

ASSOCIATION OF EXOGENOUS ESTROGENS AND CANCER IN HUMANS

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INTRODUCTION

When an epidemiologist is invited to discuss a specific carcinogen or class of carcinogens at a meeting composed primarily of laboratory scientists, the relevant human observations frequently are of great biologic interest in offering insights into basic mechanisms of carcinogenesis but of minor public health significance because of the relatively few humans exposed to the substance in question. This is often the case for therapeutic drugs which exhibit carcinogenic potential. In these circumstances the public health significance of exposure is even further diminished because the individuals exposed are often quite ill and willing to accept substantial risk of serious side effects in order to obtain the potential benefits of the therapeutic drugs in question.

Estrogenic drugs, however, certainly do not fit this description. These agents, for which there is abundant laboratory evidence of carcinogenicity, have been, and continue to be widely used at high doses for long periods of time by large numbers of healthy women. It is estimated that between 4 and 6 million Americans (mothers, daughters, sons) have been exposed to diethylstilbestrol (DES) during pregnancy.¹ A recent survey indicated that as many as 50% of all recently menopausal women in one area of the U.S. have taken estrogens for climacteric symptoms for a median duration of 10 years.² Current estimates also indicate that approximately 80 million women in the reproductive ranges throughout the world use oral contraceptives.³ Therefore, not only is the human species currently participating in a massive natural experiment to evaluate the potential carcinogenicity of these compounds, but the public health significance of even small alterations in carcinogenic risk due to these drugs is substantial.

As evidence of the level of concern over the carcinogenicity of these drugs, in the last two years I have participated in two comprehensive reviews of various aspects of this subject for two international agencies.^{3,4} However, new information is appearing at such a rapid rate, that such reviews need to be updated at least annually. In attempting to review the human

evidence with respect to potential carcinogenicity of a wide variety of estrogenic drugs, I will rely heavily on these two recent reviews, updated with subsequently published studies and the results of several studies completed but not yet published.

EARLY STUDIES

Until the mid 1960's, only a small proportion of the general population used estrogens, and only a very small proportion had used them for extended periods of time. Thus, the few early case-control studies to assess this exposure were inadequate to detect any alteration in cancer risk.⁵⁻⁷ Six follow-up (cohort) studies had been done of women intensively exposed to estrogen preparations either for symptoms of the climacteric or for treatment of osteoporosis.⁸⁻¹⁴ As recently as 1971 several of these studies were cited as indicating that estrogen replacement therapy was associated with "protection" against virtually all forms of cancer.¹⁵ If correct, the biologic and public health implications of these observations would have been impressive. However, little attention had been paid to these observations of apparent protection by either cancer researchers or the general public, a fact lamented by the author summarizing these observations in 1971.¹⁵ Perhaps part of the reason for this lack of attention arose from at least an implicit understanding of some of the deficiencies of these studies. In each of these studies, rather than identifying a group of exposed persons and then following them for cancer, information was simply extracted from existing medical records. Often a group was composed of only those patients who were followed up to a certain date; in others patients were included only up to the date they were last seen, with no additional follow-up efforts made. These methods could have resulted in underestimates of the cases of cancer occurring in these groups.

A few other comments concerning these early studies are in order since they are often resurrected in current debates over these medications. In several of the studies, only a small number of patients and/or a short follow-up period were involved. Most carcinogenic effects manifest themselves only after long latent periods, so the relevant follow-up period may occur only many years after initial exposure. Indeed, endocrine phenomena associated with changes in breast cancer risk (e.g., oophorectomy and an early age at natural menopause) do not exert their effect until about 10 years following the event.⁴ In addition, small cohorts and short follow-up make it likely that a relatively small or moderately increased risk will be

overlooked. With this in mind, it is important to note that in the 6 studies cited above: a) In two no women were followed more than 10 years^{8,14}; b) in one only 86 women were followed for more than 10 years¹¹; c) in one the number followed for 10 years could not be determined, but the longest follow-up was only 15 years¹⁰; d) in one although the longest follow-up was 25 years, the average for all 292 patients was 5 years, indicating that very few women were followed up for more than 10 years⁹; e) in one the entire study group consisted of those who had used the medication for 10 years, but the longest follow-up was only 14 years after starting use¹³. Many of these studies also suffered from flaws in their analyses. Such flaws included calculating expected numbers of cancers for periods of time during which the group was not under observation for malignancy, calculating expected numbers of cancers for organs that had been removed from these populations (e.g., the uterus and ovaries among women having undergone total hysterectomies), and making no adjustments for the protective effect against breast cancer of an oophorectomy or an early age at natural menopause.

In summary, prior to the early 1970's, the numbers of persons exposed to estrogens, particularly long term users, were too few for an adequate evaluation to be made by case-controls studies. In addition, each of the 6 early cohort studies was so deficient in either conduct or analysis that the results were uninterpretable.

DES DURING PREGNANCY

In 1971, the same year that the protective effect of estrogens was being promoted, an epidemiologic study was published which indicated that an unusual cluster of a rare form of vaginal cancer in females aged 14 to 22 years, noted at one hospital, was related to intrauterine exposure to DES by these young women.¹⁶ Later studies confirmed these results and indicated that the in-utero exposure related to an excess risk of clear cell adenocarcinoma both of the vagina and the cervix.¹ Shortly thereafter a registry of this disease in young women was established. Currently, this registry contains reports on over 350 cases of clear cell adenocarcinoma of the vagina or cervix.¹⁷ Among those cases with an available maternal pregnancy history, approximately 2/3 indicated in-utero exposure to DES or similar estrogens, such as hexestrol and dienestrol. While the relative rarity of this tumor, along with a lack of accurate estimates of exposure, have made an estimation of the actual risk of this malignancy among the exposed difficult,

a reasonable range for this estimate has been established. It appears that the incidence of clear cell adenocarcinoma is somewhere between 1 per 1,000 to 1 per 10,000 through the age of 24, among the exposed daughters.¹⁷ Since the use of DES during pregnancy was not common prior to the early 1950's, accurate estimates for what occurs beyond the age of 24 are not yet available. An analysis of the age-incidence pattern for this disease is quite interesting.¹⁷ The rates rise very sharply at age 14, peak at age 19, and then decline rapidly. The steepness of the ascending limb of this curve is noteworthy, considering that the relevant exposure occurred 15 to 20 years prior. Usually, if the latent period for a disease is long, it tends to have a wide range. In this circumstance, while the average latent period is 19 years, the range is quite constrained. It appears that something associated with puberty (perhaps the concomitant surge of endogenous estrogens) is acting as a powerful promoting agent, leading to the manifestation of this disease.

