# MORPHOMETRY OF THE ADRENALS

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A method of determining gravimetric indices of the zona glomerulosa and the combined zona fasciculata and zona reticularis and the medulla of spherical adrenal glands is described. As a result of mathematical analysis of results obtained with a plasticine model of the adrenal, with close to actual characteristics, it was found that the error of the method when used to determine the weight of the zona fasciculata et reticularis is 0.6-0.9%, of the zona glomerulosa 2.7-3.5%, and of the medulla 5.3-6.4%.

KEY WORDS: adrenal gland; gravimetric indices; zones of the adrenal gland.

Quantitative methods are being used increasingly frequently in morphology by research workers. However, as has rightly been said [1, 2], the investigator must know the limitations of the method and the degree of accuracy with which a given quantitative analysis is made.

At the present time the adrenal glands of rats, mice, and golden hamsters, which are spherical in shape, are frequent objects for morphometric analysis. In particular, a very widely used morphometric index is the width of the adrenal zones. Unfortunately, this index has been absolutized and it is regarded as a valid morphometric feature of the morphofunctional state of the organ without taking the overall size of the adrenal into account. Where such an approach can lead will be clear from the following examples in which, in order to simplify the analysis, the adrenal is regarded as a true sphere and the calculations are made for the cortical and medullary layers only.

Example 1. Let us assume that in case No. 1 the width of the cortical layer ($L_{a.c1}$) is 2 and in case No. 2 ($L_{a.c2}$) it is also 2. It would seem that the degree of development of the cortical layer is the same in both cases. However, let us assume that in case No. 1 the radius of the medulla ($R_{a.m1}$) is 1, whereas in case No. 2 $R_{a.m2} = 2$. In that case, if the volume of the cortex ($V_{a.c}$) is calculated by the equation $V_{a.c} = V_a - V_{a.m}$, the following results are obtained:

$$V_{a.c1} = \frac{4}{3}\pi \cdot (L_{a.c1} + R_{a.m1})^3 - \frac{4}{3}\pi \cdot R^3_{a.m1} = \frac{4}{3}\pi \cdot (21)^3 - \frac{4}{3}\pi \cdot 1^3 = 26 \cdot \frac{4}{3}\pi;$$

$$V_{a.c2} + \frac{4}{3}\pi \cdot (L_{a.c2} + R_{a.m2})^3 - \frac{4}{3}\pi \cdot R^3_{a.m2} = \frac{4}{3}\pi \cdot 4^3 - \frac{4}{3}\pi \cdot 2^3 = 56 \cdot \frac{4}{3}\pi,$$

i.e., $V_{a.c2}$ is more than twice as great as $V_{a.c1}$.

Example 2. In case No. 1 $L_{a.c1} = 2$, in case No. 2 $L_{a.c2} = 3$, i.e., it can be stated that the degree of development of the cortical layer in case No. 2 is 1.5 times greater than in case No. 1. However, let us assume that in case No. 1 $R_{a.m1} = 3$ and in case No. 2 $R_{a.m2} = 1$. Then

$$V_{a.c1} = \frac{4}{3}\pi \cdot 5^3 - \frac{4}{3}\pi \cdot 3^3 = 98 \cdot \frac{4}{3}\pi;$$

$$V_{a.c2} = \frac{4}{3}\pi \cdot 4^3 - \frac{4}{3}\pi \cdot 1^3 = 63 \cdot \frac{4}{3}\pi.$$

i.e., on the contrary, $V_{a.c1}$ is more than 1.5 times greater than $V_{a.c2}$.

These examples show that the index of width of the zones of the adrenal glands, when spherical in shape, has no meaning when used independently. It is essential only on conversion into an objective index of the amount of tissue in a given zone, expressed in volumetric or gravimetric units. In the writer's view, the index

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of the greatest practical importance is gravimetric, for it is technically easier to determine the quantity of tissues in the native adrenal gland of small laboratory animals by the use of gravimetric indices than volumetric. Calculation of the volume of the adrenal zones or of gravimetric indices of the adrenal zones based on the relative weight and data for the width of the zones, as has been done by Kovacs et al. [3], also runs the risk of erroneous conclusions, for during processing of the adrenal tissues they are compressed, and this gives the figures for the width of the adrenal zones purely relative significance. The results are distorted even more by the fact that it is difficult to process the material identically in different cases. However, since when calculating the gravimetric index for the adrenal zones relative values for the width of the adrenal zones are sufficient, the difficulties mentioned above disappear.

