

QUINACRINE HYDROCHLORIDE

Review and mode of action of an antimalarial, used as an occlusive agent
for transvaginal human sterilization

Eva Patek

*From the Department of Obstetrics and Gynecology, Karolinska Institutet, Huddinge University Hospital,
Huddinge, Sweden*

Abstract. Quinacrine hydrochloride, mainly used as an anti-malarial, has been used as a nontoxic chemosterilant in a transvaginal procedure in the human female. Clinical experiments indicate that Quinacrine acts as a powerful obstructive agent exclusively on the epithelium of the intramural portion of the tube without altering the histology of the endometrium.

The precise mechanism of Quinacrine's obstructive action on the mucosa of the uterotubal junction is unknown.

Its possible mode of action is binding to epithelial DNA thus forming a clot of granulomatous tissue, as Quinacrine is known to form adhesions when used in the control of neoplastic effusions.

Zinc is known to inhibit Quinacrine-DNA binding. The human endometrium, rich in Zinc is unaffected by Quinacrine, whereas the tubal cornua, with little Zinc promote the obstruction by Quinacrine DNA bondage.

The procedure is effective in 90 per cent of the cases with two instillations of Quinacrine. Further studies are essential to find agents that would potentiate the action of Quinacrine on the human Fallopian tube epithelium.

In the search for a transvaginal technique of sterilization of women where a nontoxic watersoluble substance could be applied directly into the uterine cavity, Jaime Zipper in Santiago di Chile has made extensive studies on the effect of various cytotoxic agents (42). He wanted a compound which would have a rather specific and selective effect upon the epithelium of the intramural portion of the Fallopian tubes and which would be relatively innocuous if it came in contact with the peritoneum by escape from the tubes, and to this effect he used a saturated solution of Quinacrine hydrochloride prepared in sterile distilled water. Originally a 4 ml suspension containing 1 g of the drug was instilled through a biopsy cannula introduced to the top of the endometrial cavity (45). Later, trials with more saturated solutions of Quinacrine were undertaken and he also studied the effects of chelating agents and other compounds

which would enhance the occlusive action of Quinacrine (40, 44). The procedure proved effective in 65 per cent of cases but had to be repeated once in 35 per cent in order to obstruct the tubes. A cumulative obstructive effect took place, and in 90 per cent of the cases no patency of the tubes existed after repeat insufflation and hysterosalpingography.

Spontaneous reversibility occurred infrequently, and only during the first year of treatment. Experiments in rats, however, (43) have shown that the hyperplastic reaction in the intramural portion of the tubes can be reversed by the administration of either an estrogen or a progesterone. Zipper suggests that it may be possible to reverse the reaction in the human as well as in the rat by the systemic administration of either estrogens or progesterons, or perhaps by a combination of both. If tubal patency can be restored clinically, then this procedure which was originally developed as a technique for nonsurgical female sterilization could become a reversible method of contraception, easily performed by paramedical personnel with minimum danger to the patient.

MATERIAL AND METHODS

Pharmacology. Quinacrine hydrochloride (Quinacrine, Atebrin/e/, Mepacrin/e/) is a bright yellow odourless crystalline powder with a bitter taste. Mol wt = 508.9. The compound is a derivate of acridine and has been used in the treatment of malaria since 1930. It has also been used as a remedy for at least thirty different ailments and its toxicology has been studied most extensively (9, 12, 26, 27, 29, 33). The LD₅₀ of Quinacrine hydrochloride for rats is 900 mg per kilogram at parenteral (stomach tube) administration (36). The LD₅₀ for the intraperitoneal route for rats has not been estimated, but the experiments of Keeler *et al* (21) show that it is approximately 250 mg per kilogram. They also described the development of entromegaly and scatorrhea in the rat following intraperitoneal Quinacrine hydrochloride.

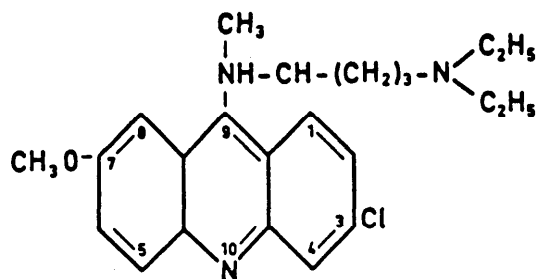


Fig. 1. Quinacrine chloride.

Morphological changes. The Fallopian tubes from 4 of Zig per's patients were removed one year after Quinacrine instillation by salpingectomy and the tubes were sectioned serially and studied microscopically (45). The histological changes were most marked in the cornual portion of the tubes and diminished rapidly and progressively for a distance of 2-3 mm. The remaining portions of the tubes were found to be normal. Within the affected area the lumen was obstructed by granulomatous fibrous tissue consisting of lightly stained cells having regular normal nuclei. No significant abnormalities were detected in the tubal musculature and Zipper found little effect upon the endometrium. So, the antifertility effect of Quinacrine hydrochloride is attributable to mechanical obstruction of the cornual portions of the Fallopian tubes (20, 34, 45).