With the enthusiasm for describing various features of clear cell adenocarcinoma associated with in-utero DES exposure, it is easy to forget that the influence of this exposure on cancers of other hormonally sensitive sites has not yet been evaluated adequately. Recent reports have suggested, and denied, that cervical and vaginal intraepithelial neoplasia (squamous cell dysplasia and carcinoma in-situ) might be more common in DES exposed women.^{18,19} The women exposed to DES in-utero are just now entering the age range where cancers of the cervix and breast begin to appear, and it will be a number of years before they reach the high risk ages for cancers of the endometrium and ovary. Therefore, the need for continued evaluation of this exposure seems to be obvious.

Male Offspring

It has been noted recently that males exposed in-utero to DES demonstrate a number of teratogenic effects.²⁰ There is a clear excess of abnormalities of the external genital tract among exposed males. These consist primarily of a history of cryptorchidism and the finding of an increased number of hypoplastic testes and epididymal cysts. In addition, single semen determinations suggest an increase in sperm abnormalities, such as low sperm counts, decreased sperm motility, and possibly an increased number of abnormal sperm forms. The implications of these findings for potential carcinogenic effects are as yet unknown. However, two small case-control studies of testicular cancer have raised the suspicion of an increased risk of this tumor associated with in-utero exposure to DES.^{1,21}

Mothers

It is often overlooked that the mothers who took DES during the pregnancy had a substantial, albeit short-term, exposure to exogenous estrogens. The dose regimen most popular in the late 1940's and early 1950's called for between 10 and 12 grams of DES to be administered during the pregnancy. Currently the only information we have concerning the potential risks to the mother associated with this exposure comes from the follow-up of women who participated in a randomized clinical trial conducted at the University of Chicago in the early 1950's.²² Among this group, more patients in the DES-exposed group developed cancer in reproductive organs than among women not treated with DES. Thirty-two cases of breast cancer were observed in the 693 women exposed to DES compared to 21 cases in the 668 women in the comparison group. More cases of ovarian cancer (4 versus 1) and cancers of the uterine cervix (7 versus 3) occurred in the DES-exposed group than the comparison group. However, fewer cases of endometrial cancer (3 versus 5) occurred in the exposed in comparison to the unexposed. Thirty-eight of the exposed women have died compared to 28 in the comparison group. All of this difference was seemingly attributable to deaths due to cancers of the breast and gynecologic organs (e.g., 12 deaths were attributed to breast cancer among the exposed versus 4 among the comparison group). These observations provide some cause for serious concern about the carcinogenic potential of a large dose of DES for the breast and gynecologic organs of the mothers taking the medication. However, there is need for caution since the differences observed in this study were based on a relatively small number of cancers and could be due to chance alone. Further studies will be needed to confirm or deny the implications of these observations.

MENOPAUSAL ESTROGENS

As noted, early studies of menopausal estrogen use failed to identify any excess risk, and indeed had implied a substantial amount of "protection" against virtually all malignancy. Since 1975 there has been a dramatic reversal in the weight of the evidence concerning the carcinogenic consequences of the use of these substances.

Endometrial Cancer

Late in 1975, two case control studies were published which indicated that the use of conjugated estrogens for symptoms of the climacteric was associated with a relatively high risk of endometrial cancer.^{23,24} The first study, conducted in a cancer clinic, indicated a 4 to 8-fold increased

risk of endometrial cancer among estrogen users compared to nonusers. The second study, from a large prepaid health plan, indicated an 8-fold excess risk overall for users of estrogen, and a dose-response relationship with duration of use, rising to 14-fold among those who had used estrogens for 7 years or more. Both of these studies were based on record reviews. A subsequent study in a large retirement community utilized both health plan records and personal interviews and obtained results similar to the first two.²⁵ In addition, this study indicated a comparable relative risk associated with the use of nonconjugated estrogens, and a dose-response relationship with the dose of the tablet usually used. Since these first reports, eight independent investigations have found similar results using a wide variety of study designs.²⁶⁻³³ Three were case-control interview studies,²⁶⁻²⁸ two were case-control studies involving a review of medical records,^{29,30} two were case-control record-linkage studies in large group practices,^{31,32} and one was a cohort study of the frequency of subsequent primary malignancies of the endometrium among breast cancer patients treated with non-steroidal estrogens.³³ Since the first reports, the methods employed in a number of these studies have been criticized and questioned in a variety of ways.^{30,34} Targets for criticism have included the following: the use of control women who had diseases with risk factors different from those of endometrial cancer; the possibility of inadequate control for endometrial cancer risk factors in the analyses; an interval between first exposure and diagnosis that was too short to be consistent with current concepts of carcinogenesis; the exclusion of women who had had a hysterectomy from control groups; the accuracy of the endometrial cancer diagnoses; and the possibility of a surveillance bias (those using estrogens being more likely to have endometrial cancer diagnosed or diagnosed earlier than those not using estrogens). In the variety of studies that have been reported since the initial papers, and in several commentaries,^{4,34,35} each of these criticisms has been addressed adequately, without altering any conclusions concerning the association.

In addition, the conclusions of these analytic studies have been supported by evidence of rising incidence rates of endometrial cancer following the dramatic increase in use of estrogens for symptoms of the climacteric in the United States.^{36,37}

In summary, a number of recent studies utilizing a variety of designs have found a consistent, strongly positive association between a number of estrogenic substances and the risk of endometrial cancer, with positive dose-response relationships both with the strength of the medication and

with the duration of use. These observations have been supported by a dramatic rise in the incidence rates of endometrial cancer in concert with the dramatic increase in the use of these medications.

While most of the important issues have been addressed adequately by the current studies, there are at least two remaining issues that need to be addressed. First, no adequate evaluation has been made of the influence on endometrial cancer risk associated of the addition of progestational agents to estrogenic compounds used for hormonal replacement therapy. It has been suggested that this addition might at least partially diminish the risk of endometrial cancer in women undergoing estrogen therapy.^{38,39} Until such time as this has been evaluated, however, it should be noted that the sequential, cyclic use of estrogen and progestins in oral contraceptives has been related to an increased risk of endometrial cancer.⁴⁰ The other major issue requiring further data concerns the risk among women who have stopped using estrogens. Very recent evidence seems to indicate a plateauing of the incidence rates of endometrial cancer, and perhaps even a slight downturn in the rates, following quite closely on the dramatic reduction in use of menopausal estrogens after the initial reports in 1975.³⁷ In fact, a recent study of individuals in a large group practice indicated that the decline in incidence rate in this group practice following the reduction in estrogen use was due to the decrease in use, since the incidence rates among those using estrogens remained at the same high level.³² Very recently, the first study to attempt to address the risk among former users has been reported.²⁸ While the numbers of relevant observations are small, two features are noteworthy. First of all, even among former users who stopped some time ago, a substantial elevation in risk remains. However, after standardization for amount of estrogen received, there is evidence of a meaningful reduction in the excess risk of endometrial cancer very soon after the women stopped using the medications. These observations are particularly exciting for their immediate relevance to cancer prevention, and for their biologic implications with respect to understanding cancer initiation and promotion.

