Radiotracers Binding to Estrogen Receptors: I: Tissue Distribution of 17α-Ethynylestradiol and Moxestrol in Normal and Tumor-Bearing Rats

A. Feenstra, G. M. J. Nolten, W. Vaalburg, S. Reiffers, and M. G. Woldring

University Hospital, Groningen, The Netherlands

Ethynylestradiol and moxestrol can be labeled with carbon-11 by introducing this positron emitter in the 17α-ethynyl group. To investigate their potential as radiotracers binding to estrogen receptors, we studied the tissue distribution of tritiated ethynylestradiol and moxestrol, with specific activities of 57 Ci/mmol and 77–90 Ci/mmol, respectively, in the adult female rat. At 30 min after injection, both compounds showed specific uptake in the uterus (% dose/g): 2.52 for ethynylestradiol and of 2.43 for moxestrol. A decrease of the specific activity to 6–9 Ci/mmol resulted in uterine uptakes of 1.60 and 2.10 respectively, for ethynylestradiol and moxestrol, at 30 min. In the female rat bearing DMBA-induced mammary tumors, specific uptake was also measured in the tumors, although the values were only 25–30% of the uterine uptake. Moxestrol showed a greater uptake selectivity in the tumors compared with ethynylestradiol. From this study we conclude that ethynylestradiol and moxestrol have good potential as tracers binding to mammary tumors that contain estrogen receptors.

J Nucl Med 23: 599–605, 1982

During the last decade the relationship between the estrogen-receptor concentration and the response to endocrine therapy in human breast cancer has become well established (*1–3*). Patients with tumors lacking estrogen receptors are not likely to respond to endocrine therapy, whereas patients with tumors that show significant receptor concentrations have a better prognosis (*4*) and a chance to respond to endocrine therapy (*5*). An in vivo scintigraphic measurement of the estrogen-receptor content of breast tumors and their metastases would have many advantages over the present in vitro methods: inaccessible metastases could be investigated and the effect of therapy on the estrogen-receptor content could be followed. Most important, tumors and metastases that contain estrogen receptors could be identified more easily and rapidly. This in vivo determination has evoked much interest in developing radiopharmaceuticals capable of binding to the estrogen receptor (*6–19*). Gatley (*19*) recently reported on the tissue distribution of 17β-(16α-^{125}I)iodoestradiol in female Fischer rats bearing mammary adenocarcinoma. This labeled estrogen, introduced by Hochberg (*9,18*), has a high affinity for the estrogen receptor relative to estradiol, and a low binding to plasma proteins. Katzenellenbogen et al. (*14*) investigated halogenated estrogens. Of the fluorinated compounds 1-fluorohexestrol appears to be the most promising. They also prepared 17β(16α-^{77}Br)bromoestradiol (*16*). This compound showed high affinity for the estrogen receptor and demonstrated a receptor-specific uptake in the uterus of immature and mature rats and into DMBA-induced mammary tumors of the rat. Previously we suggested carbon-11 (20.4 min, β^+), an isotope of carbon suitable for positron-emission tomography, as a label for estrogen-receptor-binding radiopharmaceuticals. In 1976 we described the synthesis of carbon-11-labeled 17α-ethynylestradiol (*6*) and recently the synthesis of carbon-

Received Jan. 7, 1981; revision accepted Feb. 23, 1982.

For reprints contact: W. Vaalburg, PhD, Dept. of Nuclear Medicine, University of Groningen, Oostersingel 59, 9713 EZ Groningen, The Netherlands.

11-labeled 11β-methoxy, 17α-ethynylestradiol (moxestrol) (*17*). Moxestrol and 17α-ethynylestradiol are known as potent estrogens (*20*). They are bound with high affinity to the estrogen receptor in rat uterus and stimulate uterine growth in immature rats. Jensen (*21*) investigated the in vivo uptake of ethynylestradiol and Raynaud (*20*) the in vivo uptake of moxestrol in immature rats. However, these studies are not suitable for determination of the potential of the carbon-11-labeled analogs as estrogen-receptor-binding radiopharmaceuticals. In such experiments the half-life and achievable specific activity of carbon-11, as well as the demands of the detection in vivo must be considered.

