

7. Measurement of the amount of substance

7.1 Measure Substance

Looking at the eyes is exploring light absorption characteristics. The odor detects volatile molecules and tastes are examining ion types and sugars. The estimation by such sensory function is more objective, the kind of material components is specified using a sensor (qualitative analysis), and the method of determining the contents of these components (quantitative analysis) is instrumental analysis. Machine Devices for analysis are called analytical instruments.

Analytical methods can be roughly divided into (1) spectroscopic analysis (absorption, emission, fluorescence), (2) electromagnetic analysis (X-ray, electron beam, NMR, ESR), (3) separation analysis (chromatograph, mass spectrometry) (4) Electrical analysis (ion selection electrode, electric titration, polarography), (5) others.

7.2 Principle of Instrumental Analysis

7.2.1 Spectrometry

Substances cause various interactions with electromagnetic waves depending on the movement of electrons and the movement of molecules. Spectrometry refers to examining the relation between interaction and electromagnetic wave energy and is used for various analyzes. Analytical methods that utilize light, in particular, are collectively called spectroscopic analysis. Among them, absorption spectroscopy that analyzes components using light absorption as an index and emission spectroscopy that uses emission light from a substance as an index It is often used.

In Fig.7.1, light refers to a specific wavelength range of electromagnetic waves. As shown in Fig.7.1, light having a wavelength of 400~750 nm is visible light, light of under 400 nm is ultraviolet light, and light of above 750 nm is infrared. In the infrared region, instead of the wavelength, a wave number (the number of waves per cm, 1 cm/wavelength) which is proportional to the frequency is often used. The wavelength λ is expressed by $\lambda = c / \nu$ using the frequency ν of the light and the speed c of the light.

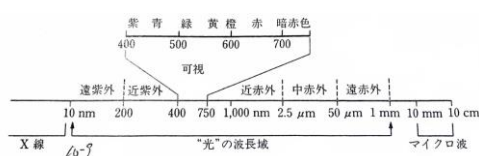


Fig.7.1 Wavelength Range of Light

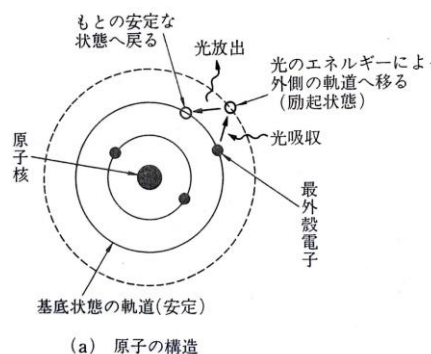
Therefore, we first explain why there is a correlation between light and matter.

A. Spectrum of Molecules and Atoms

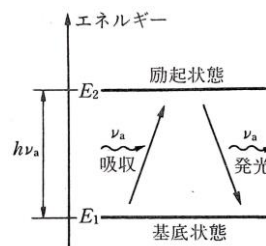
At the atom, electrons surround the atomic nucleus many times. The outermost electron is called the outermost shell electron and it has an important function to determine the nature of the atom. When light is irradiated to an atom in a stable state (ground state) as shown in Fig.7.2 (a), the outermost shell electron absorbs a part of light and becomes a higher energy state (excited state). That is, the shape of the atom slightly expands. Assuming that the energy levels of the ground state and the excited state are $E_1(J)$, $E_2(J)$, the frequency $\nu_a(\text{Hz})$ of the light absorbed at this time is $\nu_a = (E_2 - E_1)/h$ (h is Planck's universal constant), indicate specific values for atoms. This is schematically shown in Fig.7.2 (b). Absorption by such electronic level occurs in the ultraviolet~ visible region.

Atom expanse shape will immediately return to its original shape. In other words, the atoms in the excited state return to the ground state, but at that time they emit light of the frequency ν_a . Excitation of atoms occurs not only by light but also by heat and collision. The excitation and emission by heat can be explained by fireworks.

A molecule is formed by atoms gathered except for a part such as Ar and He. As shown in Fig.7.3 (a), since atoms are not rigidly coupled, they are constantly vibrating and further rotating.



(a) 原子の構造



(b) エネルギー準位

Fig.7.2 Excitation of Atoms by Light

Since the energy level of the vibrational motion is smaller than the energy level of the electron, it absorbs infrared radiation with small energy and changes its level. The change in the level of the rotational motion corresponds to far infrared and microwave, which have lower energy, and these energy levels are schematically represented as shown in Fig.7.3 (b).