Breast Cancer

A number of reports from a cohort study carried out in Nashville, Tennessee have appeared in the literature since the early 1970's.⁴¹ Although this study had some of the same faults as those described under "early studies", it was better designed and analyzed and was the first that did not describe "protection" against breast cancer. Among the 735 women who were followed for an average of 15 years, 21 cases of breast cancer were observed versus

18 expected. A criticism of the development of the expected value in the study has indicated that it may be too high. In addition, although half of the total group had undergone bilateral oophorectomy, the anticipated protection against breast cancer due to this procedure did not occur.

In 1976, another, and much larger, cohort study was reported in which 1891 women given conjugated estrogens for symptoms of the climacteric were followed for an average of 12 years.⁴² Breast cancer was observed in 49, whereas 39 were expected on the basis of rates in the general population. The relative risk of breast cancer increased with duration of follow-up, progressing to about two-fold after 15 years. In addition, after ten years of follow-up observation, two factors related to lower risk of breast cancer, nulliparity and oophorectomy, were no longer so related. In this study, estrogen use was also related to an increased risk of breast cancer among women in whom benign disease developed after they had started the drug.

A number of recent case-control studies of breast cancer have shown no significant association with estrogen use; however, none of these have been able to address the question of long-term use.⁴³⁻⁴⁸

Since the cohort studies had raised the question of excess risk in long-term users, a number of case-control studies to evaluate this association have been initiated. As yet the results from these studies have not been published. However, I am aware of the preliminary results of at least three of these studies, all three of which seem to lend some support to the estimate of a two- to three-fold excess breast cancer risk among long term users of conjugated estrogens.⁴⁹⁻⁵¹

Ovarian Cancer

In a recent study, a statistically significant excess risk of ovarian cancer was reported among a small group of women who had been treated both with DES and conjugated estrogens for symptoms of the climacteric.⁵² In this study, there was no significant elevation of risk for those women who had received only conjugated estrogens. As noted previously, in a follow-up study of women exposed to DES, four women treated with DES during pregnancy subsequently developed ovarian cancer, compared with 1 in a control group of comparable size.²² On the other hand, one recent record linkage case-control study, and one interview case-control study have not found an association between conjugated estrogen use and the risk of ovarian cancer.^{53,54} Taken in the aggregate, the human observations, together with a suggestion of an association between DES and the development of ovarian cancers in laboratory animals,^{55,56} indicate the need for further investigation. Several studies

of ovarian cancer designed to address this issue are currently reaching their analysis phase.

Other Cancers

Two cohort studies since 1971 have reported lower than expected numbers of cancers other than those of the breast and reproductive system, and especially of colon cancer.^{41,42} While the lack of any evidence of a dose-response relationship for this association weakens arguments in favor of a protective effect, additional studies need to be done to explain these observations.

Benign Breast Disease

The only benign neoplasm that has been extensively evaluated for its relationship with the use of estrogens for symptoms of the climacteric is benign breast disease. However, the results of these evaluations have been conflicting. Three case-control studies have failed to find an association between estrogen use and the risk of surgically confirmed benign breast disease,^{43,47,57} while one case-control study has found a two and one half-fold excess risk.⁵⁸

ORAL CONTRACEPTIVES

The situation for oral contraceptives is somewhat different than that for DES and other estrogens used in treatment. For these other hormones, adequate human evaluations lagged behind the use of these medications for an unfortunately long period of time. The human exposure circumstances surrounding oral contraceptive use is truly a story unique in the annals of therapeutic drug history. Prior to 1960 essentially no one had used these agents outside of the clinical trial context. In less than ten years of their introduction in 1960, fully 60% of young women in this country had had significant exposure to these potent combinations of estrogenic and progestational agents.⁵⁹ As noted previously, current estimates are that 80 million women worldwide are using these medications for contraception.³ Because of this abrupt widespread use by a healthy population of potent physiologic agents for which there was laboratory evidence of carcinogenicity, a number of people started calling quite early for appropriate evaluations to be done in women.⁶⁰ Because of this concern, the literature on the subject is quite extensive.

Benign Breast Disease

With one exception, a number of case-control and cohort studies have been consistent in finding a deficit of benign breast disease in current oral contraceptive users. (Tables 1 and 2) This deficit is consistently observed

Table 1. Case-Control Studies of Oral Contraceptive Use: Benign Breast Neoplasia

Investigator and Year	Age Range (Years)	Disease*	Number of Cases	Relative Risk By Duration of Oral Contraceptive Use (Years)					
				0	<1	1-	2-	3- 4-	
Vessey (61) 1972	16-39	FC	117	1.0	—	1.0	—	0.3	—
		FA	86	—	—	—	—	—	—
Sartwell et al. (57) 1973	20-70	FC and Misc, FA	306	1.0	—	1.3	—	0.5	—
		Misc, FA	71	1.0	—	1.0	—	1.8	—
Boston Collaborative Drug Surveillance Program (43) 1973	20-44	FC	62	1.0	—	—	0.5	—	—
		FA	30	—	—	—	—	—	—
Kelsey et al. (62) 1978	20-44	FC	209	1.0	0.9	0.7	1.2	1.1	0.4
		FA	123	1.0	0.9	1.4	0.8	0.8	0.4
Fasal & Paffenbarger (63) 1975	15-49 And Misc.	FC, FA And Misc.	446	1.0	—	1.4	—	0.4	0.5
Nomura & Comstock (58) 1977	20-49	FC	275	1.0	—	—	1.0	—	—
		FA	45	—	—	—	—	—	—
Hoover et al. (64) 1978	26-50 and Misc.	FC, FA and Misc.	342	1.0	1.2	1.1	—	0.6	—
Ravnihar et al. (47) 1979	15-49	FC	266	1.0	—	0.9	—	0.4	—
		FA	106	1.0	—	1.2	—	0.6	—