RESULTS

Effect on carcinomatosis. Since the beginning of the sixties Quinacrine hydrochloride has been used for the control of recurrent neoplastic effusions. The response to this treatment has been favorable (6, 7, 8, 9, 15, 35, 38). By local instillation into serous cavities Quinacrine produced an inflammatory reaction, resulting in adhesions that partially or completely obliterated the serous cavities. Quinacrine hydrochloride is cytotoxic to a variety of normal and tumor cells grown in tissue culture but *in vivo* experiments show that the effect is mainly the production of scrostitis. The maintenance dosage varies, according to the Council on Drugs (6), from 200 mgs to 1 g daily, depending on the location of the effusion (pleura or peritoneum) and the continued tolerance of the patient.

Biochemical effect. Whitehouse and Boström (39) have found that Quinacrine hydrochloride significantly uncouples oxidative phosphorylation and Hemker and Hülsmann (14) have confirmed Hellerman's view (13) that Atebrin (Quinacrine) exhibits the relatively unspecific activity of a nonspecific enzyme inhibitor, acting by binding generally with proteins.

Specific effect on DNA. It is well known that Quinacrine hydrochloride forms a molecular complex with DNA (22). The acridine ring of the molecule is intercalated between the base pairs of double-helical DNA (23-25), and the aliphatic diamine side chain apparently bridges complementary DNA strands across the minor groove by ionic attraction to phosphate groups (1).

Consequences of the reaction of Quinacrine hydrochloride with DNA are *i)* inhibition of the enzymatic hydrolysis of DNA (22) and *ii)* inhibition of the DNA-dependent DNA and RNA polymerase reactions (1).

Biophysical studies on the nature of the Quinacrine-DNA complex (22-25), in addition to reports on inhibitions of DNA-dependent enzymatic reactions by the drug (1, 22) are consistent with the view that the mechanism of biological action of the drug is the specific reaction of Quinacrine hydrochloride with native, double-stranded DNA. This is also confirmed by the experiments of Ciak & Hahn in Washington (5) on the effects of Quinacrine on whole bacterial cells of *E. coli*. They found that the resulting mode of action is an impairment of DNA replication and, at a cytotoxic concentration, of RNA transcription.

Irvin & Irvin (16, 17, 18, 19) have found evidence for a reversible interaction of Quinacrine with nucleotides using spectrophotometric studies. They have also postulated, that the side chain of Quinacrine hydrochloride is not an absolute requisite for the interaction of the compound with nucleoproteins.

Investigations have been made by Mortland *et al* (28) on the absorption spectra of various basic dyes including the aminoacridines with nucleic acids. They found a formation of a dye-acid complex joined by more than simple electrostatic attractions. The complex binding is stronger for those acridines which have been shown to have an affinity for cell nuclei *in vivo*. The dye is more tightly bound to DNA than to RNA.

The affinity of Quinacrine for DNA has been used by Caspersson and collaborators (3, 4) to identify chromosomes. They found that Quinacrine mustard effects selective, discrete, fluorescent labelling of both plant and mammalian chromosomes. Chemically reactive loci may be differentiated along the linear axis of chromosomes by the use of ultramicrofluorometric techniques in combination with ultramicrospectrophotometric determination of the pattern of DNA distribution. Thus the Quinacrine-DNA complex can be used for the labelling and identification of

individual chromosomes.

Efforts have been **made** to assay Quinacrine in biological **material** (2, 37), mainly by spectrophotofluorometric methods. However, no accurate method exists to measure the content of DNA-Quinacrine in individual cell nuclei (30, 31). Thus, the Quinacrine DNA complex is still incompletely understood.

DISCUSSION

The effect of trace elements on the Quinacrine **DNA** complex. Trace elements are known to act as co-factors and cation antagonists in different enzyme systems (46). Studies by Hagenfeldt *et al.* (10, 11) indicate that the human endometrium is rich in Zinc. The Zinc content of the endometrium also shows a cyclic behaviour. This is in contrast to the endosalpingeal levels of Zinc which Patek & Hagenfeldt (32) found to be considerably lower and totally lacking cyclic changes.

The "binding" of Quinacrine to DNA in the intramural epithelium of the tube is dependent on the relatively rich content of Zinc in the endometrium and its more sparse content in the tubal mucosa. The relatively low concentrations of Zinc in the endosalpinx compared with that of the endometrium might be due to different levels of Zinc metalloenzymes, e.g. carbonic anhydrase and alkaline phosphatase. Further studies on these enzyme levels are necessary to find agents that could potentiate the occlusive action of Quinacrine hydrochloride on the intramural portion of the human Fallopian tube.