Accordingly, we investigated the potential of these estrogens as estrogen-receptor seekers with the tritiated analogs (sp. act. 57–90 Ci/mmol). The tissue distribution of the steroids was determined in mature female rats and in rats bearing mammary tumors induced with dimethylbenz(a)anthracene (DMBA) within one hour after injection (~3 half-lives of C-11). The uptake in estrogen-receptor-containing tissues and the target-to-nontarget ratios were established.

We also report on the tissue distribution of tritiated 17α-ethynylestradiol and moxestrol with specific activities of 6–9 Ci/mmol. With our current facilities, this is the achievable specific activity of the carbon-11-labeled steroids, when the precursor C-11 acetylene is produced according to Crouzel (*22*).

MATERIALS AND METHODS

[6,7-^{3}H(N)]Estradiol* (51 Ci/mmol) and (11β-methoxy-^{3}H)moxestrol* (77–90 Ci/mmol) were purified on a chromosorb-ethanediol (5:1) column (5 × 0.6 cm). After washing the column with 10 ml of 7% ethyl acetate in iso-octane, estradiol(H-3) was eluted with 22.5% ethyl acetate in iso-octane. After column chromatography, the radiochemical purity was greater than 97%, as measured according to the manufacturer. 17α-[6,7-^{3}H(N)]Ethynylestradiol (53–58 Ci/mmol) was used without prior purification. The radiochemical purity was better than 98%. Other chemicals were obtained commercially: moxestrol (R 2858),* 17α-ethynylestradiol,† 17β-estradiol,† 35% aqueous formaldehyde,† Nuclear Chicago Solubilizer (NCS),‡ Picofluor,¶ Plasmasol,¶ 7,12-dimethylbenz(a)-anthracene (DMBA).§

Animals. For in vivo studies we used normal female Wistar rats (12–16 wk, 200 g). Every day a vaginal smear was made to determine the stage of the estrous cycle. Only rats that showed at least two consecutive cycles were used for this study. Mammary tumors were induced with DMBA in Sprague-Dawley rats. At age 7, 8, and 9 wk the rats received an intragastric feeding of 10 mg DMBA in 1 ml of olive oil. Animals with growing tumors 0.5–3 cm diameter were used for in vivo studies, no discrimination being made for the stage of the estrous cycle. The Wistar and Sprague-Dawley rats were housed in groups of five, maintained in air-conditioned surroundings under controlled lighting conditions, and given standard laboratory food and water ad libitum.

In vivo tissue distribution studies. The Wistar rats used in this study were at the metoestrus phase of the estrous cycle. Rats were injected through the tail vein under light ether anesthesia with a solution of the appropriate steroid in physiological saline containing 5–10% ethanol (5 μCi, 57–90 Ci/mmol). In control experiments animals received a 100-fold excess (2 μg) of nonradioactive estradiol immediately before injection of the tritiated steroid. In another experiment the radioactive steroids were diluted with the stable analogs to a specific activity of 6 Ci/mmol for 17α-ethynylestradiol(H-3) and 9 Ci/mmol for moxestrol(H-3). The injected dose was 5 μCi.

The Sprague-Dawley rats (250 g) bearing mammary tumors were used at random stages of the estrous cycle. They received 7.5 μCi of the diluted and undiluted tritiated steroids as described for the Wistar rats. In control experiments 3 μg of stable estradiol was used. At the indicated times the animals were decapitated. Blood was collected immediately in tubes containing a few drops of heparin (5000 IU/ml). Whole organs and the tissues were excised rapidly and samples of 0.05 to 0.15 g were digested overnight at 50°C in 1–1.5 ml of NCS tissue solubilizer. After cooling to room temperature, the colored samples were bleached with 0.1–0.5 ml of commercial bleaching water and neutralized with an equal volume of 35% aqueous formaldehyde. To 0.5 ml of blood an equal volume of water was added with gentle shaking, and 0.5 ml of bleaching water. After dicolorizing, 0.5 ml of 30% formaldehyde was added. All samples were counted in 15 ml of Plasmasol with an efficiency of 20–30%.

