DIVISION OF NARCOTIC DRUGS
Vienna

RECOMMENDED METHODS FOR TESTING BENZODIAZEPINE DERIVATIVES UNDER INTERNATIONAL CONTROL

MANUAL FOR USE BY NATIONAL NARCOTICS LABORATORIES

UNITED NATIONS
New York, 1988

ST/NAR/16

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INTRODUCTION

Background

At its eighth special session in February 1984, the United Nations Commission on Narcotic Drugs decided to place 33 benzodiazepines under international control. This decision significantly increased the number of substances already under control and meant that national laboratories, concerned with the identification and analysis of drugs, would have to develop or have access to available analytical methods on most if not all of these substances. The international character of drug trafficking requires the speedy exchange of analytical data between laboratories and law enforcement authorities both on the national and the international levels. Development of internationally acceptable methods of testing would contribute greatly to the achievement of these objectives and this possibility has been under consideration for some time.

The Commission, at its tenth special session, reviewed the technical and scientific assistance programme of the Division of Narcotic Drugs with special emphasis on the development of laboratory methodologies. It noted with satisfaction that the harmonization of laboratory methods and the programme on establishment of recommended methods of testing for national forensic laboratories was pursued vigorously and that such methods had already been developed for heroin, cocaine, cannabis products, opium/crude morphine, amphetamine/methamphetamine and ring-substituted amphetamine derivatives.

In emphasizing the importance of the expert group meetings organized by the Division on various scientific and technical aspects of drug control and the high practical value for national law enforcement and laboratory services of the technical manuals as the outcome of the expert meetings, the Commission strongly recommended that such meetings and the publication of laboratory manuals continue on a regular basis.

Purpose of the manual

In accordance with the recommendation of the Commission on Narcotic Drugs, a group of eleven experts was convened in June 1988 in Ottawa, Canada, by the Division of Narcotic Drugs in cooperation and with the financial support of the Government of Canada through UNFDAC. The present manual published by the United Nations Division of Narcotic Drugs reflects the conclusions of the group of experts and has been designed to provide practical assistance to national authorities by describing recommended methods to be used in forensic laboratories for the identification and analysis of benzodiazepine derivatives under international control. The manual may also serve as a guide to national authorities in assessing existing methods used within their own government and university laboratories.

This manual is one in a series of similar publications dealing with the identification and analysis of various groups of drugs under international control; it was preceded by manuals on heroin (ST/NAR/6), cocaine (ST/NAR/7), cannabis (ST/NAR/8), amphetamine/methamphetamine (ST/NAR/9), opium/crude morphine (ST/NAR/11) and ring-substituted amphetamine derivatives (ST/NAR/12). Similar manuals on LSD and methaqualone/mecloqualone are in preparation.

These manuals suggest approaches that may help the forensic analyst to select a technique appropriate to the sample currently being examined. The analyst may then choose to follow any of the methods described in the manual, as each method can be expected to produce reliable analytical information with respect to the samples to which they are applied. Each method has been used for a number of years in reputable forensic laboratories and has been published in the scientific literature. In identifying these methods, the expert group was aware that many other useful and acceptable methods produce worthwhile analysis and information for the forensic analyst, and that a number of other acceptable options are recorded in the forensic scientific literature.

Use of the Manual

Few methods are perfect, least of all in forensic drug analysis where the materials under examination are very likely to show significant variation both in their physical form and chemical composition. The choice of methodology and approach to analysis remains within the control of the analyst working within his or her own country. The analyst alone has seen the suspect material and can best judge the correct approach to the problem at hand. Furthermore, the choice of methods may necessarily depend on the availability of reference materials and of instrumentation.

Not all methods listed need to be applied to all samples suspected to contain a benzodiazepine derivative. Requirements vary, for example, as a result of local trends in samples encountered, facilities available, and the standard of proof acceptable in the prosecution system within which the analyst works. The more complex methods are needed only for certain forensic requirements, such as comparison of samples or for source determination.

In order to establish the identity of any controlled drug, it is suggested that the criteria should be at least two independent analytical parameters. The selection of these parameters in any particular case would take into account the drug involved and the laboratory resources available to the analyst. For example, two uncorrelated TLC systems would count as two parameters. Uncorrelated TLC systems in this context means that either the solvent systems or the coating on the plates are completely different. When possible, three entirely different analytical techniques should be used, for example: colour test, chromatography (TLC, GLC or HPLC) and spectroscopy (IR or UV). The actual choice of parameters is left to the discretion of the chemist.

Attention is also drawn to the vital importance of the availability of textbooks on drugs of abuse and analytical techniques. Furthermore, the analyst must continually keep abreast of current trends in analysis, consistently following current analytical and forensic science literature. Analysts should refer to these and to previous manuals in this series for general descriptions of the analytical techniques included in this manual.

It is equally important that the latest information on changes in drugs available in the illicit traffic be quickly disseminated. This may often need to be done prior to publication in specialized periodicals dealing with forensic and other chemical analyses, since these publications are available to the forensic community some two to three years after the changes become known. The value of frequently published national reports on the latest information on such changes in drugs and on work being undertaken and analytical results obtained within individual laboratories cannot be over-emphasized.

The Division of Narcotic Drugs would welcome observations on the contents and usefulness of the present manual. Comments and suggestions may be addressed to:

Division of Narcotic Drugs
United Nations Office at Vienna
Vienna International Centre
P.O. Box 500
A-1400 Vienna, Austria

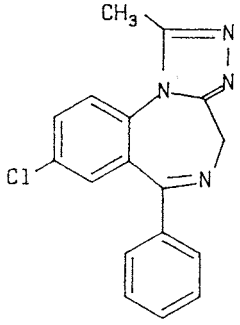
I. DESCRIPTION OF THE PURE COMPOUNDS

ALPRAZOLAM

8-chloro-1-methyl-6-phenyl-4H-s-triazolo[4,3-a][1,4]benzodiazepine

Scheduled under the "Convention on Psychotropic Substances 1971"

Alprazolam — Schedule IV



$C_{17}H_{13}ClN_4$
M.Wt. = 308.8

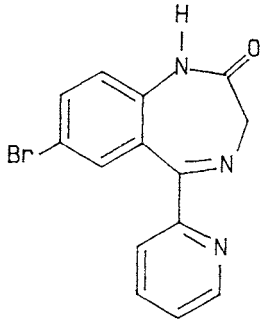
m.pt. = 228-228.5°C

BROMAZEPAM

7-bromo-1,3-dihydro-5-(2-pyridyl)-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Bromazepam — Schedule IV



$C_{14}H_{10}BrN_3O$
M.Wt. = 316.2

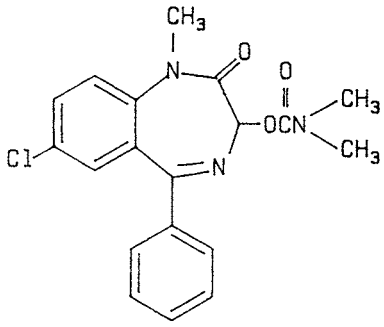
m.pt. = 237-238.5°C

CAMAZEPAM

7-chloro-1,3-dihydro-3-hydroxy-1-methyl-5-phenyl-2H-1,4-benzodiazepin-2-one dimethylcarbamate (ester)

Scheduled under the "Convention on Psychotropic Substances 1971"

Camazepam — Schedule IV



$C_{19}H_{18}ClN_3O_3$
M.Wt. = 371.8

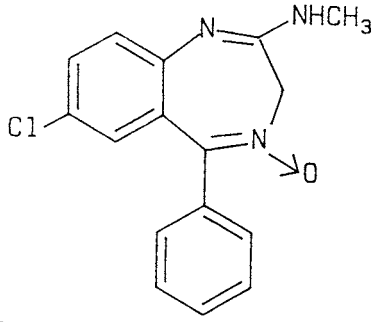
m.pt. = 173-174°C

CHLORDIAZEPOXIDE

7-chloro-2-(methylamino)-5-phenyl-3H-1,4-benzodiazepine-4-oxide

Scheduled under the "Convention on Psychotropic Substances 1971"

Chlordiazepoxide — Schedule IV



$C_{16}H_{14}ClN_3O$
M.Wt. = 299.8

m.pt. = 236-236.5°C

CHLORDIAZEPOXIDE HYDROCHLORIDE

$C_{16}H_{14}ClN_3O.HCl$
M.Wt. = 336.2

m.pt. = 212-218°C (decomp.)

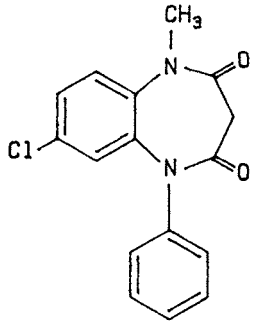
CLOBAZAM

7-chloro-1-methyl-5-phenyl-1H-1,5-benzodiazepine-2,4(3H,5H)-dione

Scheduled under the "Convention on Psychotropic Substances 1971"

Clobazam

Schedule IV



$C_{16}H_{13}ClN_2O_2$
M.Wt. = 300.7

m.pt. = 180-182°C

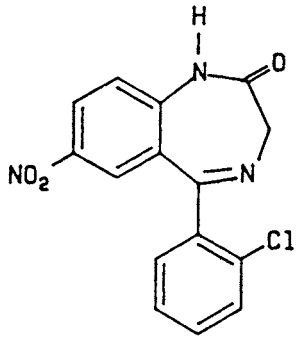
CLONAZEPAM

5-(o-chlorophenyl)-1,3-dihydro-7-nitro-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Clonazepam

Schedule IV



$C_{15}H_{10}ClN_3O_3$
M.Wt. = 315.7

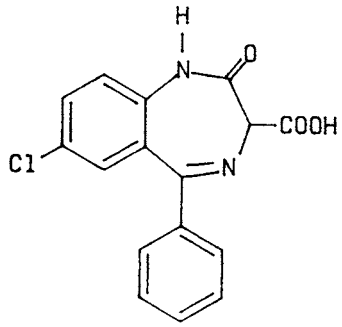
m.pt. = 238-240°C

CLORAZEPATE

7-chloro-2,3-dihydro-2-oxo-5-phenyl-1H-1,4-benzodiazepine-3-carboxylic acid

Scheduled under the "Convention on Psychotropic Substances 1971"

Clorazepate Schedule IV



$C_{16}H_{11}ClN_2O_3$
M.Wt. = 314.7

CLORAZEPATE DIPOTASSIUM

$C_{16}H_{11}ClK_2N_2O_4$
M.Wt. = 408.9 m.pt. = 209-211°C

CLORAZEPATE MONOPOTASSIUM

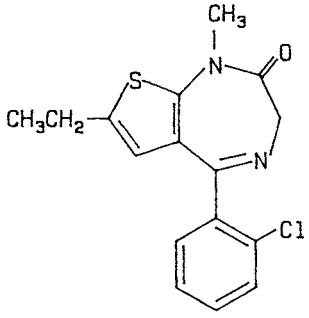
$C_{16}H_{10}ClKN_2O_3$
M.Wt. = 352.8

CLOTIAZEPAM

5-(o-chlorophenyl)-7-ethyl-1,3-dihydro-1-methyl-2H-thieno-[2,3-e]-1,4-diazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Clotiazepam — Schedule IV



$C_{16}H_{15}ClN_2OS$
M.Wt. = 318.8

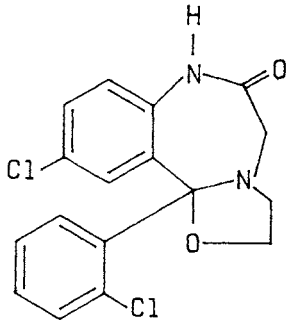
m.pt. = 105-106°C

CLOXAZOLAM

10-chloro-11b-(o-chlorophenyl)-2,3,7,11b-tetrahydro-oxazolo-[3,2-d][1,4]benzodiazepin-6(5H)-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Cloxazolam — Schedule IV



$C_{17}H_{14}Cl_2N_2O_2$
M.Wt. = 349.2

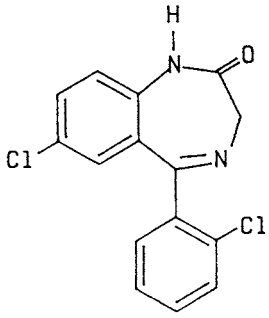
m.pt. = 202-204°C (decomp.)

DELORAZEPAM

7-chloro-5(o-chlorophenyl)-1,3-dihydro-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Delorazepam Schedule IV



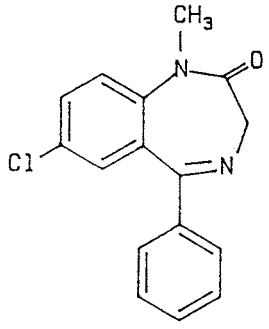
$C_{15}H_{10}Cl_2N_2O$
M.Wt. = 305.2 m.pt. = 198-199°C

DIAZEPAM

7-chloro-1,3-dihydro-1-methyl-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Diazepam Schedule IV



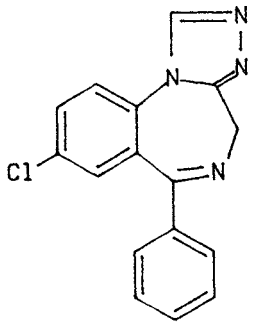
$C_{16}H_{13}ClN_2O$
M.Wt. = 284.7 m.pt. = 131-135°C

ESTAZOLAM

8-chloro-6-phenyl-4H-s-triazolo[4,3-a][1,4]benzodiazepine

Scheduled under the "Convention on Psychotropic Substances 1971"

Estazolam Schedule IV



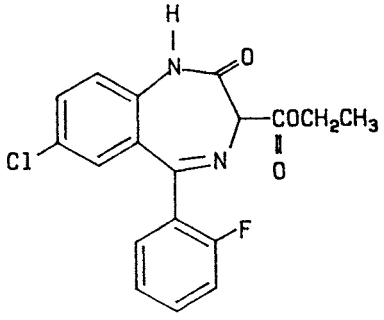
$C_{16}H_{11}ClN_4$
M.Wt. = 294.8 m.pt. = 227-233°C

ETHYL LOFLAZEPATE

Ethyl-7-chloro-5-(o-fluorophenyl)-2,3-dihydro-2-oxo-1H-1,4-benzodiazepine-3-carboxylate

Scheduled under the "Convention on Psychotropic Substances 1971"

Ethyl Loflazepate Schedule IV



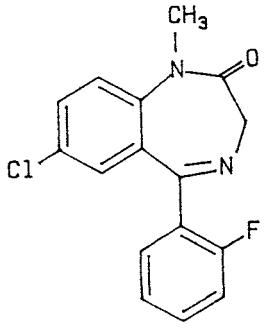
$C_{18}H_{14}ClFN_2O_3$
M.Wt. = 360.8 m.pt. = 193-194°C

FLUDIAZEPAM

7-chloro-5-(o-fluorophenyl)-1,3-dihydro-1-methyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Fludiazepam — Schedule IV



$C_{16}H_{12}ClFN_2O$
M.Wt. = 302.7

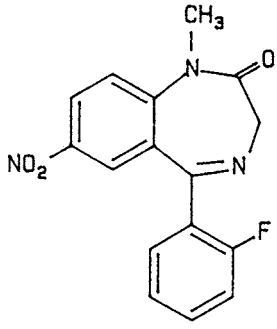
m.pt. = 88-92°C

FLUNITRAZEPAM

5-(o-fluorophenyl)-1,3-dihydro-1-methyl-7-nitro-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Flunitrazepam — Schedule IV



$C_{16}H_{12}FN_3O_3$
M.Wt. = 313.3

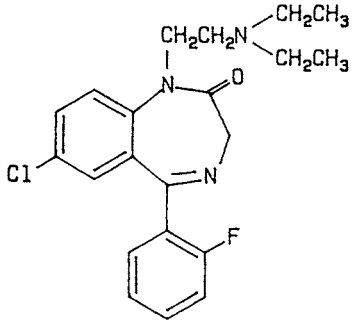
m.pt. = 166-167°C

FLURAZEPAM

7-chloro-1-[2-(diethylamino)ethyl]-5-(o-fluorophenyl)-1,3-dihydro-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Convention 1971"

Flurazepam — Schedule IV



$C_{21}H_{23}ClFN_3O$
M.Wt. = 387.9 — m.pt. = 77-82°C

FLURAZEPAM DIHYDROCHLORIDE

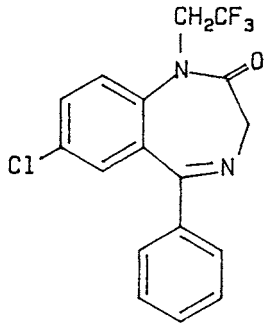
$C_{21}H_{23}ClFN_3O.2HCl$
M.Wt. = 460.8 — m.pt. = 212°C (decomp.)

HALAZEPAM

7-chloro-1,3-dihydro-5-phenyl-1-(2,2,2-trifluoroethyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Halazepam — Schedule IV



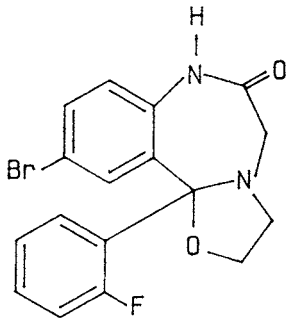
$C_{17}H_{12}ClF_3N_2O$
M.Wt. = 352.7 — m.pt. = 164-166°C

HALOXAZOLAM

10-bromo-11b(o-fluorophenyl)2,3,7,11b-tetrahydrooxazolo[3,2-d]-[1,4]benzodiazepin-6(5H)-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Haloxazolam — Schedule IV



$C_{17}H_{14}BrFN_2O_2$
M.Wt. = 377.2

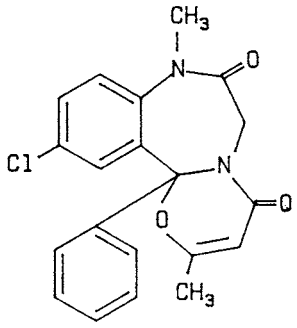
m.pt. = 185°C

KETAZOLAM

11-chloro-8,12b-dihydro-2,8-dimethyl-12b-phenyl-4H-[1,3]oxazino[3,2-d][1,4]benzodiazepine-4,7(6H)-dione

Scheduled under The "Convention on Psychotropic Substances 1971"

Ketazolam — Schedule IV



$C_{20}H_{17}ClN_2O_3$
M.Wt. = 368.8

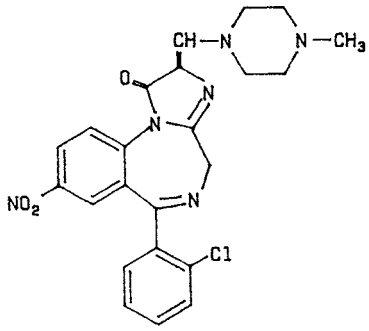
m.pt. = 182-183.5°C

LOPRAZOLAM

6-(o-chlorophenyl)-2,4-dihydro-2-[(4-methyl-1-piperazinyl)-methylene]-8-nitro-1H-imidazo[1,2-a][1,4]benzodiazepin-1-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Loprazolam — Schedule IV



$C_{23}H_{21}ClN_6O_3$
M.Wt. = 464.9

m.pt. = 214-215°C

LOPRAZOLAM MESILATE

$C_{23}H_{21}ClN_6O_3.CH_4O_3S.H_2O$
M.Wt. = 579.0

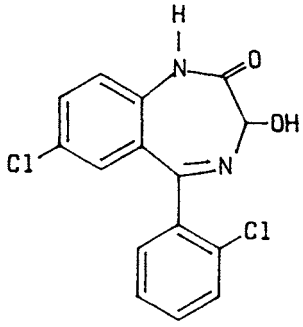
m.pt. = 242-245°C (decomp.)

LORAZEPAM

7-chloro-5-(o-chlorophenyl)-1,3-dihydro-3-hydroxy-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Lorazepam — Schedule IV



$C_{15}H_{10}Cl_2N_2O_2$
M.Wt. = 321.2

m.pt. = 166-168°C

LORAZEPAM MESILATE

$C_{15}H_{10}Cl_2N_2O_2.CH_4O_3S$
M.Wt. = 417.3

LORAZEPAM PIVALATE

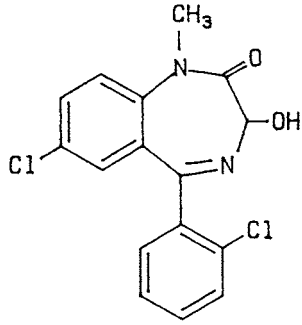
$C_{15}H_{10}Cl_2N_2O_2.C_5H_{10}O_2$
M.Wt. = 423.3

LORMETAZEPAM

7-chloro-5-(o-chlorophenyl)-1,3-dihydro-3-hydroxy-1-methyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Lormetazepam — Schedule IV



$C_{16}H_{12}Cl_2N_2O_2$
M.Wt. = 335.2

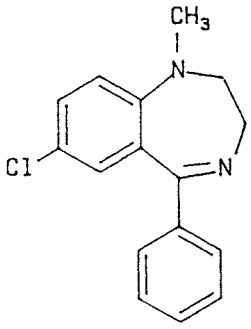
m.pt. = 209-211°C

MEDAZEPAM

7-chloro-2,3-dihydro-1-methyl-5-phenyl-1H-1,4-benzodiazepine

Scheduled under the "Convention on Psychotropic Substances 1971"

Medazepam — Schedule IV



$C_{16}H_{15}ClN_2$
M.Wt. = 270.8

m.pt. = 95-97°C

MEDAZEPAM HYDROCHLORIDE

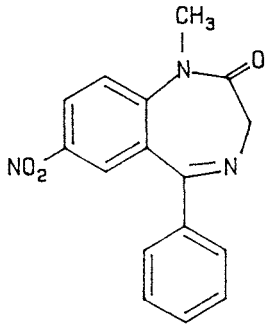
$C_{16}H_{15}ClN_2.HCl$
M.Wt. = 307.2

NIMETAZEPAM

1,3-dihydro-1-methyl-7-nitro-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Nimetazepam Schedule IV



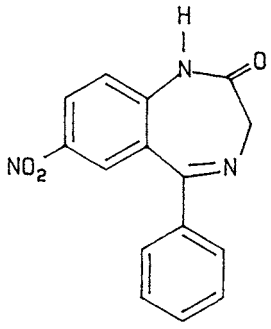
$C_{16}H_{13}N_3O_3$
M.Wt. = 295.3 m.pt. = 156.5-157.5°C

NITRAZEPAM

1,3-dihydro-7-nitro-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Nitrazepam Schedule IV



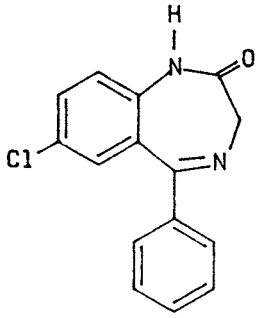
$C_{15}H_{11}N_3O_3$
M.Wt. = 281.3 m.pt. = 224-226°C

NORDAZEPAM

7-chloro-1,3-dihydro-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Nordazepam Schedule IV



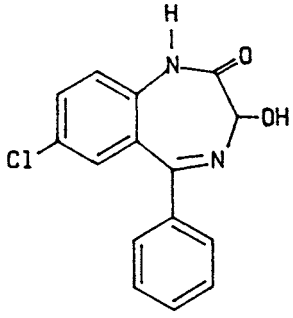
$C_{15}H_{11}ClN_2O$
M.Wt. = 270.7 m.pt. = 216-217°C

OXAZEPAM

7-chloro-1,3-dihydro-3-hydroxy-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Oxazepam Schedule IV



$C_{15}H_{11}ClN_2O_2$
M.Wt. = 286.7 m.pt. = 204-206°C

OXAZEPAM HEMISUCCINATE

$C_{15}H_{11}ClN_2O_2.1/2\ C_4H_6O_4$
M.Wt. = 345.8

OXAZEPAM SUCCINATE

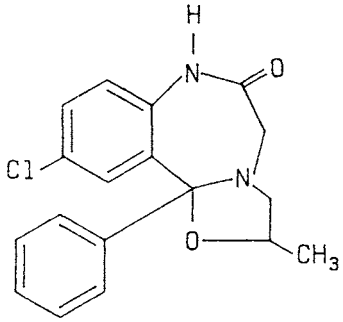
$C_{15}H_{11}ClN_2O_2.C_4H_6O_4$
M.Wt. = 404.8

