Nitrogen Determinations
in sea-water

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The determination of different nitrogen compounds: ammonia, nitrate and nitrite, is of the greatest importance when studying the nutrition factors in sea-water.

It is necessary to find methods to estimate the nitrogen in concentrations from 0 to some hundred milligrams per cu.m. of sea-water.

The analytical procedure must be simple and rapid enough for examination of a great number of samples on board ships within a comparatively short time.

Nitrite: Nitrite-nitrogen is usually determined colorimetrically by means of the Griess-Ilosway reagent. This method was first used on sea-water samples in 1902 (Geelmuyden 1902) and is so sensitive that it is possible to determine nitrite nitrogen in concentrations as low as 0.3 mg/cu.m.

Ammonia: Ammonium nitrogen may be determined directly with Nessler's reagent following a slightly modified Wattenberg method (Føyn 1950).

Nitrate: Some rapid methods of estimating nitrate nitrogen in sea-water have been developed. The method of Harvey (1929) is generally preferred. In strong sulphuric acid strychnidine produces an intensely red colour with nitrate.

The presence of organic matter in solution, or appreciable amounts of plankton organisms, interferes with the colour formation.

These, as well as other nitrate reagents, which all work in strong sulphuric acid, are unsuitable for analyses of polluted water.

For nitrate determinations in sea-water from the inner Oslofjord it was, therefore, necessary to search for a method which would eliminate these difficulties.

It is well known that when nitrate is reduced, a certain amount of nitrite is formed, and may be detected. (Hahn & Jäger 1925), (Lemoigne et al. 1937), (Bray 1945). It occurred to the author that a determination of the nitrite formed might be used as the best a measure of the amount of nitrate originally present, if this procedure were sufficiently sensitive, accurate and rapid.

Various experiments had to be done to find the best procedure. Ten mesh granulated zinc was found to be a suitable reducing reagent. The reduction had to take place in warm solution.

Both the amount of zinc and the length of time for the reduction process will influence the amount of nitrite produced. This is demonstrated in the following experiment.

Five 100 ml samples of water containing about 100 mg N/cu.m. were heated individually in a 300 ml flask for 2 minutes in a boiling water-bath with increasing amounts of zinc, and the nitrite determinated colorimetrically. Table 1 shows the results.o

Table 1.

| Zinc in g.	Nitrite, colour-intensity
2	3.0
5	5.8
10	8.0
20	13.0
50	12.0

The amount of nitrite was found to increase to a limit with increasing amounts of zinc.

The effect of the length of reduction time is demonstrated in Fig. 1. In a 300 ml flask, 100 ml of the water sample while being moderately shaken, was heated in a boiling water-bath with about 40 g zinc for different periods of time. The figure shows how the amount of nitrite formed by the reduction of the nitrate increases rapidly with increasing reduction time, reaches a maximum and then decreases. At optimal conditions the sensitivity of the method seems to be very high. But these experiments show that if the method is going to be used for quantitative determination of nitrate in sea-water, it is necessary to work under absolutely similar conditions both when reducing the samples and when preparing the calibration curve.

It was found most suitable to carry out the reduction in a flask to which 40 g of ten mesh gran. zinc had been added and which was shaken while continuously immersed in a boiling waterbath for 1.5 min.

The amount of formed nitrite which is determined in this way, represents only about 10 % of the original amount of nitrate but as the analytical method for the estimation of nitrite is so extraordinarily sensitive it is still possible to determinate as little as 5 mg/cu.m of nitrate-N in sea-water by this method.