RESULTS

In vivo distribution studies in mature female rats. The in vivo characteristics of tritiated ethynylestradiol and moxestrol were first investigated in normal female rats at metoestrus, the stage of the cycle in which the estrogen level is low (*23*). Due to the short half-life of C-11, its application in labeled compounds is limited in time. We therefore determined the distributions of the tritium-labeled compounds at 15, 30, and 45 min after injection. In Tables 1 and 2 the tissue distributions of tritiated 17α-ethynylestradiol and moxestrol (specific activities 57 and 77 Ci/mmol) in mature rats are shown. The data are expressed as percentages of the injected dose per gram of wet weight (% dose/g). Both steroids showed pronounced uptakes in estrogen target tissue such as the uterus and ovaries, compared with the uptake in nontarget tissue. Moreover, organs involved in steroid metabolism, such as liver, kidneys, and intestine, displayed high uptake percentages. The amount of tritiated eth-

TABLE 1. TISSUE DISTRIBUTION (% DOSE/g) OF 17α-ETHYNYLESTRADIOL(H-3) WITH SPECIFIC ACTIVITY OF 57 Ci/mmol IN FEMALE RATS*

Organ	15 min	30 min	45 min	Control† 30 min
Blood	0.18 (0.14–0.27)	0.08 (0.07–0.08)	0.05 (0.04–0.06)	0.06 0.05–0.06)
Uterus	2.40 (2.04–2.91)	2.52 (1.42–3.31)	2.20 (1.81–2.84)	0.48 (0.41–0.62)
Ovary	2.20 (1.43–3.31)	1.35 (1.03–1.90)	1.08 (0.79–1.30)	0.45 (0.38–0.51)
Spleen	0.60 (0.51–0.81)	0.33 (0.28–0.46)	0.25 (0.20–0.33)	0.12 (0.10–0.24)
Kidney	1.35 (1.13–1.92)	0.83 (0.70–1.17)	0.55 (0.38–0.77)	0.30 (0.23–0.43)
Adrenals	4.52 (2.58–9.11)	1.47 (1.32–1.72)	0.92 (0.81–1.26)	0.70 (0.58–0.84)
Liver	4.62 (3.27–6.63)	2.40 (1.94–2.83)	1.75 (1.54–2.07)	1.50 (1.41–1.73)
Heart	0.60 (0.49–0.96)	0.30 (0.20–0.51)	0.15 (0.10–0.23)	0.15 (0.11–0.23)
Lung	0.82 (0.52–1.32)	0.37 (0.30–0.42)	0.25 (0.29–0.31)	0.18 (0.12–0.20)
Fat	0.42 (0.24–0.61)	0.47 (0.38–0.74)	0.60 (0.32–0.84)	0.48 (0.37–0.63)
Muscle	0.28 (0.25–0.40)	0.13 (0.10–0.21)	0.10 (0.10–0.10)	0.10 (0.10–0.10)
Small intestine	10.42 (8.01–11.83)	5.13 (3.61–7.24)	3.15 (1.76–7.02)	3.68 (1.72–7.10)

* Rats were at the metoestrus phase of their cycle. Mean of four rats, with range in parentheses. Dose: 5 μCi.