Since the molecule has an electron level of a complicated structure compared to an atom, the spectrum of absorbed light also becomes complicated. In addition, because vibration and rotational motion also influence the level of electrons to broaden the absorption line, Absorption lines overlap, ultraviolet, visible absorption becomes broad curve (Fig.7.3 (c)).

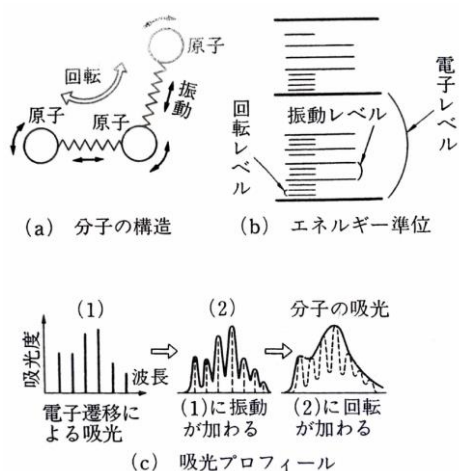


Fig.7.3 Molecular Structure and Absorption Waveform

B. Fluorescence

After the atoms or molecules are brought into the excited state (E_2) by the absorption of light, they release part of the excitation energy as thermal energy to transition to the lower excitation level (E'_2), and return to ground state (E_1), the emitted light is called fluorescence. In general, the wavelength of fluorescence is longer than the wavelength of excitation light, as shown in Fig.7.4, but the wavelength of absorbed light

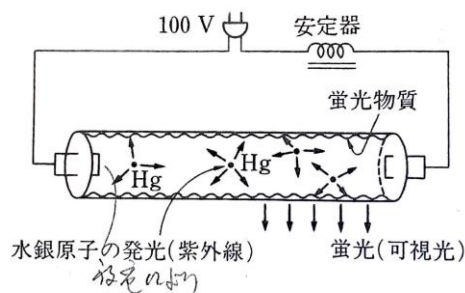


Fig.7.4 Construction of Fluorescence

(excitation light) and fluorescence may be the same in atom excitation, it is called resonance fluorescence. A method that uses such excitation light and emitted light as an indicator is called fluorometry, an example of familiar fluorescence is a fluorescent lamp, and a fluorescent substance is applied to the inner wall of the glass tube. The inside of the lamp is filled with mercury vapor, and ultraviolet rays are emitted from mercury by discharge. The ultraviolet rays excite the fluorescent material and emit visible fluorescence.

7.2.2 Spectroscopic Analysis

A. Monochromator

In order to perform spectroscopic analysis, it is necessary to disperse light by wavelength (energy), and a prism and a diffraction grating are used as dispersion elements. An apparatus that selects light of a single wavelength by using a diffraction grating or a prism is called a spectrometer or a monochromator. In the prism, the property that the refractive index of the glass becomes larger as the wavelength becomes shorter is used, and when white light is incident, it diffuses from purple to red as so-called rainbow color as shown in Fig.7.5. The lens of the camera and the microscope is a prism are smoothly connected to each other, and the change in the refractive index due to the wavelength is a change in the focal length, resulting in chromatic aberration.

For the diffraction grating, a transmission type diffraction grating formed by depositing opaque lines at equal intervals on the surface of a transparent glass plate to form multiple slits and a reflection type diffraction grating with equally spaced marks on the surface of the mirror There are. Diffraction means literally bending, light with wave property bends its path as part of the light goes out when obstacles on the straight line are on the order of the wavelength, if there is a slit surface perpendicular to the course as shown in Fig.7.6 (a), there are components that diffract at the edge and head towards various directions and components that go straight through the slit as it is. The diffracted lights of adjacent slits interfere with each other and strengthen each other when the optical path difference becomes an integral multiple of the wavelength, but if they do not,

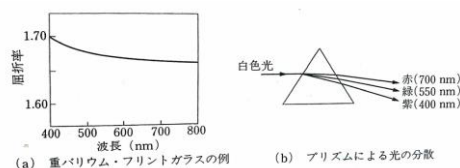


Fig.7.5 Wavelength Dependence of Refractive Index of Glass

they cancel each other out and gradually disappear.