* FC = Fibrocystic Disease

* FA = Fibroadenoma

* Misc = Miscellaneous

Table 2. Cohort Studies of Oral Contraceptive Use: Benign Breast Neoplasia

Investigator, Year and Period of Enrollment	Age Ranges of Cohorts at Enrollment (yrs.)	Disease* of Cases	Number of Cases	Relative Risk by Duration of Oral Contraceptive Use (Years)					
				0	<1	1-	2-	3- 4-	
Royal College of General Practitioners (65) 1974 (Subjects enrolled during 1968-1969)	15-49	FC,FA, and Misc (clinical diagnosis)	859	1.0	0.9	0.9	0.8	0.6	0.5
Ory et al. (66) 1976 (Subjects enrolled during 1970)	25-49	FC (histological diagnosis) FA (histological diagnosis)	499	1.0	0.9	0.7	0.4	0.5	0.5
Vessey et al. (67) 1976 (Subjects enrolled during 1968-1974)	25-39	FC,FA, and Misc. (clinical diagnosis)	263	1.0	1.2	0.5	0.5	0.5	0.5

*FC = Fibrocystic Disease

* FA = Fibroadenoma

* Misc = Miscellaneous

with respect to fibrocystic disease but has only been inconsistently linked to fibroadenoma. The apparent protective effect is related to duration of use and may persist for some time after cessation, however the association among former users has yet to be investigated adequately. For current users of oral contraceptives with a total exposure of longer than two years, the risk of being hospitalized for benign breast disease is only about 25% of those who have never used oral contraceptives.⁶¹ The cohort study from the Royal College of General Practitioners was able to take advantage of the popularity of two varieties of a particular brand of contraceptive.⁶⁸ The only difference between the two varieties was in the dose of progestin involved. This study seemed to indicate that the apparent protective effect was directly related to the strength of the progestational component.

Obviously, the important question is whether this apparent protective effect against benign breast disease will be relevant to breast cancer. Since benign breast disease identifies a group at high risk for breast cancer, these findings with respect to diminished risk of benign breast disease have been somewhat encouraging. However, a recent study indicates caution.⁶⁹ In this study, the cases of benign breast disease were reviewed and scored according to an index of ductal atypia. The marked protective effect associated with oral contraceptives seemed to apply primarily to the form of the disease associated with the least atypia (the form that may not be a risk factor for breast cancer). In fact, for the type of benign disease associated with the highest subsequent risk of breast cancer (the one with the most severe atypia), oral contraceptives were associated with an actual increased risk.

Breast Cancer

To date, studies on the relationship of oral contraceptive use to breast cancer have yielded inconclusive results.

Cohort studies have provided only limited information, due to the small numbers of incident cases observed thus far (Table 3). In one study, 16 cases have been reported and the lowest rate was among those using oral contraceptives, however, the differences were not significant.⁶⁷ In another cohort study, 31 cancers were reported, and the standardized rates were no different in users, ex-users, and nonusers.⁶⁵ It should be noted that in this study only 5% of women had used hormones for more than five years. In another cohort evaluation, hospitalization rates for breast cancers in users and nonusers based on 137 cases of cancer observed over a 30 month period yielded no significant differences between the rates in users and nonusers.⁶⁶

Table 3. Cohort Studies of Oral Contraceptive Use: Breast Cancer

Investigator and Year	Age Range (Years)	Number of Cases	Relative Risk by Duration (Years)		
			Never Used	1- 1-	2- 2-
Royal College of General Practitioners (65) 1974	15-49	31	1.0	1.1	1.1
Ory et al. (66) 1976	25-49	137	1.0	0.6	0.7
Vessey et al. (67) 1976	25-39	16	1.0	0.4	0.4

An increasing number of case-control evaluations of oral contraceptive use have been reported (Table 4). The most recent report from the ongoing case-control study at Oxford has the largest numbers of cases reported to date.⁷¹ Among the 621 total cases and their matched controls, there was no evidence of excess risk associated with ever use of oral contraceptives and no evidence of a dose-response relationship with number of years of use. When analyzed by age there was some evidence of excess risk in the oldest age group under study (ages 46-50). However, the trends in the next oldest age group (ages 41-45) were for the most part in the opposite direction and the authors interpreted this as evidence that the positive association was likely a result of chance, since they had investigated the risk in a number of subgroups. Among the 487 patients for whom clinical stage information was analyzed, those who had never used oral contraceptives had more advanced tumors at presentation than those using the pill in the year prior to diagnosis. The former pill users occupied an intermediate position with respect to clinical stage. These differences in clinical stage were reflected in differential survival patterns also. Since there was evidence that these differences were not due to a diagnostic (surveillance) bias, the authors suggested that these results may indicate that oral contraceptives may have had a beneficial effect on tumor growth and spread. Even in a study of this size, because of the recency of introduction of oral contraceptives only 3.5% of the control group had used oral contraceptives for more than 8 years (the longest duration of use category evaluated).

Additional studies have produced similarly negative results, with the same reservation as that of the Oxford study, that of a paucity of long-term users.

Two recent case-control studies may be worthy of separate note. In one, no significant differences were found in the risk of breast cancer between cases and controls who had ever used contraceptives.^{63,72} However, a positive association was noted for long-term contraceptive users who also had a history of surgically treated benign breast disease, and among a small group of women who had used oral contraceptives prior to their first childbirth. In the other study, no overall association between contraceptive use and breast cancer was noted.⁷⁰ However, among women with a natural menopause there was a consistent finding of excess risk among oral contraceptive users with evidence of a dose-response relationship for those women who had another breast cancer risk indicator (those who had a history of surgically treated benign breast disease, those who had a late age at first birth, those with a

Table 4. Case-Control Studies of Oral Contraceptive Use:
Breast Cancer (100 or More Cases)

Investigator and Year	Age Range (Years)*	Number of Cases	Relative Risk by Duration (Years) of Oral Contraceptive Use	
			Never Used	>2
Arthes et al. (5) 1971	15-75	119	1.0	0.8
Ravnihar et al. (47) 1979	20-49	190	1.0	1.0
Brinton et al. (70) 1979	PRE NAT	126 160	1.0	0.8
Vessey et al. (71) 1979	16-50	621	1.0	0.9
Paffenbarger et al. (72) 1977	15-49	452	1.0	1.1

* PRE = Premenopausal

* NAT = Natural Menopause

family history of breast cancer, and those with a late age at natural menopause). There are a variety of difficulties involved in interpreting studies in which a number of subgroups have been evaluated. Interpretation of the studies reported here is hampered by these difficulties. However, the observations of excess risk associated with long term use of oral contraceptives by women who are already at high risk because of the presence of another breast cancer risk indicator should be cause for concern, and should stimulate more intensive evaluations for possible synergistic effects. In addition, the suggestion of increased risk among oral contraceptive users who used the pills at a young age needs attention also. This time in a woman's life appears to be one when she is particularly sensitive to hormonal and other events that influence breast cancer risk.^{73,74}