† Rats received a 100-fold dose of estradiol before the injection of 5 μCi 17α-ethynylestradiol(H-3) with a specific activity of 57 Ci/mmol.

ynylestradiol and moxestrol in the liver decreases rapidly owing to delivery of metabolized steroids in the small intestine. It is essential to know whether the observed uptakes of ethynylestradiol(H-3) and moxestrol(H-3) are due to the interaction of the steroids with the estrogen receptor in the target organs or to other steroid-protein interactions. Accordingly, in control experiments animals were injected with a 100-fold dose of stable estradiol just before the injection of the tritiated steroids, to saturate the estrogen receptor present in the target organs. As shown in Tables 1 and 2, this preinjection markedly reduced the uterine uptake of the two radiotracers, in contrast to that in nontarget organs. The ovarian uptake was only slightly decreased.

In the studies mentioned above, commercial tritiated steroids were used with a specific activity of 57–77 Ci/mmol. Because of the short half-life of carbon-11 and the carrier problem in the production and synthesis of carbon-11-labeled steroids, a specific activity of 57–77 Ci/mmol is still not realizable. We therefore investigated the uptake of tritiated 17α-ethynylestradiol and moxestrol with specific activities of 6–9 Ci/mmol, which is the specific activity currently feasible for these carbon-11-labeled steroids. Again mature cycling rats at metoestrous were injected with a dose of 5 μCi. Table 3 shows the uptakes of ethynylestradiol(H-3) and moxestrol(H-3) with these specific activities. The results show that the tissue distribution of the tritiated steroids with one tenth of the former specific activity followed the same pattern as those with the high specific activity. The effect of the reduced specific activity on the uptake of these tracers by the uterus is not significant at 30 min after injection, and this is also true for the 15- and 45-min time intervals (results not shown). This suggests that the steroid concentration is increased about tenfold with a tenfold increase of injected dose. Thus the injected dose (radioactive and stable steroid) of the preparations with high specific activity did not saturate the estrogen re-

TABLE 2. TISSUE DISTRIBUTION (% DOSE/g) OF MOXESTROL(H-3) WITH SPECIFIC ACTIVITY 77 Ci/mmol IN FEMALE RATS*

Organ	15 min	30 min	45 min	Control† 30 min
Blood	0.11 (0.08–0.14)	0.10 (0.07–0.12)	0.07 (0.06–0.08)	0.04 (0.04–0.05)
Uterus	1.64 (1.12–2.31)	2.43 (1.60–3.11)	2.34 (1.93–2.60)	1.04 (0.94–1.17)
Ovary	1.02 (0.86–1.31)	1.20 (0.99–1.61)	1.12 (0.94–1.51)	0.74 (0.59–0.85)
Spleen	0.28 (0.19–0.36)	0.36 (0.23–0.54)	0.24 (0.20–0.28)	0.24 (0.18–0.33)
Kidney	0.58 (0.46–0.72)	0.70 (0.54–0.79)	0.57 (0.48–0.63)	0.44 (0.33–0.53)
Adrenals	1.21 (0.84–1.52)	0.94 (0.71–1.24)	0.81 (0.66–0.89)	0.68 (0.48–0.81)
Liver	3.75 (2.93–4.35)	2.93 (2.39–3.27)	2.55 (2.16–2.95)	4.86 (4.49–5.42)
Heart	0.33 (0.23–0.40)	0.39 (0.27–0.56)	0.18 (0.16–0.23)	0.26 (0.24–0.30)
Lung	0.32 (0.23–0.39)	0.29 (0.21–0.36)	0.28 (0.18–0.34)	0.28 (0.20–0.34)
Fat	0.26 (0.17–0.35)	0.34 (0.29–0.39)	0.34 (0.26–0.45)	0.47 (0.35–0.56)
Muscle	0.18 (0.14–0.23)	0.15 (0.10–0.20)	0.11 (0.06–0.18)	0.14 (0.12–0.16)
Small intestine	4.87 (0.24–8.21)	3.32 (1.17–5.47)	1.84 (0.83–2.53)	5.88 (0.15–9.22)

* Rats were at the metoestrus phase of their cycle. Mean of four rats with range in parentheses. Dose: 5 μCi.