The writer has developed a method of successive determination of gravimetric indices of the components of the adrenal glands from the capsule to the medulla. In this case careful determination of the weight of the adrenals in the native state is strictly essential.

The order of the operations of computation is as follows: 1) determination of the gravimetric index of the capsule; 2) determination of the gravimetric index of the zona glomerulosa (z.g.); 3) determination of the gravimetric index of the combined zonae fasciculata et reticularis (z.f.r.); and 4) determination of the gravimetric index of the medulla.

Example. Weight of the adrenal gland P = 30 mg; thickness of the capsule $L_c = 10\ \mu$; width of z.g. $L_{z.g.} = 40\ \mu$; width of z.f.r. $L_{z.f.r.} = 300\ \mu$; radius of the medulla $R_{a.m} = 300\ \mu$; total radius $R = 650\ \mu$.

1) Weight of capsule $= P.\dfrac{\frac{4}{3}\pi R^3 - \frac{4}{3}\pi (R-L_c)^3}{\frac{4}{3}\pi\cdot R^3} = P.\dfrac{R^3-(R-L_c)^3}{R^3} = 30\cdot\dfrac{650^3-640^3}{650^3} = 1.3$ mg;

2) Weight of z.g. $= (30-1,3)\times\dfrac{(R-L_c)^3-(R-L_c-Lz.g.)^3}{(R-L_c)^3} = 28,7\cdot\dfrac{640^3-600^3}{640^3} = 5.04$ mg;

3) Weight of z.f.r. $= (28,7-5,04)\ \dfrac{(R-L_c-Lz.g.)^3 - R^3_{a.m}}{(R-L_c)^3} = 23,66\cdot\dfrac{600^3-300^3}{600^3} = 20,71$ mg;

4) Weight of medulla = 23.66 − 20.71 = 295 mg.

To establish the basis of the method, to determine its accuracy, and to elucidate some of the technical recommendations when determining the gravimetric indices for the adrenal zones a series of calculations was made on a plasticine model of the adrenal simulating the three chief components of the adrenal: z.g., z.f.r., and the medulla. As a first step the materials composing the model were weighed separately, after which a cast was made of the adrenal. The gravimetric indices were found by the formula $P(R^3 - R_1^3)/R^3$. The initial calculations of the gravimetric indices on the plasticine model showed that the truest results were obtained when maximal total sections of the plasticine model and also of the medulla were analyzed. However, the plane of maximal total section when the medulla is disposed eccentrically does not coincide with the plane of maximal section of the medulla, and this must be taken into account during mathematical analysis of the results.

What is the accuracy of determination of the gravimetric indices of the components of the adrenals? To answer this question data obtained on 20 plasticine models of spherical adrenal glands, the maximal plane of section of which was chosen with the models in different random positions, were analyzed mathematically. The true weight of z.g. was 5.0 mg, of z.f.r. 21 mg, and of the medulla 3 mg. The ratio between the gravimetric indices was close to the actual ratio characteristic of female sexually mature rats. The results of the analysis are given in Table 1.

As Table 1 shows, the mean statistical error of the method when used to determine the gravimetric index for z.g. was 2.7%, for z.f.r. 0.6%, and for the medulla 5.3%.

On mathematical analysis of six randomly chosen models the maximal areas of cross section for which passed through the greatest, smallest, and middle axes, it was found that the mean probable error of determination of the gravimetric index for z.g. was 3.5%, for z.f.r. 0.9%, and for the medulla 6.4%, i.e., the results obtained by two methods of mathematical analysis were very close. It is another interesting fact that even when the ratio between the areas of maximal cross section coinciding with the largest and smallest axes of the spherical models was 1:2, the error did not exceed 5-5.5% for determination of the gravimetric index of z.g. and 0.95% for determination of the gravimetric index for z.f.r. In sexually mature rats this ratio is 1.17 ± 0.3,

TABLE 1. Results of Mathematical Analysis of Data Obtained on Plasticine Models of the Adrenal