OXAZOLAM

10-chloro-2,3,7,11b-tetrahydro-2-methyl-11b-phenyloxazolo[3,2-d][1,4]benzodiazepin-6(5H)-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Oxazolam — Schedule IV



$C_{18}H_{17}ClN_2O_2$
M.Wt. = 328.8

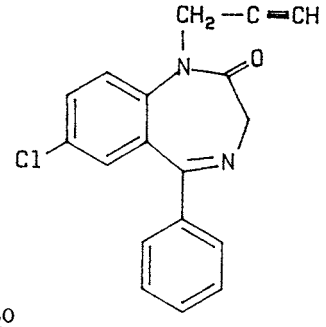
m.pt. = 186-188°C

PINAZEPAM

7-chloro-1,3-dihydro-5-phenyl-1-(2-propynyl)-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Pinazepam — Schedule IV



$C_{18}H_{13}ClN_2O$
M.Wt. = 308.8

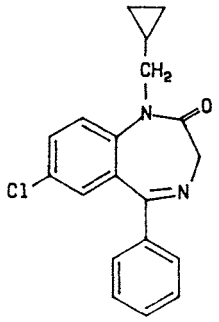
m.pt. = 140-142°C

PRAZEPAM

7-chloro-1-(cyclopropylmethyl)-1,3-dihydro-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Prazepam — Schedule IV



$C_{19}H_{17}ClN_2O$
M.Wt. = 324.8

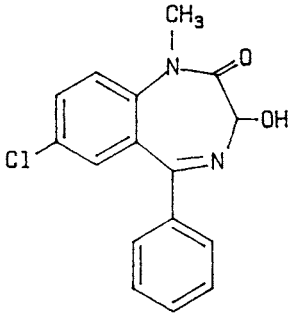
m.pt. = 145-146°C

TEMAZEPAM

7-chloro-1,3-dihydro-3-hydroxy-1-methyl-5-phenyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Temazepam — Schedule IV



$C_{16}H_{13}ClN_2O_2$
M.Wt. = 300.7

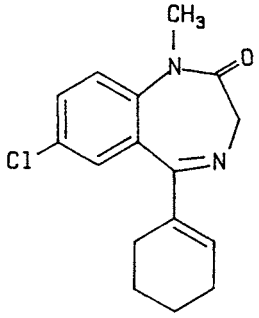
m.pt. = 156-159°C

TETRAZEPAM

7-chloro-5-(cyclohexen-1-yl)1,3-dihydro-1-methyl-2H-1,4-benzodiazepin-2-one

Scheduled under the "Convention on Psychotropic Substances 1971"

Tetrazepam Schedule IV



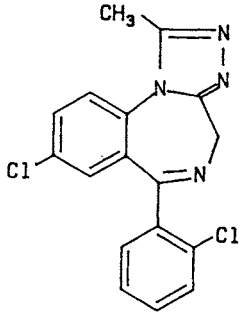
$C_{16}H_{17}ClN_2O$
M.Wt. = 288.8 m.pt. = 144°C

TRIAZOLAM

8-chloro-6(o-chlorophenyl)-1-methyl-4H-s-triazolo[4,3-a][1,4]benzodiazepine

Scheduled under the "Convention on Psychotropic Substances 1971"

Triazolam Schedule IV



$C_{17}H_{12}Cl_2N_4$
M.Wt. = 343.2 m.pt. = 233-235°C

II. PRODUCTION, PHYSICAL AND CHEMICAL CHARACTERISTICS OF BENZODIAZEPINE DERIVATIVES UNDER INTERNATIONAL CONTROL

Benzodiazepines, therapeutically used as tranquilizers, hypnotics, anticonvulsants and centrally-acting muscle relaxants, rank among the most frequently prescribed drugs and virtually all of those encountered in the illicit market result from diversion from licit sources. There is no evidence of clandestine manufacture.

The 33 benzodiazepine derivatives recently placed under international control appear in the illicit market mainly as tablets and capsules. Lorazepam is available in some countries in vials and cartridge-needle syringes for injection as well as in tablet form. Clonazepam is marketed as a tablet and as a dry powder to be dissolved in a suitable vehicle for injection. Temazepam is available in the United Kingdom in soft gelatin capsules and in a polyethyleneglycol solution for injection.

Chlordiazepoxide is marketed as the free base and hydrochloride salt in capsule and tablet form and, in some cases, in dry-filled ampoules. The hydrochloride salts of medazepam and flurazepam are the pharmaceutical forms licitly produced. The dipotassium salt of clorazepate is the most common form, although some clorazepate monopotassium is also available. The free acid is unstable and decarboxylates forming nordazepam on workup for analysis.

Diazepam, the most widely available benzodiazepine, could be found as capsules, tablets, aqueous or polyethyleneglycol solutions for injection, syrups and suppositories. In some countries, diazepam in bulk powder is used in cattle feed to immobilize the animals during transport and there exists the possibility of this form of the drug being diverted into the illicit traffic.

In a few cases, combination products such as chlordiazepoxide-amitriptyline and chlordiazepoxide-clidinium bromide also appear on the illicit market.

No laboratory would be expected routinely to encounter all 33 benzodiazepines under international control; however analysts should be aware of the particular ones available in their country and the characteristics and methodologies available for their identification and analysis. Reference should also be made to national pharmacopoeias and drug tablet and capsule identification guides for preliminary screening information. However, with many of these benzodiazepines coming out of patent protection, caution must be exercised when relying on such identicodes. Addendum I to the Multilingual Dictionary of Narcotic Drugs and Psychotropic Substances under International Control (ST/NAR/1/Add.1) recently published by UNDND includes a comprehensive listing of all brand names for these drugs.

III. THE ANALYSIS OF MATERIALS CONTAINING BENZODIAZEPINE DERIVATIVES

A. Sampling

The principal reason for a sampling procedure is to produce a correct and meaningful chemical analysis. Because most methods - qualitative and quantitative - used in forensic science laboratories for the examination of drugs require very small aliquots of material, it is vital that these small aliquots be entirely representative of the bulk from which they have been drawn. Sampling should be undertaken to conform to the principles of analytical chemistry, as laid down, for example, in national pharmacopoeias or by such organizations as the Association of Official Analytical Chemists.

There may be situations where, for legal reasons, the normal rules of sampling and homogenization cannot be followed if, for example, the analyst wishes to preserve some part of an exhibit as visual evidence. Alternatively, it may be necessary to perform separate assays on two powder items, rather than combining the powders prior to a single assay being performed on the mixture, because each has been separately exhibited by the seizing officer, and the legal system within which the analyst works requires individual results on every exhibit which is to be taken before the courts.

To preserve valuable resources and time, forensic analysts should seek, on all possible occasions, to use an approved sampling system and thereby reduce the number of quantitative determinations needed. To facilitate such an approach, the forensic analyst may need to discuss individual situations with both seizing officers and the legal personnel with whom he works.

Benzodiazepine derivative exhibits are encountered predominantly as capsules and tablets as a result of diversion from the licit market. In a few cases solutions for injection, syringes, syrups, suppositories and vials containing powders to be made up with a suitable vehicle for injection may be submitted for analysis. In some countries, diazepam bulk drug powder may be diverted from legitimate veterinary use.

1. Powders

(a) Sampling of single package items

The simplest sampling situation is where the submitted item consists of a single package of material. The material should be removed from its container or wrappings, placed in a clean clear plastic bag and the net weight recorded. The material should be thoroughly homogenized prior to the application of the sequence of chemical tests, although presumptive

testing may be applied at this stage if it is thought that the sampling or homogenization process will be lengthy and there is still some doubt as to the identity of the material. The simplest way to homogenize a powder is to shake it thoroughly within the clear plastic bag in to which it has been transferred. If the powder contains aggregates, these may be broken down by passing through successively finer sieves, or by pounding in mortar with a pestle, or by use of an adapted commercial food-mixer or food-processor.

Alternatively, the technique of coning-and-quartering can be applied, as follows: the sample is mixed by shaking or stirring. Large fragments are reduced in size if necessary; the material is then poured on a flat surface to form a cone. The "cone" is flattened and the material is then divided at right angles, forming quarters. Opposite quarters are taken for a sample; the remainder of the material is returned to the receptacle from which it was removed. Should further coning-and-quartering be desired to reduce sample size, particle sizes are further reduced, the material mixed thoroughly, poured onto a flat surface, and divided as before.

(b) Sampling of items consisting of more than one package

The analyst should examine the contents of all packages by eye, and possibly screen by using a simple colour test or TLC to determine:

1. If all packages contain suspect material, and/or

2. If one or more packages contain material different to that of the majority of packages. The simplest indicator is the physical appearance of the powder. If one or more packages obviously differ in content, these should be segregated and subjected to separate analysis.

The compositing of multiple container items is as follows:

(a) If there are less than 10 packages - all packages should be sampled.

(b) If there are 10 - 100 packages, randomly select 10 packages.

(c) If there are more than 100 packages - randomly select a number of packages equal to the square root of the total number of packages rounded to the next highest integer.

If the powders are found to be the same then the contents of a number of packages may be combined; the combined bulk material may then be homogenized in, for example, an adapted commercial food-processor. Alternately, the bulk may be subjected to coning-and-quartering.

When different types of material have been identified in the various packages, then each sub-group should be composited in an identical fashion to that previously outlined.

Sampling errors for quantitative methods are reduced if large aliquots of material are subjected to sequential dilution with the dissolving solvent. If the total sample size is large, this approach may be adopted. However, when large amounts of material are used for the first dissolution, it may be necessary to add the solvents by pipette to avoid error due to insoluble materials.

2. Tablets and capsules - Commercial or licit preparations

The preliminary determination of commercial origin is a subjective one. Clear-cut examples of products of commercial origin would be dosage units resembling descriptions as pictorial representations in national compendia of pharmaceutical preparations. Commercial preparations usually undergo quality control by the manufacturer; therefore, little useful information would be gained by screening a large number of units from each package. The amount of ingredient per tablet or capsule determined will be statistically valid for the entire lot.

(a) Single container

1. 1-50 dosage units -- Randomly select 1/2 total number of units to a maximum of 20. Determine average weight, powder to pass through a 20-mesh sieve and mix thoroughly.

2. 51-100 dosage units -- Randomly select 20 units, proceed as above.

3. 101-1,000 dosage units -- Randomly select 30 units, proceed as above.

4. Greater than 1,000 dosage units -- Randomly select a number of units equal to the square root of the total number present, rounded to the next higher integer; proceed as above.

(b) Multiple containers

Segregate containers by lot numbers and treat each group as described in 1 (b) above. Report results separately for each group.

Determine the square root of the total number of packages in each group. Randomly select a number of packages equivalent to the square root, rounded to the next highest integer.

From each of the selected packages, randomly select a number of dosage units equivalent to the square root of the total number of dosage units divided by the square root of the number of packages, rounded to the next higher integer.

Form a composite by grinding, sieving through a 20-mesh sieve and thoroughly mixing. Perform the analysis on the composite.

3. Aqueous solutions

Aqueous solutions for injection and syrups are available in some countries. Since solutions by their very nature are homogeneous, a relatively small sample (10 ml) represents the entire volume.

(a) Single container

If sample size permits, pipet an amount for assay of at least 10 ml.

(b) Multiple containers

Segregate containers by lot numbers or other characteristics and treat each group as described under 1 (b) above. Report results separately for each group.

Determine the square root of the total number of containers in each group. Randomly select a number of containers equivalent to the square root rounded to the next higher integer.

From each of the selected containers withdraw a 10 ml or larger sample (if size permits) for a composite.

If size permits, pipet at least 10 ml of the composite for assay.

4. Residues from syringes

Because of the trace amounts of drug usually present on hypodermic syringes seized from individuals, the analyst should not attempt to perform presumptive tests but should proceed directly with conclusive analytical procedures.

Wash the syringe with a minimum amount of methanol and concentrate it to dryness under a stream of nitrogen. Proceed with selected tests.

B. Extraction techniques

Since the majority of benzodiazepine derivatives appear as capsules or tablets from licit manufacture, elaborate solvent fractionation schemes are not usually necessary because they are soluble in methanol while most of the excipients are not; therefore the preparation of a simple methanol extract of the crushed tablet or capsule content usually is all that is required. The analyst should refer to relevant pharmacopoeias and formularies for the published content of medicament and adjust the amount of methanol to produce an approximate concentration of 1 to 20 mg/ml.

For quantitative analysis, the contents of a representative number of capsules or tablets are combined, as determined by the sampling procedures described above. An accurately weighed portion of the capsule or tablet contents, equal to one or more tablet or capsule full weight, is transfered to a suitably sized volumetric flask and diluted to volume with methanol to give a final concentration of approximately 1 to 20 mg/ml.

C. Presumptive tests

1. Tablet and capsule identification guides

As a first test, analysts should refer to national identification guides (pharmacopoeias, national formularies) for presumptive identification of the benzodiazepines commonly available in their country.

2. Colour tests

With such a large group of drugs containing diverse chemical functionality, no one colour test is specific for this class of drug. Therefore, colour tests are not recommended. Analysts are advised to use, as a presumptive test, a combination of TLC and colour development after spraying with selected reagents as a presumptive test. Reference is made also to the following publications for several colour tests for benzodiazepines if other methodology is not available:

1. "Rapid Testing Methods of Drugs of Abuse" (ST/NAR/13), New York, 1988.

2. The Identification and Analysis of Benzodiazepines under International Control.
   I. Colour Tests and Chromatographic Methods.
   U.N. Scientific and Technical Notes/1, Dec. 1987.

D. Thin-layer chromatography

The benzodiazepines form a diverse group of chemicals; however, the following three thin-layer chromatographic systems, when used in combination, give good separations. System C is a general system previously recommended in the manuals dealing with the identification and analysis of cocaine, opium and amphetamine/methamphetamine manuals.

PLATES

Activated silica gel G on glass-backed plates; the coating (0.25 mm thickness) contains an additive which fluoresces at 254 nm.

DEVELOPING SOLVENTS

SYSTEM A:	Chloroform	80
	Acetone	20
SYSTEM B:	Chloroform	90
	Methanol	10
SYSTEM C:	Cyclohexane	75
	Toluene	15
	Diethylamine	10

Preparation of solutions to be applied to the TLC plates

Powder:	Prepare a solution at a concentration of 5 mg per ml in methanol.
Capsules:	Remove the contents of a representative sample of capsules (see sampling procedure above) and prepare a solution containing the equivalent of approximately 5 mg of the drug per ml in methanol.
Tablets:	Grind a representative number of tablets to a fine powder and prepare a solution containing the equivalent of approximately 5 mg of the drug per ml in methanol.
Aqueous solutions:	Spot directly or the equivalent of 5 mg/ml if the concentration of the drug is known.
Standard solutions:	All made at a concentration of 5 mg per ml in methanol.

Apply 2 ul of a 5 mg per ml solution of the drug in methanol to the plate.

VISUALIZATION

The plates must be dried prior to visualization. This can be done at 120°C for 5 minutes in an oven or, more quickly, by using a hot air blower. It is important for proper colour development that all traces of diethylamine be removed from the plate.

Visualization methods

1. UV light at 254 nm
2. 2N H_2SO_4/heat/UV light at 366 nm
3. Acidified potassium iodoplatinate reagent

Spray reagent

Acidified potassium iodoplatinate reagent: Dissolve 0.25 g of platinic chloride and 5 g of potassium iodide in sufficient water to produce 100 ml. This is potassium iodoplatinate reagent; for the acidified version add 5 ml of concentrated hydrochloric acid to 100 ml of iodoplatinate solution.

METHOD

First observe the plate under short wave UV light. Spray with 2N H_2SO_4 and heat in an oven at 80°C for 5 minutes. Observe the fluorescent spots under long UV light (366 nm). The plate may then be oversprayed with the acidified iodoplatinate reagent. All benzodiazepines gave purple coloured spots when oversprayed with the reagent.

For alternative methods see:

1. Pharm. Acta. Helv. 61 (1986), pp. 167-176.
2. J. Chromatography 179 (1979), pp. 381-389.
3. J. Chromatography 194 (1980), pp. 262-269.
4. J. Chromatography 358 (1986), pp. 169-178.

RESULTS R_f x 100 values:

COMPOUND	DEVELOPING SYSTEM		
	A	B	C
Alprazolam	8	62	1
Bromazepam	13	55	3
Camazepam	73	89	11
Chlordiazepoxide	15	58	2
Clobazam	68	83	11
Clonazepam	47	60	0
Clorazepic acid*	44	65	4
Clotiazepam	75	88	34
Cloxazolam	48	81	13
Delorazepam	46	69	7
Diazepam	70	87	30
Estazolam	11	50	1
Ethyl loflazepate	63	70	1
Fludiazepam	67	84	29
Flunitrazepam	67	86	16
Flurazepam	5	48	38
Halazepam	76	88	21
Haloxazolam	8	85	13
Ketazolam**	66,70	86	12,30
Loprazolam	3	49	2
Lorazepam	31	48	1
Lormetazepam	63	81	9
Medazepam	72	89	45
Nimetazepam	45	87	12
Nitrazepam	42	64	1
Nordazepam*	44	63	5
Oxazepam	29	48	1
Oxazolam	69	63	4,15,19***
Pinazepam	81	90	27
Prazepam	79	90	35
Temazepam	63	82	10
Tetrazepam	74	88	32
Triazolam	9	52	1

* Clorazepic acid converts to nordazepam.
** Ketazolam decomposes to diazepam.
*** Multiple spots.

E. Gas-liquid chromatography

NOTE

Although gas liquid chromatography can be recommended as a suitable method for the analysis of most benzodiazepines, several of them, particularly the 3-hydroxy substituted ones, undergo thermal degradation and rearrangement. Chlordiazepoxide, cloxazolam, lormetazepam, haloxazolam, oxazolam, ethyl loflazepate and temazepam yield multiple peaks. Similarly while clorazepic acid gives a single peak having a retention index value corresponding to nordazepam, the commercial product which is the dipotassium salt does not chromatograph. Ketazolam gives a single peak corresponding to diazepam. Loprazolam does not elute from the glc columns under the experimental conditions recommended in this manual. Retention indices for benzodiazepines have been shown to be dependent on column temperature. Values should be checked before use by chromatographing a few reference standards of known retention index.

Since benzodiazepines are diverted from licit manufacture, qualitative analysis will usually suffice for identification purposes. If quantitative analysis is required, analysts are referred to earlier manuals in this series for quantitative GLC methodology.

1. Packed column technique

Operating conditions:

Detector:	FID
Column:	6 ft (or 2 m), 2 to 4 mm ID glass
Packing:	3% SE-30 or OV-1 on 80-100 Chromosorb W HP
Carrier gas:	Nitrogen
Column temperatures:	programmed from 200 to 280°C at 16°C/min.
Injector/detector temperatures:	280°C

METHOD

Solutions of the standards and samples are prepared in methanol at a concentration of 1 mg per ml according to the procedure outlined in III B above. Inject 1 to 2 ul as appropriate.

2. Capillary column technique

Operating conditions:

Detector:	FID
Column:	Fused silica, chemically bonded and cross-linked methyl silicone, such as BP-1, DB-1 or equivalent
Film thickness:	0.25 um
Length:	25 m, ID 0.25 mm
Carrier gas:	Nitrogen 1 ml/min
Split ratio:	20:1
Column temperature:	250°C
Injector/detector temperatures:	275°C

METHOD

Solutions of the standards and samples (1 mg/ml) are prepared in methanol and 1 ul injected into the gas chromatograph.

RESULTS

ELUTION PROFILES ON SELECTED COLUMNS (Retention indices)

COMPOUND	SE-30/OV-1	BP-1 Capillary
Alprazolam	3050	2936
Bromazepam	2633	2626
Camazepam	2954	2954
Chlordiazepoxide*	2799 (2453, 2530)	2815 (2531, 2646)
Clobazam	2694	2582
Clonazepam	2885	2833
Clorazepic acid*	2457	2521 (2783)
Clotiazepam	2420	2485
Cloxazolam*	2405	2344 (2604)
Delorazepam	2650	2537
Diazepam	2425	2477
Estazolam	2955	2893
Ethyl loflazepate	2195	2925 (2878)
Fludiazepam	2460	2403
Flunitrazepam	2645	2633
Flurazepam	2785	2795
Halazepam	2335	2314
Haloxazolam*	2620	2634 (2518, 2272)
Ketazolam*	2425	2475
Loprazolam*	Does not elute	
Lorazepam	2402	2448
Lormetazepam*	2674	2703 (2946)
Medazepam	2226	2287
Nimetazepam	2730	2693
Nitrazepam	2750	2755
Nordazepam	2456	2522
Oxazepam	2336	2374
Oxazolam	2590	2514 (2534, 2212)
Pinazepam	2580	2551
Prazepam	2641	2676
Temazepam	2633	2613 (2828)
Tetrazepam	2460	2463
Triazolam	3130	3025

* Thermally unstable. Minor peaks in brackets.

References:

1. "Gas-Chromatographic Retention Indices of Toxicologically Relevant Substances on SE-30 or OV-1". Second, Revised and Enlarged Edition. DFG/TIAFT, VCH Verlagsgesellschaft, Weinheim, 1985.

2. J. Chromatography 439 (1988), pp. 317-339.

3. Benzodiazepines. A Handbook. Springer-Verlag Heidelberg, 1982.

For alternative methods see:

1. J. Chromatography 169 (1979), pp. 409-411.

2. J. Chromatography 222 (1981), pp. 409-419.

3. J. Chromatography 358 (1986), pp. 169-178.

F. High performance liquid chromatography

Because of the diversity of chemical structure among the benzodiazepines, no single HPLC method will separate all of them. Nevertheless, the following methods are recommended for the qualitative and quantitative analysis of all benzodiazepine derivatives under international control. Column and solvent system routinely used by national laboratories will depend upon the particular benzodiazepines commonly available in that country.

1. Normal phase

Column:	250 mm by 5 mm ID
Packing material:	Silica HPLC grade, 5 um diameter (Spherisorb S5W or equivalent)
Mobile phase:	A: Methanol (1000 ml) containing perchloric acid (100 ul) B: Methanol-water-trifluoroacetic acid (997:2:1, v/v/v)
Flow rate:	2.0 ml per minute
Detection:	UV at 240 nm
Sample solutions:	Extract the active component from tablets and capsules according to the procedure outlined in III b (above). Dissolve the material in methanol to give an approximate concentration of 1 mg per ml.
Standard solution:	Dissolve a sufficient amount of drug standard in methanol to give a solution containing 1 mg/ml.
Injection volume:	20 ul by loop injector
Quantification:	By peak areas, external standard method

2. Reverse phase

Column:	250 mm by 5 mm ID
Packing material:	Octadecyl-silica HPLC 5 um diameter (ODS-Hypersil or equivalent)
Mobile phase:	C: Methanol: water: phosphate buffer (0.1 M), (55:25:20, v/v/v), pH 7.25 D: Methanol: water: phosphate buffer (0.1 M), (70:10:20, v/v/v/), pH 7.67 The phosphate buffer (0.1 M) is prepared by dissolving sodium dihydrogenphosphate dihydrate (14.35g, 0.092 mol) and disodium hydrogenphosphate (1.14g, 0.008 mol) in 1000 ml water.
Flow rate:	1.5 ml per minute
Detector:	UV at 240 nm
Sample solutions:	Extract the active component from tablets and capsules according to the procedure outlined in III b (above). Dissolve the material in 50% (v/v) aqueous methanol to give an approximate concentration of 1 mg per ml.
Standard solution:	Dissolve a sufficient amount of drug standard in 50% (v/v) aqueous methanol to give a solution containing 1 mg/ml.
Injector volume:	20 ul by loop injector
Quantitation:	By peak areas, external standard method

Reference: J. Chromatography 439 (1988), pp. 317-339.

For alternative methods see:

1. J. Chromatography 416 (1987), pp. 303-310.
2. J. Chromatography 268 (1983), pp. 92-98.
3. J. Chromatography 247 (1982), pp. 15-37.
4. J. Chromatography 358 (1986), pp. 169-178.