REFERENCES

- O'Brien, R. L., Olenick, J. G. & Hahn, F. E.: Reactions of Quinine, Chloroquine and Quinacrine with DNA and their effects on the DNA and RNA polymerase reactions. *Proc Nat Acad Sci US* 55:151 I, 1966.
- Brodie, B. B., Udenfriend, S., Dill, W. & Downing, G.: The estimation of basic organic compounds in biological material. *J Biol Chem* 168:311, 1947.
- Caspersson, T., Farber, S., Foley, G. E., Kudynowsky, J., Modest, E. J., Simonsson, E., Wagh, U. & Zech, L.: Chemical differentiation along metaphase chromosomes. *Exptl Cell Res* 49:219, 1968.
- Caspersson, T., Zech, L., Modest, E. J., Foley, G. E., Wagh, W. & Simonsson, E.: DNA-binding fluorochromes for the study of the organization of the metaphase nucleus. *Exptl Cell Res* 58:141, 1969.
- Ciak, J. & Hahn, F. E.: Quinacrine (Atebrin): Mode of action. *Science* 156:655, 1967.
- Council on Drugs: An agent for the palliative treatment of neoplastic effusions. Quinacrine (Atabrine) Hydrochloride *JAMA* 195: 1139, 1966.
- Dollinger, M. R., Krakoff, I. H. & Karnofsky, D. A.: Quinacrine in the treatment of neoplastic effusions. *Am Intern Med* 66:249, 1967.
- Gellhorn, A., Zaidenweber, J., Ultman, J. & Hirschberg, E.: The use of Atabrine (Quinacrine) in the control of recurrent neoplastic effusions. *Dis Chest* 39: 165, 1961.
- Goodman, L. S. & Gilman, A.: The Pharmacological Basis of Therapeutics, 2nd edition Quinacrine pp. 1167-1173. The Macmillan Co, New York, 1956.
- Hagenfeldt, K., Plantin, L.-O. & Diczfalusy, E.: Trace elements in the human endometrium. 1 Zinc, copper, manganese, sodium and potassium concentrations at various phases of the normal menstrual cycle. *Acta Endocr* 65:541, 1970.
- Hagenfeldt, K., Plantin, L.-O. & Diczfalusy, E.: Op cit. 2. Zinc, copper and manganese levels in the endometrium, cervical mucus and plasma. *Acta Endocr* 72: 115, 1973.
- Hecht, G.: Pharmakologisches **über** Atebrin. *Arch Exp Path* 170:328, 1933.
- Hellerman, L., Lindsay, A. & Bovarnick, M. P.: Flavoenzyme catalysis. Inhibition of d-aminoacid oxidase by competition with flavin-adenine-dinucleotide of Atabrine (Quinacrine), Quinine, and certain other compounds. *J Bio! Chem* 163:553, 1946.
- Hcmker, H. C. & Hülsmann, W. C.: Inhibition of enzymes by Atebrin. *Biochim Biophys Acta* 44:175, 1960.
- Hickman, J. A. & Jones, M. C.: Treatment of neoplastic pleural effusions with local instillations of Quinacrine (Mepacrine) Hydrochloride. *Thorax* 25:226, 1970.
- Irvin, J. L. & Irvin, E. M.: The interaction of antimalarials with nucleic acids. I Acridine. *Science* 110:426, 1949.
- Irvin, J. L. & Irvin, E. M.: Apparent ionization exponents of homologs of Quinacrine; Electrostatic effects. *J Am Chem Soc* 72:2743, 1950.
- Irvin, J. L. & Irvin, E. M.: The interaction of a 9 aminoacridine derivative with nucleic acids and nucleoproteins. *J Biol Chem* 206:39, 1954.
- Irvin, J. L. & Irvin, E. M.: The interaction of Quinacrine with adenine nucleotides. *J Biol Chem* 210:45, 1954.
- Israel, R.: Current concepts in female sterilization. *Clin Obstet Gynecol* 17:139, 1974.
- Keeler, R., Richardson, H. & Watson, A. J.: Enteromegaly and steatorrhea in the rat following intraperitoneal Quinacrine (Atabrine). *Lab Invest* 15:1253, 1966.
- Kurnick, N. B. & Radcliffe, I. E.: Reaction between DNA and Quinacrine. *J Lab Clin Med* 60:669, 1962.
- Lerman, L. S.: Structural considerations in the interaction of DNA and acridines. *J Molec Biol* 3:18, 1961.
- Lerman, L. S.: The structure of the DNA-acridine complex. *proc Nat Acad Sci US* 49:94, 1963.
- Lerman, L. S.: Acridine mutagens and DNA structure. *J Cell Comp Physiol* 64: 1, 1964.
- Martindale's Extra Pharmacopoeia 25th Ed.: Mepacrine Hydrochloride (cd. Norman. W. Blackow), pp. 326328. London, 1972.
- Merck Index 8th Ed. Quinacrine Hydrochloride, p. 900,

