>
<td>4.05</td>
<td>3.23</td>
<td>.75</td>
<td>.60</td>
<td>.60</td>
</tr>
<tr>
<td>6</td>
<td>2.25</td>
<td>35.94</td>
<td>13.80</td>
<td>9.85</td>
<td>9.85</td>
</tr>
<tr>
<td>7</td>
<td>no peak</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>*8</td>
<td>8.16</td>
<td>3.60</td>
<td>.56</td>
<td>no peak</td>
<td>–</td>
</tr>
<tr>
<td>*9</td>
<td>5.09</td>
<td>4.96</td>
<td>6.80</td>
<td>no peak</td>
<td>–</td>
</tr>
<tr>
<td>10</td>
<td>no peak</td>
<td>–</td>
<td>–</td>
<td>no peak</td>
<td>–</td>
</tr>
</tbody>
</table>

d.f. = 5

* = Statistically significant at $\alpha = .025$
TABLE V

Room Air Pre \((n_x = 8)\) versus Room Air Post \((n_y = 8)\)

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>X</th>
<th>S</th>
<th>X</th>
<th>S</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>-.70</td>
<td>52.66</td>
<td>12.51</td>
<td>56.85</td>
<td>9.07</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>.50</td>
<td>3.06</td>
<td>.55</td>
<td>2.87</td>
<td>.77</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>.72</td>
<td>3.20</td>
<td>1.07</td>
<td>2.99</td>
<td>3.02</td>
</tr>
<tr>
<td>6</td>
<td>2.51</td>
<td>31.27</td>
<td>8.36</td>
<td>23.97</td>
<td>6.16</td>
</tr>
<tr>
<td>7</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>1.68</td>
<td>3.58</td>
<td>2.57</td>
<td>3.06</td>
<td>2.33</td>
</tr>
<tr>
<td>9</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>-.92</td>
<td>4.90</td>
<td>13.51</td>
<td>9.99</td>
<td>10.01</td>
</tr>
</tbody>
</table>

d.f. = 7

* = Statistically significant at \(\alpha = .025\)
TABLE VI

Room Air Pre ($n_x = 8$) versus Dilute Smoke ($n_y = 5$)

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>$\bar{X}$</th>
<th>S</th>
<th>$\bar{X}$</th>
<th>S</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.52</td>
<td>52.66</td>
<td>12.5</td>
<td>29.25</td>
<td>1.25</td>
</tr>
<tr>
<td>*2</td>
<td>-21.95</td>
<td>no peak</td>
<td></td>
<td>17.80</td>
<td>2.20</td>
</tr>
<tr>
<td>*3</td>
<td>-15.02</td>
<td>3.06</td>
<td>.55</td>
<td>12.85</td>
<td>1.15</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>*5</td>
<td>-5.00</td>
<td>3.20</td>
<td>1.07</td>
<td>8.35</td>
<td>1.65</td>
</tr>
<tr>
<td>*6</td>
<td>3.85</td>
<td>31.27</td>
<td>8.36</td>
<td>6.15</td>
<td>.15</td>
</tr>
<tr>
<td>7</td>
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<td>-</td>
<td>-</td>
<td>1.45</td>
<td>.05</td>
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<td>8</td>
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<td>3.58</td>
<td>2.57</td>
<td>6.40</td>
<td>.10</td>
</tr>
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<td>*9</td>
<td>-124.05</td>
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<td>-</td>
<td>14.70</td>
<td>.30</td>
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<tr>
<td>10</td>
<td>.44</td>
<td>4.90</td>
<td>13.51</td>
<td>no peak</td>
<td>-</td>
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</table>

d.f. = 8

* = Statistically significant at $\alpha = .025$
<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>Pre (n = 8)</th>
<th>Post Expired Air (n = 2)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>X</td>
<td>S</td>
</tr>
<tr>
<td></td>
<td></td>
<td>X</td>
<td>S</td>
</tr>
<tr>
<td>*1</td>
<td>-3.41</td>
<td>52.66</td>
<td>12.50</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>1.40</td>
<td>3.06</td>
<td>.55</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>*5</td>
<td>3.14</td>
<td>3.20</td>
<td>1.07</td>
</tr>
<tr>
<td>*6</td>
<td>2.81</td>
<td>31.27</td>
<td>8.36</td>
</tr>
<tr>
<td>7</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>1.83</td>
<td>3.58</td>
<td>2.57</td>
</tr>
<tr>
<td>9</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>.44</td>
<td>4.90</td>
<td>13.51</td>
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</table>