RESULTS Capacity ratios (k' values)

COMPOUND	DEVELOPING SYSTEM			
	A	B	C	D
Alprazolam	2.54	3.87	3.35	1.28
Bromazepam	2.66	6.12	1.63	0.80
Camazepam	0.11	0.12	11.80	2.56
Chlordiazepoxide	2.43	6.72	6.68	1.65
Clobazam	0.10	0.10	4.14	1.09
Clonazepam	0.35	0.32	2.92	0.79
Clorazepic acid*	1.96	3.86	8.22	1.84
Clotiazepam	2.80	5.95	17.91	3.01
Cloxazolam	2.73	7.63	15.22	2.82
Delorazepam	1.20	1.38	7.20	1.53
Diazepam	2.39	4.23	10.41	2.29
Estazolam	1.62	1.82	4.22	1.08
Ethyl loflazepate	0.15	0.13	13.71	2.19
Fludiazepam	1.38	1.57	7.91	1.67
Flunitrazepam	0.46	0.42	3.34	0.86
Flurazepam	6.23	16.02	12.98	3.12
Halazepam	1.38	1.70	18.92	2.91
Haloxazolam	2.73	6.31	11.62	2.11
Ketazolam	2.44	4.58	10.78	2.22
Loprazolam	13.00	40.00	6.42	1.23
Lorazepam	0.16	0.13	5.16	1.14
Lormetazepam	0.16	0.16	7.19	1.46
Medazepam	3.75	10.75	41.46	6.31
Nimetazepam	1.47	2.01	3.91	1.01
Nitrazepam	1.39	2.35	3.22	0.87
Nordazepam	1.94	4.02	8.97	1.89
Oxazepam	0.75	0.78	5.42	1.25
Oxazolam**	1.45,2.40	3.62,5.57	22.43,25.14	3.51,3.87
Pinazepam	1.85	3.08	12.82	2.21
Prazepam	2.09	3.65	29.99	4.28
Temazepam	0.63	0.57	6.80	1.49
Tetrazepam	2.84	8.13	25.82	4.25
Triazolam	1.71	2.13	5.18	1.13

* Converts to Nordazepam during chromatography.

** Two peaks due to equilibrium mixture of cis and trans isomers. (see J. Medicinal Chem. 14 (1971), pp. 520-526).

G. Infrared Spectroscopy

In some countries, confirmation of identity by spectroscopic means may be required. Theoretically each substance has a unique infrared spectrum and this method should permit the unequivocable identification of any benzodiazepine. However, with licit samples, the lack of solubility in chloroform of certain benzodiazepine derivatives and the isolation of the drug in a pure form, free from pharmaceutical excipients, place limitations on this method. The following procedure is recommended. Other methods utilizing column or preparative thin-layer chromatography may also be employed. Because of solvate formation and polymorphism, it is recommended that an authentic pharmaceutical formulation be carried through the same extraction procedure and its spectrum compared with that of the sample.

ISOLATION OF PURE DRUGS FROM SAMPLE

a) Benzodiazepines as the hydrochloride salt

Chlordiazepoxide, flurazepam and medazepam are marketed as hydrochloride salts. Triturate the powders from the capsule contents with 1 to 2 ml of methanol. Filter, collect the extract and evaporate to dryness. Induce crystallization and run the infrared spectrum of the resulting benzodiazepine salt by the KBr disk method.

b) Benzodiazepine free base

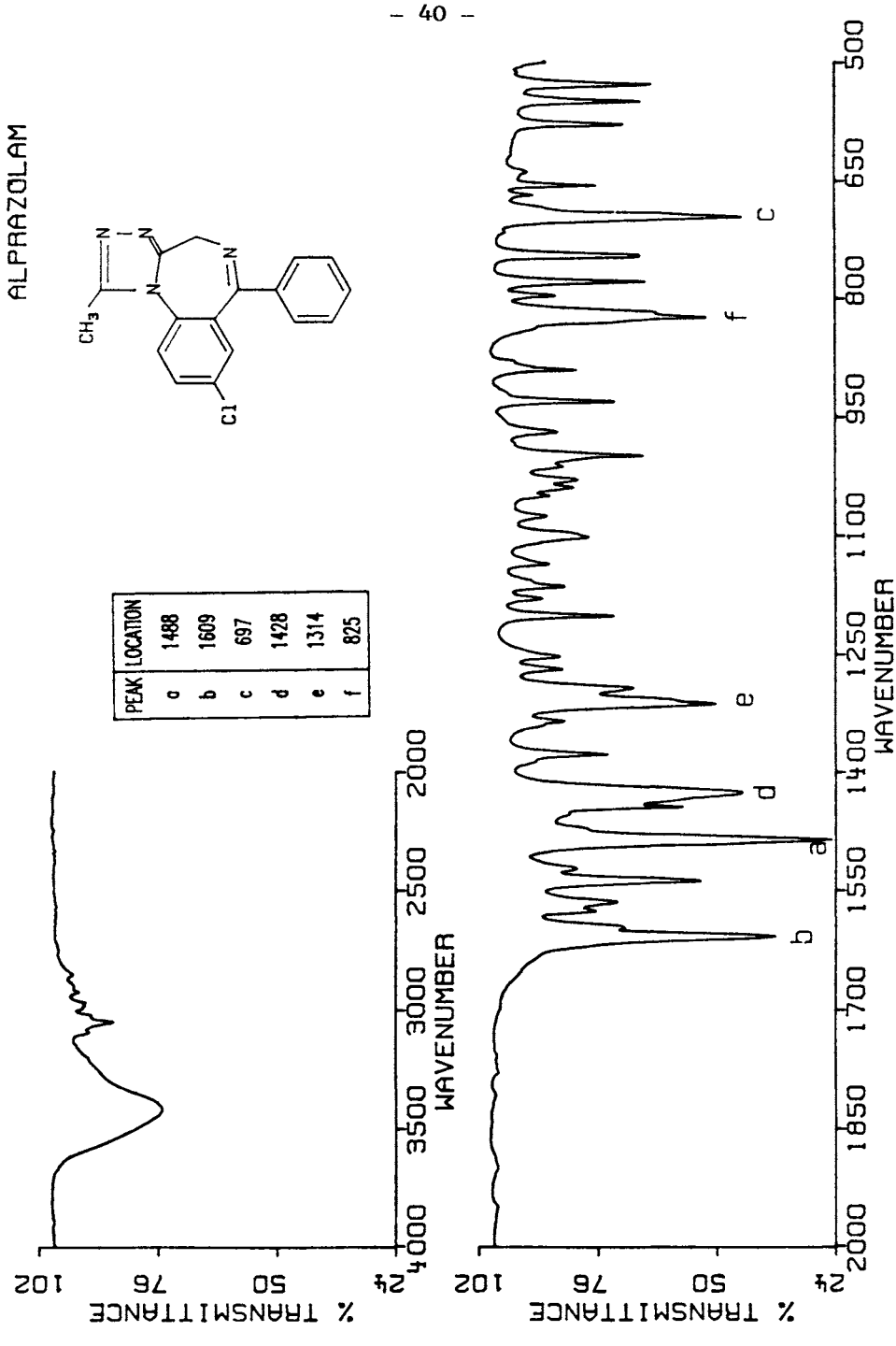
Suspend a portion of the powder or crushed tablet contents containing approximately 10 mg of drug in 1 ml of 0.1 M NaOH solution and extract with $CHCl_3$. Evaporate the $CHCl_3$ to dryness and induce crystallization.

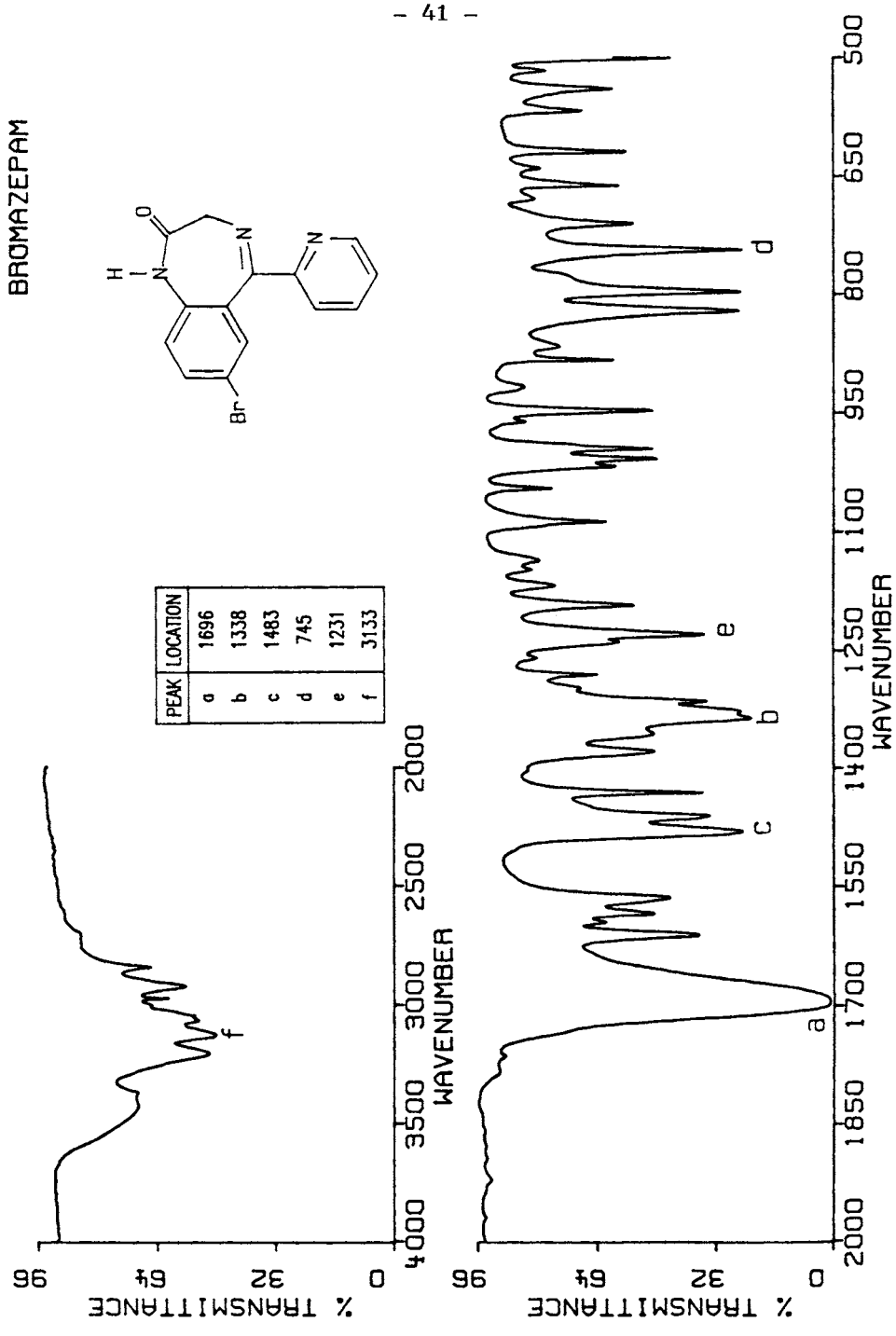
METHOD

For a description of the standard methods (halide disk, microhalide, nujol mull and thin-film techniques) see previous manuals in the series.

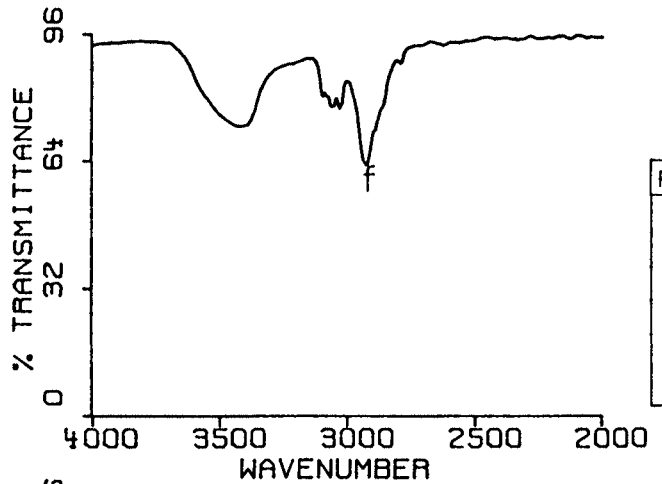
RESULTS

The following spectra were recorded at 2 cm^{-1} resolution by the KBr disk technique. Significant absorption bands are listed on the spectra.

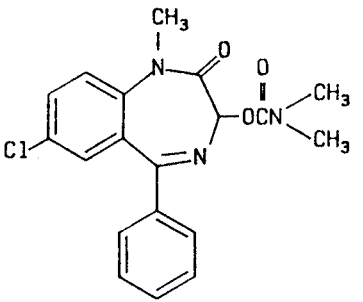

ALPRAZOLAM
CH3
N
N
N
N
Cl
PEAK LOCATION
a 1488
b 1609
c 697
d 1428
e 1314
f 825
% TRANSMITTANCE
102
76
50
24
4000
3500
3000
2500
2000
WAVENUMBER
% TRANSMITTANCE
102
76
50
24
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
a
b
c
d
e
f

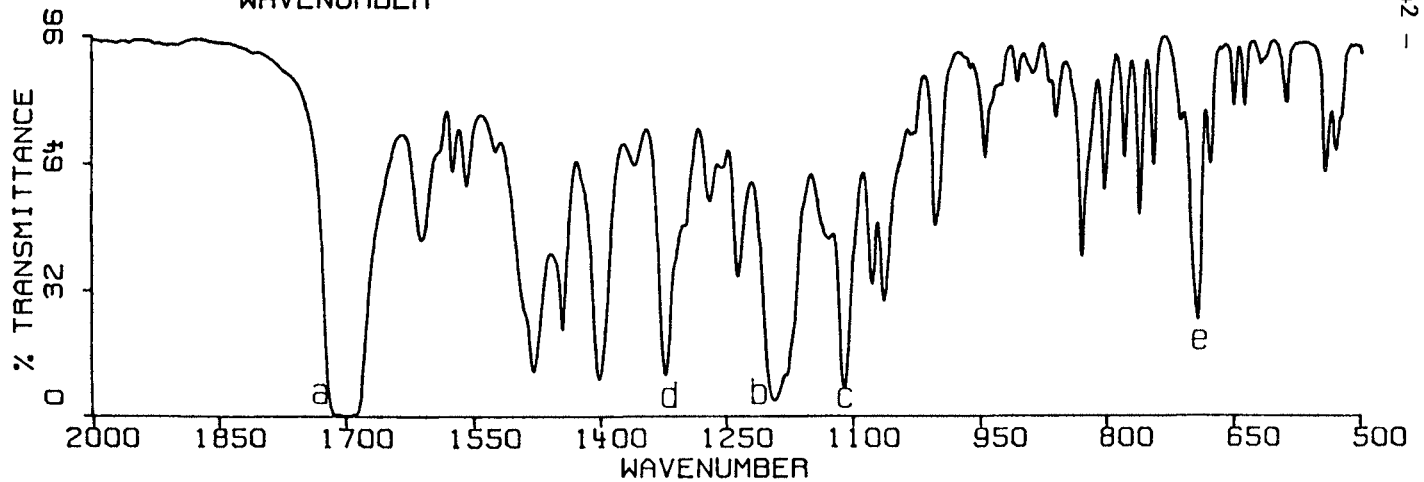

BROMAZEPAM
PEAK LOCATION
a 1696
b 1338
c 1483
d 745
e 1231
f 3133
% TRANSMITTANCE
WAVENUMBER
96
64
32
0
4000
3500
3000
2500
2000
1850
1700
1550
1400
1250
1100
950
800
650
500