Assuming that the interval between the slits is d , the wavelength is λ , and the diffraction angle is θ , the optical path difference a between diffracted light traveling in the θ direction from adjacent slits is expressed by next equation.

$$a = d \sin \theta$$

The condition under which the diffracted light is strengthened is as follows:

$$m\lambda = d \sin \theta \quad (m = 0, \pm 1, \pm 2, \dots)$$

The smaller the d (grating constant) is, the larger the dispersion becomes. For example, $d = -1 / 200 \text{ mm}$, when $\theta = 7^\circ$, $\lambda = 600 \text{ nm}$, and the light of 600 nm , 300 nm , 200 nm , ... goes in that direction. Dispersion can also be obtained by the same method for the reflection type. Fig.7.6 (b) shows the light intensity distribution of the diffraction grating when monochromatic light of wavelengths λ_1 and λ_2 is incident.

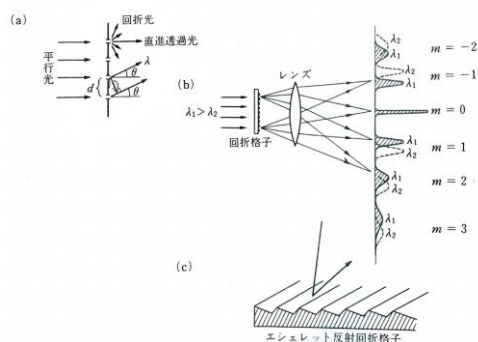


Fig.7.6 Diffraction Grating

B. Spectrophotometer

In the absorption analysis, there is the following relationship between the light absorption amount and the concentration C of the measurement substance according to the Lambert-Beer law.

$$C \propto A = -\log \frac{I}{I_0}$$

However, I_0 is light incident on the sample, I is light after passing through the sample, (I/I_0) is transmittance, A is absorbance.

Therefore, in the absorption spectrometer, as shown in Fig.7.7, monochromatic light is obtained from a white light source by a monochromator, qualitative analysis is performed by obtaining the position (peak wavelength) of the absorption waveform of the measurement sample. In the quantitative analysis, the wavelength of the monochromator is adjusted to the absorption peak wavelength of the sample, and the absorbance is determined from the ratio between the sample transmitted light amount I and the incident light

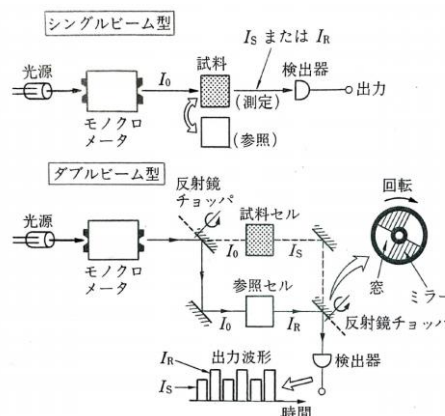


Fig.7.7 Absorption Spectrophotometer

amount I_0 . The apparatus for performing the qualitative and quantitative analysis in this manner is referred to as an absorption spectrophotometer. There are single beam type and double beam type. In actual analysis there is the influence of light absorption of measuring cell, solvent or contaminants, which needs to be corrected. Therefore, in the single-beam type, firstly the transmitted light intensity I_R of the glass cell and the solvent is obtained using a reference sample (blank cell), then the absorbance of the reference cell and the sample cell for obtaining I_S by replacing it with the measurement sample are A_R , A_S .

$$A_R = -\log \frac{I_R}{I_0}, \quad A_S = -\log \frac{I_S}{I_0}$$

The true absorbance A_C of a sample corrected for absorbance of a solvent or a cell is given by next equation.

$$A_C = A_S - A_R = -\log \frac{I_S}{I_R}$$

In the double beam method, monochromatic light is divided into two light fluxes using a reflector chopper, and I_S and I_R are alternately obtained, so that the measurement time is shortened and the intensity fluctuation of the light source is offset at the same time.

In the luminescence analysis, qualitative analysis is performed from the emission line wavelength of the emission spectrum from the sample, and since its light amount is proportional to the amount of analytical element, it is measured with a single luminous flux. In this case, in the method of scanning the monochromator to sequentially obtain the emission spectral intensity of the predetermined wavelength, the emission intensity often changes during the wavelength scanning, which hinders the quantification. Therefore, the dispersion by the highly dispersive diffraction grating, A system is adopted in which slits and

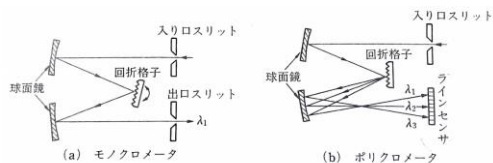


Fig.7.8 Monochromator and Polychromator

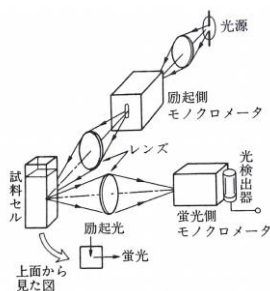


Fig.7.9 Spectrofluorometer

photosensors are installed at the positions of the corresponding element wavelengths and line sensors are attached to quantify multiple elements at the same time. Such a spectroscopic optical system is called a polychromator (Fig. 7.8).