Endometrial Cancer

Since 1975, numerous case reports have appeared concerning the development of endometrial carcinoma in young women with a history of use of sequential oral contraceptives. A report in 1977 concerning a series of 30 women under age 40 who both developed endometrial carcinoma and had a history of oral contraceptive use found that the proportion of users of sequential oral contraceptives among this group was much higher than expected from national rates of use of sequential versus combination agents.⁷⁵ This association became even stronger when women with other known risk factors for the disease, short durations of contraceptive use, or use for reasons other than contraception were removed from the analysis. In addition, women who developed endometrial carcinoma in association with sequential contraceptive use had fewer of the previously established risk factors for the disease than did a similar series of young endometrial cancer patients diagnosed prior to the introduction of oral contraceptives. The proportional exposure method used in this analysis is open to criticism. However, taken in the aggregate, there appears to be an increased, although not quantified at this time, risk of endometrial cancer among users of sequential oral contraceptives.

Cancer of the Uterine Cervix

Few data are available concerning the risk of invasive carcinoma of the cervix associated with use of oral contraceptives. The data available relate primarily to the risk of development of dysplasia and/or carcinoma in-situ of the uterine cervix. Thus, much of the evidence is made more difficult to interpret because of the various controversies in pathology and epidemiology concerning these entities.

A number of studies concerning the possible carcinogenic effects of oral contraceptive use on the uterine cervix have utilized data abstracted from programs for the cytological detection of cervical neoplasia by comparing the prevalence of cervical neoplasia in users and nonusers of oral contraceptives.⁷⁶⁻⁸⁵ These studies therefore have been based on data that were not collected with a view to research on the effects of oral contraceptives and they have yielded conflicting results and are difficult to interpret.

Four case-control studies have been conducted.⁸⁶⁻⁸⁹ Three have found no association, while the fourth⁸⁹ conducted among black women attending a screening program in Atlanta, Georgia, found a positive association with some evidence of a dose-response relationship (the risk rising to five-fold over that of nonusers for contraceptive users of three years or greater). While the results of this study were standardized for a number of factors, no information was available on a number of confounding factors directly related to sexual activity. Another problem was the substantial disagreement with the original histologic diagnosis of cancer in-situ on the part of one of the two pathologists who reviewed the slides.

Four cohort studies have thus far been reported.⁹⁰⁻⁹³ In two, no significant differences in cancer or in-situ precancerous lesions were found between contraceptive users and users of methods other than the diaphragm.^{90,91} Diaphragm users have been noted on a number of occasions to have a substantially reduced risk of cervical neoplasia. One cohort study concerning 17,942 women enrolled pre-paid health plan detected a significantly increased relative risk of cancer in-situ among oral contraceptive users, a risk which increased with duration of exposure.⁹²

A number of risk factors were taken into account into the analysis of this study, but information on risk factors related to sexual activity were not available. A subsequent investigation of these variables in this group indicated that when these factors were taken into account, the association between duration of oral contraceptive use and carcinoma in-situ remained, but was less marked.⁹⁴ This finding again illustrates the importance of sexual activity as a major confounding variable with regard to the study of cervical neoplasia and contraception.

In 1977, the results of a 7 year follow-up of a group of contraceptive users and nonusers was reported.⁹³ This study was a follow-up of patients with cervical dysplasia. Rates of progression of cervical dysplasia to carcinoma in-situ were compared for users of oral contraceptives and nonusers. Over 90% of the nonusers used intrauterine devices. The results of this

study suggested that extended oral contraceptive use (for 6 years or greater) appeared to increase by several times the rate of conversion of cervical dysplasia to carcinoma in-situ among women with dysplasia at the time they began to use oral contraceptives.

As this brief review indicates, the studies addressing the issue of cervical neoplasia and contraceptive use have been numerous, conflicting, and difficult to interpret. Detailed discussions of the methodologic issues involved could occupy a number of pages, and have been summarized elsewhere.^{3,4} The general conclusion that can be achieved at this time based on the available data would be that there is a suggestion of an increased risk of cervical dysplasia and carcinoma in-situ among long-term oral contraceptive users who also have other factors predisposing them to these conditions. However, to date, all of the potential sources of bias and confounding in these studies have not been controlled adequately, so this conclusion must remain a tentative one at this time.

Ovarian Cancer

Three recent studies have suggested that patients with ovarian cancer have a less frequent history of use of oral contraceptives than controls.^{54,95,96} It has also been noted that this apparent "protective" effect is biologically consistent with the other risk factors for ovarian cancer, which indicate that patients with "incessant" ovulatory activity tend to be a higher risk than those who have had less ovulatory activity.⁹⁶ It should also be emphasized that this apparent protective effect may be a relatively acute effect, with the long-term consequences of contraceptive use on ovarian cancer yet to be evaluated.

Liver Neoplasms

Increasing numbers of reports of hepatocellular adenomas in young women have appeared in the literature since 1973.^{97,98} These neoplasms, although benign, are highly vascular and often present as emergencies because of intrahepatic or abdominal hemorrhage with shock.

Two case-control studies have linked these tumors to the use of oral contraceptives.⁹⁹⁻¹⁰⁰ The relative risk associated with oral contraception is quite high (100 times that of nonusers for those who have used contraceptives for 3 to 5 years and over 500 times that of nonusers for those who have used for 7 years or more). The relative risk also appears to be higher for contraceptive users over age 30, and appears to be higher among contraceptive users who took pills with higher doses of estrogen and progestin. While the relative risk is high, the absolute risk does not appear to be

large for this rare tumor. Preliminary calculations suggest that the amount of hepatocellular adenoma among women under age 30 is no more than 3 per 100,000 contraceptive users per year.³ Over age 30 the absolute risk is greater, but not yet estimated.

Several malignant hepatomas of the liver have been reported among women using oral contraceptives. In one instance, such malignant tissue was found in an hepatic adenoma in a contraceptive user.¹⁰¹ Whether these reports indicate any excess risk or not is impossible to determine, since no controlled study has been conducted to date.