CAMAZEPAM

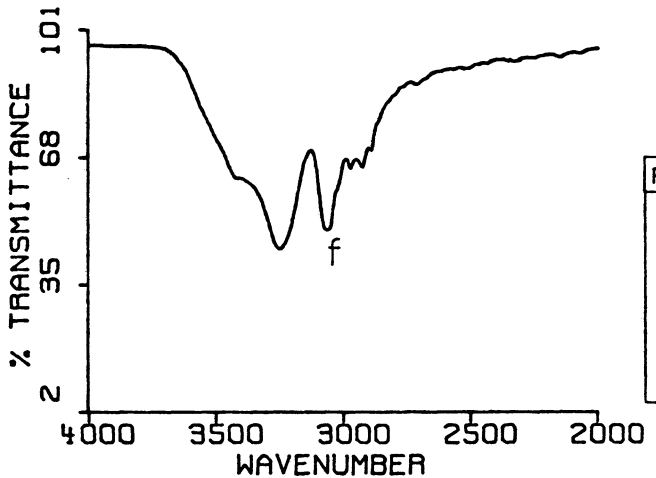


PEAK	LOCATION
a	1699
b	1194
c	1112
d	1403
e	695
f	2933

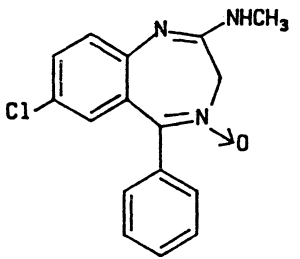


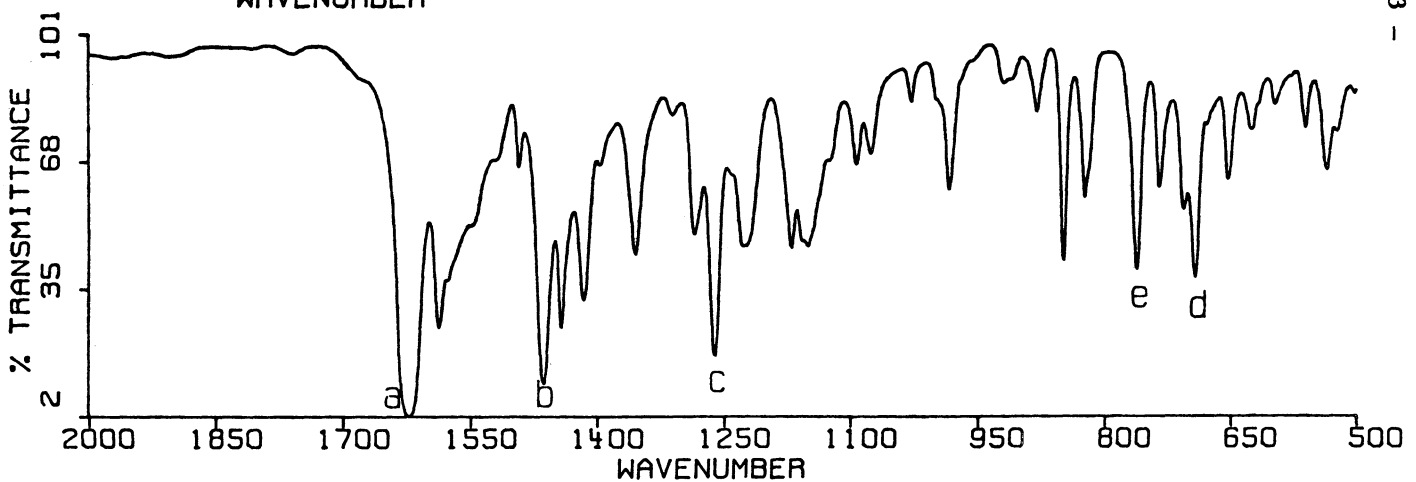


CHLORDIAZEPOXIDE

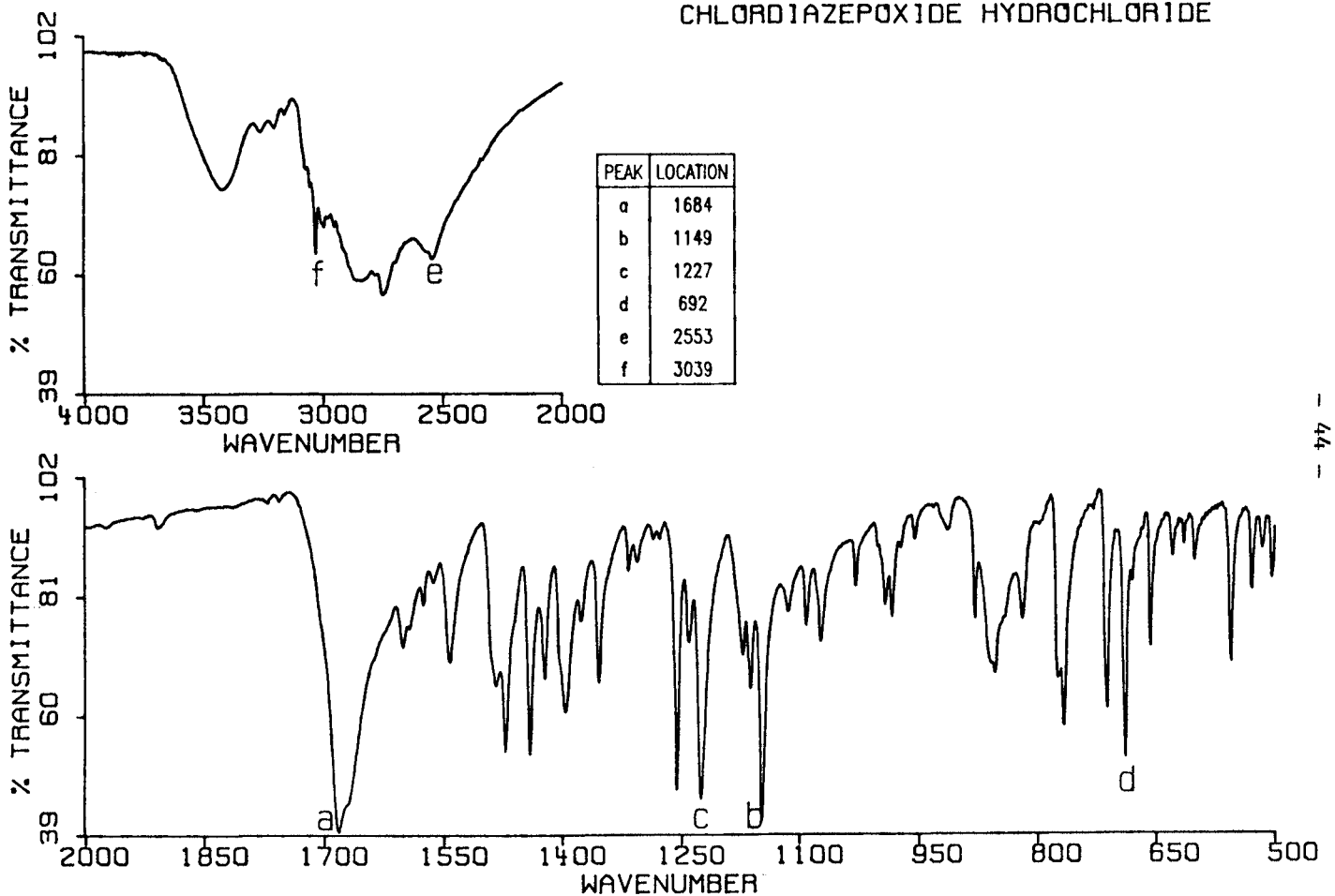


PEAK	LOCATION
a	1624
b	1465
c	1262
d	695
e	764
f	3072

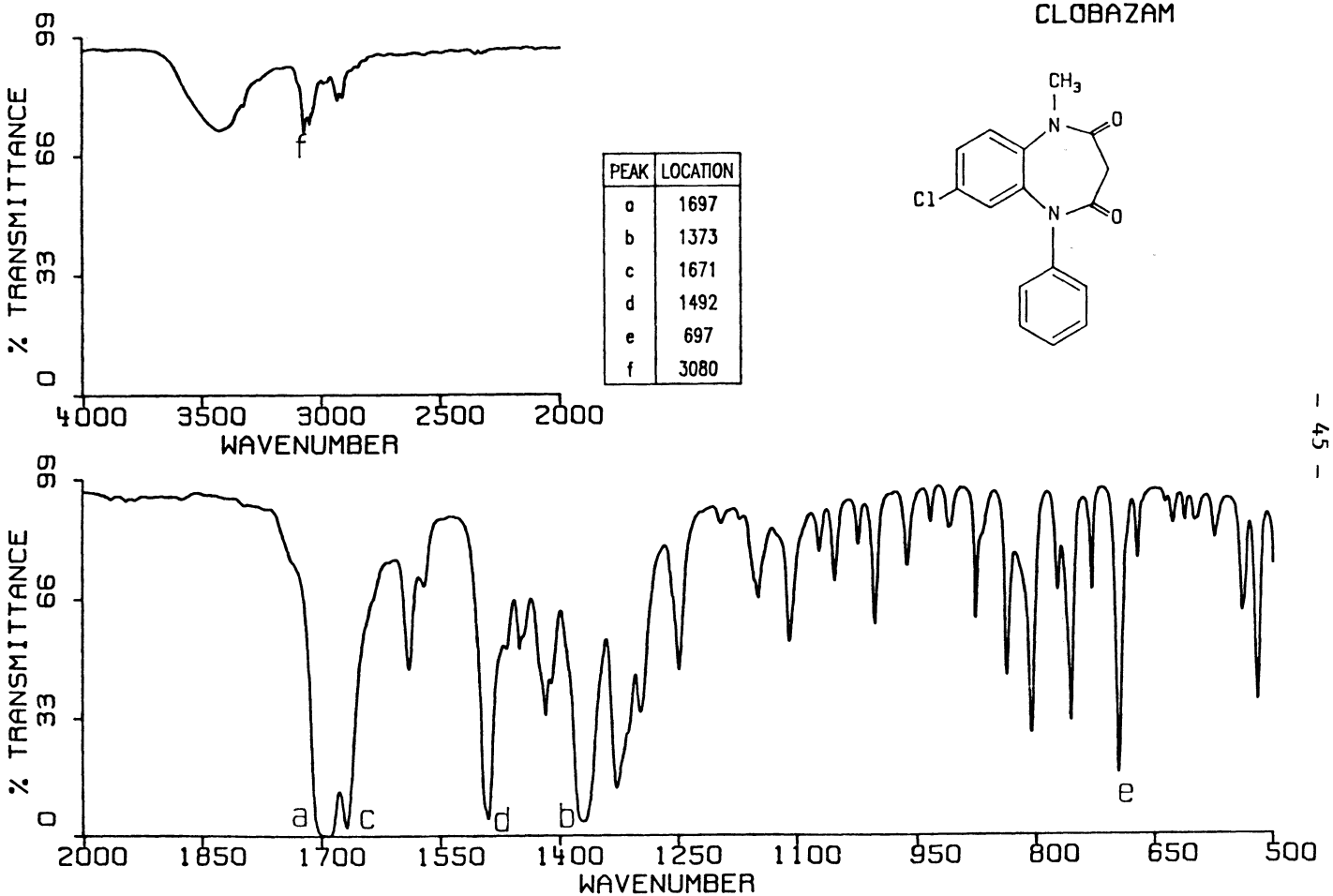




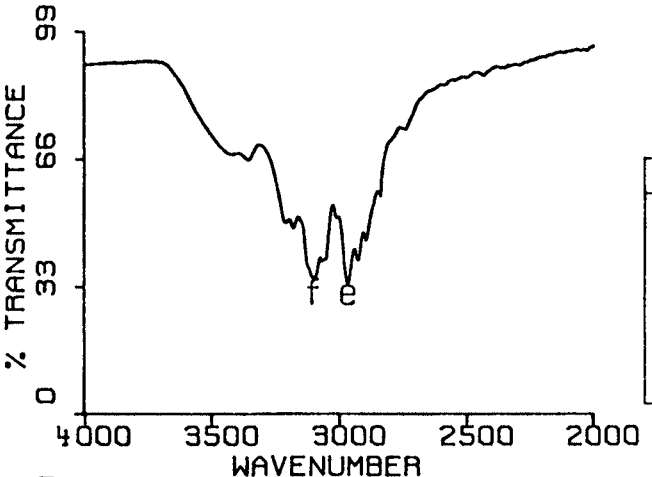
CHLORDIAZEPOXIDE HYDROCHLORIDE



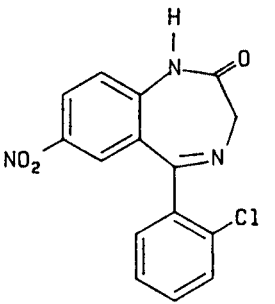
PEAK	LOCATION
a	1684
b	1149
c	1227
d	692
e	2553
f	3039

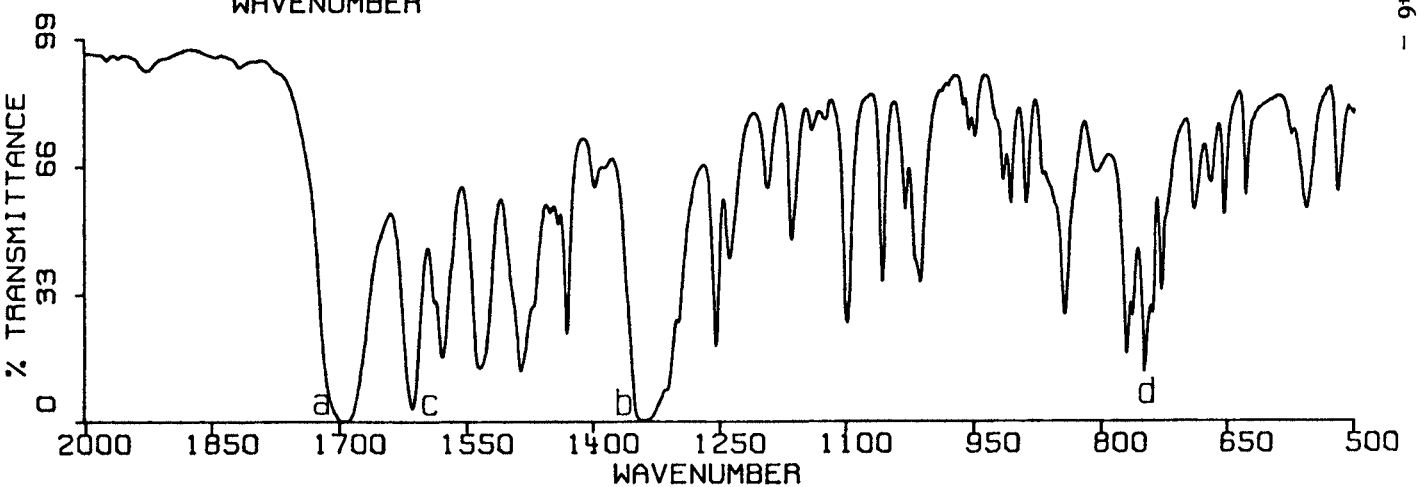

CLOBAZAM
% TRANSMITTANCE
99
66
33
0
4000
3500
3000
2500
2000
WAVENUMBER
f
PEAK
LOCATION
a
1697
b
1373
c
1671
d
1492
e
697
f
3080
CH3
N
O
Cl
N
O
% TRANSMITTANCE
99
66
33
0
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
a
c
d
b
e