Spectrofluorometer is two monochromators are used to find the excitation spectrum and the fluorescence emission spectrum, excitation and fluorescence beams are orthogonal to each other to prevent excitation light mixing into the fluorescence side detector (Fig. 7.9). Since mixing is not negligible when the wavelengths of excitation light and fluorescence are close to each other, a filter that cuts unnecessary long wavelengths is inserted on the excitation side, and a filter that cuts the excitation wavelength is inserted into the detector side.

Lambert-Beer's law

"When monochromatic light passes through the solution, monochromatic light is absorbed by the light absorbing material in the solution and its proportion is proportional to the thickness of the absorbing material layer" is the Lambert's law, "the rate of absorption is It is proportional to the concentration of absorbing molecules "is Beer's law. These two are collectively referred to as Lambert-Beer's law.

7.2.3 Separation Analysis

A. Chromatography

Crude oil in petroleum contains a lot of hydrocarbons such as methane, ethane, ethylene, etc. Since the absorption spectra of these components overlap, the components and their contents cannot be specified by spectroscopic measurement. However, each component has a difference in diffusion rate and affinity for adsorbent. A method of developing and separating

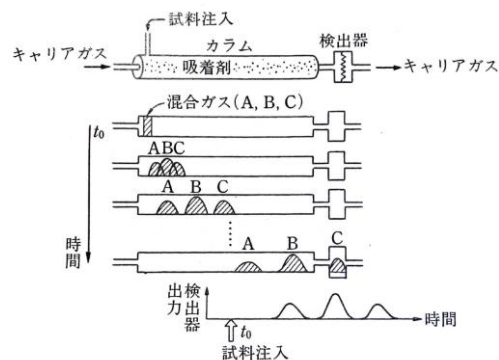


Fig.7.10 Gas Chromatography

inclusions into each component by utilizing such properties is a series of methods called chromatography.

Fig.7.10 shows the operating principle of gas chromatography. Hydrogen, helium, nitrogen, argon, etc. which do not react with the sample gas are used as the carrier gas and flowed to the column at a constant flow rate. A small amount of sample gas (about 1 ml) is instantaneously injected into the column at a certain point. The injected sample gas is swept away by the carrier gas and passes through the column, and in the process, the mixed components are sequentially separated. Since the thermal conductivity and the ionization rate of the carrier gas and the sample component gas are different from each other, when a heat conduction type gas sensor, a hydrogen flame ionization detector or the like is attached to the column outlet, the waveform of the separated component gas is outputted as an electric signal. Components are known from the waveform peak position, and content is calculated from the waveform area or peak height.

B. Mass Spectrometry

Fig.7.11 shows mass spectrometry method, which is called single convergence field type, in which electrons collide and high the measurement gas molecules ionized by applying an electric field are accelerated by an electric field of several thousand volts and made to proceed in the magnetic field. The trajectory is bent according to Fleming's left-hand rule because the advancing direction is perpendicular to the magnetic field, at this time, there is the following relationship between the ion mass m and the ion charge Z . The ions with larger mass draw a larger circle, so if you place a detector and change H or V , the ratio It is possible to separate ions by m/Z .

$$\frac{m}{Z} = \frac{eR^2H^2}{2V}$$

However, e is electric charge (1.60×10^{-19} clone), Z is charge number, V is electric