D.f. = 8

*= Statistically significant at $\alpha = .025$
### TABLE VIII

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
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<th>S</th>
<th>X</th>
<th>S</th>
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</thead>
<tbody>
<tr>
<td>1</td>
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<td>52.78</td>
<td>15.84</td>
<td>57.64</td>
<td>13.82</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>2.27</td>
<td>2.26</td>
<td>1.06</td>
<td>4.56</td>
<td>2.72</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>1.54</td>
<td>2.88</td>
<td>1.22</td>
<td>4.14</td>
<td>2.86</td>
</tr>
<tr>
<td>6</td>
<td>-1.69</td>
<td>21.45</td>
<td>8.09</td>
<td>17.38</td>
<td>4.51</td>
</tr>
<tr>
<td>7</td>
<td>-1.03</td>
<td>6.34</td>
<td>12.83</td>
<td>1.82</td>
<td>1.90</td>
</tr>
<tr>
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<td>-.12</td>
<td>9.69</td>
<td>18.67</td>
<td>9.27</td>
<td>9.78</td>
</tr>
<tr>
<td>9</td>
<td>1.98</td>
<td>.23</td>
<td>.69</td>
<td>3.30</td>
<td>4.57</td>
</tr>
<tr>
<td>10</td>
<td>-1.75</td>
<td>4.44</td>
<td>6.50</td>
<td>1.88</td>
<td>4.37</td>
</tr>
</tbody>
</table>

* d.f. = 8

** Statistically significant at $\alpha = .025$
TABLE IX

Expired Air Pre ($n_x = 9$) versus Dilute Smoke ($n_y = 2$)

<table>
<thead>
<tr>
<th>Peak</th>
<th>$t$ value</th>
<th>$\bar{X}$</th>
<th>$S$</th>
<th>$\bar{X}$</th>
<th>$S$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.09</td>
<td>52.78</td>
<td>15.84</td>
<td>29.25</td>
<td>1.25</td>
</tr>
<tr>
<td>*2</td>
<td>-21.95</td>
<td>no peak</td>
<td>-</td>
<td>17.80</td>
<td>2.20</td>
</tr>
<tr>
<td>*3</td>
<td>-11.90</td>
<td>2.26</td>
<td>1.06</td>
<td>12.85</td>
<td>1.15</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>*5</td>
<td>-5.02</td>
<td>2.88</td>
<td>1.22</td>
<td>8.35</td>
<td>1.65</td>
</tr>
<tr>
<td>*6</td>
<td>2.56</td>
<td>21.45</td>
<td>8.09</td>
<td>6.15</td>
<td>.15</td>
</tr>
<tr>
<td>7</td>
<td>-.520</td>
<td>6.34</td>
<td>12.83</td>
<td>1.45</td>
<td>.05</td>
</tr>
<tr>
<td>8</td>
<td>.23</td>
<td>9.69</td>
<td>18.67</td>
<td>6.40</td>
<td>.10</td>
</tr>
<tr>
<td>*9</td>
<td>-27.63</td>
<td>.23</td>
<td>.69</td>
<td>14.70</td>
<td>.30</td>
</tr>
<tr>
<td>10</td>
<td>.93</td>
<td>4.44</td>
<td>6.50</td>
<td>no peak</td>
<td>-</td>
</tr>
</tbody>
</table>

