

# The Aschheim-Zondek Test for Pregnancy <sup>[1]</sup>

By: Kelley, Kristin Keywords: [Reproduction](#) <sup>[2]</sup> [Fertility](#) <sup>[3]</sup>

Many different methods have been devised for the early detection of [pregnancy](#) <sup>[5]</sup>. From the time of the Ancient Egyptians, inspection of the urine has been a popular place to start. However, it was not until the discovery of [hormones](#) <sup>[6]</sup> in the early twentieth century that the development of truly reliable [pregnancy](#) <sup>[5]</sup> tests occurred. Prior to 1978, when the first home [pregnancy](#) <sup>[5]</sup> tests became available in the United States, [pregnancy](#) <sup>[5]</sup> testing was done in hospital laboratories using various methods, one of them being the Aschheim-Zondek, or [A-Z test](#) <sup>[7]</sup>.

The [A-Z test](#) <sup>[7]</sup> is a product of research into human reproduction carried out in the early twentieth century. Developed by German gynecologists [Selmar Aschheim](#) <sup>[8]</sup> and [Bernhard Zondek](#) <sup>[9]</sup> in 1927, the [A-Z test](#) <sup>[7]</sup> was one of the first [bioassays](#) <sup>[10]</sup> developed to detect early [pregnancy](#) <sup>[5]</sup>. Armed with new information about chemical messengers called [hormones](#) <sup>[6]</sup>, Aschheim and Zondek identified the anterior [pituitary gland](#) <sup>[11]</sup> as an endocrine gland that performs important roles in ovarian function. It is now known that a family of [hormones](#) <sup>[6]</sup> called gonadotropins is essential to control of the ovarian and uterine cycles and to sexual maturation. During the menstrual cycle, an increase in [gonadotropin](#) <sup>[12]</sup> levels causes a mature ovarian follicle to release its [egg](#) <sup>[13]</sup> and develop into a [corpus luteum](#) <sup>[14]</sup>.

In 1903 [Ludwig Fraenkel](#) <sup>[15]</sup> became the first to describe the human [corpus luteum](#) <sup>[14]</sup>, a glandular mass found in the ovaries of a female during [menstruation](#) <sup>[16]</sup> that has important endocrine functions. Fraenkel also named the [hormone](#) <sup>[17]</sup> [progesterone](#) <sup>[18]</sup>, which the [corpus luteum](#) <sup>[14]</sup> secretes in addition to [estrogen](#) <sup>[19]</sup>. These [hormones](#) <sup>[6]</sup> support the [endometrium](#) <sup>[20]</sup> and suppress release of [gonadotropin](#) <sup>[12]</sup> in order to prevent the maturation of other ovarian follicles. If the [egg](#) <sup>[13]</sup> is not fertilized, the [corpus luteum](#) <sup>[14]</sup> dissociates and the [endometrium](#) <sup>[20]</sup> sloughs off. Conversely, if the [egg](#) <sup>[13]</sup> is fertilized, it implants itself and secretes the [hormone](#) <sup>[17]</sup> associated with modern early [pregnancy](#) <sup>[5]</sup> detection—[human chorionic gonadotropin](#) <sup>[21]</sup> (hCG)—which signals the [corpus luteum](#) <sup>[14]</sup> to continue secretion of [progesterone](#) <sup>[18]</sup> and [estrogen](#) <sup>[19]</sup> in order to prevent the [endometrium](#) <sup>[20]</sup> from sloughing off.

