In 1934 Erwin Panofsky published an article entitled Jan van Eyck's 'Arnolfini' Portrait in the Burlington Magazine, arguing that the elaborate signature on the back wall, and other factors, showed that it was painted as a legal record of the occasion of the marriage of the couple, complete with witnesses and a witness signature. Panofsky also argues that the many details of domestic items in the painting each have a disguised symbolism attached to their appearance. While Panofsky's claim that the painting formed a kind of certificate of marriage is not accepted by all art historians, his analysis of the symbolic function of the details is broadly agreed, and has been applied to many other Early Netherlandish paintings, especially a number of depictions of the Annunciation set in richly detailed interiors, a tradition for which the Arnolfini Portrait and the Mérode Altarpiece by Robert Campin represent the start (in terms of surviving works at least). Since then, there has been considerable scholarly argument among art historians on the occasion represented.

Edwin Hall considers that the painting depicts a betrothal, not a marriage. Margaret D. Carroll argues that the painting is a portrait of a married couple that alludes also to the husband's grant of legal authority to his wife. Carroll also proposes that the portrait was meant to affirm Giovanni Arnolfini's good character as a merchant and aspiring member of the Burgundian court. She argues that the painting depicts a couple, already married, now formalizing a subsequent legal arrangement, a mandate, by which the husband "hands over" to his wife the legal authority to conduct business on her own or his behalf (similar to a power of attorney). The claim is not that the painting had any legal force, but that van Eyck played upon the imagery of legal contract as a pictorial conceit. While the two figures in the mirror could be thought of as witnesses to the oath-taking, the artist himself provides (witty) authentication with his notarial signature on the wall. Johannes de eyck fuit hic 1434 (Jan van Eyck was here. 1434).

Jan Baptist Bedaux agrees somewhat with Panofsky that this is a marriage contract portrait in his 1986 article "The reality of symbols: the question of disguised symbolism in Jan van Eyck's Arnolfini Portrait." However, he disagrees with Panofsky's idea of items in the portrait having hidden meanings. Bedaux argues, "if the symbols are disguised to such an extent that they do not clash with reality as conceived at the time ... there will be no means of proving that the painter actually intended such symbolism." He also conjectures that if these disguised symbols were normal parts of the marriage ritual, then one could not say for sure whether the items were part of a "disguised symbolism" or just social reality.

Craig Harbison takes the middle ground between Panofsky and Bedaux in their debate about "disguised symbolism" and realism. Harbison argues that "Jan van Eyck is there as storyteller ... [who] must have been able to understand that, within the context of people's lives, objects could have multiple associations", and that there are many possible purposes for the portrait and ways it can be interpreted. He maintains that this portrait cannot be fully interpreted until scholars accept the notion that objects can have multiple associations. Harbison urges the notion that one needs to conduct a multivalent reading of the painting that includes references to the secular and sexual context of the Burgundian court, as well as religious and sacramental references to marriage.

Lorne Campbell in the National Gallery Catalogue sees no need to find a special meaning in the painting: "... there seems little reason to believe that the portrait has any significant narrative content. Only the unnecessary lighted candle and the strange signature provoke speculation."
He suggests that the double portrait was very possibly made to commemorate a marriage, but not a legal record and cites examples of miniatures from manuscripts showing similarly elaborate inscriptions on walls as a normal form of decoration at the time. Another portrait in the National Gallery by van Eyck, Portrait of a Man (Leal Souvenir), has a legalistic form of signature.

Margaret Koster's new suggestion, discussed above and below, that the portrait is a memorial one, of a wife already dead for a year or so, would displace these theories. Art historian Maximiliaan Martens has suggested that the painting was meant as a gift for the Arnolfini family in Italy. It had the purpose of showing the prosperity and wealth of the couple depicted. He feels this might explain oddities in the painting, for example why the couple are standing in typical winter clothing while a cherry tree is in fruit outside, and why the phrase "Johannes de eyck fuit hic 1434" is featured so large in the centre of the painting. Herman Colenbrander has proposed that the painting may depict an old German custom of a husband promising a gift to his bride on the morning after their wedding night. He has also suggested that the painting may have been a present from the artist to his friend. In 2016, French physician Jean-Philippe Postel, in his book L'Affaire Arnolfini, agreed with Koster that the woman is dead, but he suggested that she is appearing to the man as a spectre, asking him to pray for her soul.