† Rats received a 100-fold dose of estradiol prior to the injection of 5 μCi moxestrol(H-3) with a specific activity of 77 Ci/mmol.

ceptors in the uterus. In Table 4 the uterus-to-blood ratios calculated from the data of the above mentioned experiments are shown.

In vivo studies in rats bearing DMBA-induced mammary tumors. These tumors are an acceptable model for the study of hormone dependence in human breast cancer. Most of these tumors are hormone dependent and contain estrogen receptors, although at a lower concentration than in the uterus (*24*). In rats bearing DMBA-induced mammary tumors we investigated the in vivo tissue distribution after injection of tritiated 17α-ethynylestradiol and moxestrol with two different specific activities. Rats were used at random stages of estrous cycle. The results of these experiments are summarized in Table 5. At high specific activity (57 and 90 Ci/mmol), both compounds showed tissue distributions similar to those seen in rats without tumors: high uterine uptake for ethynylestradiol (1.4%dose/g), and 2.6%dose/g for moxestrol. For both compounds the uptake in the mammary tumors is 25–30% of the uterine uptake. With 17α-ethynylestradiol(H-3) the tumor uptake is not significantly different from the uptake in fat (0.44 vs. 0.32%dose/g), but tumor uptake of moxestrol(H-3) is significantly higher than that in fat (0.67 vs. 0.22%dose/g).

Reduction of the specific activity of the tritiated steroids did not significantly decrease the uptake of radioactivity in the uterus and the tumors. As already mentioned in the previous section, this suggests that the estrogens were below the saturating amounts when high- and low-specific activity preparations were used.

As in normal rats, the estrogen specificity of the tissue distribution was investigated by preinjection of a 100-fold excess of stable estradiol. Again, preinjection of estradiol markedly reduced the uptake of the tritiated steroids in the uterus, as shown in Table 5. The reduction in tumor uptake was also significant.

TABLE 3. UPTAKE OF TRITIATED 17α-ETHYNYLESTRADIOL AND MOXESTROL (% DOSE/g), WITH SPECIFIC ACTIVITY 6 Ci/mmol AND 9 Ci/mmol RESPECTIVELY, IN FEMALE RATS*

Organ	17α-Ethynylestradiol	Moxestrol
Blood	0.06	0.10
	(0.06–0.07)	(0.09–0.12)
Uterus	1.60	2.10
	(1.11–2.21)	(1.19–2.53)
Ovary	0.75	1.01
	(0.67–0.88)	(0.64–1.40)
Kidney	0.34	0.51
	(0.28–0.39)	(0.42–0.63)
Adrenals	0.82	0.68
	(0.60–1.05)	(0.53–0.83)
Liver	1.57	3.01
	(1.20–1.94)	(1.99–4.17)
Fat	0.44	0.28
	(0.31–0.61)	(0.22–0.35)
Muscle	0.09	0.15
	(0.07–0.11)	(0.13–0.17)
Small intestine	3.88	4.87
	(1.56–7.34)	(3.87–6.12)

*Time = 30 min. Rats were at the metoestrus phase of their cycle. Mean of four rats with range in parentheses. Dose: 5 μCi.

DISCUSSION

In this work we used the tritiated analogs to investigate the potential of carbon-11-labeled 17α-ethynylestradiol and moxestrol as estrogen-receptor-binding radiopharmaceuticals. Because of the 20.4-min half-life of carbon-11, we determined the tissue distribution of tritiated 17α-ethynylestradiol and moxestrol (55 and 77–90 Ci/mmol) at 15–45 min after injection. Within this period, 17α-ethynylestradiol and moxestrol showed an estrogen-receptor-specific tissue distribution in mature female rats and in rats bearing DMBA-induced mammary tumors. Thus the use of carbon-11-labeled estrogens for in vivo studies would not be prevented by the half-life of carbon-11.