d.f. = 9

* = Statistically significant at $\alpha = .025$
<table>
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<tr>
<th>Peak</th>
<th>t value</th>
<th>$\bar{X}_x$</th>
<th>S_x</th>
<th>$\bar{X}_y$</th>
<th>S_y</th>
</tr>
</thead>
<tbody>
<tr>
<td>*1</td>
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<td>13.82</td>
<td>29.25</td>
<td>1.25</td>
</tr>
<tr>
<td>*2</td>
<td>-21.95</td>
<td>no peak</td>
<td>-</td>
<td>17.80</td>
<td>2.20</td>
</tr>
<tr>
<td>*3</td>
<td>-4.04</td>
<td>4.56</td>
<td>2.72</td>
<td>12.85</td>
<td>1.15</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
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<td>5</td>
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<td>2.86</td>
<td>8.35</td>
<td>1.65</td>
</tr>
<tr>
<td>*6</td>
<td>3.38</td>
<td>17.38</td>
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<td>6.15</td>
<td>.15</td>
</tr>
<tr>
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<td>.26</td>
<td>1.82</td>
<td>1.90</td>
<td>1.45</td>
<td>.05</td>
</tr>
<tr>
<td>8</td>
<td>.40</td>
<td>9.27</td>
<td>9.78</td>
<td>6.40</td>
<td>.10</td>
</tr>
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<td>-3.42</td>
<td>3.30</td>
<td>4.57</td>
<td>14.70</td>
<td>.30</td>
</tr>
<tr>
<td>10</td>
<td>.58</td>
<td>1.88</td>
<td>4.37</td>
<td>no peak</td>
<td>-</td>
</tr>
</tbody>
</table>

d.f. = 9

* = Statistically significant at $\alpha = .025$
TABLE XI

Expired Air Pre ($n_x = 9$) versus Big Breath Pre ($n_y = 3$)

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>$\bar{X}$</th>
<th>S</th>
<th>$\bar{X}$</th>
<th>S</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>.12</td>
<td>52.78</td>
<td>15.84</td>
<td>51.60</td>
<td>4.24</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>-.46</td>
<td>2.26</td>
<td>1.06</td>
<td>2.55</td>
<td>.27</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>-.12</td>
<td>2.88</td>
<td>1.22</td>
<td>2.97</td>
<td>.68</td>
</tr>
<tr>
<td>6</td>
<td>-2.57</td>
<td>21.45</td>
<td>8.09</td>
<td>33.83</td>
<td>1.64</td>
</tr>
<tr>
<td>7</td>
<td>.30</td>
<td>6.34</td>
<td>12.83</td>
<td>4.04</td>
<td>2.01</td>
</tr>
<tr>
<td>8</td>
<td>.41</td>
<td>9.68</td>
<td>18.67</td>
<td>5.02</td>
<td>.26</td>
</tr>
<tr>
<td>9</td>
<td>.56</td>
<td>.23</td>
<td>.69</td>
<td>no peak</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>.93</td>
<td>4.44</td>
<td>6.50</td>
<td>no peak</td>
<td>-</td>
</tr>
</tbody>
</table>

d.f. = 10

* = Statistically significant at $\alpha = .025$
**TABLE XII**

Expired Air Pre (n_x = 9) versus Small Breath Pre (n_y = 2)

<table>
<thead>
<tr>
<th>Peak</th>
<th>t value</th>
<th>X</th>
<th>S</th>
<th>X</th>
<th>S</th>
</tr>
</thead>
<tbody>
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<td>-1.01</td>
<td>52.78</td>
<td>15.84</td>
<td>58.88</td>
<td>.37</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>-.23</td>
<td>2.26</td>
<td>1.06</td>
<td>2.43</td>
<td>.17</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>.36</td>
<td>2.88</td>
<td>1.22</td>
<td>2.55</td>
<td>.43</td>
</tr>
<tr>
<td>6</td>
<td>-.02</td>
<td>21.45</td>
<td>8.09</td>
<td>21.57</td>
<td>1.16</td>
</tr>
<tr>
<td>7</td>
<td>.10</td>
<td>6.34</td>
<td>12.83</td>
<td>5.41</td>
<td>1.80</td>
</tr>
<tr>
<td>8</td>
<td>.32</td>
<td>9.68</td>
<td>18.67</td>
<td>5.08</td>
<td>.13</td>
</tr>
<tr>
<td>9</td>
<td>-2.24</td>
<td>.23</td>
<td>.69</td>
<td>4.25</td>
<td>4.25</td>
</tr>
<tr>
<td>10</td>
<td>.93</td>
<td>4.44</td>
<td>6.50</td>
<td>no peak</td>
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</tr>
</tbody>
</table>

d.f. = 9

*= Statistically significant at \( \alpha = .025 \)
TABLE XIII

Big Breath Pre \((n_x = 3)\) versus Small Breath Pre \((n_y = 2)\)