Disturbing agents as phosphates, reducing and oxidising compounds, were never found in seawater in concentrations high enough to influence

Fig. 1.
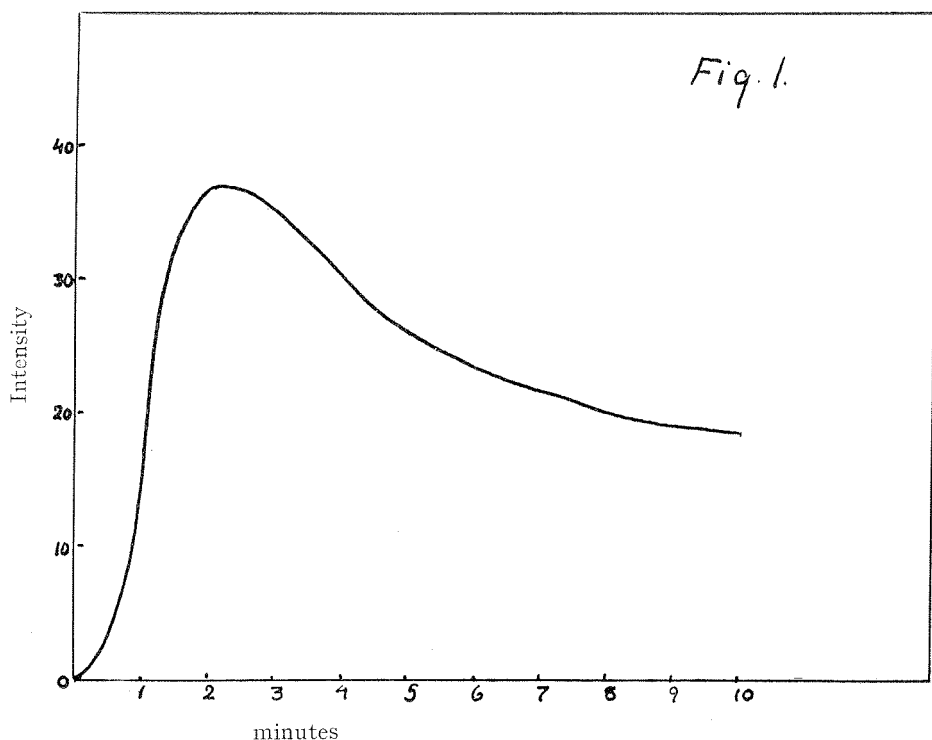
the analysis. Suspended organic matter, like plankton, does not influ-
ence the formation of colour.
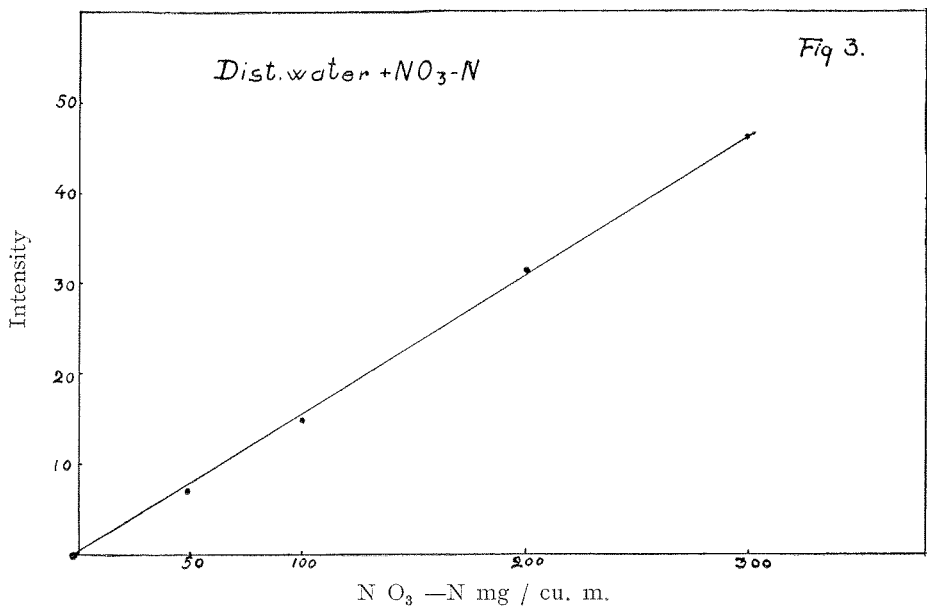
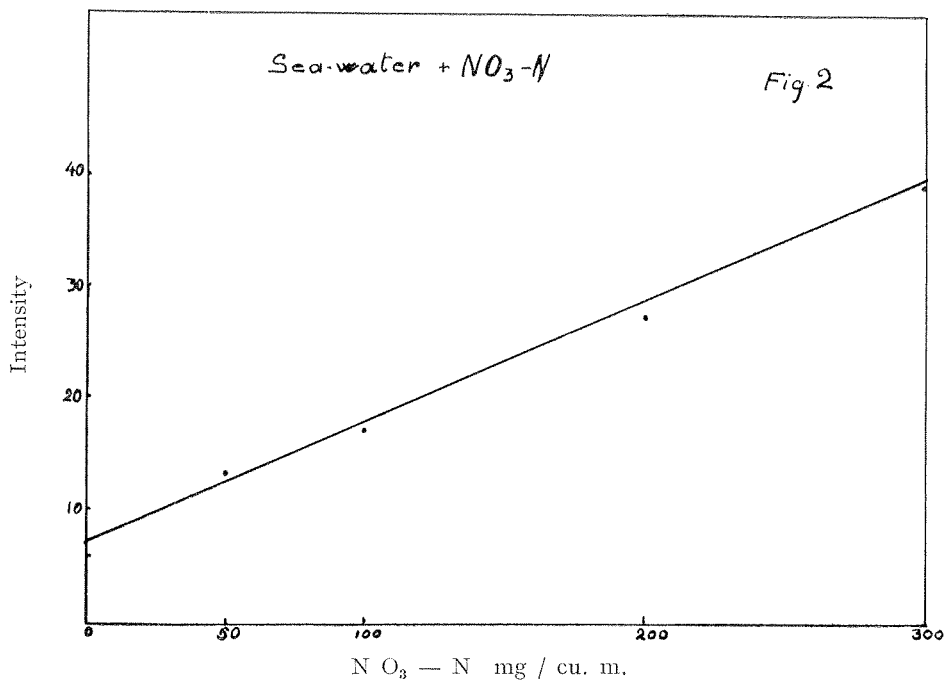When the salinity of the samples varies a great deal, it may be
necessary to make some corrections. Known amounts of KNO_3 are then
added to part of the sample and the nitrite values for this and the un-
treated part of the sample are compared and used for determination
of salt error.