In spite of endogenous circulating estrogens, high uptake percentages could be obtained in the uterus (Tables 1, 2, and 5). The tumor uptakes of 17α-ethynylestradiol and moxestrol in the DMBA-induced tumors of the rat are about 25% of the uterine uptake (Table 5). One explanation might be the lower receptor concentration (*24*); another could be the difference in vascularization and content of components binding nonspecifically. The use of tumor-bearing rats at random stages of their cycle may also have affected the accumulation in uterus and tumor. The uptake of 17α-ethynylestradiol in the tumors is not significantly different from that in fat. In this experimental model, moxestrol appears to be more selective. Tumor uptakes are well above background. The difference of tumor-to-fat ratios of 17α-ethynylestradiol and moxestrol could be explained by the very low nonspecific binding of moxestrol (*20*). This emphasizes, as indicated by Raynaud (*20*), that the in vitro affinity for the estrogen receptor alone does not fully predict the in vivo behavior of the estrogen. Therefore the Binding Selectivity Index introduced by Katzenellenbogen (*15*) should be a better predictor for in vivo behavior of potential receptor-binding radiopharmaceuticals.

In a discussion of the potential of carbon-11-labeled estrogens, the specific activity of the tracer compound must be considered. For 17α-ethynylestradiol and moxestrol, the specific activity of the radioactive precursor, C-11 acetylene, is crucial. When the method published by Crouzel et al. (*22*) is used with the cyclotron in Groningen for the production of this precursor,

TABLE 4. UTERUS-TO-BLOOD RATIOS OF TRITIATED 17α-ETHYNYLESTRADIOL AND MOXESTROL IN RATS*

	Uterus/Blood			
	17α-Ethynylestradiol		Moxestrol	
Time	6 Ci/mmol	57 Ci/mmol	9 Ci/mmol	77 Ci/mmol
15 min	7.0	13.6	12.7	13.6
	(6.4–11.3)	(10.6–15.7)	(12.0–14.4)	(11.9–15.5)
30 min	25.0	35.8	21.0	24.9
	(17.5–33.4)	(19.2–39.4)	(13.9–24.1)	(14.6–33.6)
45 min	27.2	45.2	36.8	38.0
	(24.1–33.0)	(42.7–53.4)	(33.1–42.5)	(32.6–40.2)

* Rats were at the metoestrus phase of their cycle. Mean of four rats, with range in parentheses. Ratios are calculated from the data shown in Tables 1 to 3. Dose: 5 μCi.

TABLE 5. UPTAKE OF TRITIATED 17α-ETHYNYLESTRADIOL AND MOXESTROL (% DOSE/g) IN RATS BEARING DMBA-INDUCED TUMORS*

Organ	17α-Ethynylestradiol		Moxestrol	
	6 Ci/mmol	57 Ci/mmol	9 Ci/mmol	90 Ci/mmol
Blood	0.09 (0.07–0.10)	0.10 (0.09–0.12)	0.07 (0.07–0.09)	0.07 (0.07–0.08)
Uterus	1.25 (0.80–1.99)	1.38 (1.14–1.84)	1.70 (1.09–1.99)	2.60 (1.98–5.22)
Liver	1.42 (1.20–1.65)	1.68 (1.59–1.85)	2.40 (2.26–2.61)	3.54 (3.22–4.11)
Tumor	0.33 (0.21–0.53)	0.44 (0.24–0.60)	0.43 (0.20–0.62)	0.67 (0.12–1.01)
Fat	0.22 (0.16–0.28)	0.32 (0.28–0.40)	0.23 (0.18–0.25)	0.22 (0.16–0.35)
Muscle	0.09 (0.08–0.10)	0.10 (0.07–0.15)	0.11 (0.10–0.13)	0.13 (0.10–0.16)
Controls†				
Blood		0.08 (0.08–0.09)		0.09 (0.08–0.10)
Uterus		0.23 (0.16–0.30)		0.59 (0.57–0.63)
Liver		1.40 (1.35–1.46)		3.60 (3.58–3.61)
Tumor		0.16 (0.13–0.24)		0.27 (0.19–0.37)

* Time = 30 min. Rats were used at random stages of their cycle. Dose: 7.5 μCi. Mean of four rats with range in parentheses for at least 22 tumors.