Malignant Melanoma

A possible association between oral contraceptive use and malignant melanoma of the skin was based on analysis of incidence data from a cohort of 17,942 women.¹⁰² A total of 22 cases were found during the period of observation and the age adjusted rate per 100,000 persons per year was 17.6 for those who had never used contraceptives, 24.1 for users of less than 4 years, and 29.5 for those using 4 years or longer. These differences were not statistically significant. As an adjunct to this study, an additional case-control study of 37 melanoma cases in the tumor registry of the same health plan, but not among women in the identified cohort, was conducted.¹⁰² The estimated relative risk for who had ever used oral contraceptives was 1.8, but again this excess was not statistically significant. The excess risk among users of contraceptives appeared to be localized to the lower limbs. In neither study was any information ascertained about exposure to sunlight, the most important known risk factor for malignant melanoma. If users of contraceptives are more likely to spend more time out doors than nonusers, this could have biased the results of these studies. Evaluations are underway to test this hypothesis after control for sunlight exposure.

Other Tumors

Several series of cases of adenoma of the pituitary have been reported in young women, a high proportion of whom had recently stopped using oral contraceptives.^{103,104} To date an adequate test of whether these tumors are related to contraceptive use has not been conducted.

In a review of 611 women who had been followed after the removal of a benign hydatidiform mole, approximately 10% subsequently developed an invasive mole.¹⁰⁵ Twenty-five percent of those who had taken oral contraceptives prior to the return of human choriogonadotropin levels to normal underwent this malignant transformation in comparison to about 9% of those who had not taken oral contraceptives. This suggests that increased development of invasive trophoblastic disease may be due to the use of oral contraceptives.

CONCLUSIONS

Diethylstilbestrol was first produced in 1938. That same year the occurrence of cancer was reported in animals exposed to DES.¹⁰⁶ Similar timely laboratory observations followed upon the introduction of conjugated estrogens and the various synthetic components of oral contraceptives. Unfortunately, appropriate human evaluations could not be carried out on the same agents until the proscribed latent periods associated with human tumors had elapsed. Unfortunately also, even when these latent periods had elapsed appropriate human evaluations were often not undertaken. The last eight years has seen an aggressive attempt by a number of investigators to rectify this lack of appropriate evaluations. This has led to the cascade of reports in the literature which this review has attempted to summarize. As indicated, many questions remain unanswered, new questions have been raised, and the appropriate latent periods for a number of tumors have not yet elapsed. However, a substantial leap in our understanding of the neoplastic effects of estrogenic medications in humans has occurred in this time. Unfortunately, most of the news is not good. In-utero exposure to DES has been firmly linked to vaginal and cervical clear cell adenocarcinoma. In addition, an association of this exposure with congenital malformations of the external genitalia in males has been established, and a suspicion of increased risk of testicular cancer has been raised. Similarly, suspicion of an excess risk of cancers of the breast and gynecologic organs among the mothers taking this medication has been raised. The influence of this in-utero exposure to daughters on other tumors (cervix, breast, etc.) must await the aging of the exposed cohort into the ages at high risk of these tumors.

Marketedly elevated risks of endometrial cancer have been clearly linked to use of menopausal estrogens and recent observations have also raised a distinct suspicion of increased breast cancer risk among long-term users of these medications.

The use of sequential oral contraceptives has been related to an increased risk of endometrial cancer in young women and the prolonged use of oral contraceptives have been firmly linked to benign, though definitely neoplastic, liver tumors. Suspicions have also been raised with respect to oral contraceptive use and increased risk of cancers of the breast and cervix, at least among specific groups of women (particularly high risk women). These suspicions are currently being aggressively evaluated. In addition, evidence linking these agents with the development of malignant melanoma, pituitary adenoma, and choriocarcinoma have appeared.

Even initial optimism over notations of diminished risk of benign breast disease associated with oral contraceptive use has recently been moderated with the observation that this may not apply to the premalignant form of benign breast disease. It is hoped that the initial observations of diminished risk of ovarian cancer among oral contraceptive users are also not subsequently reversed when long-term effects are evaluated adequately.

The public health consequences of the use of any medication are ultimately judged on a risk versus benefit basis. Adequate assessment of risks and benefits of estrogenic drugs will take some time to determine. In the interim, these evaluations of the numerous natural experiments underway in human beings should be utilized to their fullest to elucidate biologic mechanisms of hormonally related neoplasia. Perhaps in this way we will be able to link this material with laboratory results in order to identify those laboratory observations which are particularly relevant. Hopefully, in this way we can establish a scientific basis for evaluating the wisdom of allowing human exposure to a substance without having to wait 20 to 30 years.

1. DHEW, NIH, NCI, DES Task Force Members/Consultants (1978) DES Task Force Summary Report, September 21, 1978, pp. 1-67.
2. Stadel, B.V., and Weiss N. (1975) *Am. J. Epidemiol.* 102, 209-216.
3. Report of a WHO Scientific Group (1978) Steroid contraception and the risk of neoplasia. Technical Report Series 619. Geneva, pp. 1-54.
4. International Agency for Research on Cancer (1979) IARC Monographs on the Evaluation of Carcinogenic Risk, Vol. 21. WHO, Lyon, In press.
5. Arthes, F.G., Sartwell, P.E., and Lewison, E.F. (1971) *Cancer* 28, 1391-1394.
6. Jensen, E.I., Ostergaard, E. (1954) *Am. J. Obstet. Gynecol.* 67, 1094-1102.
7. Wynder, E.L., Escher, G.C., and Mantel, N. (1966) *Cancer* 19, 489-520.
8. Geist, S.H., Walter, R.I., and Salmon, U.F. (1941) *Am. J. Obstet. Gynecol.* 42, 242-248.
9. Wallach, S., and Henneman, P.H. (1959) *JAMA* 171, 1637-1642.
10. Schleyer-Saunders, E. (1962) *Med. Press* 244, 331-337.
11. Wilson, R.A. (1962) *JAMA* 182, 327-331.
12. Bakke, J.L. (1963) *West. J. Surg.* 71, 241-245.
13. Leis, H.P., Jr. (1966) *Int. Surg.* 45, 496-503.
14. Gordan, G.S., Picchi, J., and Roof, B.S. (1973) *Trans. Assoc. Am. Physicians* 86, 326-332.
15. Defares, J.G. (1971) *Lancet* I, 135-136.
16. Herbst, A.L., Ulfelder, H., and Poskanzer, D.C. (1971) *N. Engl. J. Med.* 284, 878-881.
17. Herbst, A.L., Cole, P., Colton, T., Robboy, S.J., and Scully, R.E. (1977) *Am. J. Obstet. Gynecol.* 128 (1), 43-50.
18. Staffl, A., Mattingly, R. F. (1974) *Am. J. Obstet. and Gynecol* 120, 666-667.