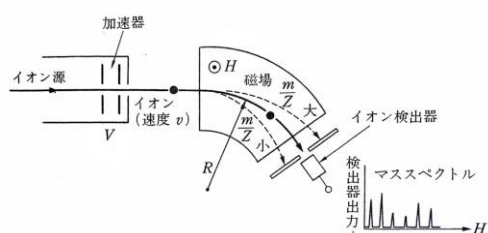


Fig.7.11 Single Convergence Field

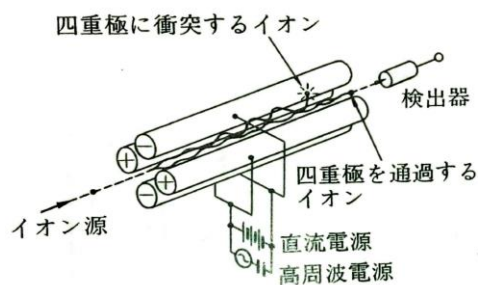


Fig.7.12 Quadra Pole

field strength, H is magnetic field strength. Quadra pole is inferior in resolving power compared with single focusing magnetic field type, it is often used for small size and capable of high speed scanning. In this system, as shown in Fig.7.12, four parallel rod electrodes By multiplying the opposing electrodes by the high-frequency electric field $\pm(A + B\cos\omega t)$ of frequency ω , the ions move while vibrating along the central axis of the electrode (However, A is the direct current voltage, B is the direct voltage, In this case, although the vibration amplitude of a specific ion can reach the detector without increasing it, other ions scatter outside the orbit. Therefore, m and Z can be selected by scanning A and B while keeping A/B constant.

7.2.4 Electrical Analysis

A. Conductometry

Salt, sulfuric acid, caustic soda, etc. can dissociate into cations and anions in minerals and show conductivity. The easiest way to measure the concentration of the electrolyte solution is to measure the electric conductivity conductance at constant temperature. Theoretically, we set the electric resistance $R(\Omega)$ by placing two face-to-face electrodes (each area is $A(m^2)$) by distance $L(m)$ in the measurement solution, conductivity $\sigma(1/\Omega \cdot m)$ is given by next equation.

$$\sigma = \frac{L}{RA}$$

Fig.7.13 shows, there is a proportional

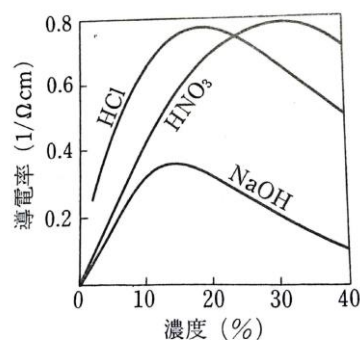


Fig.7.13 Relation of Concentration and Conductivity

relationship between concentration and conductivity within a certain range. Since the conductivity of pure water is zero and the value of σ increases as it is contaminated with ammonia water, metal ions, etc. Conductivity measurement method is used for simple water quality inspection However, It is not selective and not suitable for qualitative analysis.

B. Polarography

As a method for performing qualitative and quantitative analysis of the electrolytic solution while measuring the conductivity, there is a

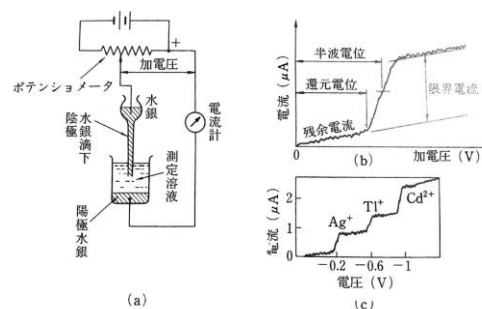


Fig.7.14 Polarography

polarograph. As shown in Fig.7.14, mercury sunk in the measurement solution is used as the anode, and mercury is dropped through the capillary tube as the cathode. When the relation between the voltage applied between the anode and the cathode and the current is recorded, Until the applied voltage reaches the reduction (or oxidation) potential peculiar to the analyte, only a very small amount of current flows (which is called residual current), but when the applied potential exceeds the reduction potential, a current flows stepwise and it is proportional to the substance concentration As the constant current value of the quantity is obtained, a waveform like the one shown in Fig.7.14 (b) is obtained Here, if we define the incremental current in the form of a step as the limiting current and the applied

voltage when obtaining the half current as the half-wave potential, the half wave potential shows the value peculiar to the measuring substance , qualitative and quantitative analysis can be performed because the limiting current value is proportional to the concentration of the substance. When there are mixed substances, it is a feature that there are step waveforms as many as the number of substances as shown in Fig.7.14 (c).