† Rats received a 100-fold dose of estradiol before the injection of 7.5 μCi 17α-ethynylestradiol(H-3) (57 Ci/mmol) or 7.5 μCi moxestrol(H-3) (90 Ci/mmol). Mean of two rats with range in parentheses for at least 11 tumors.

the specific activity of these carbon-11-labeled steroids could be 5–10 Ci/mmol. We therefore investigated the tissue distribution of tritiated 17α-ethynylestradiol and moxestrol with specific activity 6–9 Ci/mmol. We showed that with both steroids high uterine uptakes and uterus-to-blood ratios can be obtained.

The extent of uptake and the uptake selectivity of the tumors are low with the tritiated estrogens of low specific activity. Thus tracer specific activity is more critical for the tumor than for the uterus with the dose used (~0.2 μg/animal).

We conclude that both 17α-ethynylestradiol and moxestrol, labeled with C-11, have good potential as receptor-binding radiopharmaceuticals. Their application is not hindered by the short half-life of carbon-11. Both steroids show a selective tissue distribution in mature female rats and in rats bearing DMBA-induced mammary tumors, although moxestrol seems to be more selective than 17α-ethynylestradiol in the tumor-bearing rat. However, the achievable specific activity of these carbon-11-labeled estrogens (5–10 Ci/mmol) is probably too low for reliable detection of tumors by current methods, and future research should be directed toward the design of production processes that result in carbon-11-labeled 17α-ethynylestradiol and moxestrol of high specific activity.

FOOTNOTES

* New England Nuclear, Boston, MA.
† Merck, Darmstadt, FRG.
‡ Amersham.
¶ Packard.
§ Fluca, Basel, Switzerland.

ACKNOWLEDGMENTS

This research was supported by a grant from the Dutch Cancer Foundation "Koningin Wilhelmina Fonds." We are grateful to Dr. W. de Boer, Dr. M. Gruber, and Dr. D. A. Piers for helpful discussions.