CLONAZEPAM

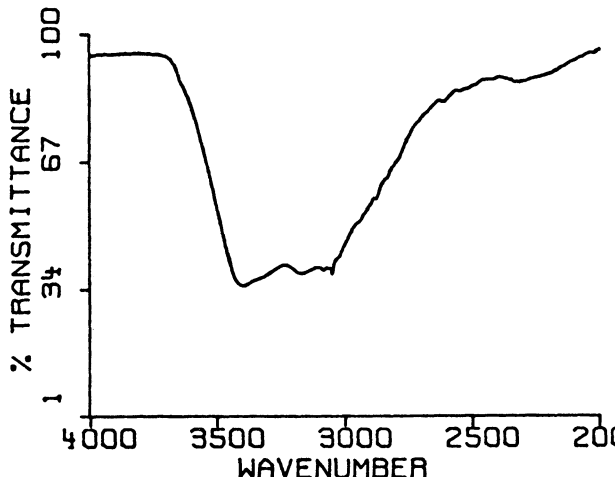


PEAK	LOCATION
a	1693
b	1339
c	1615
d	751
e	2971
f	3104

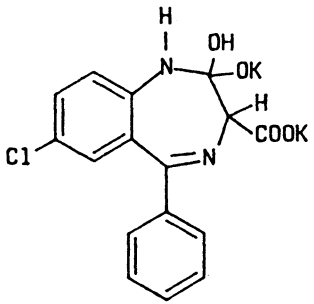


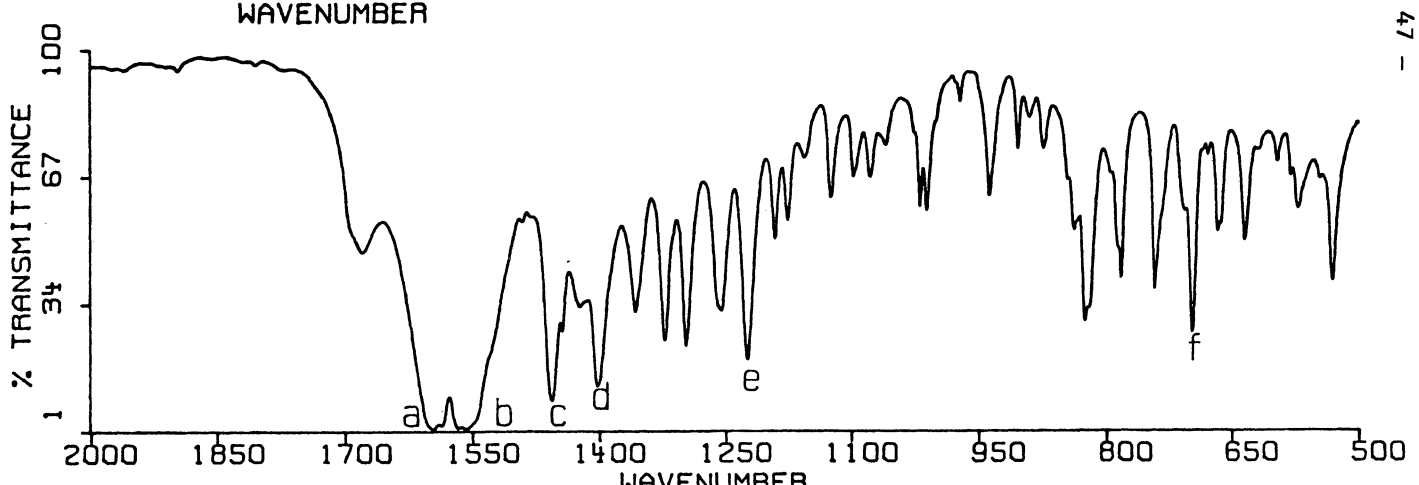


CLORAZEPATE DIPOTASSIUM

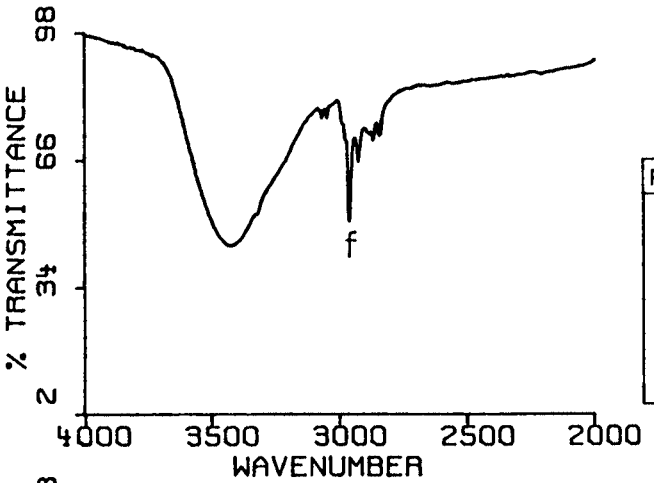


PEAK	LOCATION
a	1597&1588
b	1567&1558
c	1458
d	1404
e	1226
f	700

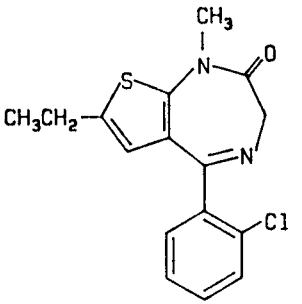


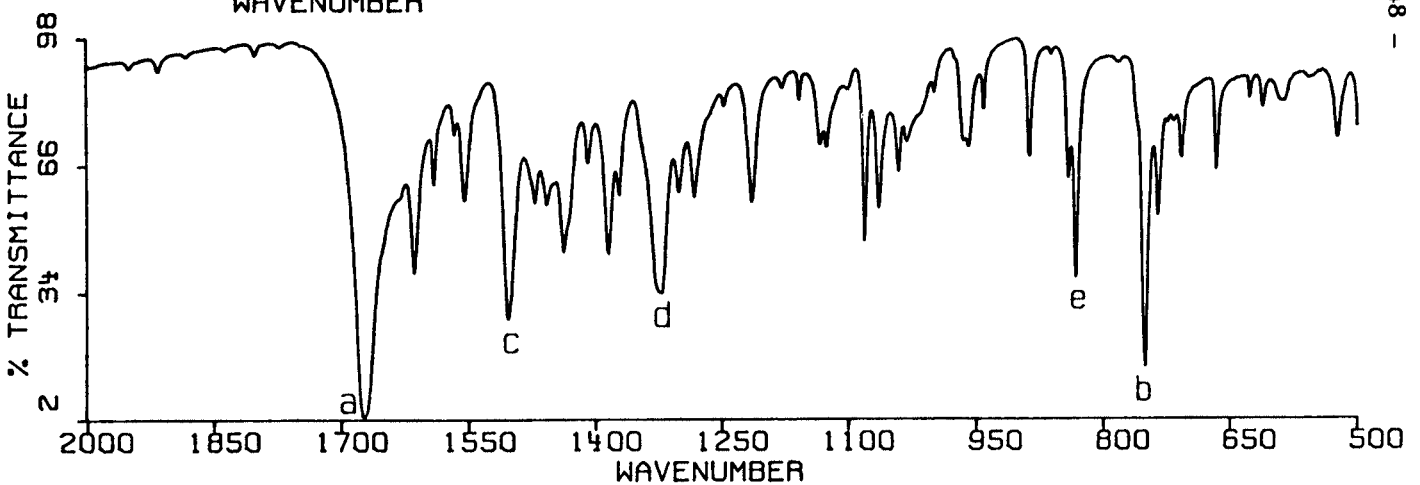


CLOTIAZEPAM

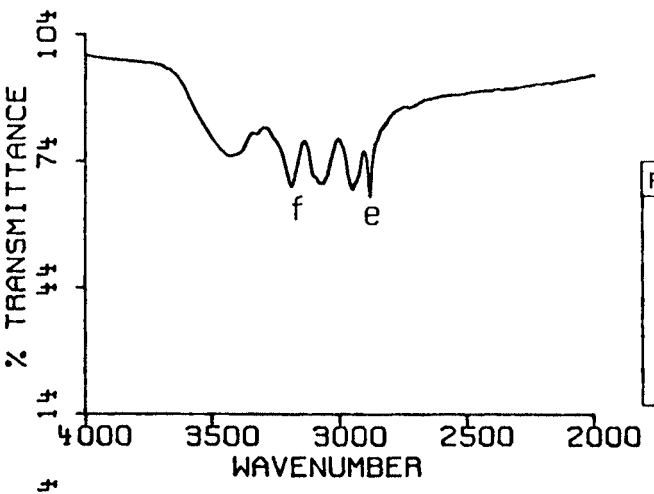


PEAK	LOCATION
a	1675
b	752
c	1506
d	1324
e	834
f	2968

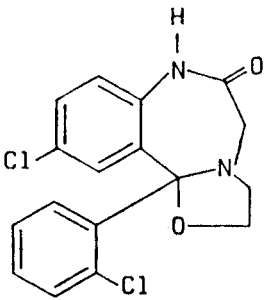


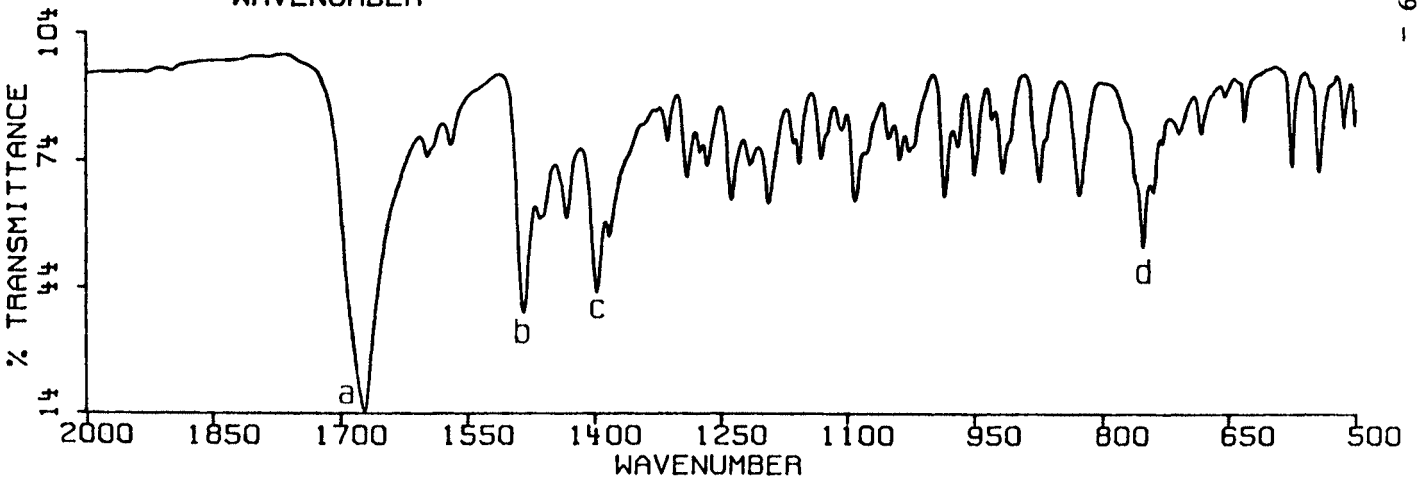


CLOXAZOLAM

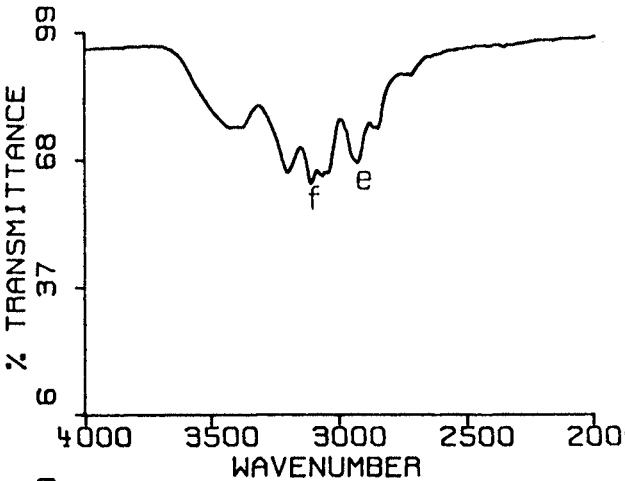


PEAK	LOCATION
a	1673
b	1485
c	1399
d	754
e	2885
f	3194

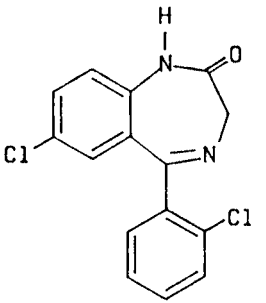


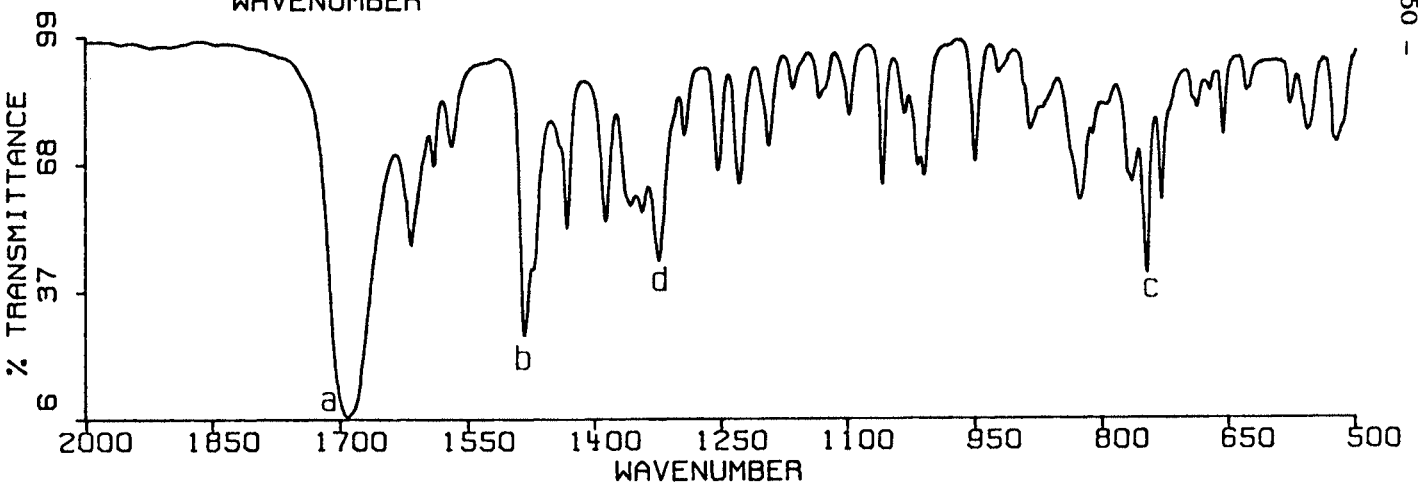


DELORAZEPAM

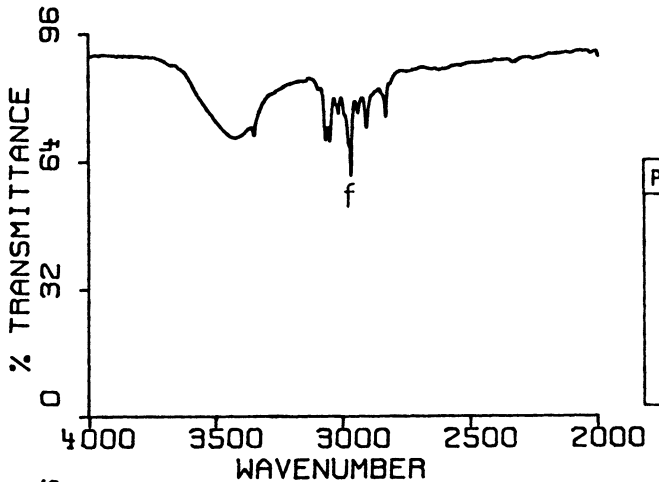


PEAK	LOCATION
a	1692
b	1485
c	749
d	1325
e	2937
f	3118

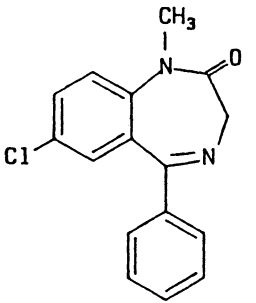


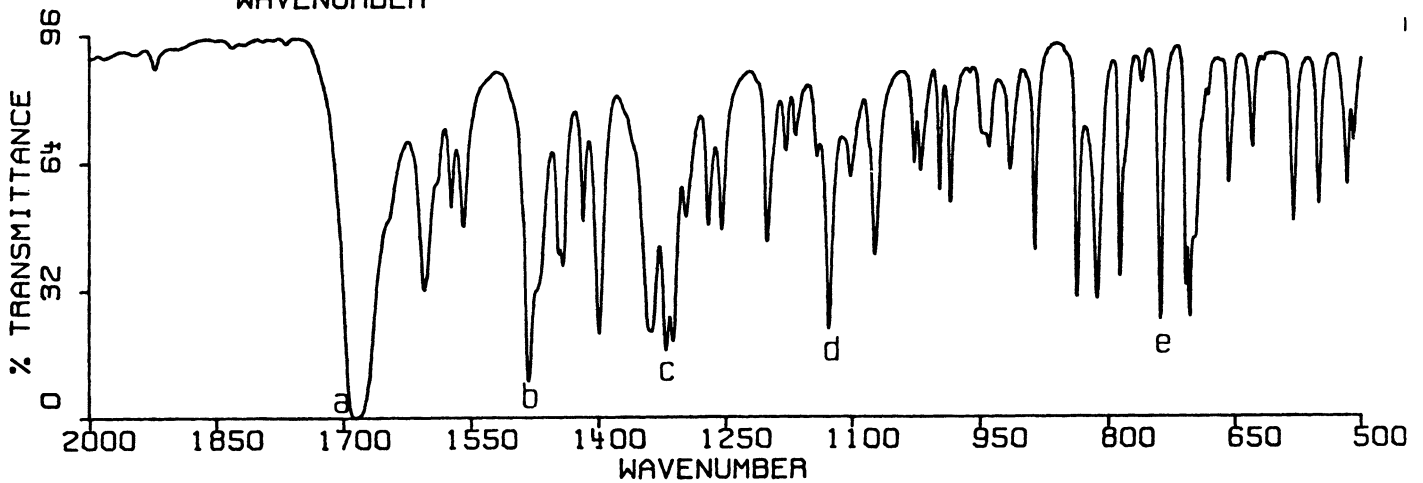


DIAZEPAM

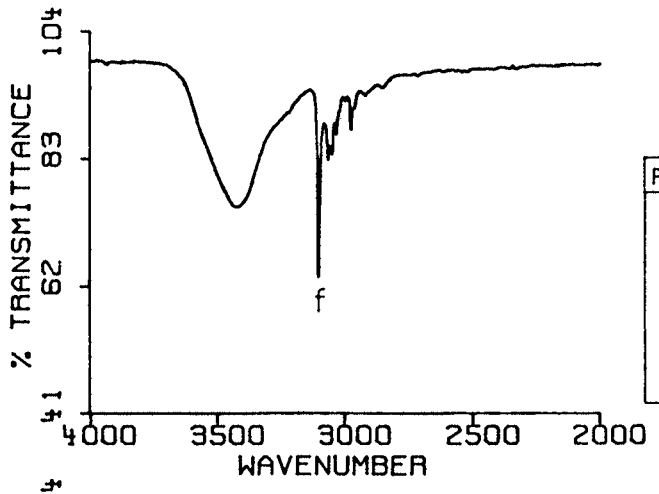


PEAK	LOCATION
a	1687
b	1484
c	1323
d	1129
e	740
f	2972

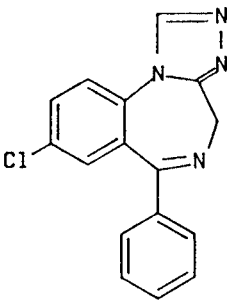


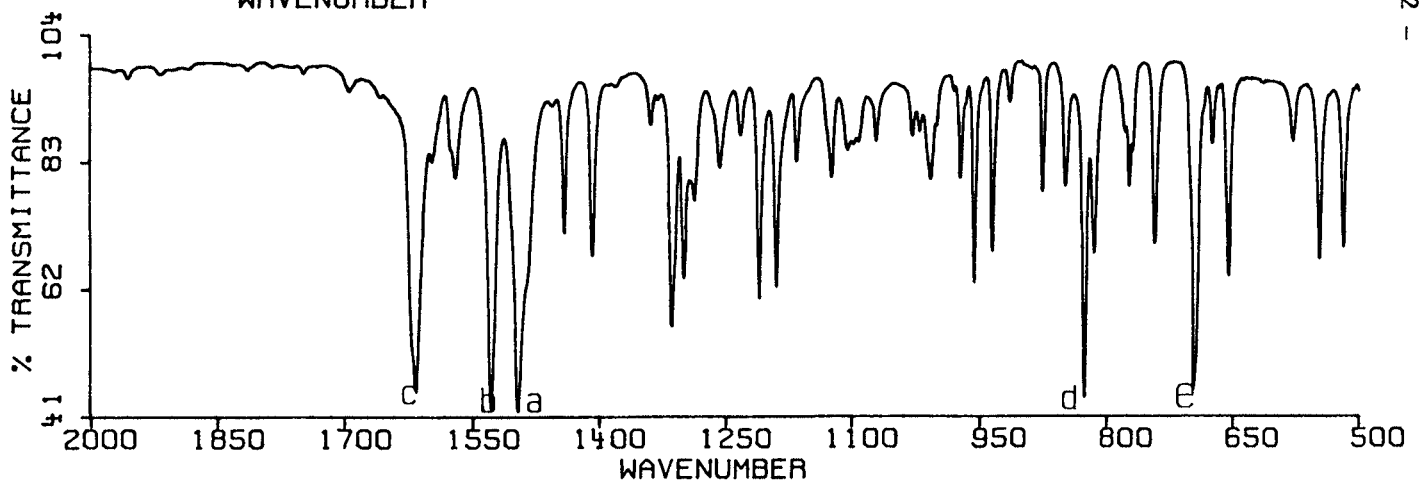


ESTAZOLAM

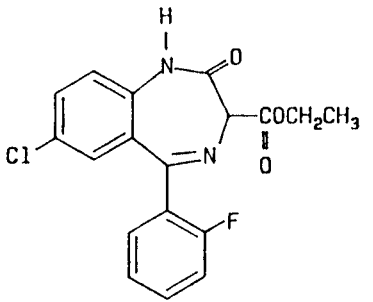


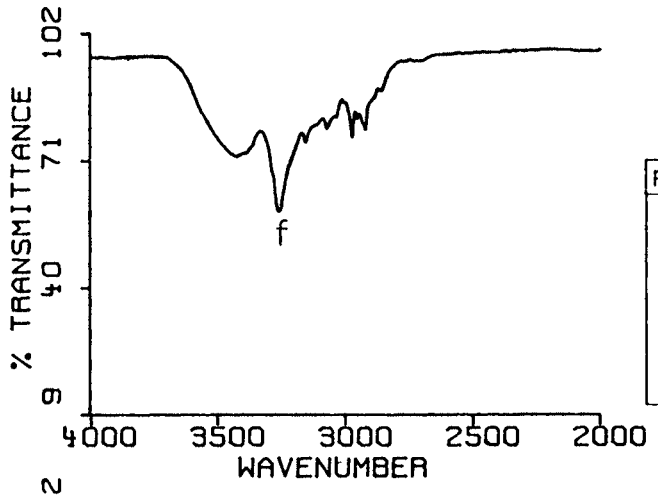
PEAK	LOCATION
a	1498
b	1529
c	1617
d	827
e	696
f	3108



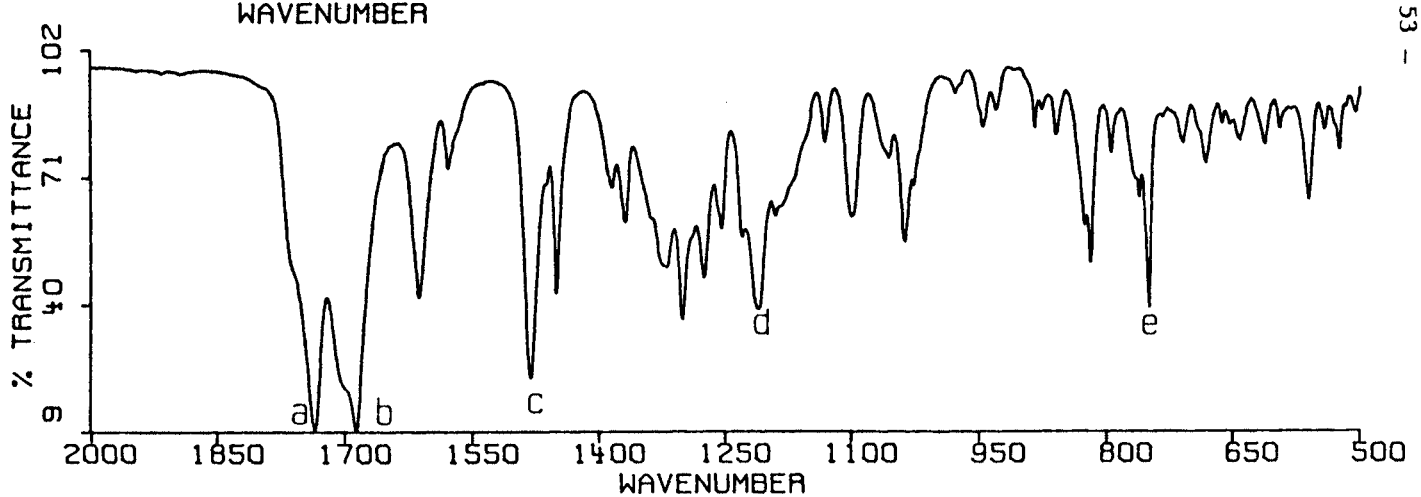


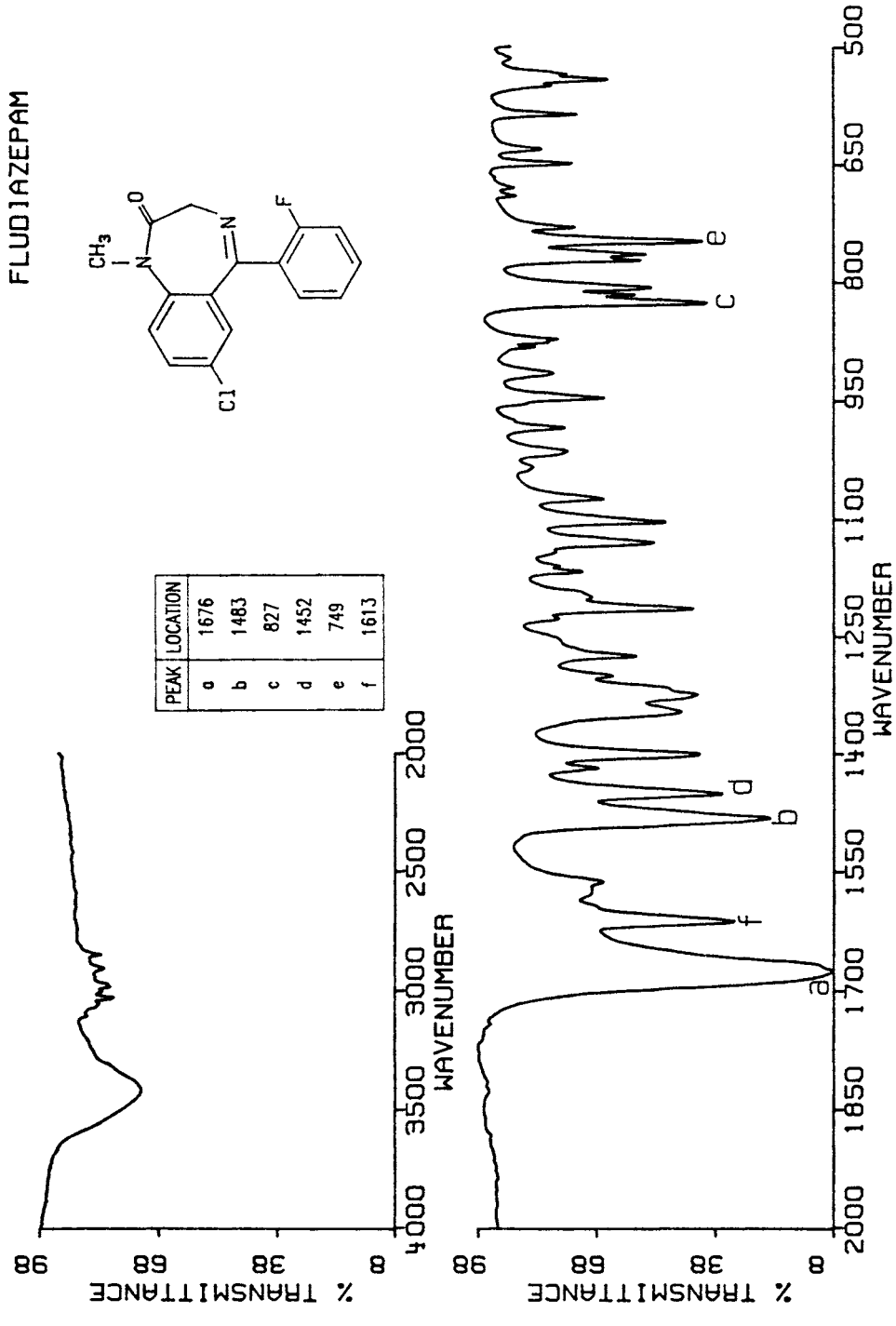
ETHYL LOFLAZEPATE



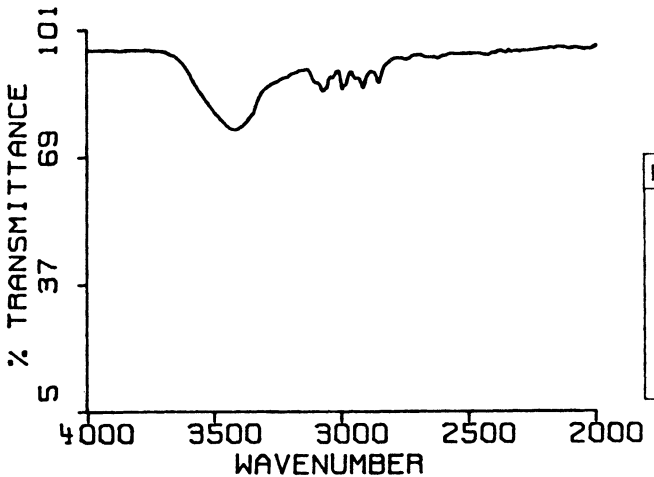


PEAK	LOCATION
a	1737
b	1688
c	1483
d	1212
e	753
f	3269

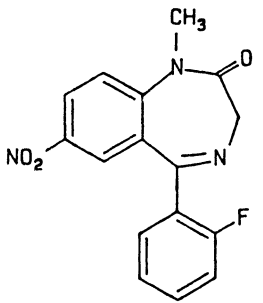


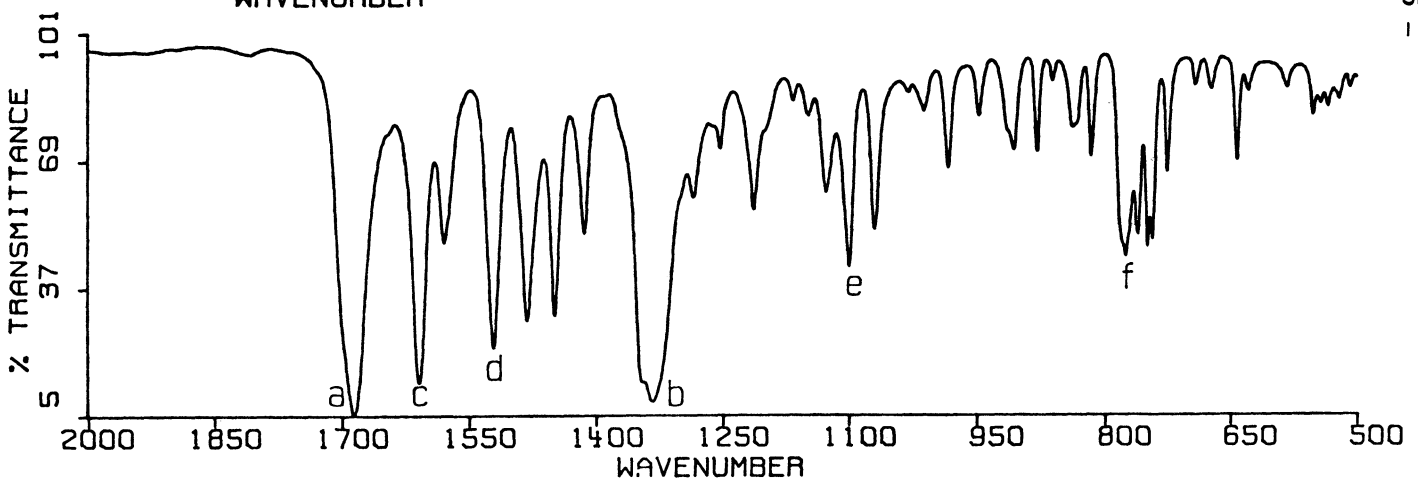

FLUDIAZEPAM
CH3
O
N
N
F
Cl
PEAK | LOCATION
a | 1676
b | 1483
c | 827
d | 1452
e | 749
f | 1613
% TRANSMITTANCE
98
68
38
8
4000
3500
3000
2500
2000
WAVENUMBER
% TRANSMITTANCE
98
68
38
8
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
a
b
c
d
e
f

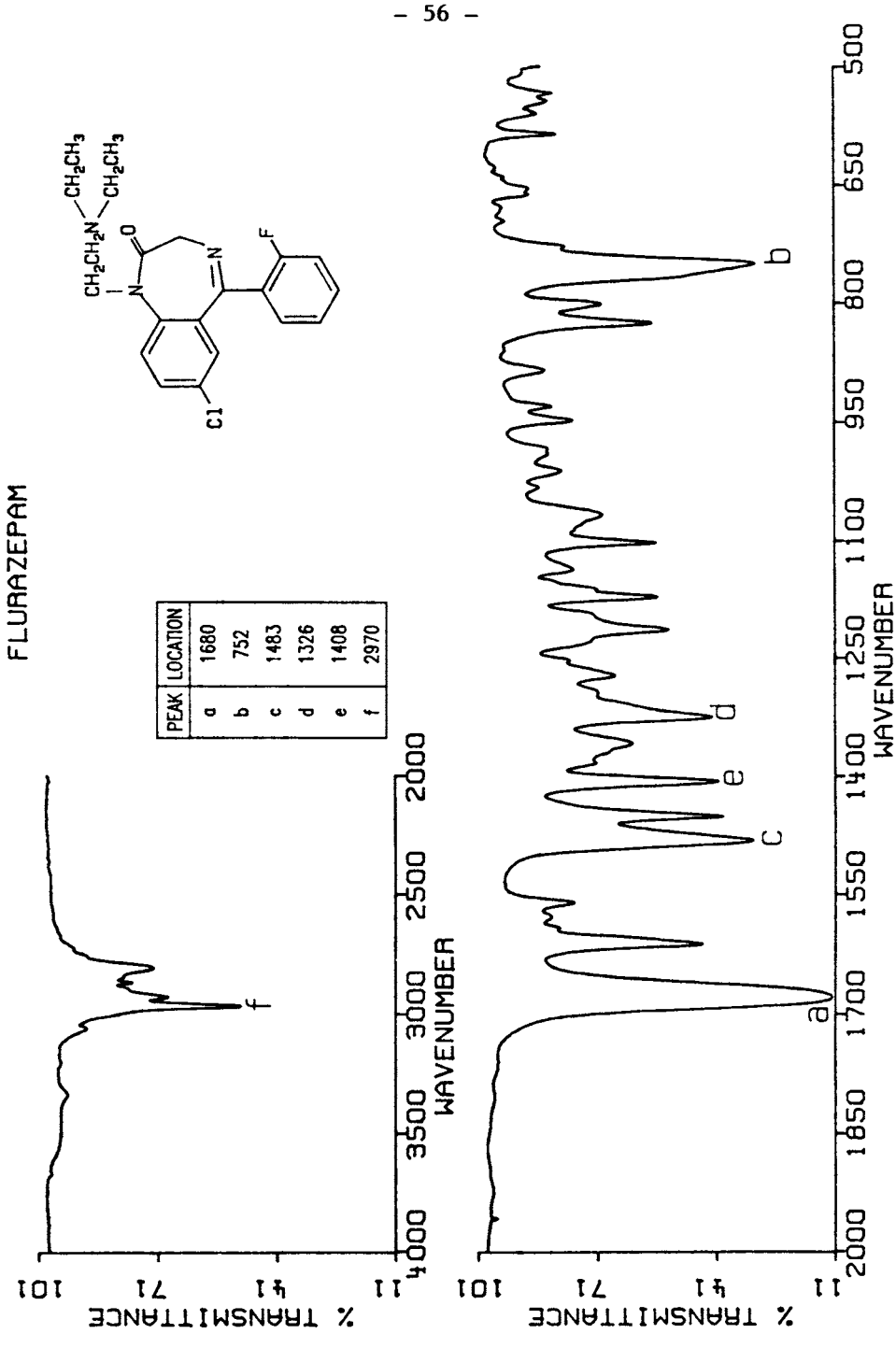
FLUNITRAZEPAM

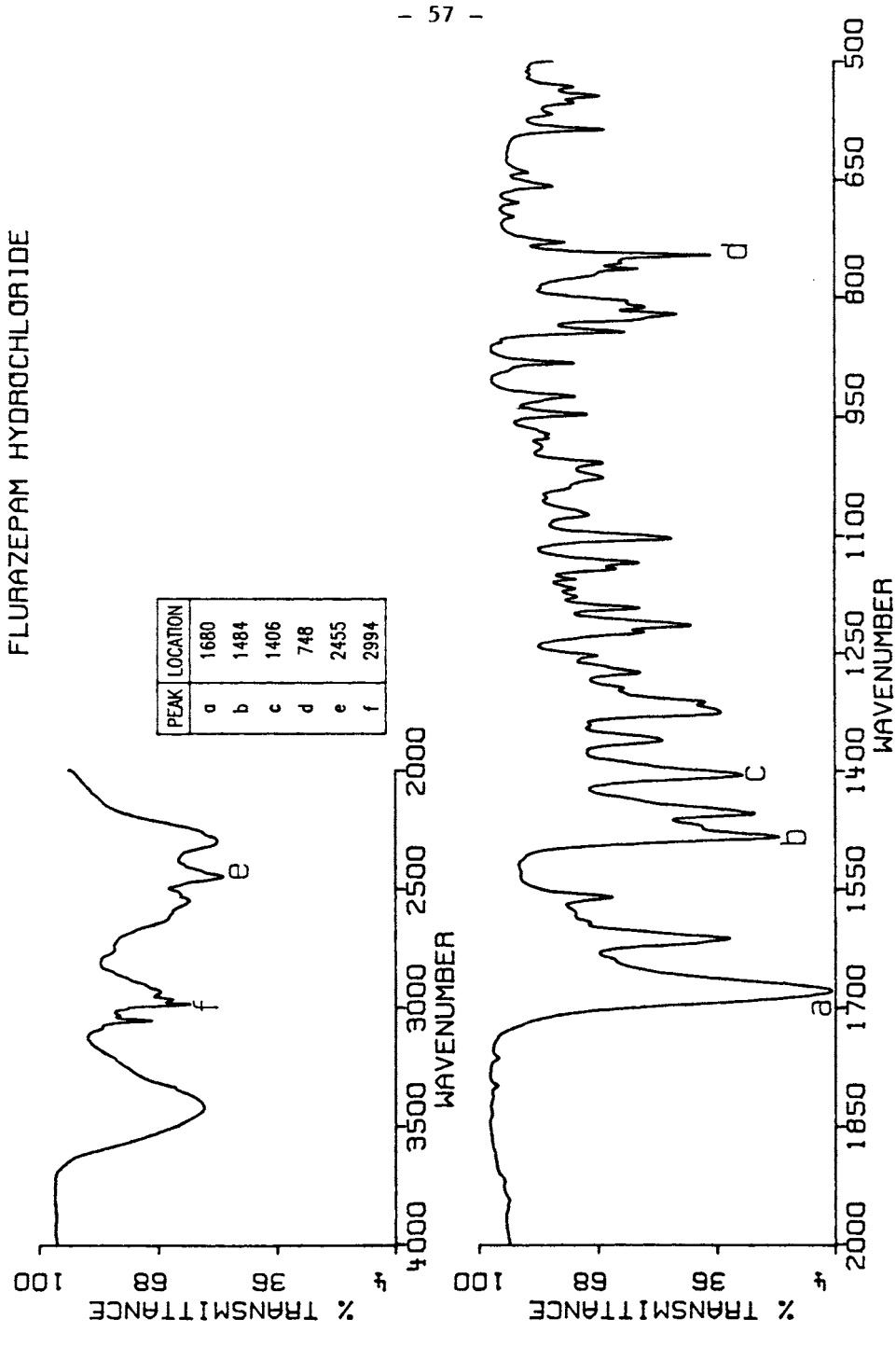


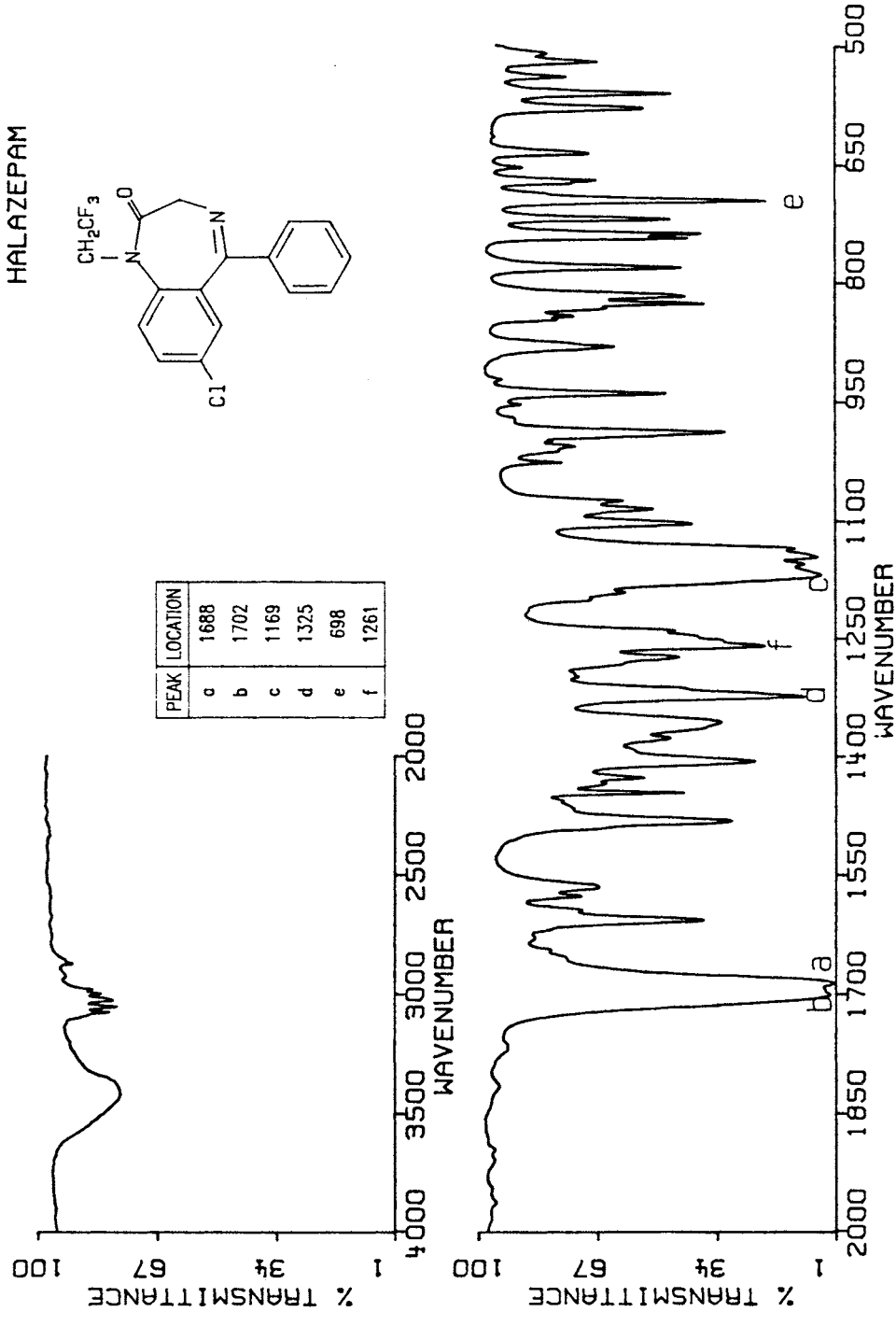
PEAK	LOCATION
a	1688
b	1336
c	1611
d	1524
e	1102
f	777