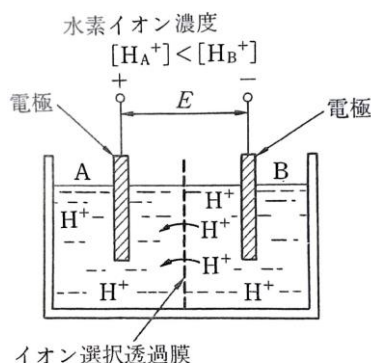


Fig.7.15 Battery sandwiching ion permeable membrane

C. Measure Ion

When a metal electrode is inserted into each interface in contact with the interface of the electrolyte solution, a battery is formed, and its output voltage is determined by the properties of the electrode and the electrolyte solution. Ions can be detected using such a battery. Specifically, as shown in Fig.7.15, a battery with a structure in which the solution tank is in contact with the ion permeable membrane sandwiched is used Here, the membrane selectively permeates only the test ion is selected. When the H^+ concentration of the solution B is larger than A, H^+ diffuses through the membrane and diffuses in the direction of decreasing the concentration difference (B to A), but with that counter ion Since an anion cannot be passed, an anion is left in B, and a potential difference E occurs on both sides of the film, and the A side is higher than the B side This is called the membrane potential. The membrane potential $E(V)$ and hydrogen the ion concentration

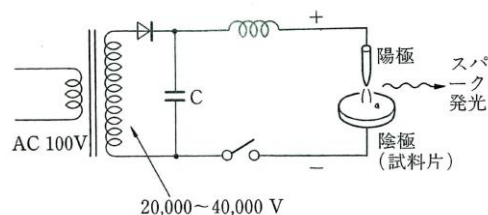


Fig.7.16 Spark Light Source

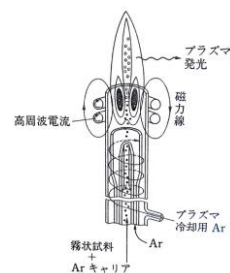


Fig.7.17 Light Emitting Part of ICP

$[H^+](mol/l)$ is expressed by next equation.

$$E = a \log \frac{[H_B^+]}{[H_A^+]}$$

However, a is unique value depending on the type of ion, for hydrogen ion $a = 0.059$ (25°C). Therefore, by setting one solution to a known concentration, the concentration of the other can be known.

7.3 Measure Elements

7.3.1 Analysis Steel Materials

The amounts of carbon, manganese, sulfur, etc. contained in the steel material are specified according to the use of the steel material, and it is extremely important to quickly measure these amounts in order to maintain the quality of the material. Spark emission spectrometry is used for analysis of representative elements other than iron contained in steel materials and their contents.

As shown in Fig.7.16, a flat surface of the specimen shall be used, and a high voltage is applied to the anode made of graphite (or the same material as the specimen) with pointed point. When the applied voltage exceeds the dielectric breakdown voltage of the gas in the electrode gap, the electrode gap becomes conductive and the charge accumulated in the capacitor C is rapidly discharged in the electrode gap, Of the resulting gas plasma, positive of the charged particles violently collide with the cathode and the surface of the cathode emits light, which is spark emission. When eventually releasing the charge and stopping the light emission, the electrode gap returns to the original insulation state. Since the energy of the spark light source is quite large, not only the metal atom but also the emission spectrum of the ion appears, and the observed spectrum becomes complicated, so a spectrometer with high wavelength decomposition is necessary to obtain individual element information.

7.3.2 Analysis of River Water

ICP (inductively coupled plasma) is used for trace element analysis, such as analysis of dissolved metals in river water, where concentration

detection in the order of ppb (10^{-9}) is required. The light emitting part of a representative ICP has a structure as shown in Fig.7.17, and the induction coil is wrapped several times around the outer periphery of the triple quartz tube called a torch. When argon gas is flowed to the torch and a high frequency current of several tens MHz (usually 27.12 MHz or 40.68 MHz, output 1~2.5 kW) is passed through the coil, a high frequency magnetic field is generated around the argon, further induced by this magnetic field A high frequency electric field is generated.

When a high field electric stimulus is given to the torch by a Teller coil or the like, the argon instantaneously discharges and the electrons generated at that time are accelerated by the high frequency induction electric field and repeatedly collide with the argon, resulting in the argon plasma. The excitation temperature Will be 5500~7000 K. When a solution sample is atomized and introduced into such a plasma torch, the sample is dissociated into atoms as ions are further ionized because of high temperature These atoms and ions are excited by the high temperature plasma to emit light, so that the emission line the type of the element in the sample can be known and the element quantity can be known from the intensity of the emitting ray.

7.3.3 Harmful Metal in Crops

In Japan, the content of cadmium per 1g of brown rice is regulated to $1\mu\text{g}$ or less (1 ppm in terms of volume ratio). In this analysis, atomic absorption spectrometry, which is one type of absorption measurement is often used instead of ICP in order to investigate a single element having a relatively high concentration. When the sample liquid is atomized and introduced into the acetylene air flame, the carom in the sample is atomized by the heat of the flame. Since the cadmium atom is in the ground state, it selectively absorbs light (228.9 nm) having a wavelength corresponding to the difference between the excitation energy level and the base energy.