- MSD, 1968.
28. Morthland, F. W., De Bruyn, P. P. H. & Smith, N. H.: Spectrophotometric studies on the interaction of nucleic acids with aminoacridines and other basic dyes. *Exptl Cell Res* 7:201, 1954.
 29. Møller, K. O.: Mepakrin. In *Farmakologi 5 Udgave*, p. 114. Nyt Nordisk Forlag Arnold Busck, København, 1958.
 30. Patek, E.: Some cytochemical properties of isolated human endosalpingeal cells. *Acta Cytol* 18:414, 1974.
 31. Patek, E.: The epithelium of the human Fallopian tube. A surface ultrastructural and cytochemical study. *Acta Obstet Gynecol Scand Suppl* 3 1, 1974.
 32. Patek, E. & Hagcnfeldt, K.: Trace elements in the human Fallopian tube epithelium. Copper, Zinc, Manganese and Potassium in the menstrual cycle. *Int J Fertil* 19:85, 1974.
 33. *Paramacopoca Nordica, Editio Svecica Vol II.: Mepacrin chloridum pp. 362-365, Apotekarsocietens förlag, Stockholm 1964.*
 34. Porter, C. W. Jr. & Hulka, J. F.: Female sterilization in current clinical practise. *Fam Plann Perspect* 6:30, 1974.
 35. Rochlin, D. B., Smart, C. R., Wagner, D. E. & Silva, A. R. M.: The control of recurrent malignant effusions using Quinacrine hydrochloride. *Surg Gynecol Obstet* 991-994, May 1964.
 36. Siegel, H. & Mushett, W.: Structural changes following administration of Quinacrine hydrochloride. *Arch Path* 38:63, 1944.
 37. Udcnfriend, S., Duggan, D. E., Vasta, B. M. & Brodie, B. B.: A spectrophotofluorometric study of organic compounds of pharmacological interest. *J Pharmacol Exptl Therap* 120:26, 1957.
 38. Ultman, J.E., Gellhorn, A., Osnos, M. & Hirschberg, E.: The effect of Quinacrine on neoplastic effusions and certain of their enzymes. *Cancer (Philad)* 16:283, 1963.
 39. Whitcousc, M. W. & Boström, H.: Biochemical properties of anti-inflammatory drugs - VI. The effects of Chloroquine (Resochin), Mepacrine (Quinacrine) and some of their potential metabolites on cartilage metabolism and oxidative phosphorylation. *Biochem Pharmacol* 14:1173, 1965.
 40. Zipper, J. A. & Insunza, S.: Pharmacological agents that potentiate or inhibit the occlusive action of Quinacrine in the rabbit tube and rat uterus. In *Female Sterilization* (eds. G. W. Duncan, R. D. Falb and J. J. Speidel), pp 131-137. Academic Press, New York & London, 1972.
 41. Zipper, J. A., Medel, M., Pastene, L., Rivera, M. & Taturu, H. J.: Human fertility control by use of trace elements. In *Control of human fertility. Proc 15th Nobel Symposium* (eds. E. Diczfalusy and U. Borell), Almqvist & Wiksell, Uppsala, 1971.
 42. Zipper, J. A., Medel, M., Pastene, L. & Rivera, M.: Intrauterine instillation of chemical cytotoxic agents for tubal sterilization and treatment of functional metrorrhagias. *Int J Fertil* 14:280, 1969.
 43. Zipper, J. A., Pragcr, R. & Medel, M.: Biological changes induced by unilateral intrauterine instillation of Quinacrine in the rat and their reversal by either estradiol or progesterone. *Fertil Steril* 24:48, 1973.
 44. Zipper, J. A., Stachetti, E. & Medel, M.: Esterilizacion Feminina, Presentado en Bogota 24.S - 4.6. 1971. Personal communication.
 45. Zipper, J. A., Stachetti, E. & Medel, M.: Human fertility control by transvaginal application of Quinacrine on the Fallopian tube. *Fertil Steril* 21 :581, 1970.
 46. Zipper, J. A., Medel, M., Pastene, L. & Rivcra, M.: Human fertility control through the use of endouterine metal antagonisms of trace elements. (EMATE). In *Control of Human Fertility* (eds. E. Diczfalusy and U. Borell), pp. 199-218. Almqvist & Wiksell, Stockholm, 1970.

Submitted for publication March 9, 1978

Eva Patek, M.D.
 Department of Obstetrics and Gynecology
 Huddinge Hospital
 S-141 86 Huddinge
 Sweden