19. Robboy, S. J., Keh, P.C., Nickerson, R. J., Helmanis, E. K., Prat, J., Szyfelbein, W. M., Taft, P. D., Barnes, A. B., Scully, R. E., Welch, W. R. (1978) *Obstet Gynec* 51, 528-535.
20. Gill, W.B., Schumacher, G.F.B., and Bibbo, M. (1977) *J. Urol.* 117, 477-480.
21. Henderson, B.E., Benton, B., Jing, J., Yu, M.C., and Pike, M.C. (1979) *Int. J. Cancer* 23, 598-602.
22. Bibbo, M., Haenszel, W.M., Wied, G.L., Hubby, M., and Herbst, A.L. (1978) *N. Engl. J. Med.* 298, 763-767.
23. Smith, D.C., Prentice, R., Thompson, D.J., and Herrmann, W. C. (1975) *N. Engl. J. Med.* 293, 1164-1167.
24. Ziel, H.K., and Finkle, W.D. (1975) *N. Engl. J. Med.* 293, 1167-1170.
25. Mack, T.M., Pike, M.C., Henderson, B.E., Pfeffer, R.I., Gerkins, V.R., Arthur, M., Brown, S.E. (1976) *N. Engl. J. Med.* 294, 1262-1267.
26. Antunes, C.M.F., Stolley, P.D., Rosenstein, N.B., Davies, J.L., Tonascia, J.A., Brown, C., Burnett, L., Rutledge, A., Pokempner, M., and Garcia, R. (1979) *N. Engl. J. Med.* 300 (1), 9-13.
27. Wigle, D.T., Grace, M., Smith, E.S.O. (1978) *Can. Med. Assoc. J.* 118, 1276-1278.
28. Weiss, N.S., Szekely, D.R., English, D.R., and Schweid, A.I. (1979) *JAMA* 242, 261-264.
29. Gray, L.A., Christopherson, W.M., and Hoover, R. (1977) *Obstet. Gynecol.* 49, 385-389.
30. Horwitz, R.I., and Feinstein, A.R. (1978) *N. Engl. J. Med.* 299, 1089-1094.
31. McDonald, T.W., Annegers, J.F., O'Fallon, W.M., Dockerty, M.B., Malkasian, G.D., Kurland, L.T. (1977) *Am. J. Obstet. Gynecol.* 127, 572-580.
32. Jick, H., Watkins, R.N., Hunter, J.R., Dinan, B.J., Madsen, S., Rothman, K.J., and Walker, A.M. (1979) *N. Engl. J. Med.* 300, 218-222.
33. Hoover, R., Fraumeni, J.F., Jr., Everson, R., Myers, M.H. (1976) *Lancet* I, 885-887.
34. Weiss, N.S. (1977) in *Epidemiological Evaluation of Drugs*, Colombo, F., Shapiro, S., Slone, D., Tognoni, G. ed., PSG Publishing Company, Inc. pp. 161-166.
35. Hutchison, G.B., and Rothman, K.J. (1978) *N. Engl. J. Med.* 299, 1129-1130.
36. Weiss, N.S., Szekely, D.K., and Austin, D.F. (1976) *N. Engl. J. Med.* 294, 1259-1262.
37. Greenwald, P., Caputo, T.A., Wolfgang, P.E. (1977) *Obstet. Gynecol.* 50, 239-243.
38. Thom, M., White, P., Williams, R. M., Sturdee, D. W., Paterson, M. E. L., Wade-Evans, T., Studd, J. W. W. (1979) *Lancet* II, 455-457.
39. Quigley, M. M., and Hammond, C. B. (1979) *N. Engl. J. Med.* 301, 646-648.
40. Cohen, C.J., and Deppe, G. (1977) *Obstet. Gynecol.* 49, 390-392.
41. Burch, J.C., Byrd, B.F., and Vaughn, W.K. (1975) in *Frontiers of Hormone Research*. Vol. 3, Basel, Karger, S. ed., pp. 208-214.
42. Hoover, R., Gray, L.A., Cole, P., MacMahon, B. (1976) *N. Engl. J. Med.* 295, 401-405.
43. Boston Collaborative Drug Surveillance Program (1974) *N. Engl. J. Med.* 290, 15-19.
44. Casagrande, J., Gerkins, V., Henderson, B.E., Mack, T., and Pike, M.C. (1976) *J. Natl. Cancer Inst.* 56, 839-841.
45. Craig, T. J., Comstock, G. W., Geiser, P. B., (1974) *J. Natl. Cancer Inst.* 53, 1577-1581.
46. Mack, T. M., Henderson, B. E., Gerkins, V. R., Arthur, M., Baptista, Jr., and Pike, M. C., (1975) *N. Engl. J. Med.* 292, 1366-1371.
47. Ravnihar, B., Seigel, D.G., and Lindtner, J. (1979) *Europ. J. Cancer* 15, 395-405.