REFERENCES

1. MCGUIRE WL, CARBONE PP, VOLLMER EP, Eds: *Estrogen Receptors in Human Breast Cancer.* New York, Raven Press, 1975
2. LIPPMAN ME, ALLEGRA JC: Receptors in breast cancer. *N Engl J Med* 299:930-933, 1978
3. KOENDERS AJM, BEEX LVM, BENRAAD THJ: Steroid hormone receptors in human breast cancer. In *Perspectives in Steroid Receptor Research.* New York, Raven Press, 1980, pp 247-259
4. ALLEGRA JC, LIPPMAN ME, SIMON R, et al: Association between steroid hormone receptor status and disease-free interval in breast cancer. *Cancer Treat Rep* 63:1271-1277, 1979
5. BYAR DP, SEARS ME, MCGUIRE WL: Relationship between estrogen receptor values and clinical data in predicting the response to endocrine therapy for patients with advanced breast cancer. *Eur J Cancer* 15:299-310, 1979
6. VAALBURG W, REIFFERS S, BEERLING E, et al: The preparation of carbon-11 labeled 17α-ethynylestradiol. *J Lab Comp Radiopharm* 13:200-201, 1977
7. KOMAI T, ECKELMAN WC, JOHNSONBAUGH RE, et al: Estrogen derivatives for the external localization of estrogen-dependent malignancy. *J Nucl Med* 18:360-366, 1977
8. ECKELMAN WC, REBA RC, GIBSON RE, et al: Receptor-binding radiotracers: A class of potential radiopharmaceuticals. *J Nucl Med* 20:350-357, 1979
9. HOCHBERG RB: Iodine-125-labeled estradiol: a gamma-emitting analog of estradiol that binds to the estrogen receptor. *Science* 205,1138-1140, 1979
10. SPICER JA, PRESTON DF, BARANCZUK RJ, et al: The biodistribution of Br-77-labeled estrone: Visualization of tissues containing increased estrogen receptors. *J Nucl Med* 20:761-765, 1979
11. ARUNACHALAM T, LONGCOPE C, CASPI E: Iodoestrogens, syntheses, and interaction with uterine receptors. *J Biol Chem* 254:5900-5905, 1979
12. FEENSTRA A, DIJKSTRA JPH, NOLTEN GMJ, et al: An approach to the in vivo detection of hormone sensitive mammary tumors. *Cancer Treat Rep* 63:1146, 1979
13. MAZAITIS JK, GIBSON RE, KOMAI T, et al: Radioiodinated estrogen derivatives. *J Nucl Med* 21:142-146, 1980
14. KATZENELLENBOGEN JA, CARLSON KE, HEIMAN DF, et al: Receptor-binding radiopharmaceuticals for imaging breast tumors: Estrogen-receptor interactions and selectivity of tissue uptake of halogenated estrogen analogs. *J Nucl Med* 21:550-558, 1980
15. KATZENELLENBOGEN JA, HEIMAN DF, LLOYD JE, et al: In vitro binding assays as a guide for optimization of the structure of steroid receptor-binding radiopharmaceuticals: estimation of receptor vs. non-receptor binding. In *Proceedings of the Third International Symposium on Radiopharmaceutical Chemistry*, 1980, pp 85-87
16. KATZENELLENBOGEN JA, SENDEROFF SG, MCELVANY KD, et al: 16α-(^{77}Br)Bromoestradiol-17β: a high specific-activity, gamma-emitting tracer with uptake in rat uterus and induced mammary tumors. *J Nucl Med* 22:42-47, 1981
17. VAALBURG W, FEENSTRA A, WIEGMAN T, et al: Carbon-11 labelled moxestrol and 17α-methylestradiol as receptor binding radiopharmaceuticals. In *Proceedings of the Third International Symposium on Radiopharmaceutical Chemistry*, 1980, pp 100-101
18. HOCHBERG RB, ROSNER W: Interaction of 16α-(^{125}I) iodo-estradiol with estrogen-receptor and other steroid-binding proteins. *Proc Natl Acad Sci USA* 77:328-332, 1980
19. GATLEY SJ, SHAUGHNESSY WJ, INHORN L, et al: Studies with 17β(16α-[^{125}I])iodo-estradiol, an estrogen receptor-binding radiopharmaceutical in rats bearing mammary tumors. *J Nucl Med* 22:459-464, 1981
20. RAYNAUD JP, BOUTON MM, GALLET-BOURQUIN D, et al: Comparative study of estrogen action. *Mol Pharmcol* 9: 520-533, 1973
21. JENSEN EV, JACOBSEN HI: Basic guides to the mechanism of estrogen actions. *Rec Prog Hormone Res* XVIII: 387-414, 1962
22. CROUZEL C, SEJOURNE C, COMAR D: Production of ^{11}C-acetylene by methane pyrolysis. *Int J Appl Radiat Isotop* 30:566-568, 1979
23. HAWKINS RA, HILL A, FREEDMAN B, et al. Oestrogen receptor activity and endocrine status in DMBA-induced rat mammary tumors. *Eur J Cancer* 13:223-228, 1977
24. TSAI T-LS, KATZENELLENBOGEN BS: Antagonism of development and growth of 7,12-dimethylbenz(a)anthracene induced rat mammary tumors by the antiestrogen U 23, 469 and effects on estrogen and progesterone receptors. *Cancer Res* 37:1537-1543, 1977