Nitrite which occurs in the seawater in much smaller amounts
than nitrate, does not influence the nitrate analyses, as the nitrite itself
is being reduced under the process.

Table 2 shows the results of six analyses of the same water to
which was added 200 mg/cu.m of nitrate—nitrogen.

Table 2.

Seawater 200 mg NO_3-N/cu.m	Intensity of colour
Sample nr. 1	37.2
2	37.1
3	37.0
4	36.9
5	37.4
6	35.7

Fig. 2

Sea·water + NO_3-N

Intensity

N O_3 — N   mg / cu. m.

Fig 3.

Dist. water + NO_3-N

Intensity

N O_3 —N mg / cu. m.

The correspondance of values for these parallel analyses seems to be sufficient good for the method to be used for determination of nitrate in seawater. The calibration curves (Fig. 2 & 3) prepared by addition of KNO_3 to seawater and distilled water also indicate the reliability of the method.

Procedure:

The following procedure was adopted: 100 ml of the water sample to be examined was placed in a 300 ml Erlenmeyer flask with about 40 gr. of 10 mesh granulated metallic zinc, which had previously been boiled in distilled water and washed with cold distilled water.* The flask was immediately placed in a boiling water-bath and then heated while being moderately shaken for exactly one and a half minute. The sample was decanted into a clean flask and the Griess-Ilosvay reagents were added. When cold, the intensity of the colour was measured with a colorimeter. The flask with the zinc was washed three times in cold distilled water, and was then ready to be used for the next sample.

All samples in a serie were always reduced in the same flask and with the same zinc and the calibration curve was prepared by reducing seawater to which were added known amounts of KNO_3. The amount of nitrate-nitrogen of a sample was then found by means of the calibration curve.

The method has now been used for the nitrate determinations in our laboratory for about two years and on several hundred samples.

Literature.

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* If the zinc previously had been treated with acid, the washing-process must be repeated several times to give constant values.