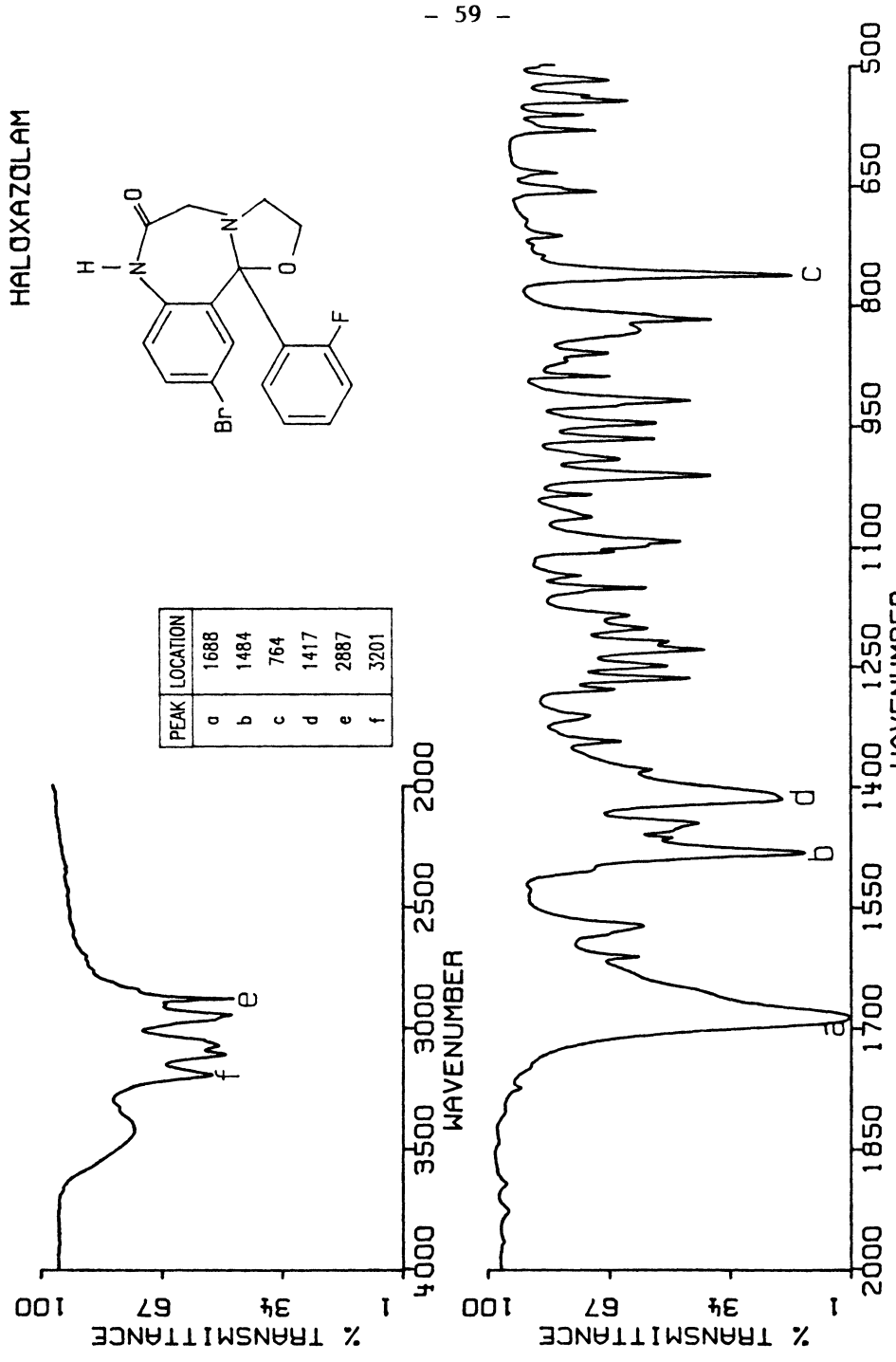


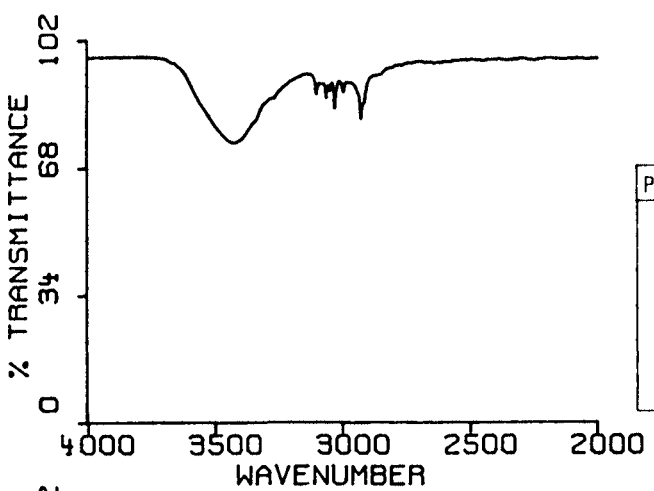



FLURAZEPAM
PEAK LOCATION
a 1680
b 752
c 1483
d 1326
e 1408
f 2970
% TRANSMITTANCE
WAVENUMBER
4000 3500 3000 2500 2000
2000 1850 1700 1550 1400 1250 1100 950 800 650 500
Cl
F
N
O
CH2CH3
CH2CH3
CH2CH2N


FLURAZEPAM HYDROCHLORIDE
PEAK LOCATION
a 1680
b 1484
c 1406
d 748
e 2455
f 2994
% TRANSMITTANCE
100
68
36
4
WAVENUMBER
4000 3500 3000 2500 2000
2000 1850 1700 1550 1400 1250 1100 950 800 650 500

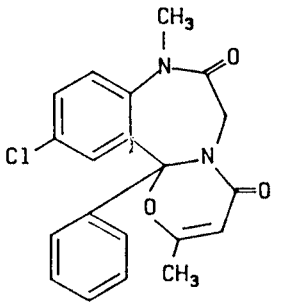

HALAZEPAM
CH_2CF_3
O
N
N
Cl
PEAK | LOCATION
a | 1688
b | 1702
c | 1169
d | 1325
e | 698
f | 1261
% TRANSMITTANCE
100
67
34
1
4000
3500
3000
2500
2000
WAVENUMBER
% TRANSMITTANCE
100
67
34
1
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
a
b
c
d
e
f

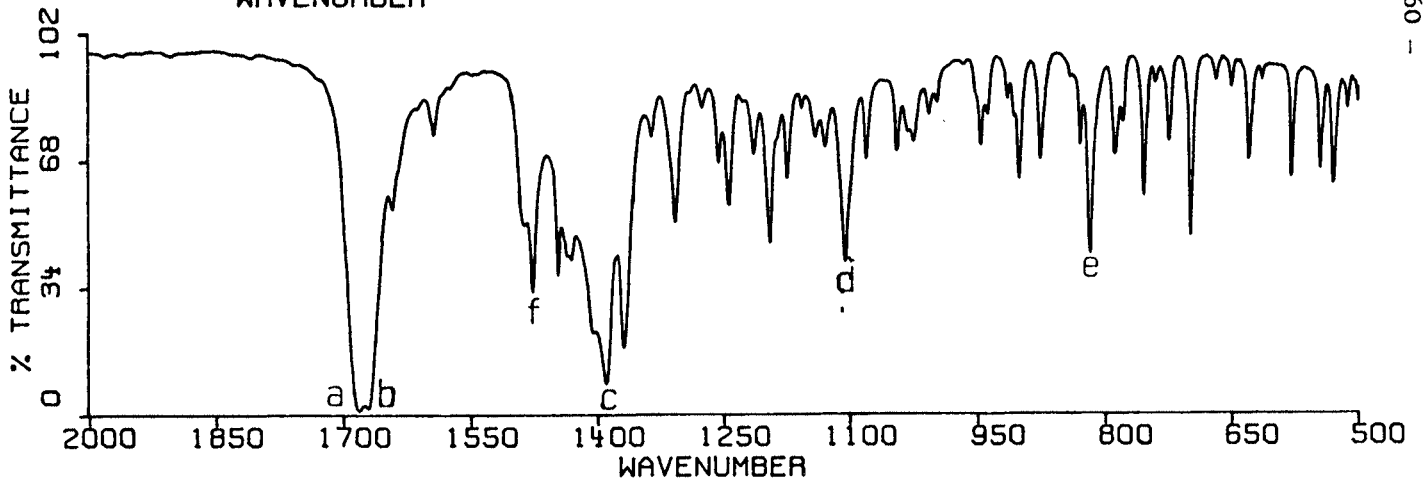

HALOXAZOLAM
PEAK	LOCATION
a	1688
b	1484
c	764
d	1417
e	2887
f	3201
% TRANSMITTANCE
100
67
34
1
4000
3500
3000
2500
2000
WAVENUMBER
2000
1850
1700
1550
1400
1250
1100
950
800
650
500

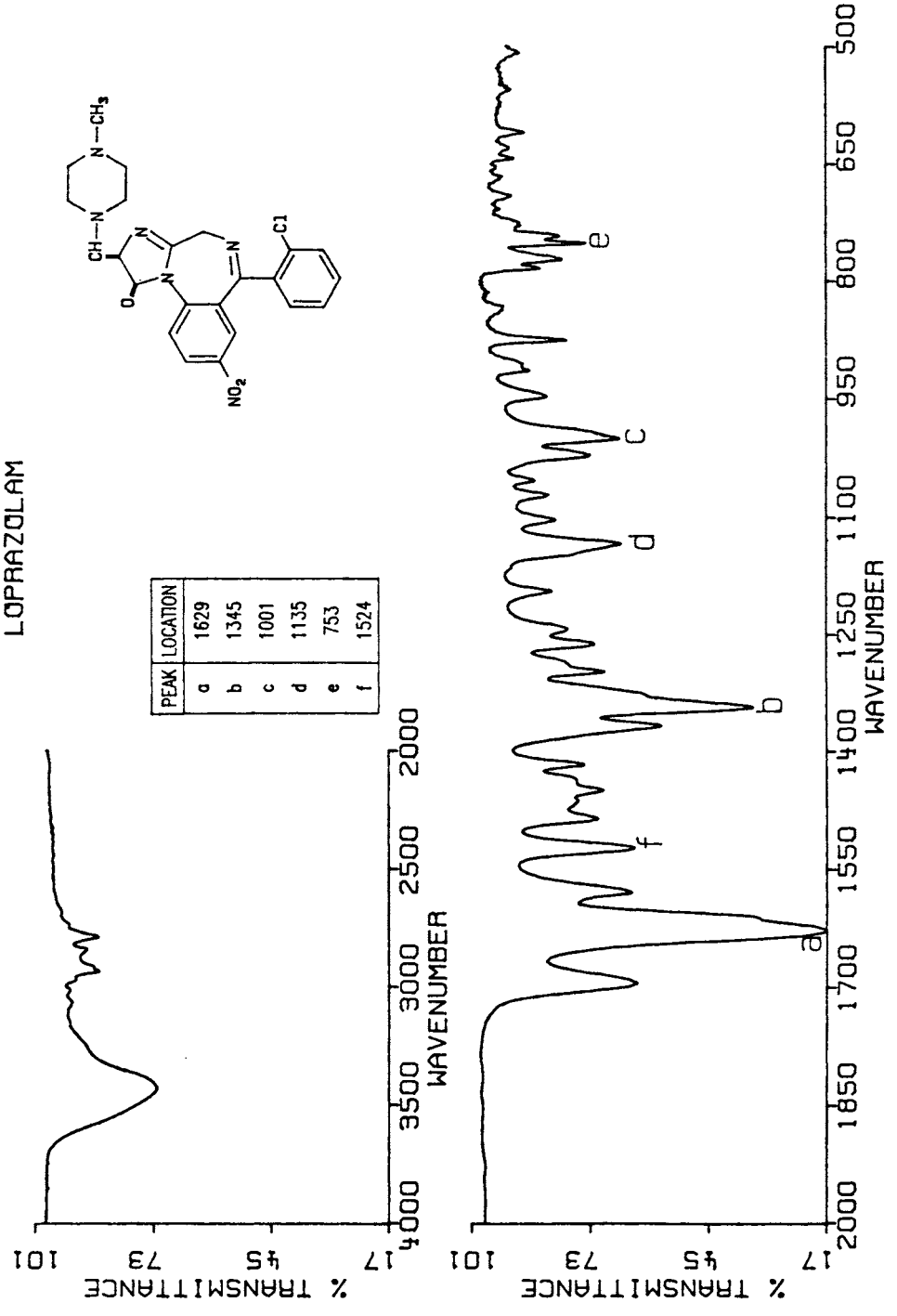


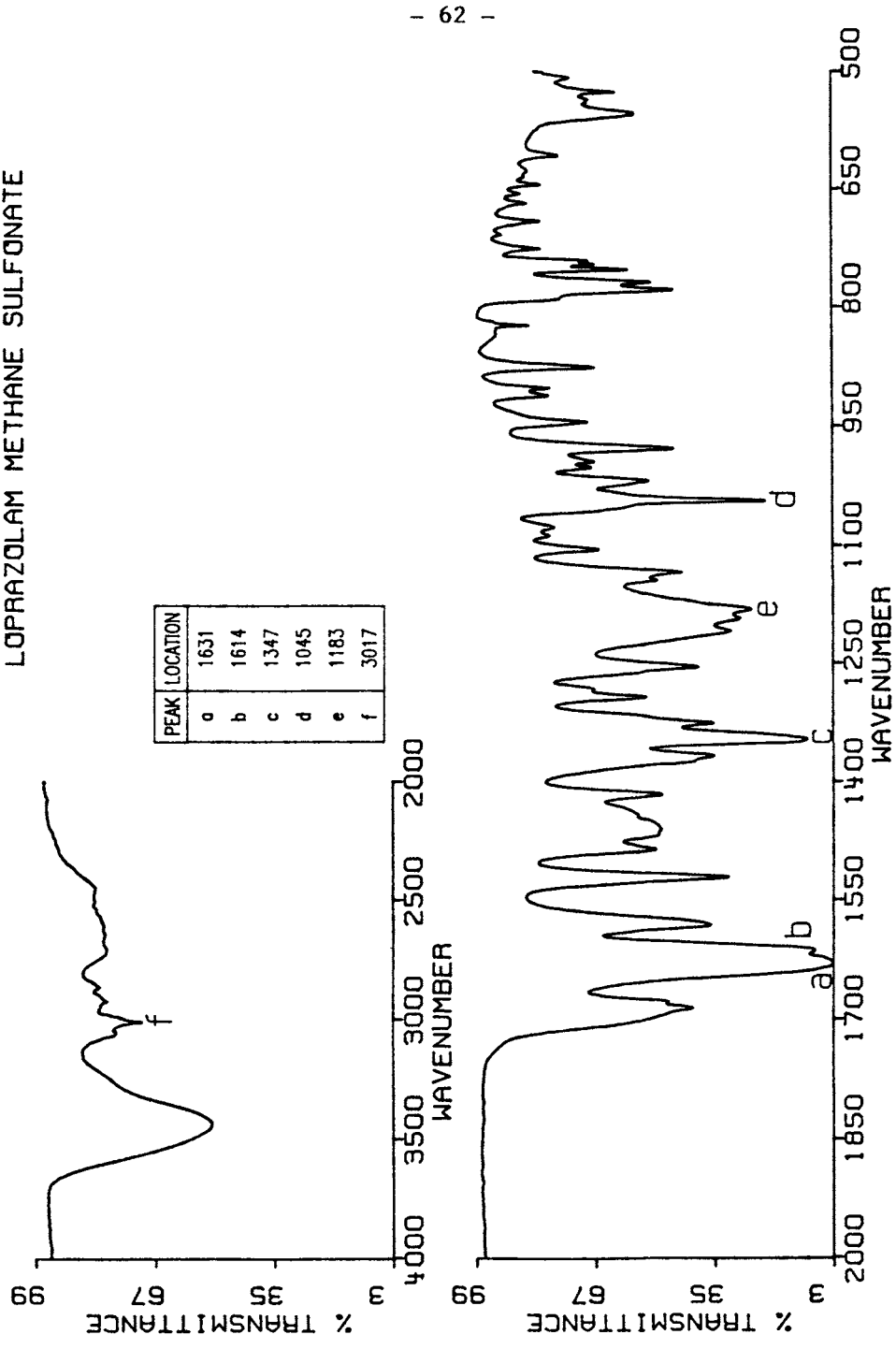
PEAK	LOCATION
a	1683
b	1672
c	1392
d	1108
e	821
f	1480