| Index | Model No. | | | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | $M \pm m$ |
| Total area | 826 | 820 | 756 | 826 | 936 | 877 | 924 | 915 | 960 | 888 | 825 | 955 | 885 | 915 | 903 | 864 | 886 | 890 | 921 | 925 | — |
| Area of z.f.r. and medulla | 720 | 716 | 660 | 720 | 830 | 774 | 816 | 806 | 850 | 776 | 723 | 840 | 780 | 807 | 795 | 760 | 783 | 787 | 814 | 818 | — |
| Area of medulla | 170 | 169 | 164 | 170 | 210 | 202 | 209 | 210 | 230 | 194 | 185 | 201 | 188 | 204 | 190 | 180 | 203 | 208 | 213 | 197 | — |
| Weight of z.g | 5,32 | 5,30 | 5,28 | 5,32 | 4,76 | 4,93 | 4,88 | 5,0 | 4,85 | 5,0 | 5,12 | 5,05 | 5,0 | 4,93 | 5,02 | 5,05 | 4,87 | 5,16 | 4,90 | 4,78 | 5,03±0,04 |
| Weight of z.f.r. | 20,95 | 20,98 | 20,77 | 20,95 | 21,16 | 20,87 | 20,9 | 20,82 | 20,75 | 21,0 | 20,8 | 21,15 | 21,15 | 21,0 | 21,17 | 21,07 | 20,95 | 20,6 | 20,89 | 21,34 | 21,03±0,04 |
| Weight of medulla | 3,73 | 2,72 | 2,95 | 2,73 | 3,08 | 3,2 | 3,10 | 3,18 | 3,40 | 3,0 | 3,08 | 2,80 | 2,85 | 3,07 | 2,81 | 2,88 | 3,18 | 3,24 | 3,21 | 2,88 | 2,94±0,05 |
| Error for z.g., % | 6,4 | 6,0 | 5,6 | 6,4 | 4,8 | 1,2 | 2,4 | 0 | 3,0 | 0 | 2,4 | 1,0 | 0 | 1,2 | 0,4 | 1,0 | 2,6 | 3,2 | 2,0 | 4,4 | 2,7±0,45 |
| Error for z.f.r., % | 0,1 | 0,1 | 1,1 | 0,1 | 0,7 | 0,6 | 0,5 | 0,8 | 1,2 | 0 | 0,9 | 0,7 | 0,7 | 0 | 0,8 | 0,3 | 0,2 | 0,3 | 0,5 | 1,6 | 0,6±0,10 |
| Error for medulla, % | 9 | 9,3 | 1,7 | 9 | 2,7 | 6,6 | 3,3 | 6 | 13 | 0 | 2,7 | 6,7 | 5,0 | 2,3 | 6,3 | 4,0 | 6,0 | 8,0 | 7,0 | 4,0 | 5,3±0,67 |

i.e., significantly less. An increase in the accuracy of determination of the gravimetric index for z.g. can be attained by allowing for the theoretically expected error which, according to our calculations, when the ratio between the areas of maximal cross section coinciding with the largest and smallest axes was 1.1-1.25, is 3%; if the ratio is 1.26-1.49 the error is 4%; and if the ratio is 1.5-2, it is 5%. Under these circumstances the adrenals must lie in a strictly assigned position when cut into sections. If the maximal plane of section coincides with the longest axis the sign of the error will be plus; if it coincides with the shortest axis, it will be minus.

Mathematical analysis of the plasticine model of a spherical adrenal gland and the ratio between the gravimetric indices of the component elements similar in actual relative proportions to the adrenals of sexually mature female rats thus revealed that the method of quantitative determination of gravimetric indices of the adrenal cortical zones and medulla is particularly accurate for measuring the weight of z.f.r. and z.g., and less accurate for determining the weight of the medulla. This method has some disadvantages. However, in the writer's view its use will give results closer to the actual values than those obtained on the basis of any other morphological feature, such as the width or area of the zones of the adrenals.

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# THE TECHNIQUE OF RESISTOGRAPHY


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To abolish the error in resistography connected with fluctuations in the animal's arterial pressure an automatic stabilizer of the input pressure (an electromagnetic valve) is suggested. To switch off the resistograph pump automatically when the blood flow into the apparatus is obstructed or stops, a monitoring device is proposed. A pressure stabilizer and monitoring device are controlled by electrical contact mercury manometers connected to the input channel of the resistograph.



KEY WORDS: resistography; design of the resistograph.


Even if all the requirements of a resistograph with external electromagnetic valves are strictly met [3], the possibility that its flow rate depends on the input pressure of the apparatus cannot be ruled out. This leads to a definite error which, according to data given by different workers [1, 3], may be 5-8%.

To reduce the error it is suggested that the external electromagnetic valves be replaced by internal valves [1], which can reduce the error in a single-channel resistograph to 1-1.5% and in a two-channel instrument to 2-3%. From the design point of view, internal valves, working compulsorily [1], are much more complicated than external electromagnetic valves, and it is particularly difficult to ensure that they operate in phase with the pump of the instrument in the extension working head. Disadvantages of internal "automatic" valves have been examined in detail by Khayutin [2, 3].


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