<table>
<thead>
<tr>
<th>Peak</th>
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<th>S</th>
<th>(\bar{X})</th>
<th>S</th>
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</thead>
<tbody>
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<td>1</td>
<td>-2.30</td>
<td>51.60</td>
<td>4.24</td>
<td>58.88</td>
<td>.37</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>.506</td>
<td>2.55</td>
<td>.27</td>
<td>2.43</td>
<td>.17</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>.13</td>
<td>2.97</td>
<td>.68</td>
<td>2.55</td>
<td>.43</td>
</tr>
<tr>
<td>*6</td>
<td>8.56</td>
<td>33.83</td>
<td>1.64</td>
<td>21.57</td>
<td>1.16</td>
</tr>
<tr>
<td>7</td>
<td>-.68</td>
<td>4.04</td>
<td>2.01</td>
<td>5.41</td>
<td>1.80</td>
</tr>
<tr>
<td>8</td>
<td>-.09</td>
<td>5.02</td>
<td>.26</td>
<td>5.08</td>
<td>.13</td>
</tr>
<tr>
<td>9</td>
<td>-1.34</td>
<td>no peak</td>
<td>-</td>
<td>4.25</td>
<td>4.25</td>
</tr>
<tr>
<td>10</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
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d.f. = 3

*= Statistically significant at \(\alpha = .025\)
TABLE XIV

<table>
<thead>
<tr>
<th>Peak</th>
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<th>$\bar{X}$</th>
<th>S</th>
</tr>
</thead>
<tbody>
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<td>29.25</td>
<td>1.25</td>
<td>50.00</td>
<td>16.00</td>
</tr>
<tr>
<td>*2</td>
<td>21.95</td>
<td>17.80</td>
<td>2.20</td>
<td>no peak</td>
<td>-</td>
</tr>
<tr>
<td>*3</td>
<td>10.63</td>
<td>12.85</td>
<td>1.15</td>
<td>2.93</td>
<td>1.10</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>1.60</td>
<td>8.35</td>
<td>1.65</td>
<td>5.93</td>
<td>1.84</td>
</tr>
<tr>
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<td>7.38</td>
<td>6.15</td>
<td>0.15</td>
<td>17.72</td>
<td>5.61</td>
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<td>1.45</td>
<td>0.05</td>
<td>2.47</td>
<td>2.30</td>
</tr>
<tr>
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<td>6.40</td>
<td>0.10</td>
<td>15.63</td>
<td>18.09</td>
</tr>
<tr>
<td>*9</td>
<td>19.54</td>
<td>14.70</td>
<td>0.30</td>
<td>0.26</td>
<td>0.99</td>
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<td>-</td>
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<td></td>
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</tr>
</tbody>
</table>

* = Statistically significant at $\alpha = 0.025$
TABLE XV

Unlit Clipped Pre \( (n_y = 2) \) versus Unlit Clipped Post \( (n_y = 2) \)

<table>
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<tr>
<th>Peak</th>
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<th>S</th>
<th>( \bar{X} )</th>
<th>S</th>
</tr>
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<td>.98</td>
<td>86.65</td>
<td>9.30</td>
</tr>
<tr>
<td>2</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>-.04</td>
<td>2.43</td>
<td>.80</td>
<td>2.40</td>
<td>1.60</td>
</tr>
<tr>
<td>4</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>-1.83</td>
<td>4.06</td>
<td>1.32</td>
<td>.60</td>
<td>.60</td>
</tr>
<tr>
<td>6</td>
<td>-.79</td>
<td>19.31</td>
<td>3.11</td>
<td>9.85</td>
<td>9.85</td>
</tr>
<tr>
<td>7</td>
<td>no peak</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
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<td>4.62</td>
<td>.32</td>
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<td>9</td>
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<td>-</td>
<td>-</td>
<td>-</td>
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<td>7.78</td>
<td>no peak</td>
<td>-</td>
</tr>
</tbody>
</table>

d.f. = 3

* = Statistically significant at \( \alpha = .025 \)
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