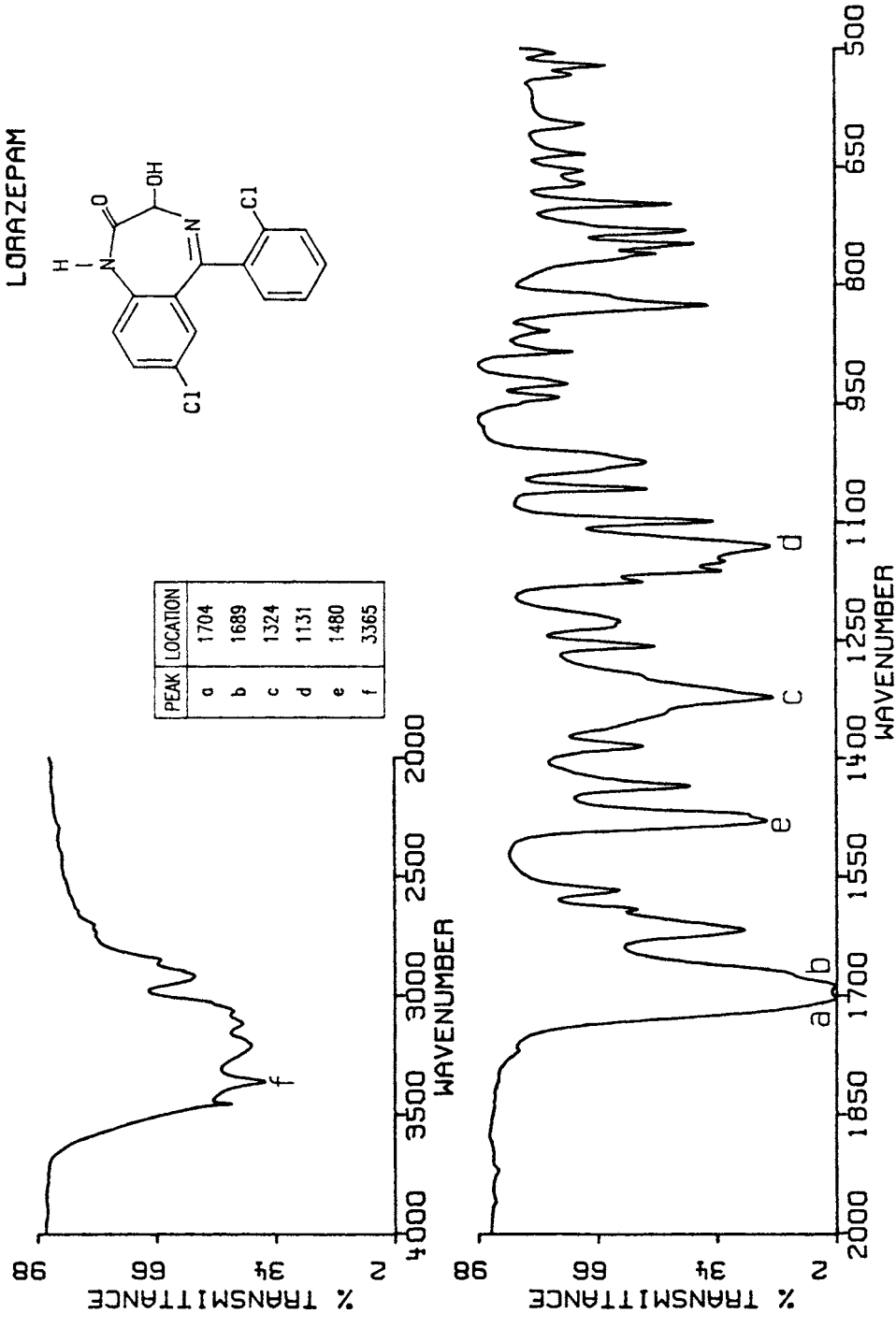
KETAZOLAM

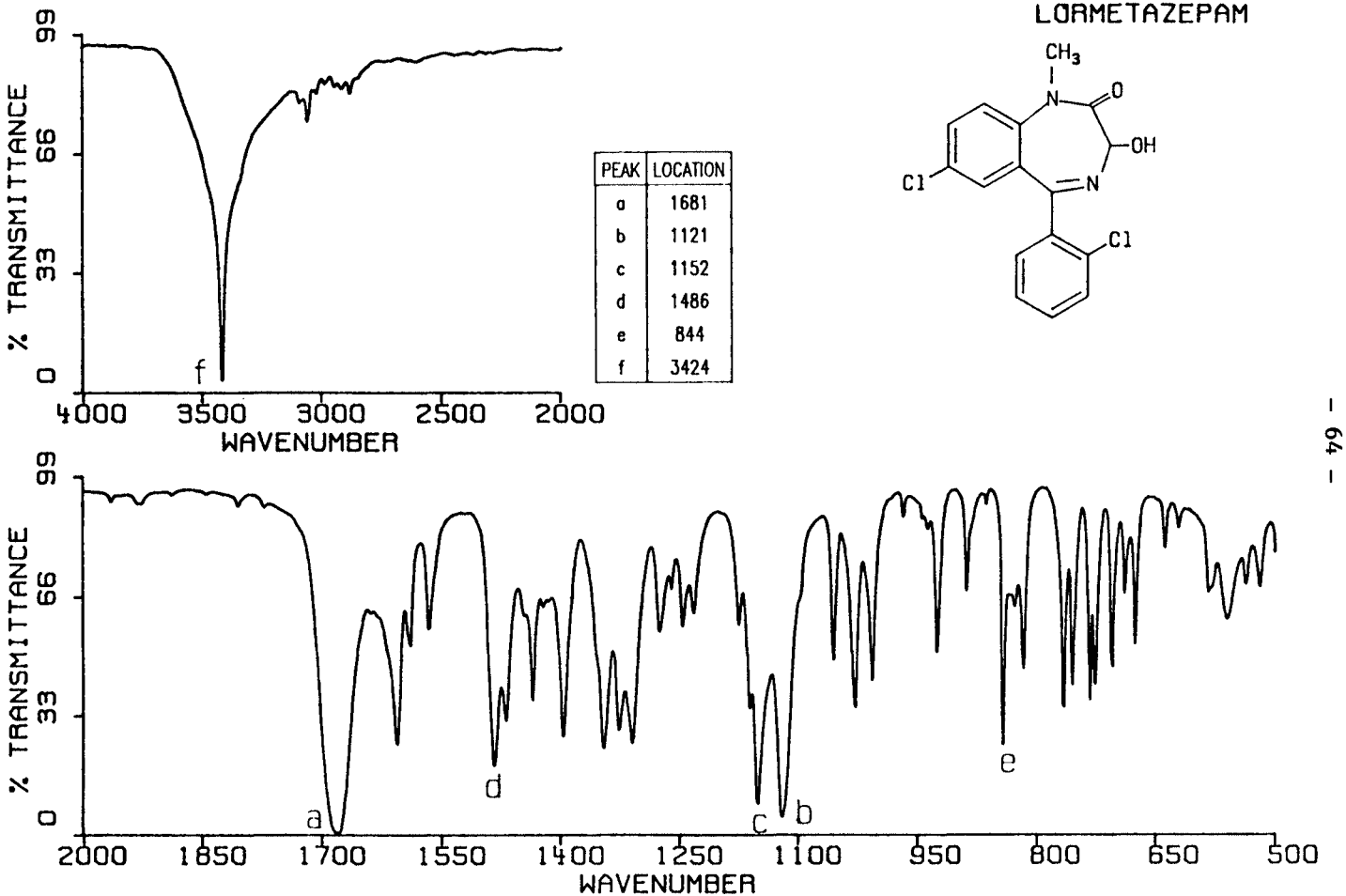


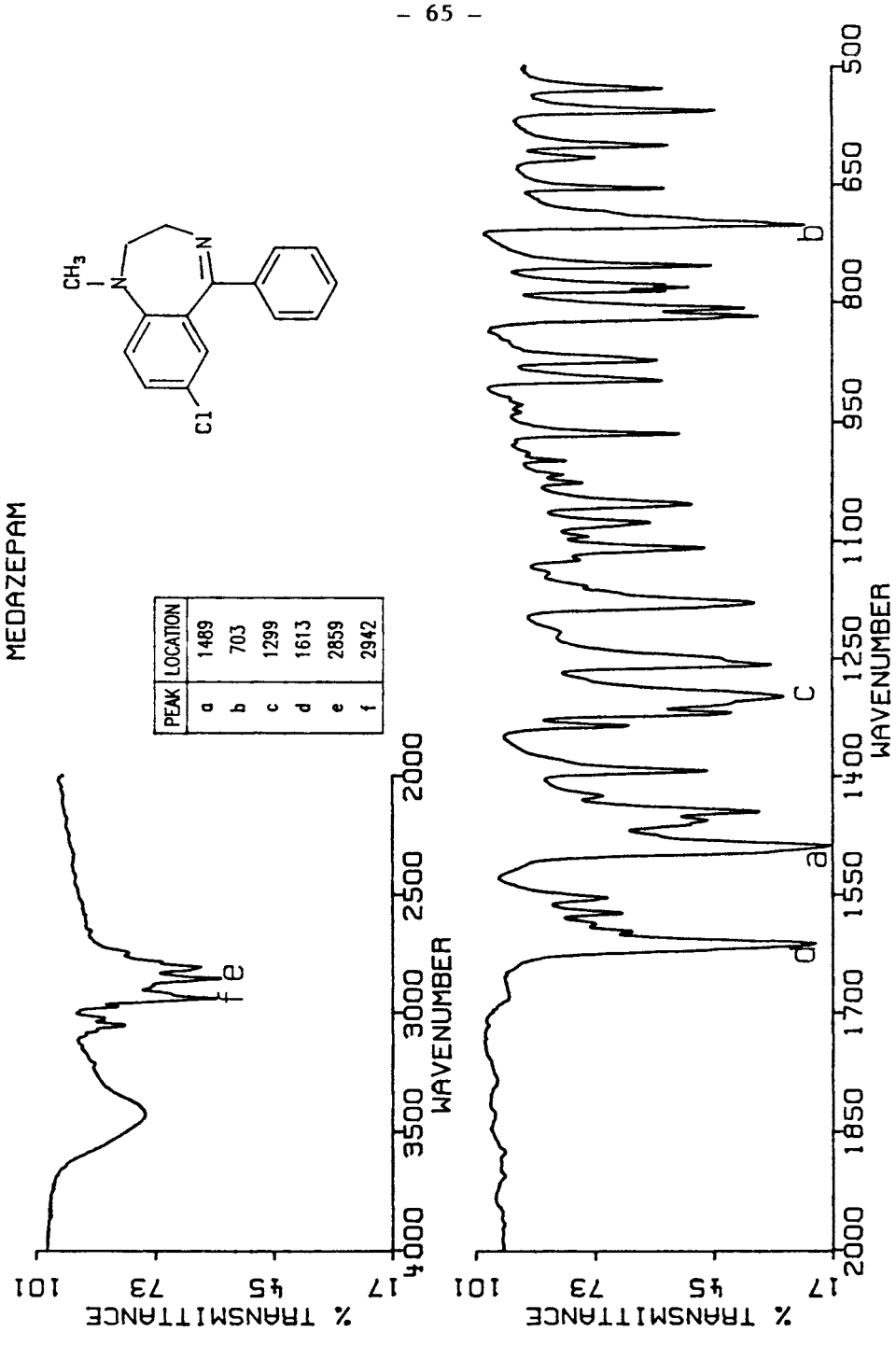


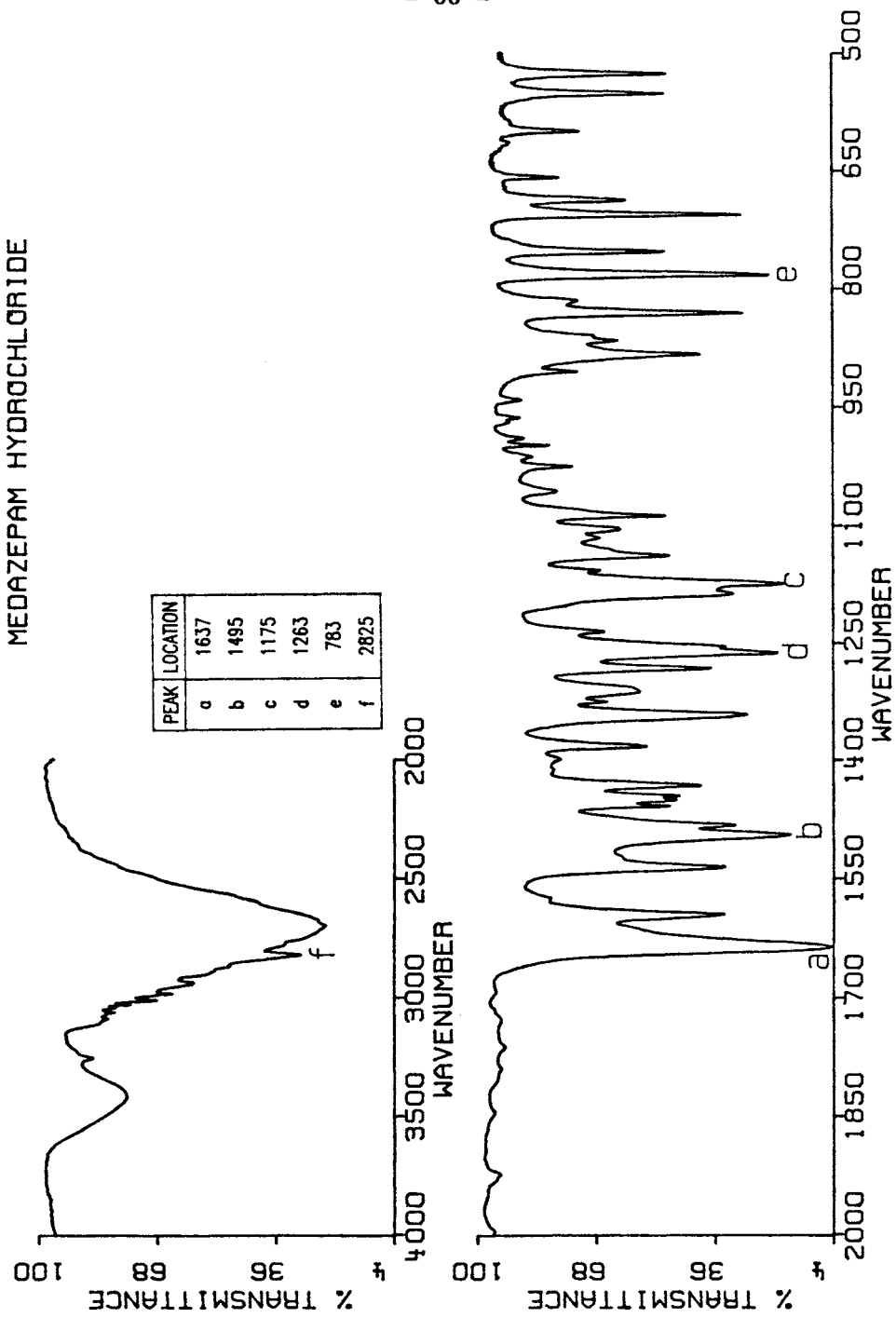

LOPRAZOLAM
PEAK | LOCATION
a 1629
b 1345
c 1001
d 1135
e 753
f 1524
% TRANSMITTANCE
101
73
45
17
4000
3500
3000
2500
2000
WAVENUMBER
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER


LOPRAZOLAM METHANE SULFONATE
PEAK | LOCATION
a | 1631
b | 1614
c | 1347
d | 1045
e | 1183
f | 3017
% TRANSMITTANCE
99
67
35
3
WAVENUMBER
4000
3500
3000
2500
2000
1850
1700
1550
1400
1250
1100
950
800
650
500

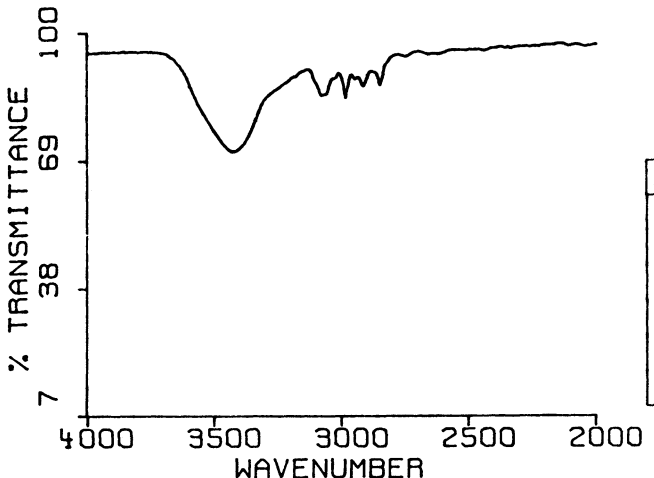

LORAZEPAM
PEAK | LOCATION
a | 1704
b | 1689
c | 1324
d | 1131
e | 1480
f | 3365
% TRANSMITTANCE
WAVENUMBER
4000 3500 3000 2500 2000
2000 1850 1700 1550 1400 1250 1100 950 800 650 500
98 66 34 2


LORMETAZEPAM
CH3
N
O
OH
Cl
N
Cl
% TRANSMITTANCE
99
66
33
0
f
4000
3500
3000
2500
2000
WAVENUMBER
PEAK | LOCATION
a | 1681
b | 1121
c | 1152
d | 1486
e | 844
f | 3424
% TRANSMITTANCE
99
66
33
0
a
d
c
b
e
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER

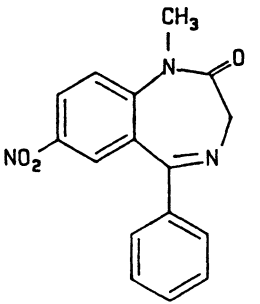

MEDAZEPAM
CH3
N
N
Cl
PEAK | LOCATION
a | 1489
b | 703
c | 1299
d | 1613
e | 2859
f | 2942
% TRANSMITTANCE
101
73
45
17
4000
3500
3000
2500
2000
WAVENUMBER
f
e
% TRANSMITTANCE
101
73
45
17
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
d
a
c
b

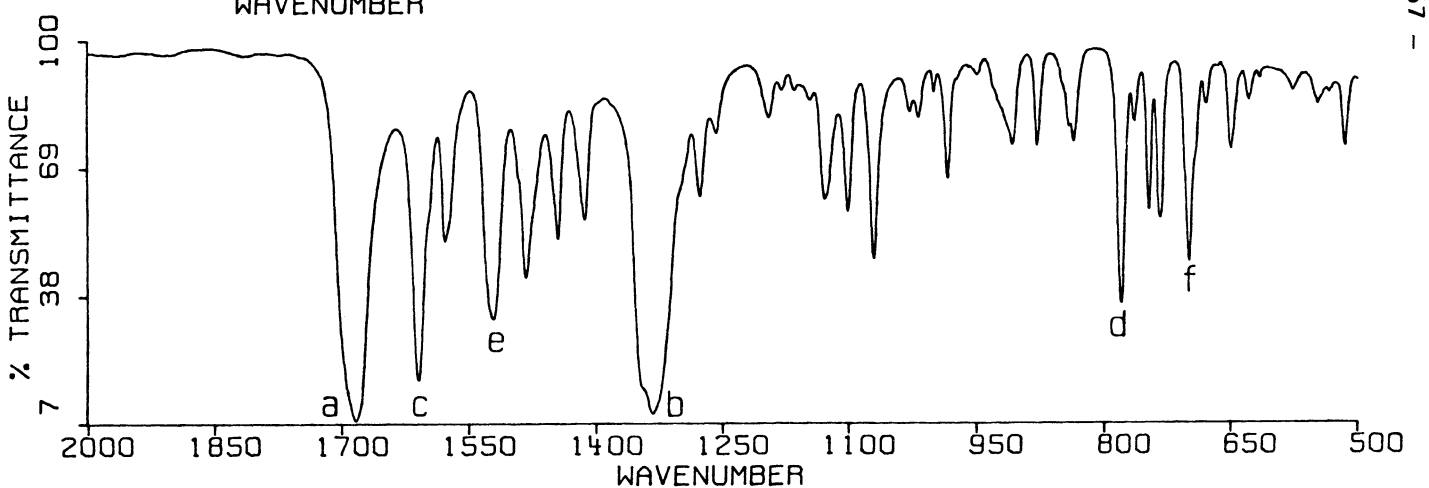

MEDAZEPAM HYDROCHLORIDE
PEAK | LOCATION
a | 1637
b | 1495
c | 1175
d | 1263
e | 783
f | 2825
% TRANSMITTANCE
100
68
36
4
WAVENUMBER
4000
3500
3000
2500
2000
1850
1700
1550
1400
1250
1100
950
800
650
500

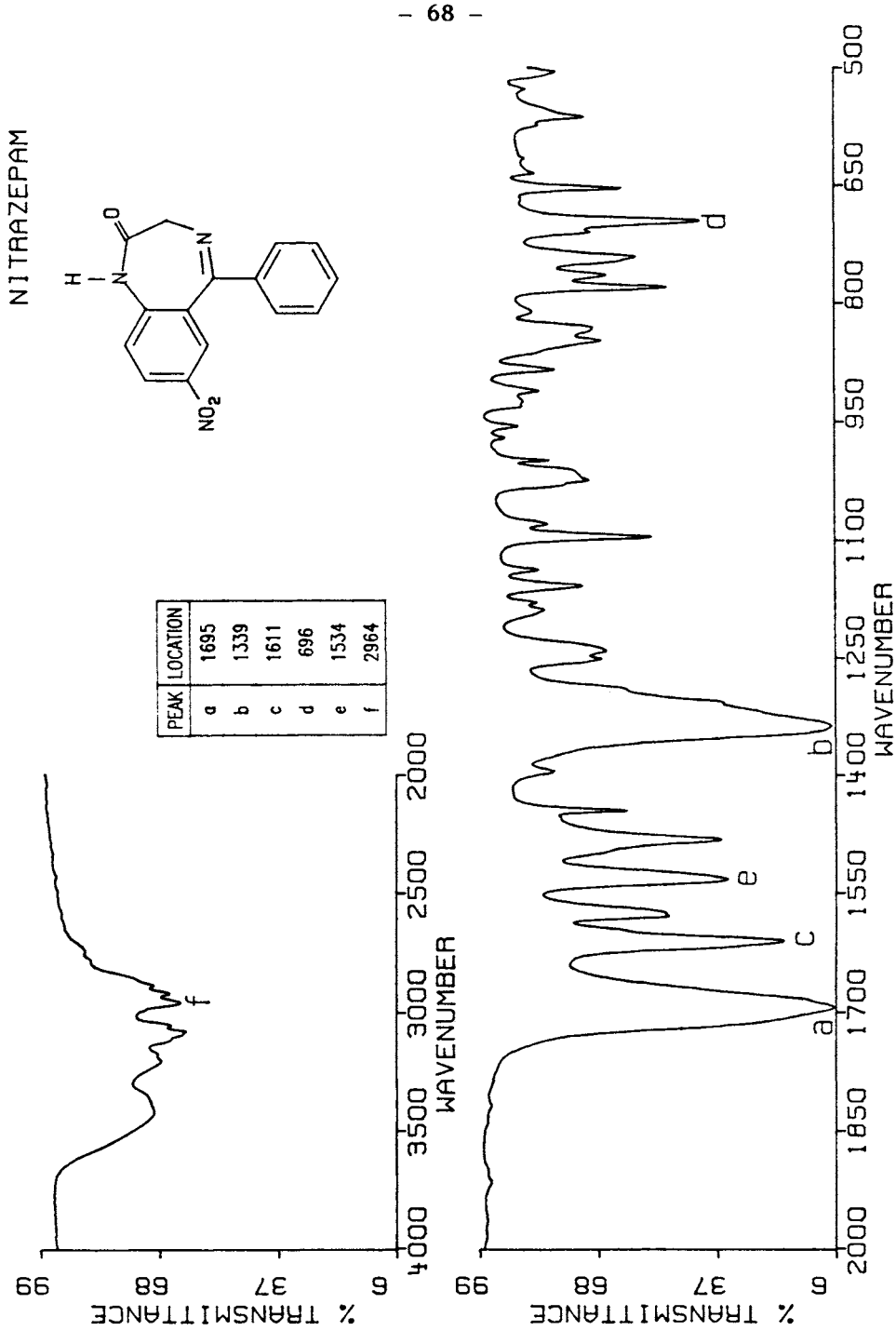
NIMETAZEPAM



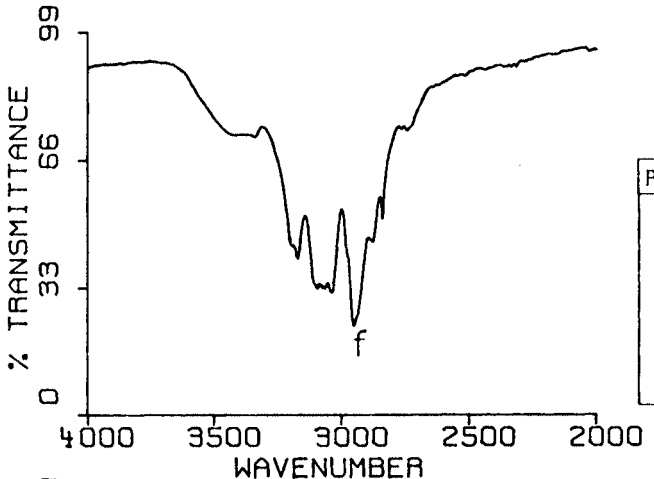
PEAK	LOCATION
a	1685
b	1335
c	1610
d	781
e	1523
f	701



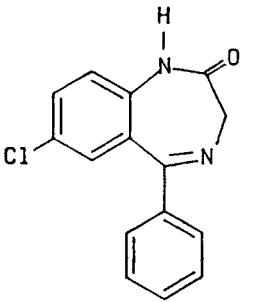


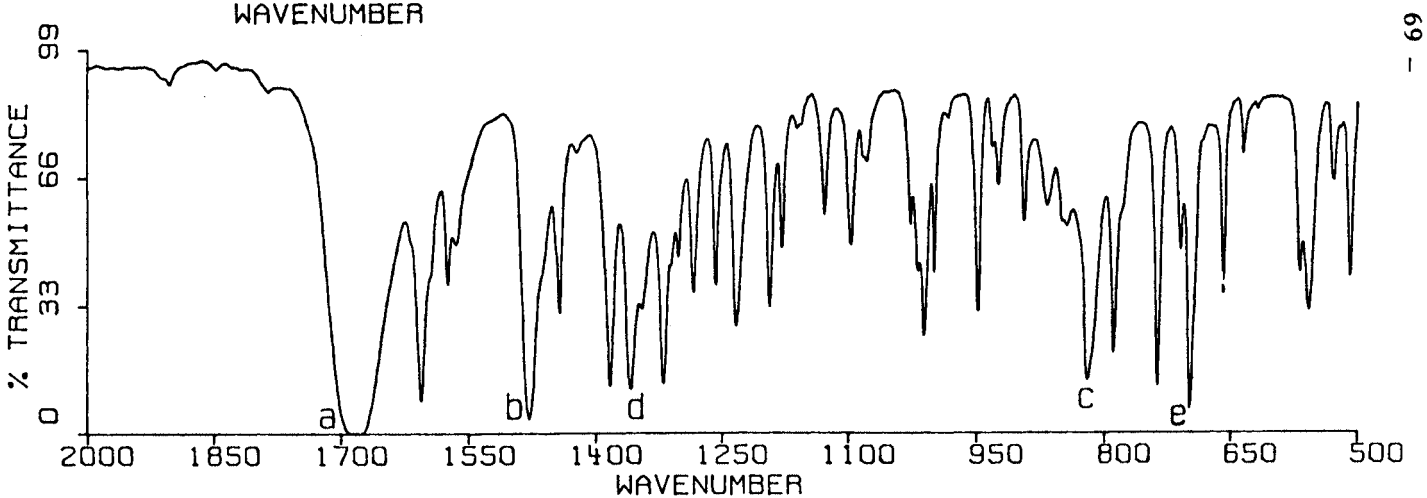

NITRAZEPAM
PEAK	LOCATION
a	1695
b	1339
c	1611
d	696
e	1534
f	2964
% TRANSMITTANCE
99
68
37
6
4000
3500
3000
2500
2000
WAVENUMBER
2000
1850
1700
1550
1400
1250
1100
950
800
650
500