7.4 Measure Gas

7.4.1 Gas Analysis

Since most gases other than one atomic molecules (A , H_e etc.) and homogeneous 2 or 3 atom molecules (H_2 , O_2 , N_2 , O_3) have absorption spectra inherent in the infrared region, absorption spectrum in the infrared region by measuring, the type and concentration of gas can be specified. Fig.7.18 shows absorption spectrum of water vapor.

If the test gas is determined beforehand, use an infrared gas analyzer as shown in Fig.7.19. The infrared ray emitted from the heater

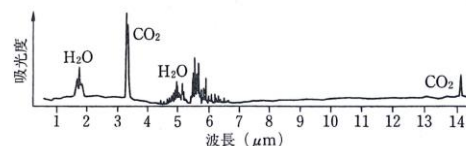


Fig.7.18 Absorption Spectrum of Water Vapor

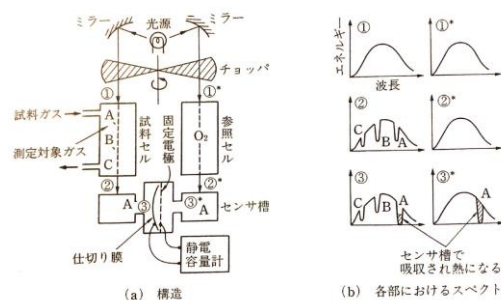


Fig.7.19 Infrared Gas Analyzer

simultaneously passes through the measurement cell and the reference cell, so if the measurement gas is put in the sample cell and O_2 etc. without infrared absorption is put in the reference cell, then in the sample cell Although it receives light absorption according to power component (A) and other components (B, C, ...), it does not absorb anything in the reference cell, so the light energy entering the sensor tank becomes larger on the reference cell side. The sensor part has a structure in which the spaces between the tanks are partitioned by a thin metal film and the gas to be measured (A) is filled in. The light transmitted through the sample cell and the reference cell enters the sensor tank Then, since the light energy is absorbed by the absorption band of the gas A, the temperature of the sensor tank rises and expands. As a result, the volume on the reference cell side of the sensor tank expands further, and the partition membrane displaces.

7.4.2 Gas leak alarm

A combustible gas that combines with oxygen and burns, such as hydrogen, carbon monoxide, or propane, is called a reducing gas. The gas leakage alarm reacts with such a combustible gas and issues an alarm. In its main body,

An n type semiconductor obtained by adding a trace amount of additives to an oxide such as SnO_2 or ZnO and sintering it to be ceramics is used. When oxygen in the air adsorbs to the surface of the semiconductor, it takes the electron from the semiconductor to become O^{2-} , which increases the electric resistance of the semiconductor. When there is a reducing gas here, O^{2-} adsorbed to the semiconductor is reduced and removed. Electrons return to the semiconductor

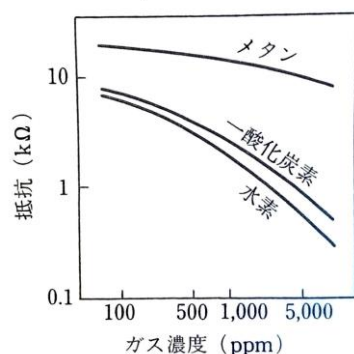


Fig.7.20 Gas Concentration Dependence of Resistance of Semiconductor Gas Sensor

and electric resistance decreases (Fig.7.20). Gas leak alarm is a gas leakage alarm that detects gas by using this change in electric resistance as an index (Fig.7.21).

7.4.3 Oxygen Sensor

By measuring the concentration ratio of fuel and oxygen in the combustion atmosphere of the automobile engine in real time and controlling the air feed amount so as to achieve optimum combustion, it is possible to reduce the harmful exhaust gas component and become a low pollution vehicle. Therefore, ZrO_2 , which is equipped with an oxygen sensor using ZrO_2 (zirconia oxide) in an automobile and is trying to improve combustion efficiency, passes oxygen ions freely and reacts with oxygen ions to generate electromotive force. If a porous electrode is attached to both sides as shown in Fig.7.22, the potential on the high oxygen concentration side is higher than that on the low oxygen side, and the battery is formed by the density of oxygen, which is represented by the logarithm of the oxygen concentration ratio on both sides. Since it is proportional, if the oxygen concentration on one side is kept constant, the concentration of the other can be measured.