Remarkably, although Aschheim and Zondek developed the [A-Z test](#) <sup>[7]</sup> in 1927, hCG itself was not discovered until the 1950s. This landmark in the history of [pregnancy](#) <sup>[5]</sup> tests simply operated under the assumption that a substance present only in the urine of pregnant women could be used to elicit some sort of measurable response in other, nonhuman, organisms. Specifically, the Aschheim-Zondek test calls for the injection of a woman's urine into an immature female [mouse](#) <sup>[22]</sup>. It was correctly hypothesized that if a woman is pregnant, the young [mouse](#) <sup>[22]</sup> will go into heat despite its young age. Ultimately, the [Friedman test](#) <sup>[23]</sup> would use rabbits rather than mice, eliciting the popular symbol of [rabbit](#) <sup>[24]</sup>-killing to describe [pregnancy](#) <sup>[5]</sup> testing as the [rabbit](#) <sup>[24]</sup> would have to be sacrificed in order to identify the presence or absence of a [corpus luteum](#) <sup>[14]</sup>. The test further evolved in efficiency in the 1950s with the use of toads rather than mice or rabbits, as these [egg](#) <sup>[13]</sup>-laying organisms do not have to be sacrificed in order to confirm a positive or negative test result.

The technical process of the [A-Z test](#) <sup>[7]</sup> is more complex than it first appears. A suitable [mouse](#) <sup>[22]</sup> for use in an [A-Z test](#) <sup>[7]</sup> needs to be three to five weeks in age and weigh between six and ten grams. For each [pregnancy](#) <sup>[5]</sup> test, three to five of these infant mice are necessary, as some of them will die before the end of the test. After a urine specimen is collected from the female, it is tested for acidity and made basic. Afterward, one or two drops of tricresol are often added in order to preserve the sample, and the urine is filtered if cloudy in appearance. Subsequently, 3 mL of the urine sample is injected subcutaneously into each [mouse](#) <sup>[22]</sup> three times per day for three days. Two days after the last injection, all of the mice are sacrificed and the ovaries are examined macroscopically. The presence of the human [gonadotropin](#) <sup>[12]</sup> [hormone](#) <sup>[17]</sup> in the urine sample is indicated by several characteristic changes in the mice.

The [A-Z test](#) <sup>[7]</sup> is said to be positive if the ovaries are enlarged (two to three times normal size) with red dots visible (due to hemorrhage into the follicles) or if luteinization occurs and several corpora lutea are visible. A [corpus luteum](#) <sup>[14]</sup> can be identified macroscopically as a small yellow dot on the ovaries. If the [uterus](#) <sup>[25]</sup> appears enlarged with no changes in the ovaries, the test is negative. The [uterus](#) <sup>[25]</sup> becomes enlarged because of other [hormones](#) <sup>[6]</sup> present in the urine, not because of hCG. However, if the first reaction is observed but the organism displays other features of heat, such as cornification of the [vagina](#) <sup>[26]</sup>, the test is repeated with a second urine sample.

The [A-Z test](#) <sup>[7]</sup> was impressively reliable. After the first 2,000 A-Z tests were performed, the test was estimated to have a 98.9% success rate (with seventeen errors being false negatives and five being false positives), according to "Aschheim-Zondek Test

for Pregnancy—Its Present Status.” Nevertheless, the [A-Z test](#)<sup>[7]</sup> was destined to become obsolete, with the introduction in 1960 of an immunoassay for [pregnancy](#)<sup>[5]</sup> testing. This was a more convenient test that did not require animal sacrifice. The [A-Z test](#)<sup>[7]</sup> was also used to test for other conditions such as [ectopic pregnancy](#)<sup>[27]</sup>, [hydatidiform mole](#)<sup>[28]</sup>, [chorion-epithelioma](#)<sup>[29]</sup>, [incomplete abortion](#)<sup>[30]</sup> and testicular tumors, which also produce [human chorionic gonadotropin](#)<sup>[21]</sup>. Although the [A-Z test](#)<sup>[7]</sup> is no longer used, it was an important step in the development of modern [pregnancy](#)<sup>[5]</sup> test kits.

## Sources

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Throughout history many different methods have been devised for the early detection of pregnancy. From the time of the Ancient Egyptians, inspection of the urine has been a popular place to start. However, it was not until the discovery of hormones in the early twentieth century that the development of truly reliable pregnancy tests occurred. Prior to 1978, when the first home pregnancy tests became available in the United States, pregnancy testing was done in hospital laboratories using various methods, one of them being the Aschheim-Zondek, or A-Z test.

### Subject

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