48. Sartwell, P.E., Arthes, F.G., Tonascia, J.A. (1977) *J. Natl. Cancer Inst.* 59, 1589-1592.
49. Hoover, R. Unpublished data.
50. Brinton, L.A. Personal communication.
51. Mack, T.M. Personal communication.
52. Hoover, R., Gray, L., and Fraumeni, J.F., Jr. (1977) *Lancet* II, 533-534.
53. Annegers, J.F., Strom, H., Decker, D.G., Dockerty, M.B., and O'Fallon, W.M. (1979) *Cancer* 43, 723-729.
54. McGowan, L., Parent, L., Lednar, W., and Norris, H.J. (1979) *Gynecol. Oncol.* 7, 325-344.
55. Jabara, A.G. (1962) *Aust. J. Exp. Biol.* 40, 139-152.
56. Owen, N.V., Pierce, E.C., and Anderson, R.C. (1972) *Toxic. Appl. Pharmac.* 21, 582-585.
57. Sartwell, P.E., Arthes, F.G., and Tonascia, J.A. (1973) *N. Engl. J. Med.* 288, 551-554.
58. Nomura, A., and Comstock, G.W. (1976) *Am. J. Epid.* 103, 439-444.
59. Rindfuss, R., and Westoff, C.F. (1974) *Demography* 11 (1), 75-87.
60. Report of a WHO Scientific Group (1968) *Hormonal Steroids in Contraception. Technical Report Series 386. Geneva*, pp. 1-28.
61. Vessey, M.P., Doll, R., Sutton, P.M. (1972) *Br. Med. J.* iii, 719-724.
62. Kelsey, J.L., Halford, T.R., White, C., Mayer, E.S., Kilty, S.E., and Acheson, R.M. (1978) *Am. J. Epidemiol.* 107, 236-244.
63. Fasal, E., and Paffenbarger, R.S. (1975) *J. Natl. Cancer Inst.* 55, 767-773.
64. Hoover, R., Bain, C., Cole, P., and MacMahon, B. (1978) *Am. J. Publ. Health* 68, 335-341.
65. Royal College of General Practitioners (1974) *Oral Contraceptives and Health*, Pitman Medical Publishing Company, London.
66. Ory, H., Cole, P., MacMahon, B., Hoover, R. (1976) *N. Engl. J. Med.* 294, 419-422.
67. Vessey, M., Doll, R., Peto, R., Johnson, B., and Wiggins, P. (1976) *J. Biosoc. Sci.* 8, 373-427.
68. Royal College of General Practitioners (1977) *Lancet* 1, 624.
69. LiVolsi, V.A., Stadel, B.V., Kelsey, J.L., Holford, T.R., and White, C. (1978) *N. Engl. J. Med.* 299, 381-385.
70. Brinton, L.A., Williams, R.R., Hoover, R.N., Stegens, N.L., Feinleib, M., and Fraumeni, J.F., Jr. (1979) *J. Natl. Cancer Inst.* 62, 37-44.
71. Vessey, M.P., Doll, R., Jones, K., McPherson, K., and Yeates, D. (1979) *Brit. Med. J.* 1, 1757-1760.
72. Paffenbarger, R.S., Fasal, E., Simmons, M.E., Kampert, J.B. (1977) *Cancer (Suppl.)* 39, 1887-1891.
73. MacMahon, B., Cole, P., Lin, T.M., Lowe, C.R., Mirra, A.P., Ravnihar, B., Salber, E.J., Valaoras, V.G., and Yuasa, S. (1970) *Bull. WHO* 43, 209-221.
74. Boice, J.D., and Stone, B.J. (1978) in *Late Biological Effects of Ionizing Radiation, Vol I. International Atomic Energy Agency, Vienna*, pp. 231-249.
75. Silverberg, S.G., Makowski, E.L., Roche, W.D. (1977) *Cancer* 39, 592-598.
76. Attwood, M.E. (1966) *J. Obstet. Gynaec. Brit. Cwlth.* 73, 662-665.
77. Berget, A., and Weber, T. (1974) *Danish Med. Bull.* 21, 172-176.
78. deBrux, J. (1974) *Sem. Hop. Paris* 50, 1491-1495.
79. Chai, M.S., Johnson, W.D., and Tricomi, V. (1970) *N.Y. State J. Med.* 70, 2663-2666.
80. Collette, H.J., Linthorst, G., deWaard, F. (1978) *Lancet* I, 441-442.
81. Kline, T.S., Holland, M., and Wemple, D. (1970) *Amer. J. Clin. Path.* 53, 215-222.

82. Liu, W., Koebel, L., Shipp, J., and Prisby, H. (1967) *Obstet. Gynecol.* 30, 228-232.
83. Melamed, M.R., Koss, L.G., Flehinger, B.J., Kelisky, R.P., and Dubrow, H. (1969) *Brit. Med. J.* iii, 195-200.
84. Pincus, G., and Garcia, C.R. (1965) *Metabolism* 14, 344-347.
85. Sandmire, H.F., Austin, S.D., Bechtel, R.C. (1976) *Am. J. Obstet. Gynecol.* 125, 339-345.
86. Thomas, D.B. (1972) *Obstet. Gynecol.* 40, 508-518.
87. Worth, A.J., and Boyes, D.A. (1972) *J. Obstet. Gynec. Brit. Cwlth.* 79, 673-679.
88. Boyce, J.G., Lu, T., Nelson, J.H., Fruchter, R.G. (1977) *Am. J. Obstet. Gynecol.* 128, 761-766.
89. Ory, H.W., Conger, S.B., Naib, Z., Tyler, C.W., Hatcher, R.A. (1977) in *Pharmacology of Steroid Contraceptive Drugs*, Garattini, S., and Berendes, H.W. ed., Raven Press, New York. pp. 211-218.
90. Melamed, M.R., and Flehinger, B.J. (1973) *Gynecol. Oncol.* 1, 290-298.
91. Wright, N.H., Vessey, M.P., Kenward, B., McPherson, K., and Doll R. (1978) *Br. J. Cancer* 38, 273-279.
92. Peritz, E., Rancharan, S., Frank, J., Brown, W.L., Huang, S., Ray R. (1977) *Am. J. Epidemiol.* 106, 462-469.
93. Stern, E., Forsythe, A.B., Youkeles, L., Coffeit, C.F. (1977) *Science* 196, 1460-1462.
94. Ramcharan, S., Personal Communication.
95. Newhouse, M.L., Pearson, R.M., Fullerton, J.M., Boesen, E.A., Shannon, H.S. (1977) *Br. J. Prev. Soc. Med.* 31, 148-153.
96. Casagrande, J.T., Pike, M.C., Ross, R.K., Louie, E.W., Roy, S., and Henderson, B.E. (1979) *Lancet* II, 170-173.
97. Baum, J.K., Bookstein, J.J., Holtz, F., and Klein, E.W. (1973) *Lancet* II, 926-929.
98. Mahboubi, E., and Shubik, P. (1976) *Cancer Letters* 1, 331-338.
99. Edmondson, H.A., Henderson, B., Benton, B. (1976) *N. Engl. J. Med.* 294, 470-472.
100. Rooks, J.B., Ory, H.W., Ishak, K.G., Strauss, L.T., Greenspan, J.R., Tyler, C.W. (1977) *Int. J. Gynaecol. Obstet.* 15, 143-144.
101. Davis, M., Portmann, B., Searle, M., Wright, R., Williams, R. (1975) *Br. Med. J.* 496-498.
102. Beral, V., Ramcharan, S., Faris, R. (1977) *Br. J. Cancer* 36, 804-809.
103. Chang, R.J., Keye, W.R., Jr., Young, J.R., Wilson, C.B., Jaffee, R.B. (1977) *Amer. J. Obstet. Gynecol.* 128, 356-363.
104. Sherman, B. M., Harris, E.E., Schlechte, J., Duello, T., Holmi, N.S., VanGilder, J., Chapler, F.K., Gronner, D.K. (1978) *Lancet* II, 1019-1020.
105. Stone, M., Dent, J., Kardana, A., Bagshaw, K.D. (1976) *Br. J. Obstet. Gynecol.* 83, 913-916.
106. Lacassagne, A. (1938) *Comptes Rendus des Seance de la Societe de Biologie* 129, 641-643.