content of traces of rhenium. In all cases the rhenium effect was lacking, proving that commercial manganous salts contain certainly less than 1 part of rhenium per 1,000,000 of manganese, so that the steps at $-1{\cdot}0$ v. and $-1{\cdot}2$ v. shown on polarograms of manganous solutions (Curve 1), as well as the lines of the X-ray spectrum[1], must have been due to coinciding effects of other elements than 75.

The large current, provoked by the presence of perrhenate in the buffer solution, is probably due to the deposition of hydrogen, catalysed by a sulphide compound of rhenium[6].

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Biological Synthesis of Ascorbic Acid

IN a previous letter[1] we pointed out that the liver tissues of the rat, rabbit and pigeon are able to synthesise ascorbic acid from mannose *in vitro*, while those of the guinea pig and monkey are unable to do so. Further experiments indicate that this power is not common to all animals independent of an external supply of ascorbic acid. The liver tissues of the ox, cat and fowl, for example, cannot convert mannose into ascorbic acid *in vitro*. This might mean either that some other organs in their body are able to effect this transformation or that some entirely different means (for example, bacterial) is employed in these animals for the synthesis of the vitamin.

The *in vitro* experiments with rat tissues have now been confirmed by experiments *in vivo*. It has been found that the intravenous injection of mannose (20 mgm.) into rats is followed by a rise in the ascorbic acid content of the tissues investigated, the animals being killed 5 hours after injection. Similar injections of glucose (20 mgm.) also increase the ascorbic acid content of the adrenal gland, though less strikingly, as shown by the average figures given in the following table. Subcutaneous injection of mannose (20 mgm.) daily for three successive days also leads to a similar rise in the ascorbic acid content of the tissues.

Nature of experiment	Ascorbic acid (mgm.) formed per gm. of tissue			
	Adrenal	Small intestine	Kidney	Liver
Controls	2·53	0·26	0·17	0·18
Mannose (intravenous)	4·83	0·30	0·21	0·22
Glucose (intravenous)	3·75	0·25	0·17	0·18

Another point of interest to which we wish to refer is that in preliminary experiments we have found

Nature of tissue	Ascorbic acid (mgm.) formed per gm. tissue
Guinea pig embryo	0·14
Ovary of the pregnant guinea pig	0·05
Ovary of the adult non-pregnant monkey	0·30

that embryonic guinea pig tissue at an early stage of development, ovarian tissue of the pregnant guinea pig and ovarian tissue of the adult non-pregnant monkey are also capable of converting mannose into ascorbic acid *in vitro* on incubation for 5 hours at *p*H 7·4 in a mixture of phosphate buffer and Ringer-Locke solution at 37°. This is shown in the accompanying table.

The guinea pig embryo has been found to lose this power of converting mannose into ascorbic acid gradually with its development. In these experiments carried out under the stated conditions, the replacement of mannose by glucose does not lead to an appreciable synthesis of ascorbic acid.

The above observations (especially those with the intravenous injection of glucose) indicate that while glucose is the ultimate precursor of ascorbic acid, it has probably to pass through the intermediary stage of mannose or some mannose-like configuration.

The experiments with the ovarian tissue of the monkey appear to have implications concerning the human species, while those with the guinea pig embryo seem to be interesting from the point of view of the theory of recapitulation.

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Estimation of Ascorbic Acid by Titration

IN the course of an investigation of the ascorbic acid content of raw and cooked Ontario foodstuffs, employing a modification of the titration procedure outlined by Birch, Harris and Ray[1], we observed that, in the case of cauliflower, carrots, parsnips, beets and potatoes, the titration value was higher in the cooked than in the raw food. This was reported in October 1934 at a meeting of the Toronto Biochemical Society[2]. Ahmad[3] has recently reported an increase in the case of cabbage, which we found to give only a decrease after heating for short periods.

We have made determinations at regular intervals when two of the above vegetables were heated under constant conditions. The increase in titration value against phenolindophenol is very rapid at first, reaching a maximum within five minutes in most cases if oxidation is retarded by the addition of cyanide, or by heating in an atmosphere of nitrogen or carbon dioxide. If oxidation is not inhibited there is not so great an increase in titration value. Following the increase there is a gradual decline as heating is continued. In the case of Ontario cauliflower, the increase is 60 per cent of the value of the raw vegetable.

It is unlikely that this increase is due to cellular disintegration as a result of heating, and consequently more thorough extraction of ascorbic acid. We were at first impressed with the likelihood of the increase being caused by the liberation of a sulphhydryl compound. However, this explanation was shown to be erroneous since colorimetric tests for cystine and cysteine are almost negative in these cooked vegetables. The character of the curves secured by plotting titration values against time of heating is such that we believe the increase is due to the setting free of bound ascorbic acid, perhaps from an ester.

In the case of certain plant tissues, then, a simple extraction and titration procedure does not give the