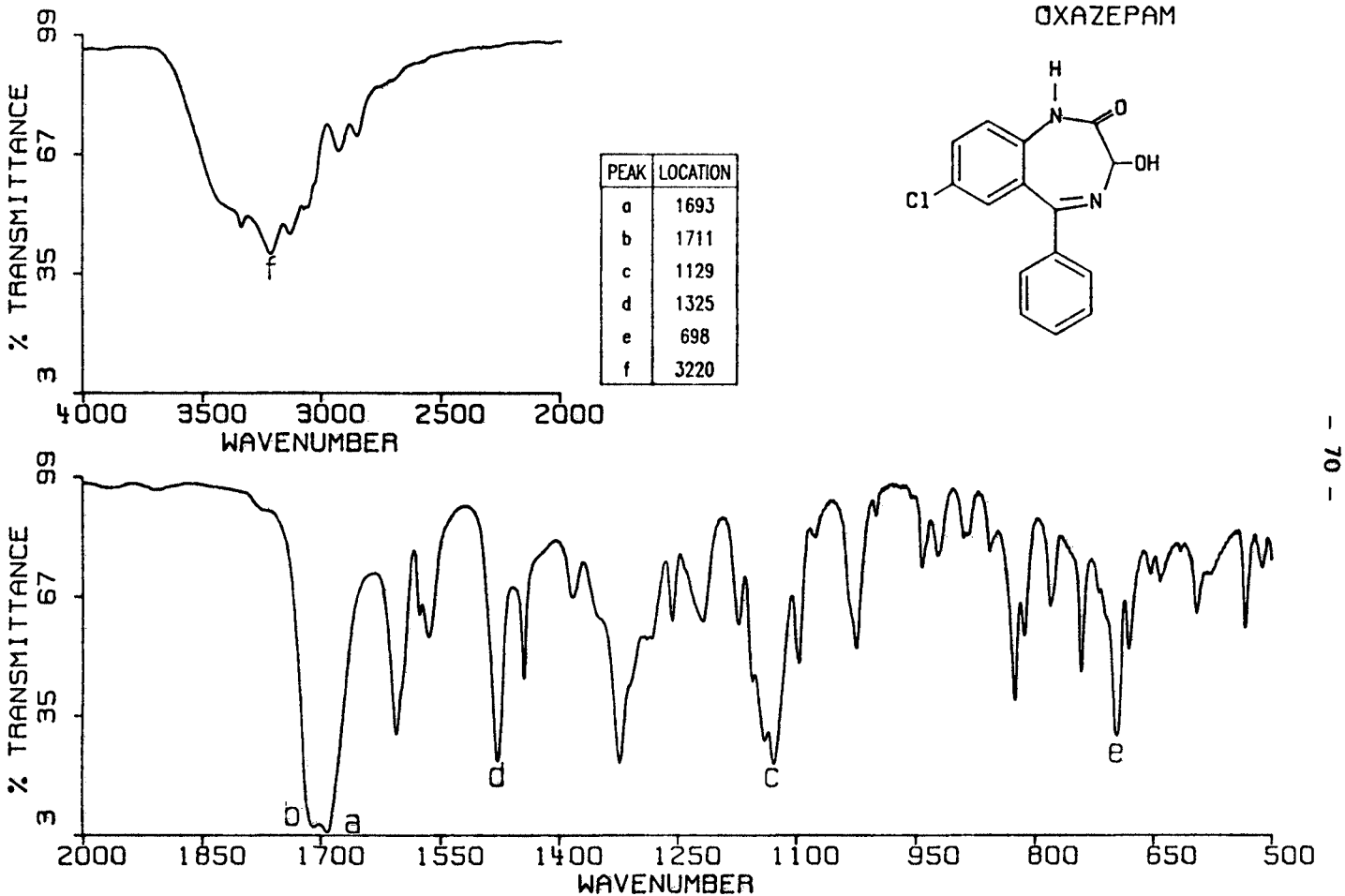
NORDAZEPAM

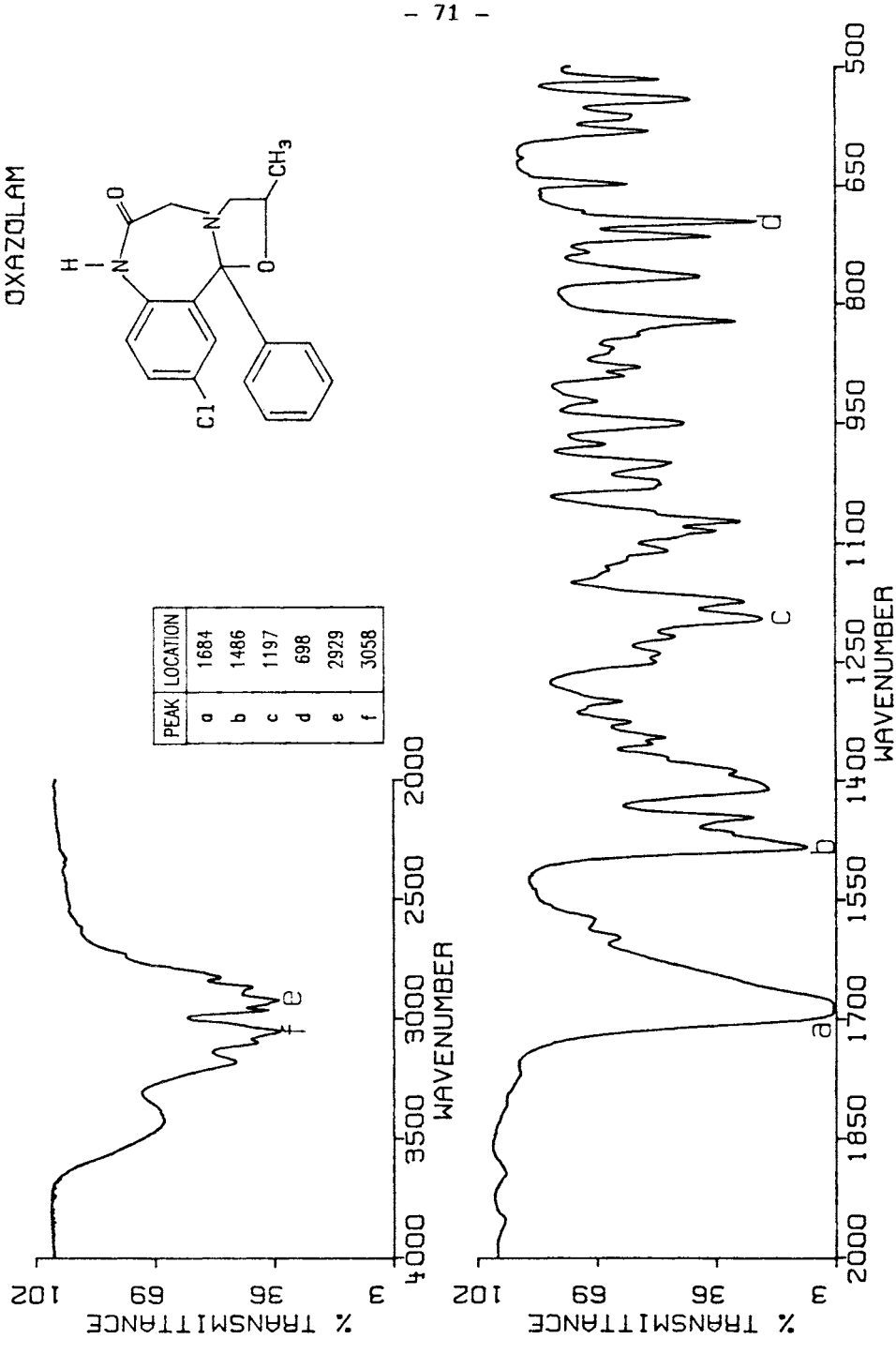


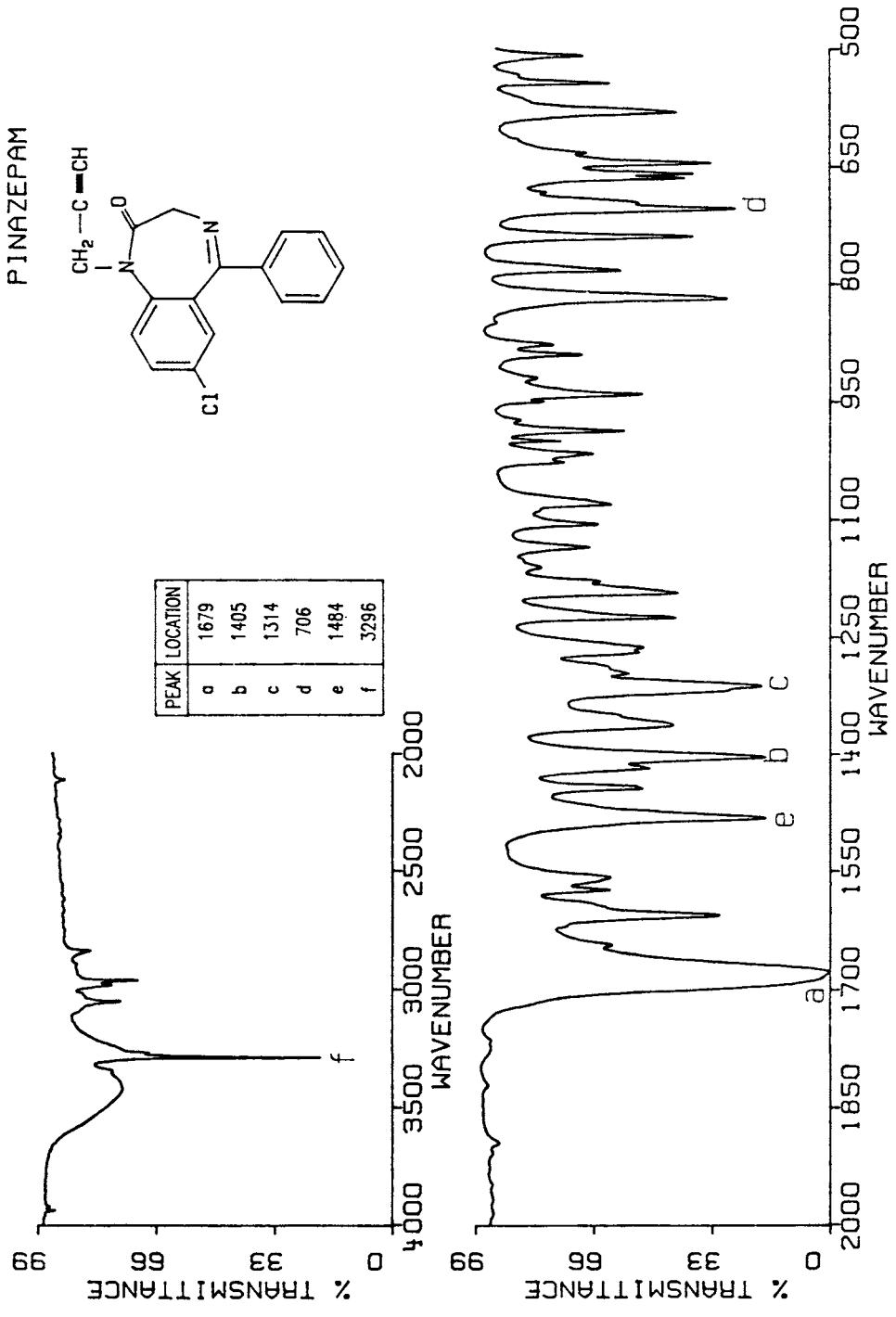
PEAK	LOCATION
a	1679
b	1480
c	822
d	1361
e	701
f	2958



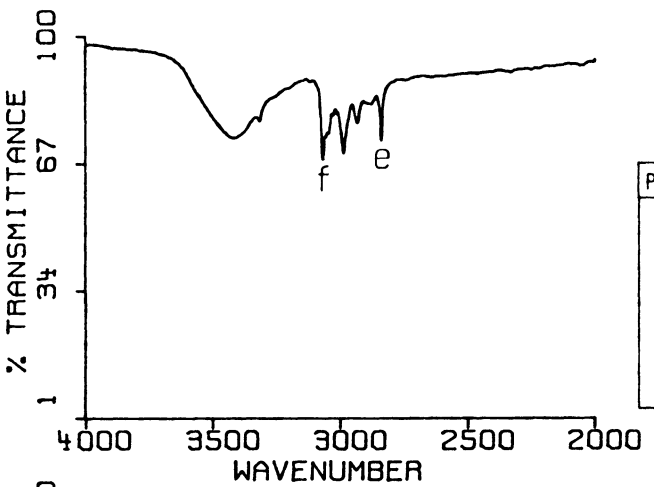



OXAZEPAM
% TRANSMITTANCE
99
67
35
3
4000
3500
3000
2500
2000
WAVENUMBER
f
PEAK | LOCATION
a | 1693
b | 1711
c | 1129
d | 1325
e | 698
f | 3220
H
N
O
OH
N
Cl
% TRANSMITTANCE
99
67
35
3
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
b
a
d
c
e

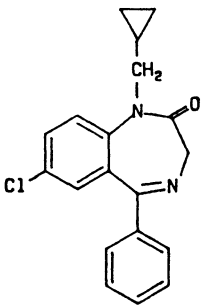

OXAZOLAM
PEAK | LOCATION
a | 1684
b | 1486
c | 1197
d | 698
e | 2929
f | 3058
% TRANSMITTANCE
102
69
36
3
WAVENUMBER
4000
3500
3000
2500
2000
1850
1700
1550
1400
1250
1100
950
800
650
500

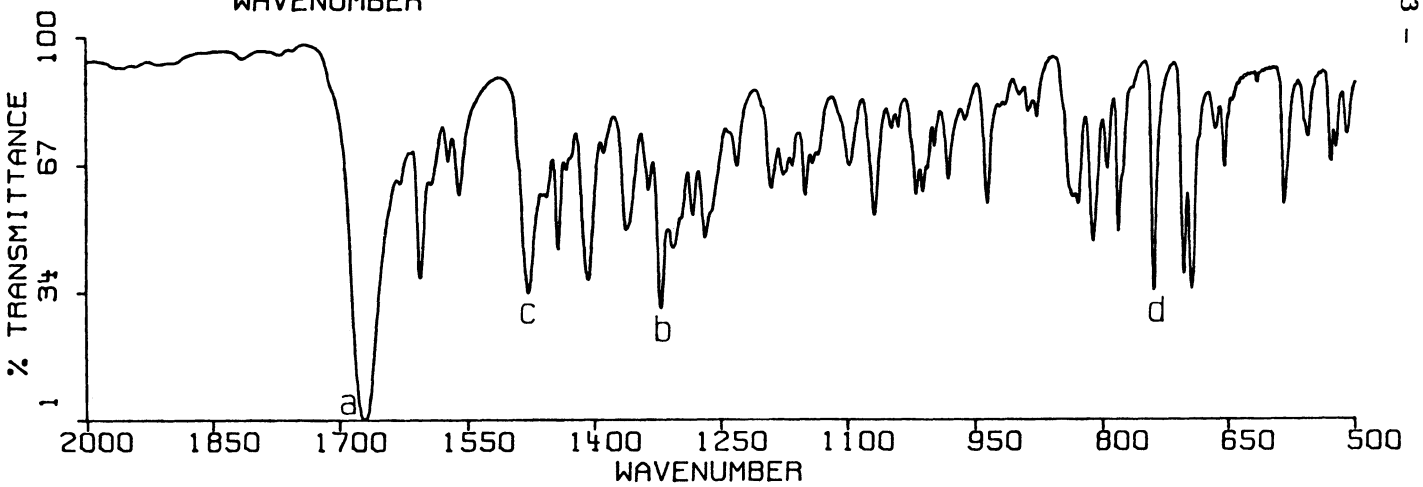

PINAZEPAM
CH2—C≡CH
O
N
N
Cl
PEAK | LOCATION
a | 1679
b | 1405
c | 1314
d | 706
e | 1484
f | 3296
% TRANSMITTANCE
99
66
33
0
4000
3500
3000
2500
2000
WAVENUMBER
f
2000
1850
1700
1550
1400
1250
1100
950
800
650
500
WAVENUMBER
a
e
b
c
d

PRAZEPAM

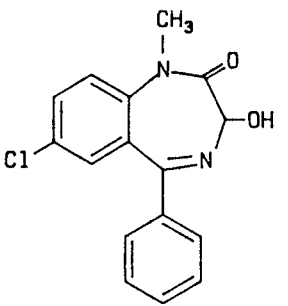


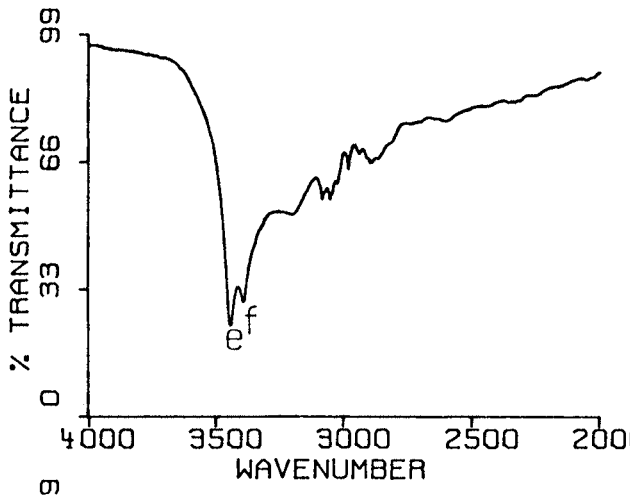
PEAK	LOCATION
a	1672
b	1323
c	1480
d	740
e	2846
f	3076



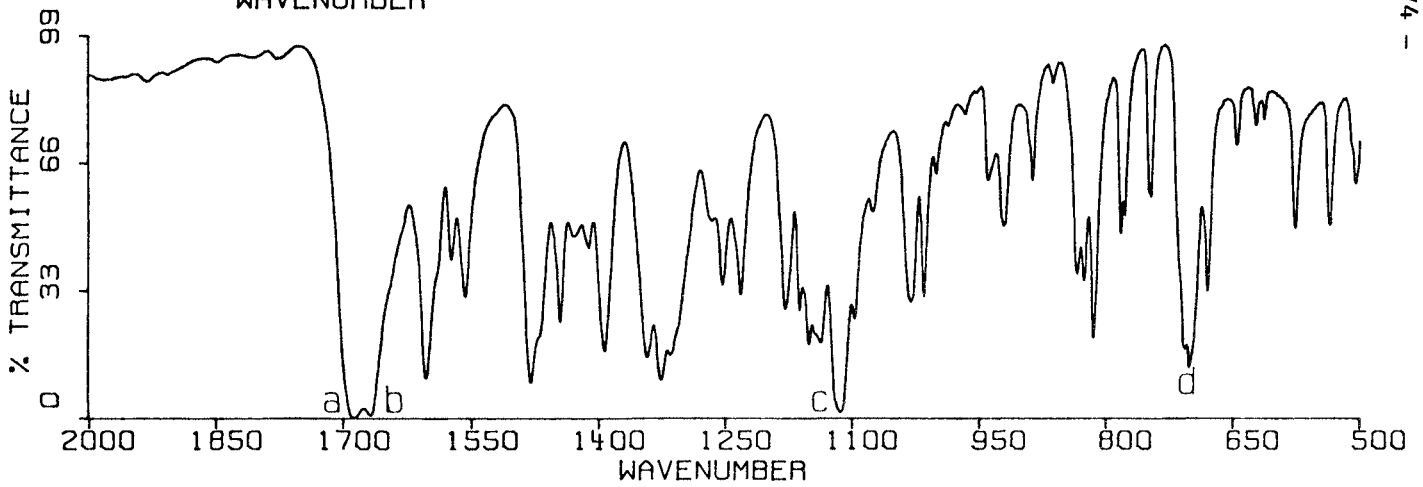


TEMAZEPAM

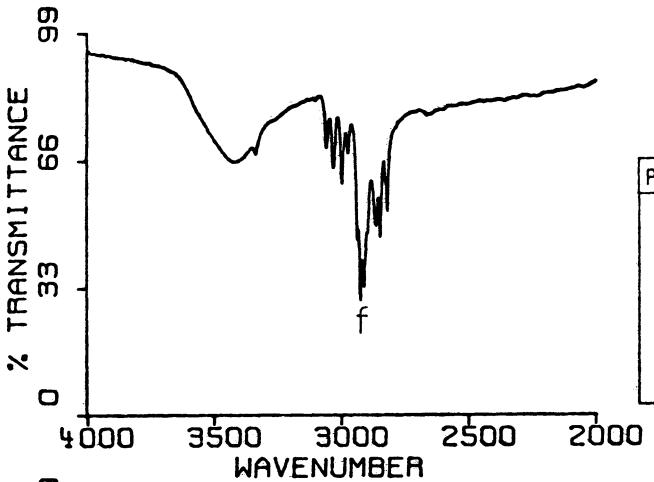




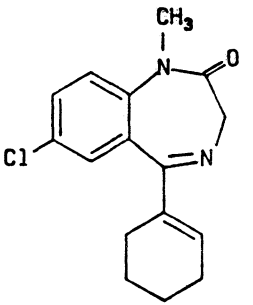
PEAK	LOCATION
a	1689
b	1670
c	1115
d	704
e	3450
f	3401

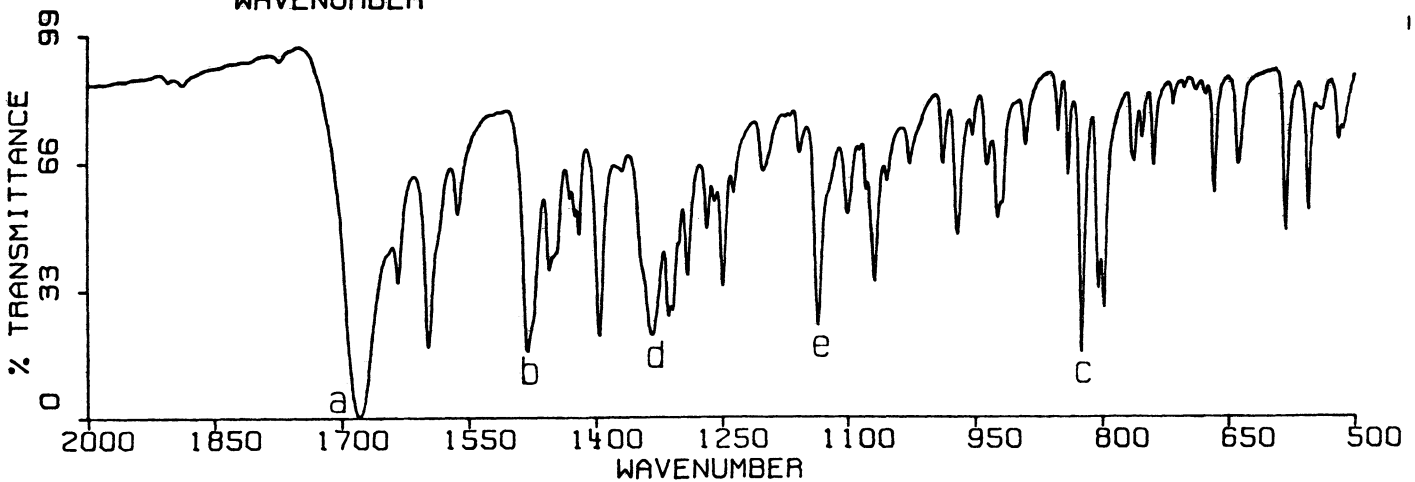


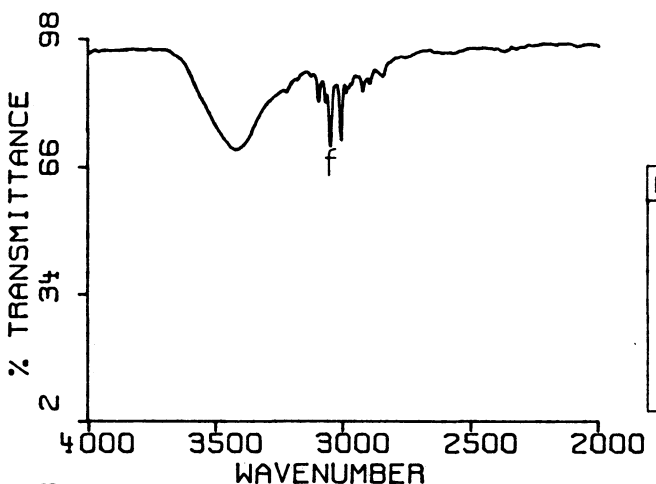
TETRAZEPAM



PEAK	LOCATION
a	1680
b	1483
c	826
d	1335
e	1136
f	2932







PEAK	LOCATION
a	1490
b	1427
c	1622
d	754
e	838
f	3060

TRIAZOLAM