7.4.4 Atmospheric Remote Sensing

A device for measuring in real time the amount of

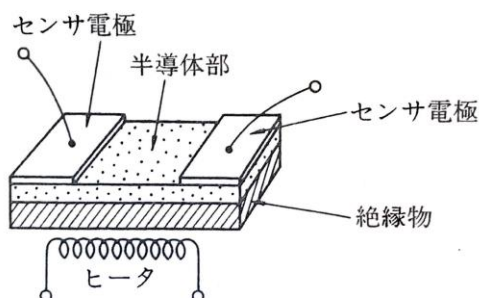


Fig.7.21 Semiconductor Gas Sensor Part of Gas Leak Alarm

dust, steam, carbon dioxide, etc. drifting over the sky and its change in real time is called a laser radar, and it is called a laser radar. It is used for detection of fine particles with a particle size of $0.1 \sim 1 \mu\text{m}$, collectively called aerosols such as dust and fine water droplets in the atmosphere. The basic configuration of the laser radar is shown in Fig.7.23. The pulsed light emitted from the laser toward the atmosphere is scattered by the aerosol and returned to the back side. The light returned in this manner is collected by the telescope and the light Obtain strength. Although this method cannot identify components, the entire aerosol is an important indicator of air pollution, while for CO_2 , NO_x , O_3 etc., the wavelength selectivity of light absorption, fluorescence or Raman scattering molecule Can be identified by a spectroscopic technique

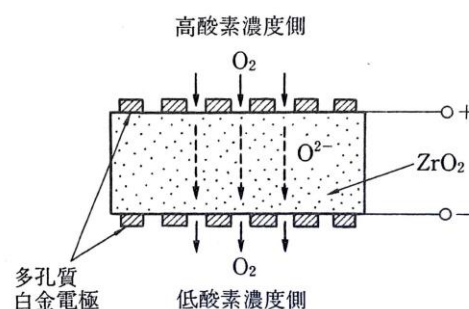


Fig.7.22 Oxidized Zirconia Oxygen Sensor

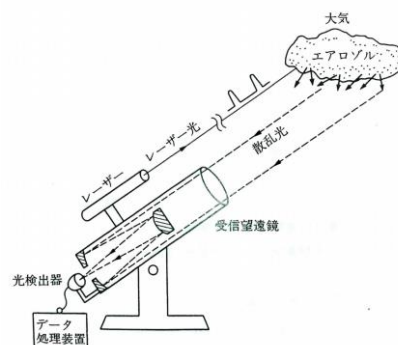


Fig.7.23 Construction of Laser Radar

utilizing. In this way, air pollution information and weather information are remotely sensed.

7.5 Measure Complex Compounds

7.5.1 Medical Biochemical Examination

From body fluids such as urine and blood, various information for obtaining health status can be obtained. In particular, from the blood tests, it is important to use a lot of biochemical components such as electrolytes (Na^+ , K^+ , Cl^- , etc.), blood glucose (glucose), lipids (cholesterol, neutral fat etc.), enzymes (amylase, GOT, GPT etc.). It is an

important criterion for the diagnosis of diseases because it shows the change. It is a device for automatically outputting these information with blood collection of about 5ml with an automatic

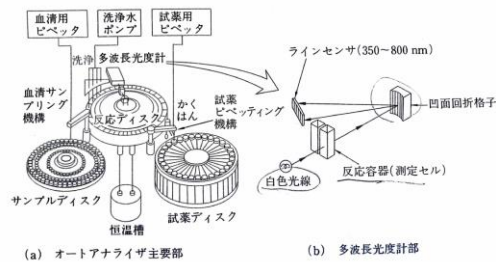


Fig.7.24 clinical autoanalyzer

blood clinical chemistry analyzer (generally called clinical autoanalyzer) is there. Fig.7.24 shows its main part, with a large number of measuring cells arranged on the circumference. After collecting the blood for a while, obtain supernatant (serum) by centrifugation. When this serum is injected into the autoanalyzer, serum is dispensed to the measurement cell in an amount of several tens μl with an automatic sampler. For the other substances, predetermined reagents are sequentially injected into the sample cell and agitated. This reagent reacts specifically with the measurement item components and develops color or opacity. Therefore, by measuring its absorbance, the concentration of the target component can be known. When the light flux passes through the measurement cell, the absorbance is measured. Since the peak wavelength of the light absorption differs depending on the inspection item, the maximum sensitivity can be obtained by setting the optimum measurement wavelength in advance.